FINAL REPORT

Control and Mitigation of Aquatic Invasive Species in Pacific Island Streams

SERDP Project RC-2447

MAY 2020

Michael Blum
Tulane University

J. Derek Hogan
Texas A&M University – Corpus Christi

Peter B. McIntyre
Cornell University

SERDP
DOD ▪ EPA ▪ DOE

Page Intentionally Left Blank

Page Intentionally Left Blank

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE *(DD-MM-YYYY)*
25-05-2020

2. REPORT TYPE
SERDP Final Report

3. DATES COVERED *(From - To)*
June 2015 – May 2020

4. TITLE AND SUBTITLE
Control and Mitigation of Aquatic Invasive Species in Pacific Island Streams

5a. CONTRACT NUMBER
W912HQ-15-C-0006

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
Michael J. Blum, J. Derek Hogan, Peter B. McIntyre

5d. PROJECT NUMBER
RC-2447

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
Tulane University
400 Lindy Boggs Building
New Orleans, LA 70118

8. PERFORMING ORGANIZATION REPORT NUMBER
RC-2447

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES) Strategic Environmental Research and Development Program
4800 Mark Center Drive, Suite 16F16
Alexandria, VA 22350

10. SPONSOR/MONITOR'S ACRONYM(S)
SERDP

11. SPONSOR/MONITOR'S REPORT NUMBER(S)
RC-2447

12. DISTRIBUTION / AVAILABILITY STATEMENT
DISTRIBUTION STATEMENT A. Approved for public release: distribution unlimited.

13. SUPPLEMENTARY NOTES

14. ABSTRACT
Experimental AIS removals showed that (1) AIS can be controlled with nominal collateral injury and mortality of native species. Removals also (2) resulted in significant and sustained reduction of AIS densities, with the magnitude of reductions varying by target species and stream discharge. AIS removals also (3) triggered a pulse in recruitment and sustained increases in growth and body condition in *A. stamineus*. Though island-wide trends in *A. stamineus* life history (4) tracked discharge and densities of invasive Poeciliid live-bearers, AIS removals only led to a modest shift in the balance of local variation. Nonetheless, (5) genomic diversity increased following AIS removals, likely reflecting shifts in recruitment and life history. While AIS removals (6) did not elicit sweeping changes in ecosystem processes, some conditions did become more favorable for native species, like reductions in total suspended sediment (i.e., clearer water). Finally, model simulations illustrated functional trait differences can be strategically utilized to not only restore local populations but also to achieve metapopulation-wide benefits through demographic spillover.

15. SUBJECT TERMS
amphidromy; armored catfish; *Awaous stamineus*; ecosystem restoration; goby; guppies; Hawaii; invasive species removal; oceanic islands; stream monitoring; water chemistry; watershed management

16. SECURITY CLASSIFICATION OF:

a. REPORT	**b. ABSTRACT**	**c. THIS PAGE**	**17. LIMITATION OF ABSTRACT**	**18. NUMBER OF PAGES**
UCLASS	UCLASS	UCLASS	SAR	178

19a. NAME OF RESPONSIBLE PERSON
Michael J. Blum

19b. TELEPHONE NUMBER *(include area code)*
504-214-9964

Page Intentionally Left Blank

Table of Contents

LIST OF TABLES

LIST OF FIGURES

List of Acronyms

AC	– Alternating current
AIC	– Akaike Information Criteria
AIS	– Aquatic invasive species
AMOVA	– Analysis of molecular variance
ANOVA	– Analysis of variance
DAPC	– Discriminant analysis of principal components
DAR	– State of Hawaii Division of Aquatic Resources
DC	– Direct current
ddRAD	– Double digest restriction-site associated DNA
DoD	– Department of Defense
ER	– Ecosystem respiration
GPP	– Gross primary production
HYCOM	– 1/25th degree Hawaii HYbrid Coordinate Ocean Models
IACUC	– Institutional Animal Care and Use Committee
ICP-MS	– Inductively coupled plasma – mass spectrometer
LA	– Laser ablation
LA-ICP-MS	– Laser ablation inductively coupled plasma mass spectrometry
LLC	– Limited Liability Company
LOESS	– Locally estimated scatterplot smoothing
MANOVA	– Multivariate analysis of variance
NDS	– Nutrient diffusing substrates
perMANOVA	– Permutational multivariate analysis of variance
SERDP	– Strategic Environmental Research and Development Program
SAP	– Special activity permit
SCP	– Systematic conservation planning
SNP	– Single nucleotide polymorphism
SON	– Statement of Need
TER-S	– Threatened, endangered, and at-risk species
USGS	– United States Geological Survey
VIE	– Visual implant elastomer tag

List of Abbreviations

Ar	– Argon
Ba	– Barium
bp	– Base pairs
C	– Carbon
°C	– Centigrade degree
Ca	– Calcium
Cr	– Chromium
Cs	– Cesium
cm	– Centimeter
CV	– Coefficient of variance

d – Day
df – Degrees of freedom
DNA – Deoxyribonucleic acid
FW – Freshwater
g – Gram
GLS – Generalized least squares
H – Hydrogen
He – Helium
H_e – Expected heterozygosity
Hz – Hertz
K – Gas exchange with the atmosphere
L – Liter
Li – Lithium
µg – Microgram
µl – Microliter
µm – Micrometer
µS – Microsecond
m – Meter
mg – Milligram
Mg – Magnesium
min – Minutes
ml – Milliliter
mm – Millimeter
mM – Millimolar
Mn – Manganese
π – Nucleotide diversity
n – Number
N – Nitrogen
NH_4 – Ammonium
nm – Nanometer
NO_3 – Nitrate
NP – Nitrogen and Phosphorus
O_2 – Oxygen
Q – Discharge
P – Phosphorus
PAR – Light
Pb – Lead
ppm – Parts per million
PO_4 – Phosphate
s – Seconds
SD – Standard deviation
SE – Standard error
Sr – Strontium
SRP – Soluble reactive phosphorus
SW – Marine
TL – Total length
TSS – Total suspended sediment
U – Unit

USA – United States of America
V – Volt
W – Watt
Zn – Zinc

Keywords

amphidromy; armored catfish; *Awaous stamineus*; coupled biophysical modeling; ecosystem restoration; electrofishing; genetic differentiation; genetic diversity; goby; guppies; Hawaii; hydrology; immigration; impairment; invasive species; invasive species removal; life history; Loricariids; mark-recapture; natural flow restoration; nutrient loading; Oahu; oceanic islands; ocean-stream connectivity; otolith microchemistry; Pacific islands; Poeciliids; population connectivity; population density; Single Nucleotide Polymorphisms; snorkel survey; species diversity; stream monitoring; water chemistry; water diversion; watershed land use; watershed management; water quality

Acknowledgements

We would like to acknowledge the funding support made available for this project from the Department of Defense Strategic Environmental Research and Development Program (SERDP). Additional acknowledgement of support is made to the many partners who provided permission to conduct this research in the State of Hawaii and who provided access or assistance to gain entry to restricted areas including: Alton Miyasaka, Glenn Higashi, and Catherine Gewecke of the Hawaii Division of Aquatic Resources; Kaeo Duarte, Imiola Lindsey and Joey Char of Kamehameha Schools; Laurent Poole of Waimea Valley, Hi'ipaka Limited Liability Company (LLC); Lance Bookless of the US Marine Corps Base Hawaii; Gordon Smith of the US Fish and Wildlife Service; representatives of Papahana Kuaola, Bayview Golf Course, Moore & Moore Properties, as well as Sharon Peterson Cheape, Bart Potter, Anne and Richard Stack, and the Fraiola family.

We thank Shelley Meaux for assistance with project administration, and we would like to thank Paul and Susan King for providing logistical support for this project. Additionally, we would like to acknowledge the support made available to this project from the senior researchers, postdoctoral associates, graduate students, staff, and undergraduate students who contributed to the research that was conducted, including: Nicolette Beeken, Nathan Bickford, Christopher Bird, Alexandra Del Favero Campbell, Kauaoa Fraiola, Kelley Fritz, Roderick Gagne, Jayson Gallatin, Alissa Ganley, James Gilliam, Susannah Halbrook, Ellen Hamann, Christine Hayes, Rebecca Hazen, Aaron Friedman-Heiman, Heidi Heim-Ballew, Virgilio Hermoso, Galen Holt, Aiyzah Javaid, Timothy Lambert, Simon Linke, Peter Lisi, John Morgan, Kristine Moody, Annessa Musgrove, David O'Connor, Christopher Patrick, Kyle Piller, David Reznick, Katherine Roach, William Rosenthal, Elizabeth Runde, Avery Scherer, Sean Schoville, Jason Selwyn, Kim Sparks, Emily Stanley, Jennifer Summers, Jennifer Whitt, and Hannah Wilson for field assistance. We also would like to thank Heidi Heim-Ballew, Galen Holt, Peter Lisi, and Kristine Moody for assistance with preparing the final project report.

1 Abstract

INTRODUCTION AND OBJECTIVES: Advancing stewardship of stream ecosystems altered by aquatic invasive species (AIS) can better ensure the continuity and long-term sustainability of Department of Defense (DoD) activities on Pacific islands. Because eradication of AIS is not always feasible, consideration must be given to alternative management approaches, including control efforts that can promote conditions favorable to at-risk native species. It is not clear how this can be achieved, however, because little work has been done on AIS control in oceanic island stream ecosystems. We addressed this deficit by evaluating genes-to-ecosystem outcomes of experimental AIS removals across a stream flow gradient in concordance with the development of a model-based decision tool to better evaluate AIS management under different hydrological and climate regimes.

TECHNICAL APPROACH: Experimental AIS removals were conducted in 13 watersheds spanning a hydrological gradient across the Koʻolau Range on the Hawaiian island of Oʻahu, which is home to a dense array of DoD installations and at-risk native species. Following a before-after, control-impact study design, AIS removals were executed following a protocol tailored for Hawaiian streams that included two tiers of precautionary steps to prevent collateral injury to electro-sensitive native species. Responses to removals were tracked for a period of up to two years, with adjacent upstream and downstream reaches serving as references. We examined whether removals yielded conditions favorable to native species by conducting snorkel surveys of population densities and community composition, as well as mark-recapture demographic assessments of *Awaous stamineus*, a native species of conservation concern. Additionally, we examined *A. stamineus* otolith microchemistry to determine how life-history varies according to AIS densities and hydrology, and whether removals influenced the balance of variation in local populations. We also assessed whether removals elicited an increase in genomic diversity by SNP genotyping *Awaous stamineus* in each study reach. We also conducted in-stream and stable isotope assays to determine whether removals influenced nutrient availability and trophic structure. Data from this work was used to parameterize a coupled ocean-watershed biophysical model to evaluate local-to-archipelago scale outcomes of AIS removal for prioritizing interventions that would maximize return on investment.

RESULTS: Experimental removals showed that (1) AIS can be controlled with nominal collateral injury and mortality of native species. Removals also (2) resulted in significant and sustained reduction of AIS densities, with the magnitude of reductions varying by target species and stream discharge. AIS removals also (3) triggered a pulse in recruitment and sustained increases in growth and body condition in *A. stamineus*. Though island-wide trends in *A. stamineus* life history (4) tracked discharge and densities of invasive Poeciliid live-bearers, AIS removals only led to a modest shift in the balance of local variation. Nonetheless, (5) genomic diversity increased following AIS removals, likely reflecting shifts in recruitment and life history. While AIS removals (6) did not elicit sweeping changes in ecosystem processes, some conditions did become more favorable for native species, like reductions in total suspended sediment (i.e., clearer water). Finally, model simulations illustrated functional trait differences can be strategically utilized to not only restore local populations but also to achieve metapopulation-wide benefits through demographic spillover.

BENEFITS: Development of innovative approaches and actionable information for managing AIS can substantively improve DoD stewardship of stream ecosystems that cross DoD lands, especially on islands like Oʻahu where installations harbor native species under federal or state protection. This project not only empirically demonstrated that modified electrofishing can be a valuable tool for AIS control in Hawaiian streams, it also illustrated how managers can maximize benefits of AIS control through careful consideration of target species and climate-driven hydrological conditions. Likewise, the project delivered a model-based decision support tool that can serve as a defensible and transparent analytical framework for prioritizing interventions according to local, regional and archipelago-scale gains.

2 Executive Summary

INTRODUCTION

Ensuring the continuity and long-term sustainability of Department of Defense (DoD) activities on Pacific islands requires effective stewardship of ecosystems that have been altered by the introduction of non-native species. Species invasions are threatening the evolutionary legacies of oceanic islands across the Pacific, and key ecological processes are being compromised by the presence of non-native species or the loss of native species. Advancing DoD stewardship of Pacific island ecosystems therefore requires understanding how ecological processes have been altered by non-native species and validating restoration approaches that foster the recovery of threatened, endangered, or at-risk species (TER-S).

Management of aquatic invasive species (AIS) in Pacific Island stream ecosystems is particularly challenging because traditional methods for control or mitigation fail to account for characteristics of insular streams and societal sensitivities associated with water resources and TER-S management. AIS are consistently identified as a primary threat to aquatic biodiversity in oceanic island streams, but little work has been done to evaluate the feasibility or outcomes of removal or mitigation. Similarly, no consistent effort has been made to develop new protocols or modify existing approaches for AIS management. Development of actionable information and innovative approaches for managing AIS therefore could substantively improve stewardship of stream ecosystems that cross DoD lands, especially on islands where installations harbor native species under federal or state protection.

OBJECTIVES

Herein we report findings from research designed to provide DoD resource managers with knowledge and tools to recover key ecological processes and TER-S in Pacific island streams impacted by AIS. Our primary research objectives were to (1) demonstrate a novel approach to AIS management in Pacific island streams (Statement of Need (SON) research interest #2); and to (2) advance basic understanding of ecosystem and TER-S responses to AIS removal from Pacific island streams (SON research interest #3). Recognizing that complete removal of AIS is not always feasible, we also undertook complementary studies to (3) determine whether careful consideration of target species and climate-driven hydrological conditions could increase the efficacy of AIS control to promote the recovery of ecological processes and TER-S (SON research interests #2, #3, #5). Information gained from this work was then leveraged to parameterize a decision-support tool to (4) help DoD managers design interventions that minimize resource expenditures and maximize in-stream conditions favorable to native species (SON research interest #2, #3, #5).

TECHNICAL APPROACH

Experimental AIS removals were conducted in 13 watersheds spanning a hydrological gradient across the Koʻolau Range on the Hawaiian island of Oʻahu (Fig. ES1), which is home to a dense array of DoD installations and at-risk native species. Following a before-after, control-impact (BACI) study design, AIS removals were executed following a protocol tailored for Hawaiian streams that included two tiers of precautionary steps to prevent collateral injury to electro-sensitive native species. Responses to removals were tracked for a period of up to two years, with adjacent upstream and downstream reaches serving as references. We examined whether removals yielded conditions favorable to native species by conducting snorkel surveys of population densities and community composition, as well as mark-recapture demographic assessments of *Awaous stamineus*, a native species of conservation concern. Additionally, we examined *A. stamineus* otolith microchemistry to determine how life-history varies according to AIS densities and hydrology, and whether removals influenced the balance of variation in local populations. We also assessed whether removals elicited an increase in genomic diversity by SNP genotyping native *Awaous stamineus* in each study reach. Finally, we conducted in-stream and stable isotope assays to determine whether removals influenced nutrient availability and trophic structure. Data from this work was used to parameterize a coupled ocean-watershed biophysical model to evaluate local-to-archipelago scale outcomes of AIS removal, with the aim of evaluating cost and benefit functions for selecting watersheds to maximize return on investments in AIS management.

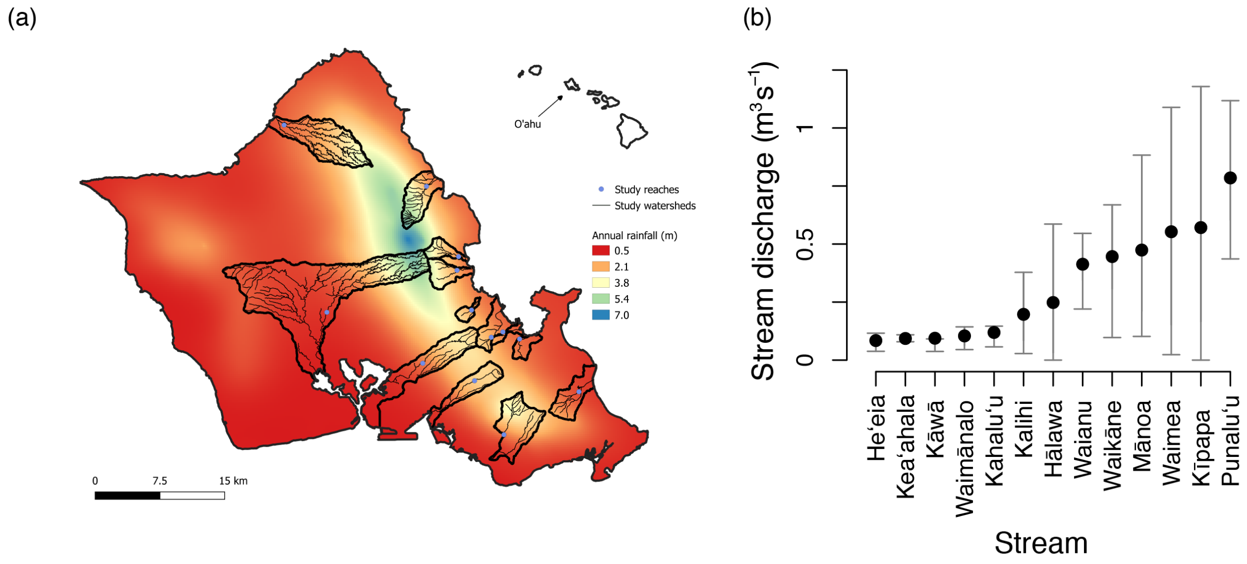


Figure ES1. (A) Map of Oʻahu delineating our study watersheds and study reaches relative to precipitation conditions; (B) Study streams ordered by increasing median discharge (solid points, Q_{50}, $m^3 s^{-1}$). Whiskers span low (Q_{90}) and high discharge (Q_{10}) derived from standard exceedance percentiles of daily discharge.

RESULTS AND DISCUSSION

Efficacy and community outcomes of AIS removal from Hawaiian streams.

Preferred methods of AIS management, such as toxicants and electrofishing, are generally considered unacceptable in the region because of the potential harm to at-risk and culturally important native species. As a result, few attempts have been made to control AIS and little effort has been made to develop protocols designed to protect at-risk species in tropical and subtropical Pacific island streams.

In this study, we assessed the effectiveness and outcomes of implementing an AIS removal protocol tailored for Hawaiian Island streams. The protocol combines hand-capture and streamside sequestration of native species with AIS removal via three-pass electrofishing with amperage and waveform combinations set to minimize harm to remaining native species. Experimental AIS removals were conducted in 13 watersheds on the island of Oʻahu (Fig. ES1) following a before-after, control-impact (BACI) design to assess how removal efficacy and post-removal reassembly vary by surface flow and target species of interest. This approach enabled us to determine whether particular flow conditions improve removal efficacy and promote conditions that favor native species like lower AIS recolonization rates.

We found that removals resulted in a sharp reduction in AIS densities without changing native species densities (Fig. ES2). The most widespread and abundant AIS were livebearing fishes (Poeciliidae) and armored catfishes (Loricariidae), which together accounted for 66% of all AIS removed. Removal efficacy was greatest in streams with intermediate discharge irrespective of pre-removal AIS densities or target group. Long-term monitoring also demonstrated that removals led to highly persistent shifts in community composition reflecting sustained reductions in AIS densities. These findings indicate that electrofishing can be a valuable tool for AIS management in tropical and subtropical Pacific island streams, so long as steps are taken to reduce risk to native species. Careful consideration of surface flow and pre-removal biotic conditions also can help maximize desirable outcomes of removal efforts.

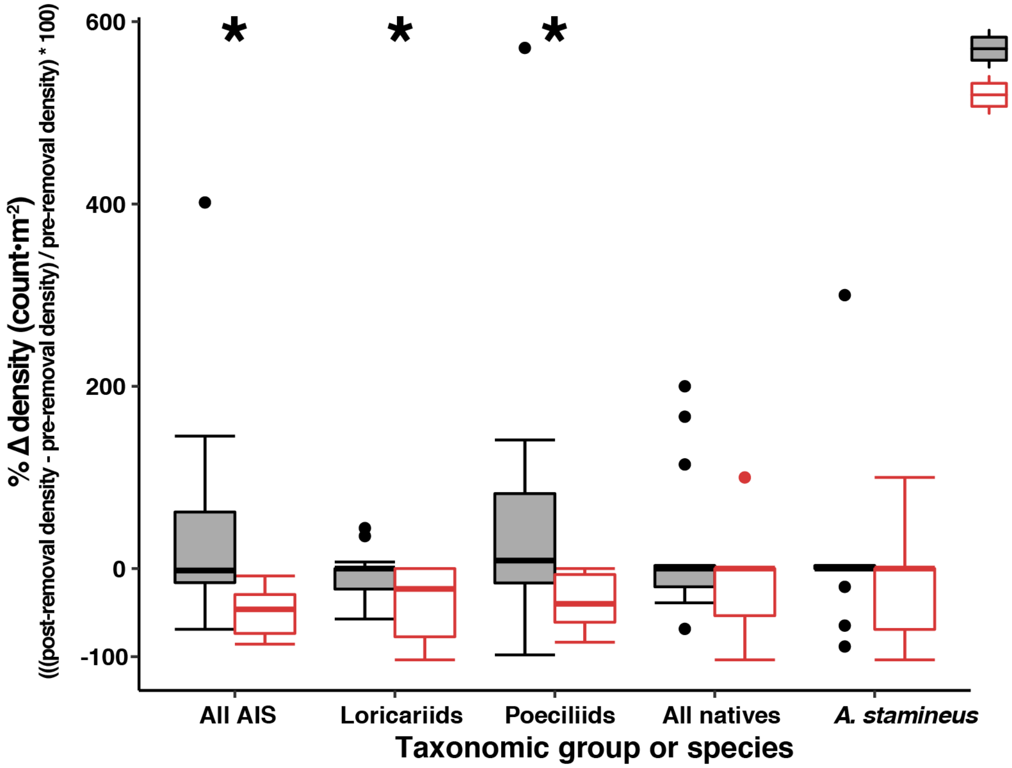


Figure ES2. The immediate percent density change (((post-removal density – pre-removal density)/pre-removal density) *100) for each group of interest between removal reaches (red) and control reaches (grey). Whiskers are minimum and maximum values relative to the first and third quartiles, respectively, whereas median values are denoted with bars. * = significant difference between reaches according to Welch's Two Sample t-tests.

Demographic responses to AIS removal from Hawaiian streams.

Understanding ecological outcomes following AIS removal can help promote evidence-based decision-making for the recovery of TER-S. In this study, we assessed the demographic responses of *Awaous stamineus*, an at-risk native Hawaiian fish, to AIS removal in study watersheds on the island of Oʻahu. Populations of *A. stamineus* were monitored before and after the removals in both treatment and control reaches. Demographic metrics including population size, the proportion of recruits, growth rates, body condition and in-stream movement.

Comparisons revealed that demographic measures like population size and recruitment were highly correlated between treatment and control reaches both before and after AIS removal, indicating that there was little to no demographic independence among reaches within streams. This pattern is consistent with inferences from snorkel surveys that AIS removals can create ecological ‘voids’ (i.e., areas where resources become newly available), which are then ‘filled’ by individuals moving in from outside of removal reaches (Koehn et al. 2000), resulting in the establishment of a new demographic equilibrium of lower overall densities in the removal and control reaches. However, we found that population sizes in the first 6 months after AIS removals were not significantly different than pre-removal population estimates. Movement of adults and juveniles into the treatment reach also did not change. However, the proportion of recruits in the populations significantly increased 3-6 months following removals. Compared to pre-removal measures, mean growth rates (measured by weight, not length) and body condition were also greater 0-3 months, 3-6 months, and >6 months following removals (Fig. ES3). The magnitude of the shift in recruitment was significantly positively correlated with the removal of Poeciliids from the study reaches. Growth and body condition responses were positively correlated with the removal of Loricariids from the study reaches and negatively correlated with mean discharge. While the increase in recruitment was short-lived, the increase in growth and body condition persisted for the duration of the study.

These findings indicate that AIS removal from Hawaiian streams can promote the recovery of native fish populations. If increases in recruitment and growth rates can be sustained, then population sizes should increase over the longer term. If AIS are not extirpated, periodic maintenance of removals (i.e., every 3-6 months) may be necessary to sustain demographic recovery.

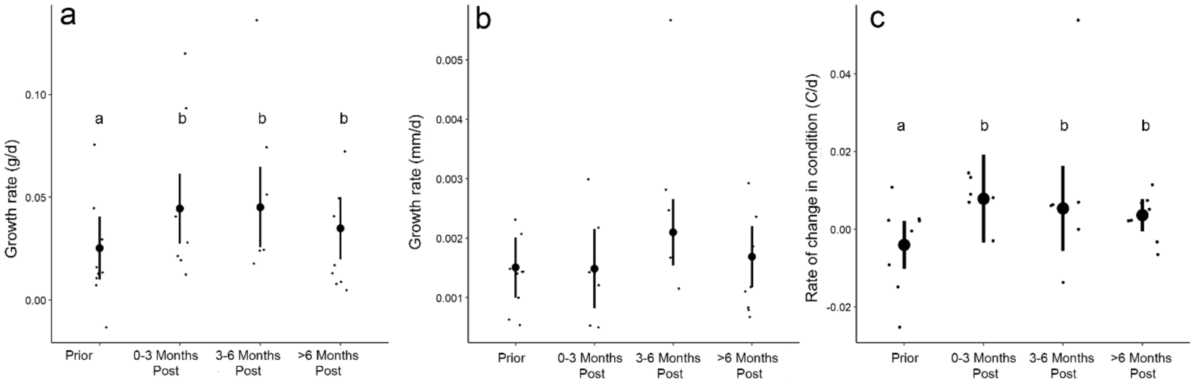


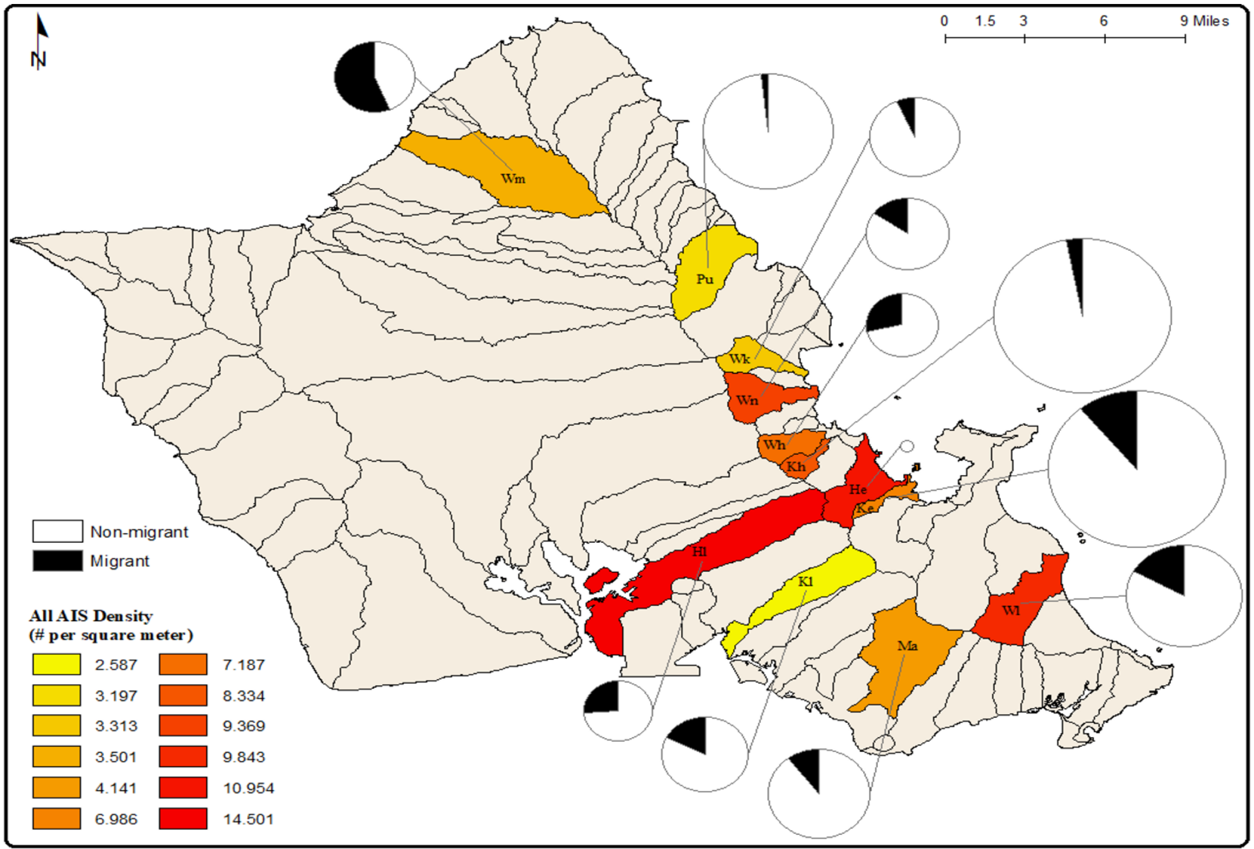
Figure ES3. Mean growth rates (large black dots) in (a) grams per day and (b) mm per day, as well as (c) rate of change in condition (*C*) per day in each study population (small black dots) prior to AIS removal, and 0-3, 3-6, and >6 months post AIS removal. Black vertical bars indicate 95% confidence intervals. Letters above each box plot indicate significant differences among periods identified by sequential contrasts post-hoc tests.

Life history variation in relation to biotic and abiotic gradients across Hawaiian streams.

Altered water quality, reduced habitat, as well as competition and predation from AIS can impose pressures that moderate the life history of native migratory species in oceanic island stream ecosystems. To properly manage oceanic island stream ecosystems, it is necessary to understand how AIS and other anthropogenic pressures influence the balance of migratory life history variation in at-risk native species. For partially migratory amphidromous species like *Awaous stamineus*, early life-stages may be spent in either a marine or natal freshwater environment, raising the possibility that the balance of ‘migrants’ to ‘residents’ (i.e., fish that remain in their natal habitat for the duration of their life cycle) in local populations might vary according to in-stream conditions.

We examined life history variation in populations of *Awaous stamineus* across in-stream biotic and abiotic gradients on the island of O‘ahu. We found that populations exhibited reduced proportions of residents when stream flow was low and where nutrients were limited (Fig. ES4). This suggests that inorganic nitrate, flow variability, and soluble reactive phosphorus likely constrain the survival of resident larvae. Similarly, we found that populations exhibited reduced proportions of residents in streams with high densities of non-native Poeciliid predators (Fig. ES4).

These findings suggest that improvement of in-stream conditions would likely increase the proportion of residents in local populations of *A. stamineus*, possibly increasing population stability and persistence. Steps could be taken, for example, to sustain discharge or restore natural flow regimes. Likewise, moderating nutrient availability and removing AIS would likely promote survival and persistence of *A. stamineus* and other native migratory fishes that are of conservation concern.



Watershed	🟨 Kalihi (Kl)	🟨 Punaluu (Pu)	🟨 Waikane (Wk)	🟧 Waimea (Wm)	🟧 Manoa (Ma)	🟧 Keaahala (Ke)	🟧 Waihee (Wh)	🟧 Kahaluu (Kh)	🟥 Waianu (Wn)	🟥 Waimanalo (Wl)	🟥 Heeia (He)	🟥 Halawa (Hl)
N	28	59	30	25	38	101	21	100	26	48	1	19
Non-migrants	23	58	28	11	34	90	15	97	22	40	1	14
Migrants	5	1	2	14	4	11	6	3	4	8	0	5
Poecillid Density	1.428	2.95	2.63	0.001	1.225	4.268	3.406	4.604	9.114	7.153	3.66	13.943
Loricariid Denisty	0.883	0.001	0.0331	0	0.499	2.577	3.628	3.45	0	0.065	5.83	0

Figure ES4. Proportions of migrant and non-migrant (i.e., resident) *Awaous stamineus* relative to overall invasive species density (# / m^2) in each study watershed, with reference to sample size (N), Poeciliid density, and Loricariid density.

Genomic and life history responses to AIS removal from Hawaiian streams.
Theory predicts that management interventions can trigger genetic rescue— with or without demographic rescue— which can bolster the potential for at-risk populations to persist via increases in immigration. We tested this prediction by assaying genomic and life history responses for comparison to demographic responses of the facultative amphidromous goby, *Awaous stamineus*, to experimental AIS removals. This involved comparing snorkel survey and mark-recapture based estimates of demography with ddRAD SNP assays of genomic variation and otolith microchemistry analysis of migratory life history. This approach allowed us to infer the nature and magnitude of responses to AIS removals as well as possible underlying ecological and evolutionary pathways of rescue. We detected increased genomic diversity following AIS removals (Fig. ES5) as well as an increase in population densities corresponding to the reproductive cycle of *A. stamineus*. We also found that immigrant post-larval juveniles (i.e., new recruits) had higher genomic diversity compared to non-migrant post-larval juveniles (Fig. ES5) , which suggests that an increase in immigration might be driving observed genomic and demographic shifts. However, contrary to this prediction, we detected an increase in the number of resident, not immigrant, post-larval juveniles following AIS removals. This indicates that, at least for *A. stamineus*, AIS control can result in genetic rescue via local demographic gains in resident populations rather than gains due to an increase in immigration. If so, then conservation management strategies aiming to promote the well-being of resident populations may be as or more effective than elevating gene flow in at-risk species with flexible migratory life histories.

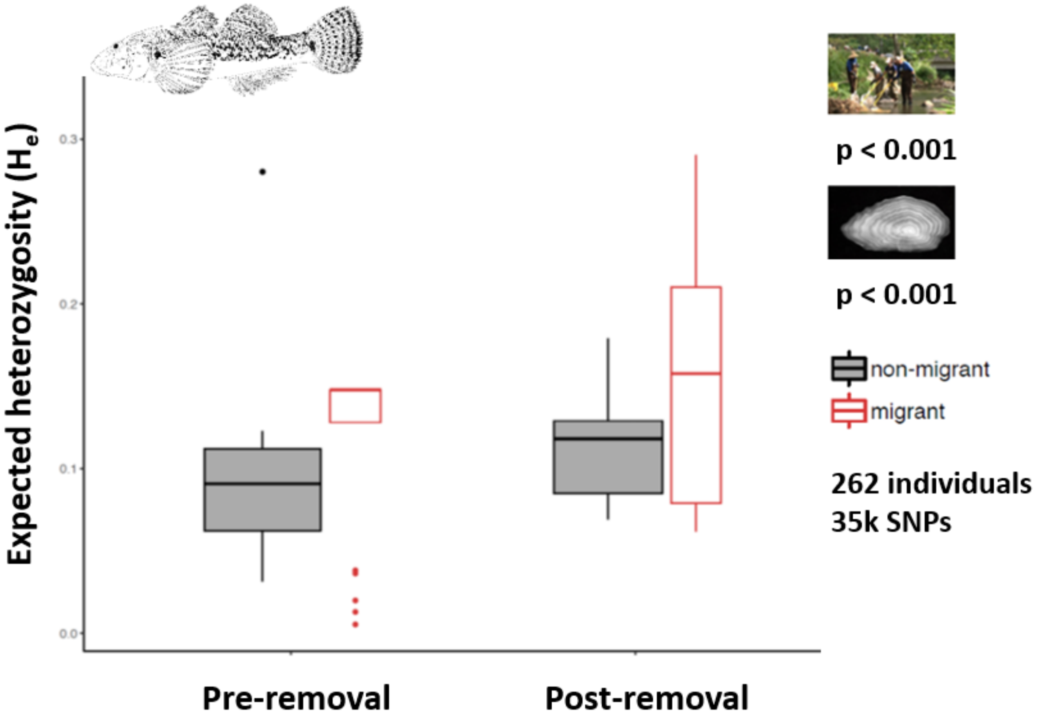


Figure ES5. Overall, *A. stamineus* migrants exhibit higher genomic diversity, and genomic diversity increased after AIS removals. Non-migrants exhibited a greater increase in diversity, however, which is consistent with demographic evidence of a post-removal rise in recruitment and an increase in the proportion of residents in the study populations following AIS removals. Whiskers are minimum and maximum values relative to the first and third quartiles, respectively, whereas median values are denoted with bars. Significant differences according to AIS removal (i.e., time period) and migratory life history determined according to Welch's Two Sample t-tests.

Ecosystem responses to AIS removal from Hawaiian streams.

AIS have reconfigured the structure and diversity of stream communities across the Hawaiian archipelago. Biotic transformation is often accompanied by altered ecosystem process rates, but it can be difficult to determine whether AIS are directly responsible for ecosystem changes or simply coincide with anthropogenic physical and chemical disturbances. In this study, we quantified the direct effects of AIS on nutrient and carbon dynamics by assaying ecological associations and experimentally removing AIS from 13 streams circumscribing broad gradients of hydrological dynamics, habitat conditions, and nutrient pollution on Oʻahu (Fig ES1).

Prior to AIS removals, ecosystem gross primary productivity was suppressed by introduced Loricariid armored catfishes even when there was an abundance of light and nutrients (Fig. ES6). Bioassays indicated that algae in several streams became nutrient-limited following removal of AIS, but otherwise we did not detect a consistent shift in nutrients that limit algal growth, concentrations of dissolved nutrients, leaf-litter breakdown rates, or whole-stream metabolism. However, differences across streams in effect sizes on ecosystem processes were related to the AIS biomass removed, background nutrient concentrations, stream size, and watershed land use. Evidence that contextual biotic and abiotic variation can mitigate or mask ecosystem responses to costly AIS removal efforts suggests that pre-removal assessments should be conducted to help prioritize restoration sites. Our results also indicate that a combination of interventions, such as improving water quality and habitat alongside AIS control, may be required to restore ecosystem conditions that favor native species.

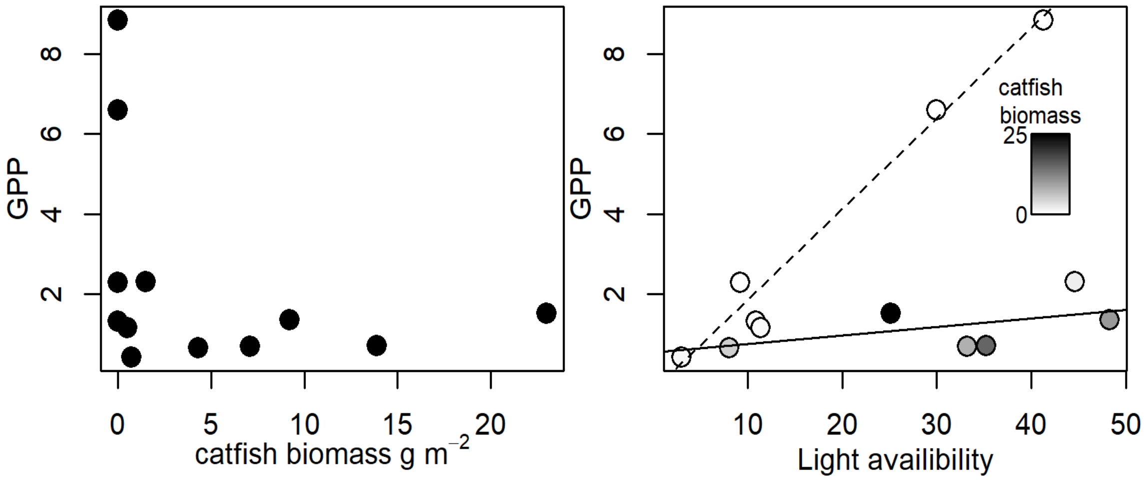


Figure ES6. (A) Reach-scale gross primary production (GPP) measured before AIS removals (0.27 to 9.82 mg O_2 m^2 d^{-1}), as a function of armored catfish (catfish) biomass (0 to 23 .0 g m^{-2}) in removal reaches. (B) GPP as a function of percent light availability in the removal reaches (inverse percent canopy cover). Pearson correlation analysis demonstrated that GPP is positively associated with percent light availability, and that the association was stronger in streams without armored catfish (open points, dashed line) compared to streams with armored catfish (points shaded, solid line).

Coupled oceanic-watershed modeling of whole-watershed AIS removal.

Even though AIS are consistently identified as a primary threat to aquatic biodiversity in oceanic island streams, little guidance is available to prioritize control or eradication of the most harmful invaders. It thus should come as no surprise that eradication attempts often fail because management campaigns do not adequately scale suppression efforts or do not adequately target high-impact populations or areas where interventions might yield disproportionate benefits beyond local populations.

Understanding that deep-rooted evolutionary differences in life history traits can sometimes buffer native species against AIS control measures, we have conducted coupled ocean-watershed model simulations showing that differences in life history can be strategically leveraged to not only restore local populations but also achieve metapopulation-wide benefits through demographic spillover.

The endemic stream fauna of the Hawaiian archipelago and other tropical oceanic islands is dominated by amphidromous species that naturally recolonize streams following oceanic larval dispersal. In contrast, the non-native fauna in Hawaiian streams (and insular streams elsewhere) is dominated by non-migratory species that are obligately bound to freshwater conditions. Accounting for this dichotomy, modeling showed that removal of both invasive (lethally) and native (lethally or non-lethally) species throughout a selected watershed can reliably convert demographic sinks into productive source populations that provide recruits to streams across the archipelago (Fig. ES7). Model simulations also indicate that this approach is both low-risk (local increases in natives always occur quickly) and high-reward (spillover somewhat enhances all other populations), but requires targeting removals to large, highly-invaded watersheds that are well connected by ocean currents (Fig. ES7).

While our model has been parameterized to illustrate restoration outcomes across Hawaiian watersheds, the proposed 'clean-slate' restoration approach is broadly applicable to situations where native species can recolonize more easily than invaders following mass mortality stemming from control measures.

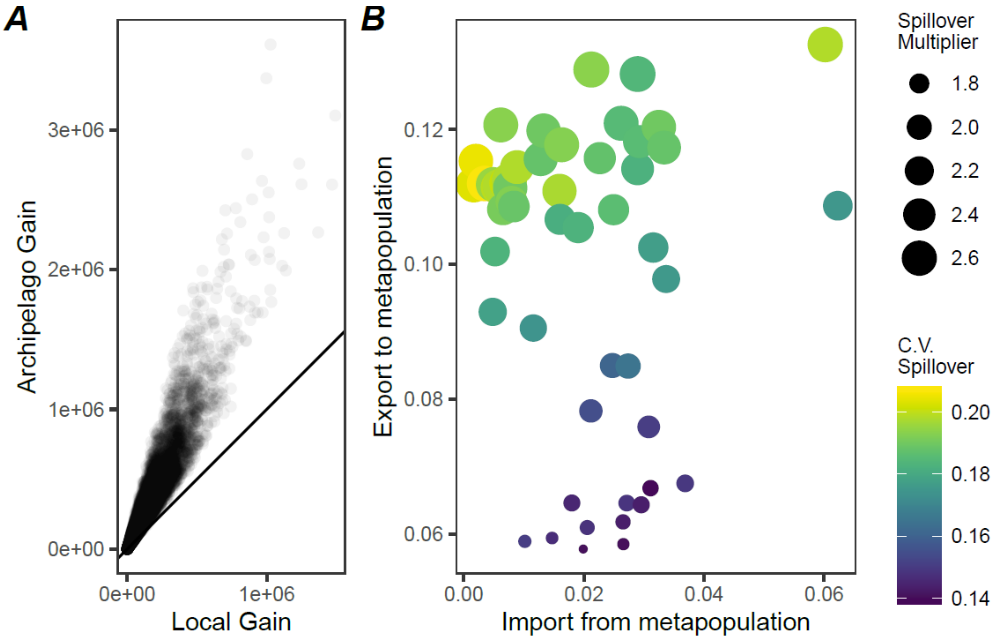


Figure ES7. (A) Following extermination of invasive species from a single watershed, growth of the metapopulation across the Hawaiian archipelago greatly exceeds local gains alone. In no case does the metapopulation decline. Spillover from 'clean-slate' restoration is a robust outcome when populations are recruitment limited: it occurs in all study streams. If recruitment is not limited, no spillover occurs, and all points fall on the black line, indicating that gains are solely local. (B) Connectivity via larval dispersal varies widely among watersheds, which strongly affects spillover (ratio of archipelago:local population gains). Fitted to simulation results depicted in (A), each of which has a distinct combination of the import (proportion of all

successfully dispersing larvae arriving at a given watershed) and export (proportion of all larvae from a given watershed that successfully disperse to any watershed) values estimated from a passive dispersal model applied to observed ocean currents from 2007-2011. Differences in spillover among streams depend primarily on export, so long as import is sufficient to ensure growth in the local population following eradication of invasive species. Spillover tends to be highest with high export.

IMPLICATIONS FOR FUTURE RESEARCH AND BENEFITS

Development of innovative approaches and actionable information for managing AIS can substantively improve DoD stewardship of stream ecosystems that cross DoD lands, especially on islands like Oʻahu where installations harbor native species under federal or state protection. This project not only empirically demonstrated that modified electrofishing can be a valuable tool for AIS control in Hawaiian streams, it also illustrated how managers can maximize benefits of AIS control through careful consideration of target species and climate-driven hydrological conditions. Likewise, the project delivered a model-based decision support tool that can serve as a defensible and transparent analytical framework for prioritizing interventions according to local, regional and archipelago-scale gains.

Development of innovative approaches and actionable information for managing AIS has yielded several key findings and important conclusions, including:

- **AIS control is feasible and effective** with a tailored electrofishing protocol that minimizes collateral harm to TER-S
- **Control efforts can result in sustained reductions** in overall AIS densities and densities of targeted AIS (where removal efficacy is mediated by AIS densities and hydrology)
- **AIS removal can increase recruitment**, growth, and body condition of TER-S
- **AIS removal can increase genomic diversity** and composition of local TER-S populations (where gains are mediated by migratory life history)
- **Whole watershed ‘clean-slate’ restoration can lead to demographic spillover** that benefits populations elsewhere in the archipelago (leveraging life history differences between AIS and TER-S)

With an expanded knowledge base and toolkit, DoD resource managers and partner organizations should be able to achieve more effective watershed management and conservation of at-risk aquatic species to ensure the sustainability of military operations in the Hawaiian Islands and other oceanic islands in the Pacific.

Several complementary approaches have been taken to transfer information and tools to practitioner communities in Hawaiʻi and elsewhere. To initiate the translation process, a core group of Oʻahu-based resource managers were engaged to demonstrate new methods for AIS control in coordination with ongoing AIS management activities. This involved, for example, conducting experimental AIS removals in study reaches located in management areas. This also involved conducting field demonstrations with federal and state resource managers. To support future, independent endeavors, we have developed a technical ‘user’ guide for resource managers to undertake AIS removals in Hawaiian streams. The guide not only offers summary and detailed overviews of protocols, it also provides supporting technical documents, such as template data sheets. Finally, we also have disseminated project findings to the broader scientific community and general public through publication of peer-review papers and seminars at regional workshops and national conferences, as well as through popular press articles and community outreach.

3 Objectives

Ensuring the continuity and long-term sustainability of Department of Defense (DoD) activities on Pacific islands requires effective stewardship of ecosystems that have been altered by the introduction of non-native species. Species invasions are threatening the evolutionary legacies of oceanic islands across the Pacific, and key ecological processes are being compromised by the presence of non-native species or the loss of native species. Advancing DoD stewardship of Pacific island ecosystems therefore requires understanding how ecological processes have been altered by non-native species and validating restoration approaches that foster the recovery of threatened, endangered, or at-risk species (TER-S).

Management of aquatic invasive species (AIS) in Pacific Island stream ecosystems is particularly challenging because traditional methods for control or mitigation fail to account for characteristics of insular streams and societal sensitivities associated with water resources and TER-S management. AIS are consistently identified as a primary threat to aquatic biodiversity in oceanic island streams, but little work has been done to evaluate the feasibility or outcomes of removal or mitigation. Similarly, no consistent effort has been made to develop new protocols or modify existing approaches for AIS management. Development of actionable information and innovative approaches for managing AIS therefore could substantively improve stewardship of stream ecosystems that cross DoD lands, especially on islands where installations harbor native species under federal or state protection.

This project was undertaken to provide DoD resource managers with knowledge and tools to recover key ecological processes and TER-S in Pacific island streams affected by AIS. Our primary research objectives were to:

(1) Demonstrate a novel approach to AIS management in Pacific island streams (Statement of Need (SON) research interest #2)

(2) Advance basic understanding of ecosystem and TER-S responses to AIS removal from Pacific island streams (SON research interest #3).

Recognizing that complete removal of AIS is not always feasible, we also undertook complementary studies to:

(3) Determine whether careful consideration of target species, climate-driven hydrological conditions, and surface flow management can increase the efficacy of AIS control to promote the recovery of ecological processes and TER-S (SON research interests #2, #3, #5).

Information gained from this work was then leveraged to parameterize a decision-support tool to:

(4) Help DoD managers design interventions that minimize resource expenditures and maximize in-stream conditions favorable to native species (SON research interest #2, #3, #5).

4 Background

4.0 PROBLEM STATEMENT

Species invasions have resulted in the loss of native species and alteration of ecological processes on islands across the Pacific (Brasher 2003). Oceanic islands harbor evolutionary legacies that have proven highly susceptible to non-native species intentionally or accidentally introduced as a consequence of human occupation. For example, the brown tree snake has decimated native and endemic bird populations after being introduced on Guam (Wiles et al. 2003). Similarly, the intentional release of predatory fishes into Hawaiian streams for sport (e.g., small mouth bass) and mosquito control (e.g., guppies and other Poeciliids) has been linked to extirpation of endemic damselflies (Polhemus 1993, Polhemus and Asquith 1996). By altering ecological processes, species invasions also can create conditions that directly or indirectly prevent recovery of native species. For example, non-native fishes in Hawaiian streams that target early life stages as prey can depress native invertebrate and fish populations by reducing juvenile recruitment (Walter et al. 2012, Hain et al. 2019). By modifying nutrient availability, non-native coqui frogs can shift competitive balances to favor non-native versus native plants in Hawaiian forests (Beard et al. 2002, Sin et al. 2008). Species invasions can also precipitate the disintegration of whole ecosystems- for instance, by eliminating native birds that disperse seeds, invasive brown tree snakes have been altering the distribution, prevalence, and composition of forests on Guam (Caves et al. 2013, Wandrag et al. 2017, Egerer et al. 2018).

Species invasions that threaten biodiversity and valuable ecosystem services can complicate DoD activities on Pacific islands (e.g., use of testing and training areas). Ensuring mission continuity and long-term sustainability of DoD interests on Pacific islands therefore requires effective stewardship of ecosystems that have been altered by the introduction of non-native species. While complete eradication is often the desired management endpoint, it is rarely feasible because of cost and logistical constraints. Well-conceived control and mitigation efforts can, however, potentially (re)establish ecological functions, boost valued ecosystem services and promote the recovery of native species, including TER-S.

Achieving these endpoints is more likely if management approaches are based on rigorous understanding of individual, population, community and ecosystem outcomes that influence the persistence of native species (Townsend 2003, Lepak et al. 2006, Marks et al. 2009). Similarly, management approaches must consider attendant societal concerns of potential risks to TER-S and the integrity of ecological or cultural resources. Management of aquatic invasive species (AIS) in Pacific Island stream ecosystems is particularly challenging because no methods for control or mitigation have been developed for insular stream conditions. AIS are a widely recognized threat to native and endemic species (Brasher 2003), yet no consistent effort has been made to develop protocols for management in oceanic island streams. As a result, AIS are rampant and poorly understood on Pacific islands (Englund and Filbert 1999). Little is known about the feasibility or potential outcomes of AIS removal and even less is known about mitigative watershed management approaches, such as deliberate regulation of stream hydrology (Englund and Filbert 1999, Kiernan et al. 2012), to sustain native biota in the presence of AIS.

We addressed this deficit by evaluating genes-to-ecosystem outcomes of experimental AIS removals across a stream flow gradient in concordance with the development of a model-based decision tool to better evaluate AIS management under different hydrological and climate regimes. Herein we report findings from research designed to provide DoD resource managers with actionable information and innovative approaches for managing AIS that will enable DoD to substantively improve stewardship of Pacific island streams and resident TER-S. Below we lay out the motivations and technical approaches for the study, as well as findings describing (1) a novel approach to AIS management in Pacific island streams; and (2) knowledge gained about ecosystem and TER-S responses to AIS removal from Pacific island stream, including how (3) careful consideration of target species and climate-driven hydrological conditions can increase the efficacy of AIS control to promote the recovery of ecological processes and TER-S. We also illustrate how information gained from experimental AIS removals allowed us to parameterize a decision-support tool to (4) help DoD managers design interventions that minimize resource expenditures and maximize in-stream conditions favorable to native species.

4.1 NATIVE SPECIES AND AQUATIC INVASIVE SPECIES (AIS) IN PACIFIC ISLAND STREAMS

The diversity of AIS is often much higher than native species diversity in Pacific island streams. Very few species are capable of long-distance dispersal through the open ocean to reach streams on oceanic islands. As a result, the native fauna of Pacific island streams features low species diversity, high endemism, and strong representation of obligate and facultative amphidromous species sustained by ocean-stream connectivity. Amphidromy is a form of diadromy. Obligate amphidromy involves growth and reproduction in freshwater, with hatchling larvae drifting downstream to marine environments where they disperse before recruiting back to freshwater as postlarvae. Larvae of facultative amphidromous species do not necessarily undergo marine dispersal; rather, some individuals may elect to instead remain in natal watersheds. The diversity of AIS in Pacific island streams reflects historical legacies of accidental or intentional introductions by humans. The dominance of AIS is particularly severe in streams across the Hawaiian archipelago, which is home to a dense array of DoD installations. Non-native freshwater fishes, for example, occur in nearly every watershed in Hawaiʻi as a consequence of mosquito control, recreational fishing, household release, control of invasive plants, and aquaculture. In streams on Oʻahu, where the majority of DoD installations are located, the number of non-native fishes can be an order of magnitude higher than that of native fishes (Eldredge 2000, Yamamoto & Tagawa 2000).

The prevalence of AIS can directly and indirectly constrain the diversity and abundance of native species in Pacific island streams (Brasher 2003, Walter et al. 2012). Through predation and competition, AIS can result in local extinction and prevent (re)colonization of streams by native species (Sax and Gaines 2008). The introduction of non-native freshwater fishes- with over 60 species from 18 families now established on Pacific islands- has proven to be particularly problematic (Nico and Walsh 2011). For example, the introduction of Mozambique tilapia (*Oreochromis mossambicus*) precipitated the collapse of traditional milkfish (*Chanos chanos*) aquaculture on Nauru and Kiribati (Nelson and Eldredge 1991, Spennemann 2002). In the Hawaiian archipelago, extirpations of native *Megalagrion* damselflies, several of which are now listed or candidates for federal protection under the Endangered Species Act, have been attributed to predation by introduced guppies and other Poeciliid live-bearers (Polhemus 1993, Polhemus and Asquith 1996, Yamamoto and Tagawa 2000, Englund 2001). Though cause and effect has been difficult to establish, non-native armored catfish (Loricaridae) likely have depressed co-occurring native fish populations by increasing stream siltation, preying on fish eggs, and possibly by limiting the availability of nutrients essential for growth and reproduction (Vanni et al. 2002, Rowe 2007, Capps and Flecker 2013). The introduction of Poeciliids has also led to the spread of non-native parasites to four of the five native amphidromous fishes in Hawaiʻi (Font 2003).

4.2 LESSONS LEARNED: AIS CONTROL AND ERADICATION ON PACIFIC ISLANDS

Though direct and indirect suppression of AIS populations is challenging, many biogeophysical features of Pacific island streams increase the odds of success. For example, most drainage basins on oceanic islands are small, narrow, and steep by continental standards (Kinzie 1988, Brasher 2003, Covich et al. 2003, Oki and Brasher 2003, Smith et al. 2003), hence populations of AIS are often spatially restricted because spread is limited by waterfalls, rapids, or other barriers (that can be surmounted by native species with the capacity to climb). The absence of watershed connectivity also minimizes the odds of recolonization following removal, and while some AIS can escape to coastal waters (e.g., tilapia), many streams exhibit terminal waterfalls that can function as barriers to re-invasion.

Surprisingly few attempts have been made to eradicate, control, or mitigate AIS in Pacific island stream ecosystems, and outcomes of most past management actions have been poorly documented (Nico and Walsh 2011). Nonetheless, several important inferences can be drawn from available records on efforts to manage non-native fishes (Englund and Filbert 1999, Nico and Walsh 2011) in tropical and subtropical Pacific island streams. AIS management in New Zealand also affords some useful perspectives, as many at-risk native fishes in New Zealand are amphidromous. First, eradication efforts have largely been unsuccessful (Nico and Walsh 2011). Second, nearly all attempts to manage AIS have involved methods designed for continental stream conditions (Englund and Filbert 1999, Nico and Walsh 2011). Third, AIS management must account for societal concerns about access to water resources and erosion of cultural traditions (Englund and Filbert 1999, Nico and Walsh 2011). Finally, management that accounts for insular conditions can promote the recovery of at-risk native species (Nico and Walsh 2011).

Eradication of entire populations is not often a viable management option (Kolar et al. 2010), so it is not surprising that efforts to eradicate non-native fishes on Pacific islands have proven unsuccessful. Efforts may be hindered by the type, abundance and distribution of the target species, as well as the size and complexity of the invaded environment (Kolar et al. 2010). Nico and Walsh (2011) identified 11 separate campaigns to eradicate non-native fishes on tropical and subtropical Pacific islands. All but one were carried out in small closed systems (e.g., ponds). Though eradication in open systems is widely considered unfeasible, the Guam Division of Aquatic and Wildlife Resources has been attempting to eliminate chevron snakeheads (*Channa striata*) from the Ajayan River on Guam. Complete eradication has not been clearly achieved in this or any other documented campaign (Nico and Walsh 2011).

Nearly all of the documented campaigns on Pacific islands have relied on toxicants (piscicides or poisons) that are the preferred method for eradicating nuisance fish worldwide (Kolar et al. 2010, Nico and Walsh 2011). Chemical toxicants, such as rotenone and antimycin-a, are typically diluted liquid solutions released into the water and then neutralized or left to naturally degrade over a period of days (Dinger and Marks 2007, Moore et al. 2008). Use of toxicants can fail to eradicate target species without proper evaluation, planning and implementation (Donlan and Wilcox 2007). Although toxicants have been used extensively in New Zealand for non-native fish eradication, (e.g., Lintermans 2000, Chadderton et al. 2003, Pham et al. 2013), no comparable effort has been made to develop protocols for tropical and subtropical Pacific islands. Prior implementation appears to have followed general principles developed for aquatic ecosystems elsewhere (Nico and Walsh 2011).

Prior campaigns have not made extensive use of electrofishing, even though it has become the foremost non-lethal capture method for freshwater fish management (Murphy and Willis 1996, Nico and Walsh 2011). Electrofishing provides the greatest return for the least effort among fish capture methods, making it especially useful for non-native fish removal. The principle of electrofishing is to produce an electric field around electrodes in water to temporarily stun nearby fish (Snyder 2003). The approach has been extensively used for removal of non-native fish across North America using backpack units for small streams and boat-mounted units for larger water bodies (Kolar et al. 2010). Plunge pools, boulders, and other complex habitat characteristics make electrofishing challenging on Pacific islands (Baker and Foster 1992), though studies conducted on Puerto Rico have demonstrated that electrofishing can be highly effective for capturing fish in insular streams (Kwak and Cooney 2008).

Many of the most widely used control and eradication methods- including electrofishing and chemical toxicants- are now generally considered unacceptable for AIS management on Pacific islands because of potential harm to native species (Britton et al. 2008, Kolar et al. 2010). Rotenone and antimycin-a may affect native fishes and aquatic invertebrates more strongly than targeted species (Finlayson et al. 2002, Dinger and Marks 2007, Schofield and Nico 2007). Electrofishing also can result in non-target injuries and mortality (Snyder 2003), though complications can be reduced through appropriate amperage and waveform combinations (Holliman et al. 2003). Use of toxicants has been banned by some Pacific island governments, including Hawaiʻi where it is a criminal felony. The State of Hawaiʻi Division of Aquatic Resources (DAR) also has established a general moratorium on electrofishing (an exception was granted for this study in order to advance understanding of the risks and effectiveness of electrofishing for AIS management in Hawaiian stream ecosystems).

Few attempts have been made to carry out AIS control or eradication on Pacific islands that have involved methods set to reflect protective controls, but at least one documented case has demonstrated that precautionary steps can be taken that protect and promote the recovery of native species. As described by Nico and Walsh (2011), rotenone was used to eradicate a reproducing population of Nile tilapia in a freshwater crater lake in the Galapagos archipelago. Approximately 40,000 tilapia were removed from the lake. Before application of rotenone to the lake, aquatic invertebrates were collected and held in refuge tanks. The captive invertebrates were released back into the lake after the rotenone had degraded in order to promote recovery of native communities possibly affected by exposure to the toxicant (Nico and Walsh 2011). This example– and other examples of sequestering and restocking native fauna in New Zealand and elsewhere (e.g., Buktenica et al. 2013, Pham et al. 2013)– indicates that AIS management can incorporate methods that reduce risks, offset potential non-target impacts, and promote the survival of at-risk species on Pacific islands.

4.3 LESSONS LEARNED: MITIGATING AIS THROUGH FLOW MANAGEMENT ON PACIFIC ISLANDS

Managing stream flow conditions can be an effective approach for AIS management, where populations of native species are enhanced either through the recovery of critical ecological and fluvial processes and/or by suppression of AIS. The objective of managing surface flow is to mimic components of natural variability, such as magnitude, frequency, and duration. Different aspects of surface flow regimes can be critical for the completion of key life history stages of riverine fishes, including spawning, migration and recruitment. If one or more life history stage of a native species is dependent on specific stream flow attributes, then hydrological management can help sustain or enhance at-risk populations (Probst and Gido 2004, Scoppetone et al. 2005, King et al. 2009, Kiernan et al 2012). However, while there is evidence from continental rivers that altered stream flow (e.g., impoundment or diversion) can favor non-native fishes (Minckley and Meffe 1987, Baltz and Moyle 1993, Marchetti and Moyle 2001), it remains unclear whether managing flow regimes is effective for suppressing non-native fish populations, particularly in oceanic island streams (Probst and Gido 2004, Marks et al. 2009).

Managing surface flow regimes has been recognized as a potential approach for fostering ecosystem recovery and suppression of AIS in Pacific island streams, especially in Hawaiʻi , but it has not been carefully evaluated (Fitzsimons and Nishimoto 1996, Fitzsimons et al. 1997, Englund and Filbert 1999). Surface flow has been altered in at least 58% of the 366 perennial streams in Hawaiʻi (Parrish et al. 1978, Brasher 2003). Stream diversions can capture a sufficient portion of base flow to disrupt "ridge to reef" ocean-stream connectivity (Gingerich and Wolff 2005). Loss of connectivity can disrupt contributions to the marine larval pool and post-larval recruitment to freshwater, while also reducing stream habitat required for adult growth and reproduction (Holmquist et al. 1998, Benstead et al. 1999, Iida et al. 2010). (Re)establishing ocean-stream connectivity (e.g., by removing or reducing water diversions) can thus potentially foster ecological and fluvial conditions favorable to native species. Managing surface flow regimes also might suppress AIS populations either by shifting environmental conditions (e.g., temperature) or by flushing during high flow events (Marchetti and Moyle 2001, Englund and Filbert 1999). Unlike native species, most AIS do not exhibit adaptations (e.g., fused pelvic fins for attachment to substrates) to persist during high flow conditions that are characteristic of many Hawaiian streams.

While the influence of reduced surface flow is well understood for Hawaiian streams (see Brasher 2003), only a few studies have been conducted on outcomes of managing surface flow to promote the well-being of native species (Englund and Filbert 1999, Englund 2001, Kinzie et al. 2006, Sherwood et al. 2009). Nonetheless, several studies suggest that increasing flow in Hawaiian streams can aid in the recovery of native species. Kinzie et al. (2006), for example, found that ecosystem productivity increased in reaches where flow was increased in association with removal of a diversion dam on Kauai. Similarly, Sherwood et al. (2009) found that macroalgal diversity substantially increased with the return of water following closure of an agricultural diversion on Hawaiʻi. Long-term monitoring of the only population of Orangeblack Hawaiian damselfly (*Megalagrion xanthomelas*) on Oʻahu, located in a stream crossing Tripler Army Medical Center, showed that augmented stream flow increased abundance by an order of magnitude over a 3 year period (Englund 2001). It remains unclear, however, whether managing surface flow suppresses AIS populations, in part because only one study has examined how AIS have responded to modification of surface flow conditions in a Hawaiian stream. Englund and Filbert (1999) found that invasive fish remained abundant and native fish diversity slightly declined over a nine year period following the cessation of agricultural diversions in Waikele stream (Englund and Filbert 1999). In contrast, studies conducted elsewhere have found that flow modification can be a powerful tool for AIS management and native fish conservation, especially when combined with other efforts. It has been demonstrated, for example, that managing surface flow can encourage greater responses of native fish to the removal of invasive fish by increasing carrying capacity and favorable spawning conditions (Marks et al. 2009).

4.4 ADVANCING CONTROL AND MITIGATION OF AIS IN PACIFIC ISLAND STREAMS

Developing actionable information and innovative approaches for managing AIS could substantively improve DoD stewardship of insular streams, especially on islands where installations harbor native species under federal or state protection. Native fishes and invertebrates are becoming increasingly imperiled in Pacific island streams, including streams that cross DoD installations on Oʻahu and elsewhere in Hawaiʻi . Two species of native damselflies (the Pacific Hawaiian damselfly (*Megalagrion pacificum*), and the Flying Earwig Hawaiian damselfly (*Megalagrion nesiotes*)) were recently added to the federal register of endangered species. Nearly two dozen other damselflies are candidates for listing, including the Orangeblack Hawaiian damselfly that inhabits the grounds of the Tripler Army Medical Center on Oʻahu. AIS, and Poeciliids in particular, are a primary cause of endangerment of native damselflies. At least three of the five native migratory stream fishes also are now rare on Oʻahu. AIS and reduced surface flow are thought to be primary drivers of population decline of native fishes on Oʻahu. Yet, DoD resource managers currently have a limited tool kit to improve conditions for the benefit of TER-S in Pacific island streams. Not only is there great need to rigorously characterize outcomes of AIS management, approaches are needed that conform to public sentiment and protections for native species. Accordingly, the overarching goal must be to develop a repeatable approach that reduces AIS with little (or no) risk to native species.

While there are no standard protocols for AIS removal in Hawaiʻi or other Pacific islands, it has been demonstrated that innovative protocols that incorporate precautionary steps can be implemented to reduce risks and promote the survival of species of concern (Nico and Walsh 2011). In addition, there is reason to believe that surface flow management can potentially suppress AIS populations (Winder et al. 2011, Kiernan et al. 2012) and enhance the recovery of at-risk native species by altering nutrient availability, predation, and other key ecological processes (Benstead et al. 1999). Stream flows also can be managed to provide conditions favorable to the dispersal, reproduction, and survival of native species (Kiernan et al. 2012).

Setting appropriate installation-specific and regional objectives also requires improved methods for evaluating outcomes of management interventions. Model-based decision tools that couple information on hydrology and demography of native species could help DoD managers establish best practices that minimize costs (e.g., the number and magnitude of interventions) and maximize benefits (e.g., larger shifts toward in-stream conditions favorable to native species). For instance, if adaptive management of surface flow hydrology conflicts with other mission-critical uses of water resources, modeling could be conducted to illustrate strategies that protect sensitive life history stages of native species at little cost or conflict. If regulating water use becomes too disruptive as climate conditions shift (Oki 2004, Diaz et al. 2005, Chu and Chen 2005, Safeeq and Fares 2011), modeling could also prove useful for updating approaches for sustaining at-risk native species.

4.5 AIS REMOVAL FROM PACIFIC ISLAND STREAMS

We assessed the feasibility, effectiveness and outcomes of AIS removals executed following an electrofishing protocol that included two tiers of precautionary steps to prevent collateral harm to electro-sensitive native species. The protocol combines hand-netting and streamside sequestration of native species with AIS removal via three-pass electrofishing with amperage and waveform combinations set to minimize potential harm to any remaining species of concern (Holliman et al. 2003). This approach balances the high return on investment offered by electrofishing with the assiduous encumbrance of measures taken to protect native species of concern.

The principle of electrofishing is to produce an electric field around electrodes in water to temporarily stun nearby fish, which can then be gathered for data collection or removal (Snyder 2003). Electrofishing is most efficient in water bodies with clear, highly conductive water and simple substrate. Properly conducted, electrofishing generally results in negligible mortality and injury rates (McCrimmon and Bidgood 1965). However adverse physiological effects can include spinal damage and hemorrhaging (e.g., Sharber and Carothers 1988, Sharber et al. 1994), reduced swimming stamina (Horak and Klein 1967, Mitton and McDonald 1994), and physiological and behavioral abnormalities (Mitton and McDonald 1994, Schreck et al. 1976, Mesa and Schreck 1989). The incidence and severity of collateral risks are typically related to the type of electrical output used (alternating current (AC), continuous direct current (DC), pulsed DC) as well as the voltage (V), frequency (Hz), and pulse width (ms). Use of continuous or low frequency pulsed DC is thought to present the least risk of harm (Whaley et al. 1978, McMichael 1993, Sharber et al. 1994).

Electrofishing has generally been considered inappropriate for AIS management in Pacific island streams because of the dual challenges of complex substrates and the sensitivity of at-risk native species. Plunge pools, boulders, and other complex habitat characteristics common in oceanic island streams, including Hawaiʻi, can make electrofishing less effective than when used in continental streams (Baker and Foster 1992). However, studies conducted across Hawaiʻi (Maciolek and Timbol 1980), Puerto Rico (Kwak et al. 2007), and Indonesia (J. Derek Hogan, unpublished data) have demonstrated that electrofishing can be highly effective for capturing fish in oceanic island streams. Nonetheless, physiological sensitivities of native species make them particularly susceptible to collateral injury and mortality at instrument settings typically used in continental streams (Snyder 2003). Many of the native species in Hawaiian streams, for example, are more sensitive to electrofishing than co-occurring AIS (Maciolek and Timbol 1980). Although fish and other fauna are generally less sensitive to electrofishing at smaller and larger size classes, this is not necessarily true of native Hawaiian species. For example, the sleeper goby *Eleotris sandwicensis* is considerably more sensitive than other native species and becomes more susceptible to injury at larger sizes, as larger fish experience greater risk of injury and mortality due to adverse effects of muscle twitch induced by pulsed current, particularly at higher voltages.

Instrument settings like voltage are typically determined according to water and tissue conductivity, which can govern capture efficacy. Thus abiotic conditions like conductivity should always be tested and checked against recommended baseline settings to better ensure optimal instrument efficacy. Smith-Root electrofishers, for example, can provide sufficient current at realistic power levels in potable water sources across the United States ranging in conductivity from 20 to 2000 µS/cm. Tissue conductivity and temperature of target species also must be considered when using electrofishing instruments. For example, carp conductivity varies from 372 µS/cm at 5°C to 1969 µS/cm at 25°C, which is similar to the conductivity of other common continental stream fish (e.g., trout, perch, carp, gudgeon) that ranges from 814 µS/cm to 1220 µS/cm. Species-specific conductivity has not been carefully determined for most of the fauna native to Pacific island streams, including Hawaiian fishes, although species generally expected to have higher conductivity, such as *E. sandwicensis*, appear to be more vulnerable to electrical current.

Adverse complications of electrofishing can be addressed by taking precautionary steps, including pre-electrofishing sequestration and post-electrofishing restocking of native species (Nico and Walsh 2011). Potential exposure of native species to electrical current can be greatly reduced by sequestering and restocking native species. Before electrofishing begins, efforts can be made to carefully capture all native fishes and invertebrates in target reaches by exhaustive hand-netting. Captured individuals can then be sequestered streamside at low densities in shaded containers filled with aerated stream water and monitored for stress during electrofishing passes. After electrofishing is completed, native species can be allowed to recover from possible stress from handling and then returned to their location of capture.

As a further precaution, standard operating procedures for electrofishing can be modified to reflect the hydrology and geochemistry (e.g., conductivity) of study streams and the sensitivity of native species (Holliman et al. 2003). Modification of instrument settings like amperage and waveform combinations can be coordinated to reduce the likelihood of injuries and mortality of any remaining native species, particularly during the first electrofishing pass (i.e., when native species are most likely to encountered). Procedures developed for warm-water streams on Puerto Rico (Kwak et al. 2007) and sensitivities of

Hawaiian species documented by the Hawaiʻi Cooperative Fishery Research Unit (Maciolek and Timbol 1980) indicate that use of continuous current presents the least risk for injury relative to other setting options. Combined with low voltage, use of unpulsed DC current reduces the likelihood of twitch reactions, the most common cause of spinal injury, particularly for larger native fishes that are at a higher risk of spinal injury from muscle twitches and spasms. Thus, it is recommended that unpulsed DC current be used during initial passes with voltage set to *ca*. 100 V, although adjustments (e.g., to 150 V or 200 V) might be necessary depending on abiotic conditions. Notably, capture rates are typically lower at these settings, and thus it is likely that many of the less sensitive AIS species as well as smaller fish will not be caught during an initial pass. Settings can be adjusted to target the capture of smaller and otherwise less sensitive AIS species during second and third passes, when risk to native species is significantly reduced. For later passes, pulsed current might be used, with the frequency set at 60 Hz, duty cycle set at 12%, and voltage set within a range of 150 V to 275 V. Voltage used during third passes might be held constant or increased slightly depending on the effectiveness of the second pass. Elevated voltage during third passes can potential improve capture of smaller fish missed in the first two passes, but performing a third pass is not always necessary, as low densities and high efficacy during earlier passes can preclude the need for further electrofishing.

4.6 STUDY SYSTEM

The research described herein was conducted in the Hawaiian Islands, a remote archipelago in the Pacific positioned 3,860 km from the nearest continent. The archipelago has long served as a model for understanding oceanic island geomorphic development (Carson and Clague 1995), ecosystem functioning (Vitousek 2004), island biogeography (e.g., Gruner 2007), and aquatic species conservation (e.g., Brasher 2003). As a result, Hawaiian streams have been studied more intensively than streams on other oceanic islands (Resh and de Szalay 1995, Brasher 2003, Brasher et al. 2006, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). Assessments focused on watersheds across Oʻahu to ensure direct relevance to DoD watershed management practices, building on prior SERDP-funded studies that examined biotic and abiotic conditions across the island in comparison to watersheds elsewhere in the archipelago (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018, Hain et al. 2019).

Some geomorphological features of Hawaiian streams favor implementation of AIS control. Like watersheds on islands elsewhere in the Pacific, most drainage basins across the Hawaiian Islands are small, narrow, and steep by continental standards (Kinzie 1988; Brasher 2003; Covich et al. 2003; Oki and Brasher 2003; Smith et al. 2003). Consequently, basins are often drained by a single mainstem stream (The Hawaiian Watershed Atlas; www.hawaiiwatershedatlas.com). Populations of introduced species also are often spatially restricted within a stream due to features that limit longitudinal spread (Brasher et al. 2006), such as insurmountable waterfalls, rapids, or other barriers that can only be overcome by native species (Kinzie, 1988; Blob et al. 2008, 2010; Moody et al. 2017). The absence of watershed connectivity also reduces the likelihood of recolonization following removal, as most AIS found in Hawaiian streams do not have the physiological capacity to survive exposure to marine conditions, and those that do (e.g., some Poeciliids; Chervinsky 1984) likely do not have the physiological response to cue towards freshwater and migrate upstream, as this is thought to be an evolutionary adaptation of amphidromous species derived from a marine ancestor (McDowall 2007). Moreover, some streams have terminal waterfalls that can function as barriers to re-invasion by AIS (e.g., tilapia) that can inhabit coastal waters.

We focused on interactions between non-native fishes and native amphidromous fishes because of the severe asymmetries in fish assemblage structure on Oʻahu (Brasher et al. 2006, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). The native fish assemblage of Hawaiian streams, including those on Oʻahu, is unusually depauperate, even in comparison to other Pacific island archipelagos (McDowall 2003a). The assemblage consists of *A. stamineus* (formerly known as *A. guamensis*; Lindstrom et al. 2012), *Lentipes concolor*, *E. sandwicensis*, *Sicyopterus stimpsoni*, and *Stenogobius hawaiiensis* (McDowall 2003a). All are amphidromous members of the goby family (Gobiidae) and all are endemic to the Hawaiian Islands, though *A. stamineus* may occur elsewhere in the Pacific (Hoese and Gill 1993; McDowell 2007; Lindstrom et al. 2012; Hogan et al. 2014). As important ecological and cultural resources, all are protected by the Hawaiʻi DAR, though none are listed as threatened or endangered under the Endangered Species Act (Devick et al. 1995; Brasher 2003; Walter et al. 2012). Given their status as species of special concern, the presence and relative abundance of native fishes are currently used as key indicators of stream integrity for the purposes of watershed management (Brasher 2003, Blum et al. 2014). Recent stream surveys have revealed that populations of all native amphidromous species are depressed across Oʻahu, despite considerable variation in land use and stream habitat quality (Blum et al. 2014; Moody et al. 2017; Lisi et al. 2018; Hain et al. 2019). The surveys also indicate that three (*L. concolor, S. stimpsoni, S. hawaiiensis*) of the five native migratory fishes are now rare on Oʻahu (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). Additionally, the surveys showed that the highest densities of AIS (including non-native fishes) occur in streams on Oʻahu, including streams that cross DoD installations. AIS densities can be an order of magnitude higher than native fish densities in streams on Oʻahu (Eldredge 2000, Yamamoto and Tagawa 2000, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018).

We also focused on drift-feeding Poeciliid live-bearers (i.e., guppies, mollies and swordtails) and algae-eating Loricariid armored catfish because they are the two most abundant non-native fishes on Oʻahu (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018), and because they impact native fishes through different ecological pathways. Poeciliids can affect native goby fishes via competition for food resources and habitat (Holitzki et al. 2013, El-Sabaawi et al. 2016), and parasitism (Font 2003, Gagne et al. 2015, 2016, 2018). Surveys indicate that nitrogen concentrations are higher, benthic invertebrate communities differ, and native fish abundances are lower in streams where Poeciliids are present (Holitzki et al. 2013). Surveys and mark-recapture studies also indicate that Poeciliids (and perhaps other AIS) constrain native fish densities by limiting recruitment of juveniles, likely through a combination of predation and competition (Bell 1994, Blum et al. 2014, Holitski et al 2013, Hain et al. 2019). Armored catfish, on the other hand, compete with native species for basal algal food resources and can restrict the availability of phosphorus and other essential nutrients (Kido et al. 1993, Kido 1996, Capps and Flecker 2013).

4.7 ASSESSING THE INFLUENCE OF AIS AND RESPONSES TO AIS REMOVAL

AIS can exert influence on a range of conditions and processes in oceanic island stream ecosystems, thus the effects of AIS removal might manifest in several ways. For example, because Poeciliids prey on native fish larvae and compete with native fish juveniles and adults for food resources (Holitski et al. 2013, Hain et al. 2019), removal of Poeciliids might therefore result in a downward shift in the population size structure of native fish like *A. stamineus*, shifts in the abundance and diversity of invertebrate prey, or isotopic shifts in native fish tissue indicating greater consumption of basal food resources. Because armored catfishes feed on algae, sequester nutrients (e.g. phosphorus), and erode stream banks, removal of Loricariids might increase water clarity (i.e., a reduction in total suspended solids (TSS)), increase stream substrate heterogeneity (e.g., a reduction in sediment deposition), increase stream bank stability or vegetation, and possibility lead to shifts in the abundance and diversity of benthic diatoms or isotopic shifts in native fish tissue indicating greater consumption of invertebrate prey food resources. Shifts might also occur in indicators of whole stream metabolism like rates of decomposition (i.e., of allochthonous organic material like leaf litter). Notably, a diverse range of cascading effects may differ stream to stream because removal efficacy might vary due to differences in pre-removal invasive densities and other in-stream characteristics. Accordingly, we characterized genes-to-ecosystem variation in relation to AIS prevalence and AIS removal with reference to surface flow conditions.

Assessments of stream community structure and composition according to AIS removal.
It is well understood that AIS can directly or indirectly influence stream community structure and composition through negative (e.g,. competition, predation, parasitism; Holitzski et al. 2013, Gagne et al. 2015, Gagne et al. 2016) and also possibly through positive (e.g., facilitation) interactions. Accordingly, we assessed a suite of metrics to understand community-level outcomes of AIS removal, including measures of species richness, relative abundance and community composition. This not only afforded perspective on the sensitivity and response capacity of at-risk native species to AIS removal, it also offered insight into recolonization, community (re)assembly, and wholesale shifts that can arise as a result of AIS removal. As populations of all native amphidromous species are depressed across Oʻahu, it was expected that AIS removals would not elicit much change in the richness of native species communities. Likewise, it was expected that AIS removals likely would not elicit much change in overall population densities of native species. Rather, responses to AIS removal might manifest in other community and associated demographic measures.

Assessments of native species demography according to AIS removal.
Aquatic species introductions can change ecological interactions such as predation and competition which can alter native species population demographics. For example, predation by introduced Pacific lionfish (*Pterios volitans/miles*) has resulted in reductions in abundance (Albins 2015) and recruitment (Albins & Hixon 2008) of native Caribbean fishes which are naïve to the novel predator (Anton et al. 2016). Competition for food with invasive round goby (*Apollonia melanostomus*) resulted in depressed body masses of native scuplin (*Cottus cognatus*) and logperch (*Percina caprodes*) in experimental feeding trials (Bergstrom & Mensinger 2009). Competition for shelters by invasive gobies resulted in displacement of native fishes from their preferred habitats (Van Kessel et al. 2011).

Invasive species management is motivated by the desire to reduce impacts on native species and to effect ecological recovery of native populations. The removal of invasive predators and competitors can restore ecological interactions to a precursor (i.e., natural) state, which has been shown to lead to demographic recovery of some native species. For example, the eradication of invasive topmouth gudgeon (*Pseudorasbora parva*) from a small lake in the United Kingdom resulted in greater abundance and growth rates of native fishes (Britton et al. 2009). Experimental eradication of invasive lionfish from small reefs in Panama resulted in a significant increase in recruitment of native reef fishes compared to control sites with lionfish (Palmer et al. 2017). In some cases, total eradication is not required, rather suppression of the invader is enough to trigger significant demographic recovery, as has been observed with lionfish control in the Bahamas (Green et al. 2014).

A recent review (Prior et al. 2018) of responses to invasive species removal found that positive outcomes were only documented in 51% of case studies. Ecological recovery was much more likely in animal populations, and animal communities recovered more often when managed areas were more connected (i.e., less isolated) with surrounding habitats (Prior et al. 2018). This prompted the hypothesis that small, simple native communities that are characterized by strong trophic links, like those on islands (Jones et al. 2016) and in aquatic ecosystems (Gallardo et al. 2016), may be more likely to recover after invasive species removal. While the recent review of case studies found no differences according to ecosystem (Prior et al. 2018), comparisons did not include studies of insular freshwater systems, particularly small streams ecosystems like those on oceanic islands that are characterized by depauperate communities and strong trophic linkages (e.g., Coat et al. 2009).

Given the high densities of AIS on Oʻahu, the simple community of native species present there, and the expectation of strong trophic linkages in these small, simple systems, we expected that AIS removal could restore species interactions and trigger demographic recovery. We hypothesized that the alleviation of predation pressure from Poeciliids and competition from Loricariids would elicit an increase in abundance and recruitment of juveniles, as well as elevate the growth rate and condition of native fishes. As removal of competitors is also expected to increase habitat availability, we also hypothesized that native fish would move into remediated areas following AIS removal.

To test these hypotheses, we conducted a multiyear, individual-based mark-recapture study of *A. stamineus* responses to AIS removal (no individuals were sacrificed to complete this work). The mark-recapture data was used, for example, to derive site and watershed-specific estimates of individual adult growth rate, body condition, reproductive status, and approximate age distributions, as well as apparent survival and recruitment over time in addition to population size and density.

Assessments of life history variation in relation to AIS, nutrients, and hydrology.

The conditions experienced during early life-history stages can determine the number of surviving individuals that recruit to adult populations (Hjort 1914, Hjort 1926, Cushing 1974, Houde 2008). Mortality rates that affect the level of recruitment may be altered by the quality of local habitat conditions (McDowall 2007a) including conditions determined by environmental factors. Habitat quality has been shown to influence the dominant recruitment type (migrant or residents) in partial migrant species (Taylor and Norris 2007), and environmental factors can influence the representation of either contingent in some diadromous fishes (Huey et al. 2014, Watanabe et al. 2014). For example, in Australia, streams with steeper gradients and higher flow rates harbored populations with more migrants than slower, sluggish streams (Huey et al. 2014) presumably because high flow rates promote the migration of larvae after hatching. Large-scale climate drivers like changes in rainfall and groundwater availability can interact with and modify local environmental conditions (Mosley 2000, Cole et al. 2002) that affect the survival and recruitment rates of contingents (Barber and Chavez 1983), particularly in riverine environments (Walter et al. 2012). For instance, more migrants of white perch, *Morone americana*, were found in populations when stream flow was high, and no migrants were found during drought years (Kerr and Secor 2010). This may have been the result of increased nutrient delivery and improved environmental conditions or a larger larval cohort originating during a period of high surface flow conditions (i.e., more spawning may have occurred during high flow conditions; Kerr and Secor 2010).

Excess nutrient delivery and addition (i.e., from agricultural activities, sewage treatment, and commercial land-based practices) can similarly affect water and habitat quality (Cole et al. 2004, Fisher et al. 1992). Though phosphorus is typically limited in freshwater systems, it is important for maintaining balanced water quality and promoting primary production in lotic ecosystems (Elwood et al. 1981; Elser et al. 1990). Excess orthophosphate (reactive phosphate) can alter the natural balance of nutrient availability in freshwater systems, which can sometimes lead to an overproduction of phytoplankton, plants and algae, and other biota (Fisher et al. 1992). This can cause eutrophication and decreased dissolved oxygen that can exert additional stress and mortality on organisms that reside in those habitats (Mallin et al. 2006; Bilotta and Brazier 2008). The same can happen with nitrate addition. By changing the base of food webs, nutrient inputs can also alter higher trophic interactions (Vitousek et al.

1997; Woodward and Hildrew 2002), which can modify the pressures acting on early life-history stages of migratory fishes, including those arising from AIS.

Introductions of invasive species can further alter trophic dynamics and interspecific interactions with native species, including native species that are of conservation concern (Brasher 2003, Lisi et al. 2018). Hawaiian stream systems have proven to be especially vulnerable to biological invasions, in part because the communities are naturally depauperate (and thus present vacant niche spaces) and feature highly specialized native species, shorter food webs, and lower functional redundancy (Vitousek 1988, D'Antonio and Dudley 1995, O'Dowd et al. 2003). The distributions and prevalence of AIS in Hawaiʻi have also been shaped by human densities as well as urban and agricultural land use (Brasher 2003, Walter et al. 2012, Moody et al. 2017, Lisi et al. 2018). By increasing predation, competition, and parasitism (Jenkins et al. 2010, Morrison 2016, Gagne et al. 2015, Gagne and Blum 2016), AIS have contributed to the decline in native species across the Hawaiian archipelago (Holitzki et al. 2013, Lisi et al. 2018, Hain et al. 2019). Alteration of basal food webs and physical habitat likely further destabilizes the resiliency and persistence of native fish populations (Smith et al. 1999, Lundberg and Moberg 2003, Brasher et al. 2003, Holitski et al. 2013, Blum et al. 2014, Lisi et al. 2018).

Native fishes in Hawaiian streams are exposed to a portfolio of stressors arising from shifts in hydrology, land use, and interspecific interactions. Watersheds span gradients in rainfall, urbanization, and AIS introductions within and among islands (Brasher 2003, Blum et al. 2014, Moody et al. 2017). Additionally, anthropogenic modification of surface flows can overlay natural hydrological gradients, as most streams have managed surface flows for flood prevention or water abstraction (Smith et al. 1999, Brasher 2003, Gopalakrishnan 2005). Dewatering can reduce base flows and promote subsidence of stream banks (Brasher 2003), which can further reduce habitat quality and quantity. Similarly, intensifying watershed land use has been accompanied by physical and chemical changes to stream conditions, which can result in eutrophication (Lisi et al. 2018).

Though it is well understood that the diversity and abundance of native Hawaiian migratory fishes are shaped by natural gradients and gradients of anthropogenic modification (Brasher 2003, Moody et al. 2017, Lisi et al. 2018), it is not known whether fishes also exhibit life-history responses. It is possible that natural and anthropogenic pressures influence the proportions of migrants and residents in populations of species with facultative life histories. All five of the endemic Hawaiian stream fishes are amphidromous (McDowall 2007b; McDowall 2003b). Four of the five species exhibit partial migration (i.e., facultative amphidromy), where there are both migrant and resident forms within adult populations (Hogan et al. 2014, Heim-Ballew et al. 2019). It has been hypothesized that a flexible migratory life-history may help buffer against ecosystem alteration (Lisi et al. 2018), particularly for the most abundant native freshwater fish in Hawaiʻi, *Awaous stamineus* (Eydoux and Souleyet 1850), which is relatively insensitive to anthropogenic pressures (Lisi et al. 2018).

We have analyzed life-history variation from the microchemistry of post-larval otoliths to determine whether the ratio of residents to migrants in *A. stamineus* populations varies across environmental and AIS density gradients. Focusing on our 13 study watersheds, the goal of the study was to examine how human alteration to Hawaiian streams influences differential survival of residents. We hypothesized that the proportion of residents would be lower in streams with (1) higher AIS densities due to increased competition and predation; and (2) lower mean discharge; and (3) higher discharge variation due to greater probability of desiccation, lowered habitat quantity and quality, and reduced access to nutrition sources. Consistent with these hypotheses, we expected that more pristine streams, indicated by low nutrient concentrations and low AIS densities, would have a higher proportion of residents since less stressful in-stream conditions would be more favorable for survival of resident larvae.

Assessments of genomic and life history variation according to AIS removal.
Habitat alteration is a primary driver of population size reduction and restricted dispersal in natural populations (Haddad et al. 2015). Over time, reduced connectivity can reduce genetic diversity via genetic drift and inbreeding, contribute to inbreeding depression, and undermine the capacity of populations to adapt to disturbances such as invasive species, pollutants, and climate change (Allendorf et al. 2013, Frankham et al. 2017). Thus interventions that increase or restore levels of population connectivity can in turn increase population viability and reduce the risk of local extirpation or extinction (Seddon et al. 2014, Haddad et al. 2015).

Management interventions can achieve greater population connectivity and viability through two synergistic mechanisms. The first mechanism is demographic rescue, which occurs when an influx of individuals increases local abundance, effectively buffering against demographic stochasticity and related phenomena like Allee effects (Brown and Kodric-Brown 1977, Hufbauer et al. 2015). The second mechanism is genetic rescue, which occurs when there is an increase in population fitness (i.e., growth) owing to an influx of new alleles (Whiteley et al. 2015). Genetic rescue can result in a positive and lasting impact on populations by increasing genetic diversity, effectively reversing consequences of isolation (Allendorf et al. 2013, Frankham 2016, Frankham et al. 2017). Demographic rescue and genetic rescue can occur in concert, as an eco-evolutionary feedback, or they can be decoupled from each other, with potentially varying degrees of contribution to population persistence. Concurrence or decoupling of demographic and genetic rescue can depend on life history, including migratory life history variation.

Variation in migratory life history is widely documented in natural populations and can manifest as a plastic response to prevailing environmental conditions, underlying heritable variation, or an interaction thereof (Dodson et al. 2013). Environmental conditions like resource availability (Olsson et al. 2006), conspecific or heterospecific density-dependence (Marco-Ruis et al. 2013), and predation risk (Chapman et al. 2011b) can influence whether individuals or populations tend to migrate or remain resident (Chapman et al. 2011a, Chapman et al. 2012b). Migration behavior can also be heritable (Liedvogel et al. 2011, Hale et al. 2013), where alternative life histories reflect distinct evolutionary strategies (Jonsson and Jonsson 1993, Karlsen et al. 2013). Both single gene effects and whole chromosomal inversions have been shown, for example, to underlie migratory ecotypes in salmonids (Lemopoulos et al. 2018). Environmental and genetic components influencing migration may interact to give rise to conditional strategies, where there is a heritable threshold to migration (e.g., size or body condition) and where environmental conditions exert selection pressure on the threshold trait itself and concurrently affect the likelihood of an organism surpassing that threshold (Pulido 2011). For example, larger and faster growing individuals of rainbow trout (*Oncorhynchus mykiss*) are more likely to undergo anadromous migration (i.e., become steelhead). However, this size threshold varies across populations based on rearing conditions (Satterthwaite et al. 2010, Satterthwaite et al. 2012, Phillis et al. 2016) even though there is underlying heritable genetic architecture for size in *O. mykiss* (Carlson and Seamons 2008, Hecht et al. 2015). Migratory flexibility and corresponding genetic divergence can be cryptic, as migratory ecotypes do not always exhibit diagnostic phenotypic traits. This is well illustrated by recent genomic studies of Atlantic cod that have detected highly differentiated yet cryptic migratory ecotypes occurring in sympatry across a broad geography (Thorsteinsson et al. 2012, Barney et al. 2017, Barth et al. 2017, Sodeland et al. 2016), which serves to illustrate the value of coordinated analysis of genomic and life history variation to prevent misinterpretation or misattribution.

In theory genetic rescue is a particularly desirable management outcome because it can induce secondary beneficial demographic responses, like higher population growth rates and size (Brown and Kodric-Brown 1977, Tallmon et al. 2004), that can increase the likelihood of evolutionary rescue through adaptation to prevailing conditions. In practice, however, genetic rescue is not often set as a conservation or management goal (Mills and Allendorf 1996, Edmands 2007, Frankham et al. 2011, Whitely et al. 2015, Bell et al. 2019), even though the likelihood, relative magnitude and outcomes of genetic and demographic rescue remain relatively poorly studied in natural populations. It is unclear, for example, whether and how either form of rescue occurs in species with a facultative migratory life history. Misdiagnosis runs the risk of leading management efforts astray, potentially resulting in more harm than good.

Determining the relative importance of demographic and genetic rescue could promote more effective conservation and management, but it can be challenging to identify and disentangle one from the other. In facultatively migratory species like Atlantic cod, it is possible that an influx of migrants may cryptically replace resident ecotypes, elevating genetic variation (Willi 2005, Fauvergue et al. 2012) with little to no evidence of demographic change. On the other hand, elevated recruitment of cryptic resident types might foster demographic change with little to no evident change in genetic variation. Elevated recruitment of cryptic residents might nonetheless elicit genetic rescue by reinforcing local adaptation (Pickup et al. 2013). The likelihood and magnitude of each form of rescue can also be context dependent, and thus be highly heterogeneous among populations. For example, the magnitude of demographic rescue can depend on population size, potentially being more important in smaller populations (Lande 1988, Willi 2005). Likewise, genetic rescue may be more effective in reducing extinction risk in smaller populations, perhaps more so that demographic rescue (Hufbauer et al. 2015).

Field-scale manipulations can be a valuable approach for studying genetic and demographic rescue. Habitat restoration, invasive species removal, transplantation, and assisted migration can potentially elicit either or both forms of rescue. Invasive species removal is a particularly tractable experimental approach, especially in aquatic ecosystems with great potential for demographic recovery and high gene flow, like streams on Oʻahu that harbor high densities of AIS and low densities of native migratory fauna (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). Alleviating direct and indirect pressures imposed by AIS on native amphidromous fishes and invertebrates could, for example, promote genetic rescue by increasing immigration via restoring migratory pathways and reducing predation intensity on larvae and post-larvae passing through migratory gauntlets (Hain et al. 2019). AIS removal might also promote demographic rescue of species with migratory flexibility by elevating the survival and reproductive success of resident types (e.g., due to greater availability of limiting resources or nutrients).

Identifying and differentiating outcomes of AIS removal requires longitudinal monitoring of local populations (i.e., before and after interventions), with consideration given to conditions that govern contextual variation. Building on demographic studies (described above and below), we have examined genomic and life history variation in the facultatively amphidromous goby *A. stamineus* (Hogan et al. 2014, Heim-Ballew 2019, Heim-Ballew et al. 2020) to determine whether AIS removal fosters genetic rescue, demographic rescue, or some combination thereof. Evidence that genetic variation in *A. stamineus* is inversely related to AIS densities in streams across the Hawaiian archipelago (Blum et al. 2014), suggests that AIS removals will elicit a gain in genomic diversity. The magnitude of change might be mediated by life history, however, if genomic diversity differs between residents and migrants. This might occur because migrant recruits are drawn from admixed regional pools (Alda et al. 2016) or because of genetic differentiation among migratory types (i.e., like Atlantic cod), despite the potential for broader environmental influences on migration behavior. Evidence that the proportion of resident types declines with increasing AIS densities across the study watersheds also suggests that AIS removals should elevate the proportion of residents in local populations. Consequently, there might be little observable gain in genomic variation, even if AIS removals elicit greater recruitment (Holitski et al. 2013, Hain et al. 2019).

We evaluated these hypotheses by first quantifying genomic variation according to ddRAD-generated single nucleotide polymorphisms (SNPs) for both resident and migratory *A. stamineus* across 13 watersheds on Oʻahu where we conducted experimental AIS removals. We also assayed the proportional representation of resident and migratory *A. stamineus* in local populations before and after AIS removals to draw further inferences about the processes underlying genomic and demographic outcomes of AIS removals.

Assessment of ecosystem outcomes of AIS removal.

It remains unclear whether introduced species alter the abundance, biodiversity, and functioning of native communities by modifying ecosystem properties or processes (Elton 1958; Dudgeon et al. 2006; Gallardo et al. 2016). High densities of aquatic invasive species (AIS) are often associated with changes in turbidity and nutrient cycling, as well as shifts in food web structure and species interactions (Cucherousset and Olden 2011). Typically, heavily invaded ecosystems are also subject to habitat degradation or other forms of anthropogenic disturbance, leading to uncertainty about whether AIS directly cause ecosystem changes or instead are taking advantage of opportunities provided by prior degradation of physical and chemical conditions (Walsh et al. 2005; Dudgeon et al. 2006; Booth et al. 2016). Accordingly, determining whether AIS represent a cause or consequence of ecosystem alteration can improve stream restoration efforts intended to benefit native species.

AIS are often targeted for extermination under the presumption that it is beneficial to native species of concern. Examples include introduction of biocontrol agents to reduce Eurasian watermilfoil (*Myriophyllum spicatum*, Sheldon and Creed 1995); poisoning of sea lamprey larvae (*Petromyzon marinus*, Lawrie 1970) in Great Lakes tributaries; mobilizing anglers in the Caribbean to harvest lionfish (*Pteroinae sp*., Green et al. 2017); intensive trapping to remove rusty crayfish from lakes (*Faxonius rusticus*, Hein et al. 2007); and thermal manipulations to remove invasive rainbow smelt (*Osmerus mordax*, Gaeta et al. 2014). The success of such attempts is typically judged based on the degree to which the visibility of the target species is reduced—and for how long— because it is nearly impossible to fully eradicate AIS once established (Vander Zanden et al. 2010). The scale of local removals reflects a balance between logistical constraints versus aspiring to achieve widespread, lasting reductions in AIS densities.

A key justification for undertaking laborious and expensive AIS removals is to restore ecosystem processes to a pre-invasion state. For instance, common carp (*Cyprinus carpio*) facilitate the release of sediment-bound phosphorus through bioturbation, leading to reduced water clarity (Cahn 1929; Schrage and Downing 2004; Lathrop 2007). It has been shown that removing common carp can dramatically increase water clarity in both lakes and rivers, thereby helping to meet water quality goals (Beklioglu et al. 2003; Lathrop et al. 2013). However, ecosystem responses to carp removals may be more limited in watersheds where anthropogenic loading ensures abundant nutrients regardless of carp numbers (Schrage and Downing 2004). This suggests that ecosystem goals should be set according to assessments of AIS suppression conducted when other problematic stressors remain unchanged (Neeson et al. 2016).

Oceanic island streams can be excellent natural laboratories for assessing potential ecosystem responses to AIS removals because they often burden with high AIS densities and profound alteration of habitat conditions and water chemistry. Because oceanic islands streams are naturally depauperate (i.e., species-poor), introductions of AIS can add new functional groups that come to dominate faunal communities, which increases the likelihood that AIS alter ecosystem attributes and processes in oceanic island streams. For instance, algae-eating Loricariid armored catfish have been widely introduced in tropical and sub-tropical streams, including streams across the Hawaiian Islands, where they compete with native grazers for food and may affect nutrient availability by excreting nitrogen while also sequestering phosphorus to build dermal armor plates (Holitzki et al. 2013; Capps and Flecker 2013; Capps et al. 2015). In addition, Loricariid catfish physically disturb stream substrate, leading to increased turbidity, microbial activity, and organic decomposition rates (Capps et al. 2015). Live-bearing Poeciliid fishes— including guppies (*Poecilia reticulata*), swordtails (*Xiphophorus maculatus*), mollies (*Poecilia sp.*), and mosquitofish (*Gambusia affinis*)— are also popular aquarium fishes whose omnivorous diets (Gido and Franssen 2007) allow them to act as both predators on native species and competitors for benthic resources (Holitski et al. 2013, Hain et al. 2019). This raises the possibility that removal of dominant AIS like Loricariids and Poeciliids (Lisi et al. 2018) might not only alleviate direct pressures on native species, but also indirectly result in ecosystem conditions that are more favorable for native species of concern.

AIS removals alone might not be enough to restore ecosystem functioning if oceanic island streams suffer from other impairments or exhibit hydro-geomorphological attributes that can hinder recovery. Heavily invaded streams tend to suffer from a syndrome of watershed alteration and chemical pollution (Lisi et al. 2018). For instance, nutrient inputs from urban sewage overflows (Kirs et al. 2017, Lisi et al. 2018) and leaky septic systems (Whittier and El-Kadi 2014), which often co-vary with AIS abundance (Moody et al. 2017, Lisi et al. 2018), can potentially limit ecosystem responses to AIS removals (Schrage and Downing 2004). High-frequency disturbance regimes that often prevail in oceanic island streams can also mediate and mask responses to AIS removal (Walter et al. 2012). Non-seasonal flashy flow regimes (Lau and Mink 2006) can, for example, “reset” the benthos by scouring the streambed, potentially altering or masking expected outcomes of removing non-native grazers (Power et al. 1988, Poff and Ward 1989). Consequently, it might also be challenging to separate outcomes of AIS removals from contextual variation.

In this study, we assessed whether AIS removals modified ecosystem processes in 13 watersheds on the island of O’ahu, Hawai’i using a rigorous BACI approach. The depauperate freshwater fauna of Hawaiian streams features high endemicity of culturally significant species that are threatened by AIS (Brasher 2003, Walter et al. 2012, Holitski et al. 2013, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018, Hain et al. 2019). Accordingly, AIS removal is often advocated as a measure to protect imperiled native species in streams across the archipelago (Brasher 2003, O’Dowd et al. 2003, Walter et al. 2012, Moody et al. 2017, Lisi et al. 2018). Our aim was to promote evidence-based intervention strategies by assessing whether restoration of ecosystem functioning can be achieved solely through local suppression of non-native species. To do so, we first characterized pre-removal AIS assemblages, hydrology, nutrient availability, and ecosystem process rates in each watershed. We deployed nutrient and leaf litter bioassays to document baselines for carbon fixation and decomposition, and used whole-stream metabolism to estimate reach-scale ecosystem respiration and production (Odum 1956). After characterizing baseline conditions, we removed AIS from a treatment (i.e. ‘removal’) reach in each watershed, and then remeasured process rates in both removal and upstream control reaches. We expected to see a rapid shift in water chemistry (e.g., higher N:P ratio; Capps and Flecker 2013), increased algal responsiveness to nutrients, and higher primary production after depressing grazer densities, as well as decreases in ecosystem respiration, leaf breakdown, and suspended solids due to lack of detrital processing. However, we also expected that outcomes of AIS removal would vary according to contextual gradients in hydrology and nutrient availability across the study region.

Coupled ocean-watershed modeling of whole-watershed AIS removal.

Invasive species are a leading driver of biodiversity loss, yet most efforts to eradicate invaders are both expensive and unsuccessful (Mack et al. 2000, Simberloff 2009). Many of the most impactful invaders share traits that enhance colonization ability, competitive dominance, and release from predators (Blackburn et al. 2009, Ricciardi et al. 2013, Kleunen et al. 2015, McKnight et al. 2017), but shared life histories also can create vulnerabilities to management interventions. Many native species have evolved traits and life histories to withstand extreme events that characterize their natural habitats (e.g. McMahon 2002; McDowall 2010) that fortuitously also confer resilience to methods used for eradicating invasive species. This raises the possibility that life history differences might be leveraged to concurrently eliminate invasive species and restore populations of native species.

Implementing a proverbial ‘clean-slate’ restoration strategy represents a novel application of the widely-recognized ‘storage effect’ in demography (Chesson 1994). ‘Storage’ refers to life history stages or habitats that buffer a population against mortality elsewhere in the life cycle (Chesson 2000). In the context of controlling invasions, the goal is to identify a resilient life stage in native species that enables them (and them alone) to recolonize after local extermination of invaders—even when other life stages of the native population are intentionally or inadvertently depleted. ‘Stored’ recruits could be derived from a buried seed bank, long-distance propagule transport, or use of a distinct habitat that is unaffected by control measures. Generally, these facets of life history will have evolved to enable re-establishment after natural high-mortality events (e.g., fires, floods), hence they constitute eco-evolutionary adaptations that can enable recolonization after aggressive extermination measures (e.g., pesticides, controlled fires) are taken to eliminate invasive species.

Both invasive and native species are often structured as metapopulations, wherein a local increase in one population can have spillover benefits arising from propagule exchange across a network of connected populations (Shmida and Wilson 1985, Cottenie 2005). Following extermination of dominant invaders (i.e., ‘clean-slate’ removal), recolonization of a native species could easily convert a former metapopulation sink into a source. Regional spillover arises initially from boosting the supply of recruits from the restoration site to connected populations, then becomes amplified as growth of each recipient population enhances its export of propagules elsewhere across the metapopulation. Together, recolonization mediated by storage life history and regional spillover mediated by metapopulation connectivity offer the potential for successful elimination of invaders at one site to produce widespread benefits for native species (Fig. 1; general demonstration in Appendix B).

In this study, we integrated demographic data and contrasting dispersal potential of native and invasive fishes in Hawaiian streams to illustrate how a clean-slate approach could rescue rare native species from extinction. Using a general analytical model (Appendix B) and model simulations, we assessed the response of individual populations to complete removal of invaders and quantified the magnitude of subsequent spillover benefits to the metapopulation. Hawaiian streams are ideal candidates for clean-slate restoration; all native fishes and invertebrates have convergently evolved an amphidromous life history, wherein an oceanic larval stage enables dispersal among distant populations (McDowall 2010). Amphidromy is adaptive on islands because it allows metapopulation persistence as the presence and habitability of streams fluctuates due to volcanism, erosion, climatic shifts, and extreme weather (Schoenfuss et al. 2004, Alda et al. 2016). In contrast, invasive fishes rely entirely on humans for dispersal among watersheds and islands (Nico and Walsh 2011). Once established, invasive species almost invariably dominate stream communities and depress populations of native species (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018, Hain et al. 2019). By comparing local- and metapopulation-scale benefits of clean-slate restoration among 51 potential watersheds, we also elucidated which stream attributes govern archipelago-wide benefits of eradicating non-dispersing invaders from a single watershed.

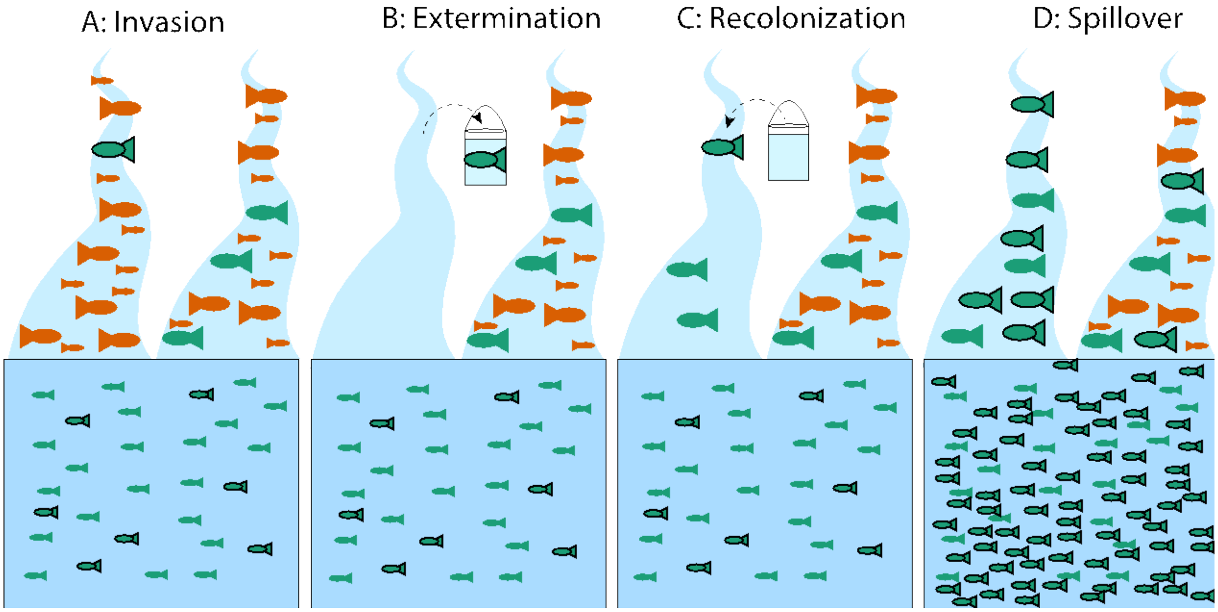


Figure 1. Conceptualization of a clean-slate restoration approach that leverages the amphidromous life history unique to native species (green) to enable recolonization of Hawaiian streams (light blue) by larvae dispersing through the ocean (dark blue) following extermination of invasive species (orange). The metapopulation is expected to subsequently grow via demographic spillover from restored sites. Native larvae (small) may recruit to any watershed. Dark outlines indicate fish originating in the focal watershed (left). Adults and larvae of invasive species are restricted to freshwater. A: Invaders variably depress the density of native species. B: Extermination of invasive species throughout the focal watershed (left) does not affect native larvae at sea, and native adults are captured prior to extermination to conserve any local adaptations (indicated by fish in bucket). C: Larvae from many sources recolonize the invasive-free watershed (storage life history), enabling rapid population growth. D: Burgeoning natives in the restored stream boost aggregate larval production, alleviating dispersal limitation and spilling over to enhance the entire metapopulation.

5 Materials and Methods

5.0 AIS REMOVAL FROM HAWAIIAN STREAMS

We evaluated the efficacy and outcomes of AIS removal using a before-after-control-impact (BACI) study design in 13 spatially compact watersheds that have a shared geological history on the island of Oʻahu (Fig. 2, Table 1). Located on the windward and leeward sides of the Koʻolau Range, the study watersheds were selected to capture the range of surface flow conditions (i.e., mean discharge, discharge variability; Fig. 2) as well as variation in the distribution and abundance of all AIS, but especially Poeciliid live-bearers and Loricariid armored catfish, on the island of Oʻahu (waterdata.usgs.gov/hi/nwis/rt, www.hawaiiwatershed atlas.com, Blum et al. 2014; Moody et al. 2017, Lisi et al. 2018). Notably, many Koʻolau watersheds have a strong military presence or have supported military activities in the recent past (e.g., Waikāne watershed was a Marine Corps training area).

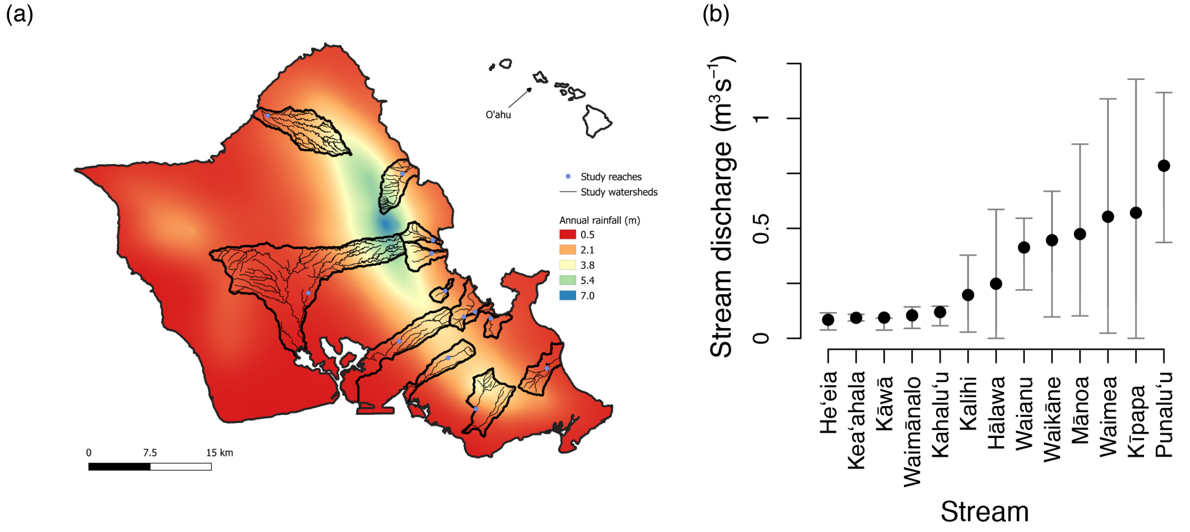


Figure 2. (A) Map of Oʻahu delineating our study watersheds and study reaches relative to precipitation conditions; (B) Study streams ordered by increasing median discharge (solid points, Q_{50}, $m^3\ s^{-1}$). Whiskers span low (Q_{90}) and high discharge (Q_{10}) derived from standard exceedance percentiles of daily discharge.

Daily stream discharge was characterized in each study watershed from May 2015 to March 2018 (Table 2). The United States Geological Survey (USGS) monitors continuous daily stream flow in 10 of the 13 study streams (Table 2). In Waianu stream, we estimate discharge based on a power relationship between direct measurements of discharge in Waianu and a USGS gage in the main stem of Waiāhole stream (USGS site no. 16294100, ~1.6 km downstream of measurements taken in Waianu stream). In streams without USGS gauging stations (Keaʻahala, Kāwā, and Kīpapa streams), water level was monitored hourly using HOBO U20 water level data loggers in the removal reach (Onset Computer Corporation, Bourne, MA, USA), corrected for atmospheric pressure monitored by Hobo U20L data logger in the adjacent riparian zone. Water level measurements were used to estimate discharge following standard methods based on a power function relationship between water level and direct measurements of discharge (Gore and Banning 2017). We characterized general hydrographic patterns in each stream by calculating the mean, coefficient of variation (CV), and the 95th, 90th, 50th, 10th, or 5th exceedance percentiles from mean daily discharge (Table 2). Standard exceedance percentiles, such as Q_{90}, can be interpreted as the mean daily discharge that is exceeded 90% of the time.

Field-based assessments of AIS removals began in May 2015 and ended in February 2018. For 12 watersheds, the study period ran from May 2015 to February 2018, whereas the study period for the remaining watershed (Punaluʻu) ran from October 2016 to February 2018 (Table 2). Following well established BACI experimental design precedents (reviewed by Schwarz (2015) and Rytwinski et al. watersheds (Fig. 2, Table 1), in each watershed we first conducted a 9-month assessment of baseline biotic and hydrological conditions in a 100 m 'removal' reach, as well as two 100 m 'control' reaches, one upstream and one downstream from the removal reach. A 25 m buffer zone separated control and removal reaches. Each set of buffer zones and reaches were located in the mainstem stream in the lower region of the study watersheds (Fig. 2, Table 1). After the 9-month baseline assessments, AIS removals were then conducted according to the protocol described below. Post-removal conditions were subsequently tracked in the removal and control reaches for a period of up to 2 years, except for the watershed with the offset study period (Punaluʻu) in which post-removal conditions were tracked for only 8 months. This approach not only allowed us to quantify and differentiate effects resulting from AIS removal relative to natural environmental and biotic variability (Osenberg et al. 1994; Schwarz 2015; Rytwinski et al. 2019), it also accounted for gradients in underlying conditions that might influence outcomes of AIS removals (Blum et al. 2014; Moody et al. 2017; Lisi et al. 2018).

Table 1. Locations of removal reaches in study watersheds across the Koʻolau Range on Oʻahu.

Stream (watershed)	**Latitude and longitude coordinates of removal reaches in each watershed**
N. Hālawa Stream (Hālawa)	(downstream terminus: 21.38349, -157.89941; upstream terminus: 21.38437, -157.89899)
Haiku Stream (Heʻeia)	(downstream terminus: 21.41009, -157.82266; upstream terminus: 21.40929, -157.82304)
Kahaluʻu Stream (Kahaluʻu)	(downstream terminus: 21.43867, -157.84447; upstream terminus: 21.43831, -157.84518)
Kalihi Stream (Kalihi)	(downstream terminus: 21.36485, -157.84166; upstream terminus: 21.36546, -157.84093)
Kāwā Stream (Kāwā)	(downstream terminus: 21.40808, -157.79108; upstream terminus: 21.40727, -157.79128)
Keeahala Stream (Keeahala)	(downstream terminus: 21.41558, -157.80965; upstream terminus: 21.41526, -157.8105)
Kīpapa Stream (Waikele)	(downstream terminus: 21.43744, -158.00613; upstream terminus: 21.43826, -158.00613)
Mānoa Stream (Ala Wai)	(downstream terminus: 21.30823, -157.80933; upstream terminus: 21.30903, -157.8093)
Punaluʻu Stream (Punaluʻu)	(downstream terminus: 21.56833, -157.89384; upstream terminus: 21.56747, -157.89398)
Waianu Stream (Waihole)	(downstream terminus: 21.48034, -157.86034; upstream terminus: 21.48016, -157.86118)
Waikāne Stream (Waikāne)	(downstream terminus: 21.49462, -157.85857; upstream terminus: 21.49510, -157.85899)
Waimānalo Stream (Waim.)	(downstream terminus: 21.35246, -157.72527; upstream terminus: 21.35188, -157.72603)
Waimea Stream (Waimea)	(downstream terminus: 21.63338, -157.05267; upstream terminus: 21.6332, -158.05157)

The "100 m stream reach" has long served as the standard unit of observation for estimating population densities of AIS and native stream fauna in Hawaiʻi (Higashi and Nishimoto 2007). With some notable exceptions (e.g., high waterfalls), it is generally sufficient to capture habitat variability in tropical and subtropical Pacific island streams, which can exhibit successive and repeated shifts from runs to pools to riffles within 100 m or less. It is also succinct enough to provide for replication within a watershed without incurring complications from confounding factors such as shifts in geomorphology. Upstream and downstream control reaches served as references for all of the response variables assessed for this study, with reference to the upstream control reach offering a basis for identifying and addressing the possibility that AIS removals influenced downstream conditions. Buffer zones served to increase independence of control and removal reaches.

AIS removals were conducted in 12 watersheds in March 2016 and in one watershed (Punaluʻu) in July 2017. Removals were carried out following a protocol that includes steps taken to reduce risk or injury to native species of concern. First, native amphidromous fishes and macroinvertebrates (e.g., *Macrobrachium* shrimp and *Neritina* snails) in the removal reaches were hand-netted, transferred to shaded streamside refuge tanks containing aerated stream water, and held in low density groups organized by capture location (demarcated with weighted flagging tape). Importantly, all *E. sandwicensis* were held in a parallel set of tanks to prevent predation on other species. AIS were then captured by three-pass electrofishing under Special Activity Permits (SAP 2016-14, SAP 2017-52) issued by the Hawaiʻi DAR. All native species captured during electrofishing passes were sequestered, checked for injury, and monitored in refuge tanks. All native species captured by hand-netting and electrofishing were counted, identified, assessed for body condition (length and mass), and photographed (Tulane University IACUC protocol #0463). All collateral harm (i.e., injury, mortality) was documented for all native species captured during electrofishing passes. All sequestered individuals were returned to their original capture location following completion of electrofishing passes.

Table 2. Daily discharge (Q) of each study stream summarized by the mean, coefficient of variation (CV: ratio of the standard deviation relative to the mean), and the 95th, 90th, 50th 10th or 5th exceedance percentiles of stream discharge over the time frame of interest. Stream discharge was monitored by USGS gages or with stationary HOBO U20 pressure loggers. Start and end dates define the temporal extent to which each stream was monitored and the analysis was conducted.

USGS site	Stream	Mean	CV	Q95	Q90	Q50	Q10	Q5	Start	End
16275000	Heʻeia	0.084	1.87	0.034	0.038	0.062	0.116	0.181	5/1/2015	3/31/2018
16226200	Hālawa	0.248	3.15	0	0	0.031	0.586	1.232	5/1/2015	3/31/2017
16283200	Kahaluʻu	0.119	1.22	0.038	0.057	0.099	0.146	0.245	5/1/2015	3/31/2018
16229000	Kalihi	0.197	2.20	0.023	0.028	0.084	0.378	0.674	5/1/2015	3/31/2018
HoboU20	Kāwā	0.094	2.56	0.035	0.037	0.045	0.091	0.300	11/13/2015	3/31/2017
HoboU20	Keaʻahala	0.093	0.13	0.075	0.079	0.091	0.109	0.117	5/8/2015	1/22/2018
HoboU20	Kīpapa	0.571	2.47	0	0	0.165	1.179	2.134	12/8/2015	3/31/2017
16241600	Mānoa	0.474	2.18	0.085	0.102	0.210	0.883	1.617	5/1/2015	3/31/2018
16301050	Punaluʻu	0.785	0.91	0.400	0.436	0.602	1.118	1.576	5/1/2015	3/31/2018
16294100	Waianu	0.413	1.06	0.200	0.220	0.344	0.546	0.712	5/1/2015	3/31/2017
16294900	Waikāne	0.446	1.82	0.082	0.097	0.306	0.669	1.135	5/1/2015	3/31/2018
16249000	Waimānalo	0.104	2.00	0.04	0.045	0.063	0.143	0.228	9/25/2015	3/31/2018
16330000	Waimea	0.553	4.29	0.012	0.023	0.151	1.089	1.750	5/1/2015	3/31/2018

The three-pass electrofishing protocol implemented for capturing AIS involved precautions intended to reduce the likelihood of non-target injuries and mortality of native species that were not hand-netted out of removal reaches. Instrument (Smith-Root LR-24) settings reflected protocols developed for streams on Puerto Rico (Kwak et al. 2007), sensitivities of Hawaiian species to electrical current as documented by the Hawaiʻi Cooperative Fishery Research Unit (Maciolek and Timbol 1980), and the hydrology and geochemistry of Hawaiian streams (e.g., discharge, depth, conductivity). It was expected that most native species remaining in the removal reach would be encountered and captured during the first electrofishing pass. Thus we always used unpulsed DC for the first electrofishing pass, as unpulsed DC and low frequency pulse current are thought to present the least risk of harm to sensitive species

(Whaley et al. 1978; Maciolek and Timbol 1980; McMichael 1993; Sharber et al. 1994; Dalbey et al. 1996). Voltage was generally set at 100 V during the first pass, though it was sometimes set at 150 V or 200 V depending on conductivity conditions (Table 3). Settings in the second and third passes targeted the capture of less sensitive AIS. For both passes, we used pulsed current with the frequency set to 60 Hz, although a setting of 30 Hz was used in some instances (Table 3). Duty cycle was set at 12%, with rare exceptions when it was set between 30% and 50%. For the second pass, voltage was generally set to 200 V, but ranged from 150 V to 275 V depending on conductivity. Voltage was unchanged or raised by 25-50 V for the third pass, depending on the outcome of the second pass (Table 3). Counts, species identification, and body size (length and mass) were recorded for all captured AIS within 24 hours after euthanasia with MS-222 following Tulane University IACUC protocol #0463.

Table 3. Instrument settings used for each stream and electrofishing pass. DC = direct current, SP = standard pulse.

Stream		Pass 1	Pass 2	Pass 3
He‘eia	Waveform	DC	SP	SP
	Voltage (V)	100	200	225
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	12%	12%
Hālawa	Waveform	DC	SP	SP
	Voltage (V)	100	200	275
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	12%	12%
Kahaluʻu	Waveform	DC	SP	
	Voltage (V)	100	200	
	Frequency (Hz)	N/A	60	
	Duty Cycle	N/A	12%	
Kalihi	Waveform	DC	DC	SP
	Voltage (V)	100	200	200
	Frequency (Hz)	N/A	N/A	60
	Duty Cycle	N/A	N/A	12%
Kāwā	Waveform	DC	SP	SP
	Voltage (V)	100	175-200	200
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	12%	12%
Keaʻahala	Waveform	DC	DC	SP
	Voltage (V)	100	200	200
	Frequency (Hz)	N/A	N/A	60
	Duty Cycle	N/A	N/A	12%
Kīpapa	Waveform	DC	SP	

	Voltage (V)	100	200	
	Frequency (Hz)	0	60	
	Duty Cycle	0	12%	
Mānoa	Waveform	DC	DC	SP
	Voltage (V)	100	200	200-275
	Frequency (Hz)	N/A	N/A	60
	Duty Cycle	N/A	N/A	12%
Punaluʻu	Waveform	DC	SP	SP
	Voltage (V)	200	200	150
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	50%	30%
Waianu	Waveform	DC	DC	SP
	Voltage (V)	100	200	200
	Frequency (Hz)	N/A	N/A	60
	Duty Cycle	N/A	N/A	12%
Waikāne	Waveform	DC	DC	SP
	Voltage (V)	100	200	200
	Frequency (Hz)	N/A	N/A	60
	Duty Cycle	N/A	N/A	12%
Waimānalo	Waveform	DC	SP	SP
	Voltage (V)	100	175	200
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	12%	12%
Waimea	Waveform	DC	SP	SP
	Voltage (V)	100	200	275
	Frequency (Hz)	N/A	60	60
	Duty Cycle	N/A	12%	12%

Additional steps to before and after AIS removals to better understand efficacy and outcomes in of the study watersheds. Accordingly, AIS removals involved the following steps, completed in the order presented (Fig. 3):

(1) Collection of water samples to assess pre-removal stream chemistry and TSS,
(2) Snorkel surveys to assess pre-removal stream communities,
(3) Collection and sequestration of native species by hand-netting,
(4) Measurement of standard length and weight of native species,
(5) Elastomer tagging of *A. stamineus* for mark and recapture to measure post-removal organismal and demographic responses,

(6) Fin clipping of *A. stamineus* for genetic analysis, sacrifice of limited numbers of *A. stamineus* for otolith-based microchemical analysis of life history variation and isotopic analyses of trophic position,
(7) Electrofishing to remove AIS,
(8) Return of native species to their place of capture,
(9) Additional collection of water samples for chemistry and TSS analysis,
(10) Post-removal snorkel surveys to assess changes in AIS and native species communities

Figure 3. All steps involved in electrofishing-based removal of non-native species as listed in the text.

Protocols and data sheets for all portions of removal events in this study have been included as a supplementary appendix. These should serve as useful resources and guidance for conducting AIS management in Pacific island streams via electrofishing, which requires standard protocols and data collection minimally for snorkel surveys, sequestering native species, and electrofishing. Practitioners may also require additional protocols, such as those provided for measuring water chemistry, to assess the success and processes underlying AIS removal.

5.1 COMMUNITY RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Biotic surveys.

Using the point-quadrat method employed by the Hawaiʻi DAR (Higashi and Nishimoto 2007), we conducted visual snorkel surveys to quantify the densities (i.e., abundance) of native and non-native fishes, crustaceans, mollusks, and amphibians in the study reaches. Surveys were undertaken on an approximately monthly cycle (weather permitting), as well as the day immediately before and after AIS removals. Calibration studies have demonstrated that estimates of benthic and limnetic species demography derived from snorkel surveys are comparable to estimates derived from removal methods such as electrofishing (Mullner et al. 1998, Ebner et al. 2015) and mark-recapture (Hain et al. 2016). Following Higashi and Nishimoto (2007) and Hain et al. (2016, 2019), snorkel surveys were conducted by two team members swimming upstream to each of 30 randomly placed 1 x 1 m quadrats longitudinally distributed across the length of each 100 m reach. Within each quadrat, summary counts of all fishes, crustaceans, gastropods, and amphibians were recorded after an acclimation period of one to three minutes. Densities were calculated as the total number of individuals per species observed throughout the site survey area divided by the total surface area of the surveyed quadrats.

Statistical analysis of removal efficacy: did removal efforts lead to an immediate change in AIS and native species densities?

Unless otherwise stated, all variables were not transformed and all statistical analyses were conducted in R (R Core Team 2013), with significance determined at $\alpha = 0.05$.

Because densities of native species are low across Oʻahu, we used both species-level and community-level metrics to characterize native species responses to AIS removals. Species level metrics focused on *A. stamineus* because it is more widely distributed and occurs at higher densities relative to other native stream species at low-to-mid elevations in streams on Oʻahu (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018, Hain et al. 2019). Similarly, we used species-level and community-level metrics to characterize AIS densities. We assessed shifts in AIS densities and shifts in the densities of drift-feeding Poeciliids (guppies, mollies, and swordtails) and algae-eating Loricariid armored catfish to illustrate how capture varied according to categorical biotic differences (e.g., limnetic versus benthic habitat use). Densities were calculated as the total number of individuals per species observed throughout the site survey area divided by the total surface area of the surveyed quadrats. The change in density of AIS following removals was calculated as the difference between the densities the day before AIS removal and the day after. We calculated the percent change in density (((post-removal – pre-removal) / pre-removal) *100) of all AIS, Poeciliids, Loricariids, all native species, and *A. stamineus* from snorkel surveys conducted in the removal reaches the day before and following the removal events. Using Welch's Two Sample t-tests, we assessed if the percentage of density change was greater in the removal reaches compared to the control reaches for all AIS, Poeciliids, and Loricariids, respectively. We likewise tested for differences in densities of all native species and separately for *A. stamineus*, expecting no significant decreases in percent density change in the removal reaches compared to the control reaches.

Statistical analysis of removal efficacy: did AIS removal differ according to hydrology and pre-removal densities?

Multiple linear regressions were conducted to assess whether the number and biomass of AIS, Loricariids and Poeciliids captured in removal reaches varied according to mean discharge, discharge variability, and pre-removal estimates of AIS densities based on snorkel surveys conducted the day before removal events. We also used multiple linear regressions with AIC and backward model selection to quantify differences in percent density changes in AIS, Loricariids and Poeciliids, respectively, in response to: mean discharge, discharge variability, the number and biomass of AIS removed, and pre-removal estimates of AIS densities based on snorkel surveys conducted the day before removal events.

Statistical analysis of post-removal re-assembly: Were temporal responses to AIS removal ephemeral or persistent?

We assessed whether responses to AIS removal were temporally ephemeral or persistent (i.e., long-lasting) by evaluating changes in population densities between removal reaches, upstream control reaches, and downstream control reaches across the entire duration of the study. First, with the entire dataset grouped with reference to AIS removals (i.e., all pre-removal data were binned separately from all post-removal data), we constructed separate generalized least squares (GLS) models for each taxonomic group of interest (all AIS, Loricariids, Poeciliids, all natives species, and *A. stamineus*) to determine if changes in population density were best explained by the effect(s) of: period (before, after

AIS removal), reach (removal, upstream control, downstream control), and the interaction between period and reach. We also constructed separate GLS models for all other native and non-target AIS species. All GLS modeling was conducted using the package nlme (Pinheiro et al. 2019) with maximum likelihood estimation according to an autoregressive correction model with a lag of 1 to account for repeated measures (i.e., temporal autocorrelation) between snorkel surveys grouped by reach and watershed (corr = corAR1(form ~1 | Watershed/Reach). We accounted for unequal variances between pre- and post-removal sampling in the models with weight = varIdent (form ~ 1| BA) when it improved model fit. Model selection was determined using log-likelihood ratios and analysis of variance was conducted on the best-fit model to obtain parameter estimates and *P*-values, accounting for the possibility of type III errors. Post-hoc tests were conducted in the emmeans package (Lenth et al. 2019) and adjusted for multiple comparisons (adjust = “mvt”).

Second, we assessed the timeline of density changes and the effect of environmental and biotic covariates on population densities. We constructed separate GLS models for all AIS, Loricariids, Poeciliids, all natives species, *and A. stamineus* densities, respectively, to determine whether the effects on density varied by reach, cycle, reach x cycle interaction, discharge mean, discharge variability (coefficient of variation of daily discharge), AIS count removed, AIS biomass removed, and pre-removal AIS densities based on snorkel surveys conducted the day before removal events. Each GLS model was conducted using maximum likelihood estimation according to an autoregressive correction model with a lag of 1 to account for repeated measures (i.e., temporal autocorrelation) between snorkel surveys grouped by each reach and watershed (corr = corAR1(form ~1 | Watershed/Reach). Model selection was determined using log-likelihood ratios. And analysis of variance was conducted on the best-fit model to obtain parameter estimates and *P*-values, accounting for type III errors. Post-hoc tests were conducted in the emmeans package (Lenth et al. 2019) and adjusted for multiple comparisons (adjust = “mvt”). The removal event conducted in July 2017 (Punaluʻu) was preliminarily analyzed both with an offset timeline and with the timeline shifted to coincide with the pre- and post-removal cycles of surveys conducted in the other 12 watersheds. There were no differences found between the outcomes of these analyses, so we retained the shifted models for direct comparisons across all watersheds in subsequent analyses.

We also assessed whether AIS removals resulted in persistent shifts in community composition by comparing Bray-Curtis dissimilarity matrices between removal reaches, upstream control reaches, and downstream control reaches. Bray-Curtis dissimilarity matrices for each reach were calculated by comparing species occurrences during each snorkel survey cycle to an estimate of pre-removal species occurrence averaged across all pre-removal snorkel survey cycles. This comparison was intended to determine whether and when the composition of post-removal communities ‘returned’ to resemble the composition of pre-removal communities. We conducted separate GLS models for whole (both AIS and native species), AIS only, and native species-only ‘communities’ to determine how community dissimilarity changed through time (i.e., snorkel survey cycle), and according to: the interaction of reach and cycle, mean discharge, discharge variability (coefficient of variation of daily discharge), the number and biomass of AIS removed, and pre-removal estimates of AIS densities based on snorkel surveys conducted the day before removal events. Each GLS model for community dissimilarity was conducted with maximum likelihood estimations according to an autoregressive correction model with a lag of 1 to account for repeated measures (i.e., temporal autocorrelation) between snorkel surveys grouped by each reach and watershed (corr = corAR1(form ~1 | Watershed/Reach). All models accounted for unequal variances between cycles weight = varIdent (form ~ 1| Cycle). Model selection was determined using log-likelihood ratios. An analysis of variance was conducted on the best-fit model to obtain parameter estimates and *P*-values, accounting for type III errors. Post-hoc test were conducted in the emmeans package (Lenth et al. 2019) and adjusted for multiple comparisons (adjust = “mvt”). As in other analyses, the removal event conducted in July 2017 (Punaluʻu) was preliminarily analyzed both with the timeline shifted to coincide with the pre- and post-removal cycles of surveys conducted in the other 12 streams. There were no differences found between the outcomes of these analyses, so we retained the shifted models for direct comparisons across all watersheds in subsequent analyses.

5.2 DEMOGRAPHIC RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Study species and study design.

We assessed the demographic response of *A. stamineus* (Fig. 4)*,* one of the five migratory species endemic to Hawaiian streams (Lindstrom et al. 2012). *A. stamineus* inhabit low- to moderate-energy areas with sand, gravel, or small rocks as substrate in lower- and middle-elevation reaches of Hawaiian streams (Kinzie 1988), which are among the reaches most highly inundated by AIS (Moody et al. 2017). *Awaous stamineus* is an omnivore capable of feeding on green algae, diatoms, and aquatic invertebrates including dipterans and trichopterans (Kido 1997). The species exhibits wide dietary flexibility ranging from predominantly algal food sources to largely carnivorous diets (Lisi et al. 2018). Carnivory increases with increasing urbanization and the abundance of introduced fishes, suggesting possible diet displacement due to competition with AIS (Lisi et al. 2018).

Like all other native migratory fishes, *A. stamineus* are a species of conservation concern across the Hawaiian archipelago. Historically, *A. stamineus* were sufficiently abundant to support an artisanal fishery and thus have long been a culturally important resource for indigenous Hawaiian s, but range-wide population decline has led to state-wide restrictions on fishing and protection by the Hawaiʻi Division of Aquatic Resources (Devick et al. 1995; Ha and Kinzie 1996; Brasher 2003; Walter et al. 2012). There is particular concern for populations of native amphidromous species on Oʻahu. While there is evidence that the densities of all species are depressed across the island, recent surveys suggest that *A. stamineus* are relatively more resilient to pressures arising from land use intensification and AIS (Lisi et al. 2018).

Figure 4. Assays of native species demographic responses to AIS removal focused on *Awaous stamineus* (photo by Keoki Stender)

Awaous stamineus spawn in streams laying adhesive eggs on rocks that are defended by males until hatching, which typically occurs within 24 to 48 hours of spawning (Ego 1956). After hatching, migration commences with passive downstream larval “drift” (Fitzsimmons et al. 2007) that begins around sunset, with larvae capable of reaching the ocean within a few hours (Lindstrom 1998). Emigrant larvae spend on average 118 days (~4 months) in the ocean before metamorphosis to post-larvae (Hogan et al. 2014). Oceanic migration allows larvae to disperse among watersheds and islands, contributing to high genetic connectivity across the archipelago (Alda et al. 2016). Marine dispersal of the native species permits the possibility of demographic rescue of populations (Hanski 1998) via recruitment from distant populations. However, *A. stamineus* is known to be facultatively migratory with the majority of larvae forgoing oceanic migration; larvae instead remain in their natal stream (Hogan et al. 2014), which suggests that AIS removal might elicit a rapid response in local demography that is not driven by immigration.

Population recovery and recruitment dynamics following AIS removal should be contingent on surface flow conditions. Marine migration is governed by stream flow- if there is not enough surface flow at larval hatching, larvae will become locally entrained (Lindstrom 1998). Additionally, if surface flow is low, recruitment from the marine environment will be reduced, as ingressing larvae will not be able to detect and recruit to the stream (Engman et al. 2017). Thus, the amount of recruitment as well as the source of recruits (migrant or non-migrant) is expected to be determined in part by stream flow (Heim-Ballew 2019).

We evaluated the demographic response of *A. stamineus* to AIS removals following a BACI study design in 12 of the 13 study watersheds (Fig. 2, Table 1). One watershed (Kīpapa stream) did not contain native fishes and so was removed from analyses in this study. For 11 watersheds, the study period ran from May 2015 to February 2018. The study period for the remaining watershed (Punaluʻu) ran from October 2016 to February 2018. In each watershed, we first conducted a 9-month assessment of baseline conditions in a 100 m ‘removal’ reach, as well as two 100 m ‘control’ reaches, one located upstream and one located downstream from the removal reach separated by a 25 m buffer area between reaches. Upstream and downstream control reaches served as references for all of the response variables assessed for this study. Following the 9-month baseline assessments, AIS removals were then conducted following the protocol described below. Post-removal conditions were subsequently tracked in the removal and control reaches for a period of up to 2 years, except for Punaluʻu in which post-removal conditions were tracked for only 8 months. This approach enabled us to not only assess conditions prior to and following AIS control in removal reaches, it also allowed us to compare conditions in the removal reach to reference conditions to better differentiate outcomes of AIS removals from natural variability within and among streams (Osenberg et al. 1994).

Visual surveys.

Conducting point-quadrat snorkel surveys (Higashi and Nishimoto 2007) allowed us to quantify the densities (i.e., abundance) of all aquatic fauna in the study reaches and allowed us to assess demographic measures of *A. stamineus* responses to AIS removals, such as juvenile recruitment. Sizes of all *A. stamineus* observed were visually estimated to the nearest 10mm increment. Observers were trained in size estimation underwater through an observation and capture method whereby observers estimated the size of fish underwater and subsequently captured the same individual and measured them with calipers. This method allowed observers to calibrate size estimates and increases consistency among observers (Bell et al. 1985). All individuals 45 mm and smaller were characterized as recruits to the populations. This is the size that fish become easily observable and catchable in streams and thus was the minimum size at which individuals were tagged for mark-recapture. We estimate that fish ≤ 45 mm fish have been in the stream for 3 months or less based on the following size at age relationship determined by an otolith aging study (Heim-Ballew 2019): $y = 3.5447(TL) + 80.621$; where TL is the total length of the fish in mm.

Mark-recapture.

In addition to conducting snorkel surveys, we employed a mark-recapture approach to assess trends and changes in population size, recruitment, individual growth rates and body condition as well as immigration, and emigration of *A. stamineus* in each experimental stream reach before and after AIS removal. Fish were collected within study reaches. In most streams, *A. stamineus* were collected in the removal and upstream control reaches only; but in streams where fish were sparse, additional collections were conducted in the downstream control reach. Each reach was exhaustively sampled to ensure the capture of a large proportion of the local population. Collections were made on an approximately monthly cycle (weather permitting).

Following Tulane University IACUC protocol #0463, fish were collected by snorkelers with hand nets and placed into aerated buckets on the stream side placed near the location of collection (demarcated with weighted flagging tape). To reduce stress and minimize injury due to handling fish were individually anesthetized with a dilute dose of sodium bicarbonate buffered MS-222 for 30-60 s until breathing slowed. Once properly anesthetized, body mass (g) was measured using a battery powered scale with a precision of 0.01 g, and total length (mm) was measured using calipers. Each fish was then marked with visual implant elastomer tags (VIE) with unique color code tags for individual identification. Between individuals, the VIE needle was sterilized using 95% ethanol to prevent the spread of diseases among fish. Fish under 45 mm in length were not tagged. Fish were allowed to recover in aerated buckets with antifungal medication added to the bucket to inhibit infection. Fish were held until their breathing rate and behavior returned to normal. Fish were returned to the stream in the same location from which they were collected to minimize disturbance. On subsequent visits any recaptured fish were noted, their VIE tag recorded and mass and total length re-measured.

Statistical analysis of demographic responses to predation release: did *A. stamineus* populations sizes increase following AIS removals?

Unless otherwise stated, all variables were not transformed and all statistical analyses were conducted in R (R Core Team 2013), with significance determined at $\alpha = 0.05$.

Predation by AIS, particularly Poeciliids, is expected to depress *A. stamineus* populations primarily by reducing recruitment of juveniles to the populations. We relied on Lincoln-Peterson population estimates to test whether AIS removal resulted in an increase in population size. This approach has been demonstrated to be a reliable method of estimating population size of *A. stamineus* (Hain et al. 2016). Population sizes were estimated for four time windows centered around the removal date, “Prior”, “0-3 Month Post”, “3-6 Months Post”, and “>6 Months Post”. All data prior to AIS removal was pooled into one time-window to increase the sample size of recapture events in the prior period. Analyses of snorkel survey data found evidence of an increase in *A. stamineus* population densities in the first 6 months following AIS removal, with densities subsequently returning to pre-removal levels (described below). Therefore, we focus our mark-recapture analyses on 0-3 months and 3-6 months following AIS removals to test for short-term responses to AIS removals. Population increases in the first 3 months following removals may indicate movement of adults and juveniles into the remediated sites due to an increase in available habitat. In contrast, increases 3-6 months following removals are likely indicative of recruitment from a reproductive event following AIS removal, as the larval duration of *A. stamineus* is 3-5 months long. Data collected >6 months following removals was pooled to examine longer-term population sizes and to detect possible effects from increased sample sizes.

We calculated population estimates of both control and removal reaches separately and used linear regression to determine if the reaches were demographically independent. We then calculated population estimates of the whole stream (i.e., control and removal reaches). Analysis of variance (ANOVA) with a Tukey’s post-hoc test was conducted to determine if post-removal population estimates were significantly different from estimates from prior to the removal. Two streams, Hālawa and Kāwā , were removed from population estimate analysis because of a lack of recaptured individuals either during the pre-removal period, post-removal period, or both periods.

We tested the hypothesis that the magnitude of the response in population size was related to the effectiveness of AIS removals. From the ANOVAs, we first identified time periods that differed from the pre-removal period. We calculated the difference in Lincoln-Peterson population estimates between the pre-removal period and post-removal time periods that were found to be significantly different. We used regression analysis to determine if the pre- to post-removal change in population size was correlated with the percent density change (i.e., pre to post) in Poeciliids, Loricariids, and all other AIS. We expect that if AIS removal efficacy was driving demographic responses, then we should find a positive correlation between response and effect size. Furthermore, we tested the hypothesis that the the magnitude of the response in population size was related to stream hydrology. We used regression analysis to determine if the pre- to post-removal change in population size was correlated with the mean and variability (CV) of stream discharge, which influences AIS removal efficacy. As described below, the percent density change in Poeciliids and all AIS were greatest at intermediate mean discharge and more Poeciliids were removed from streams with greater discharge variability. Accordingly, we also tested for the interactive effects of hydrology and removal effectiveness on the response in population size, if the main effects were significant.

Statistical analysis of demographic responses: was there an increase in recruitment of *A. stamineus* following AIS removals?

We calculated the proportion of recruits in each population as the proportion of observed fish that were ≤ 45 mm to the total number of fish observed in monthly biotic surveys. We used survey data to estimate recruitment rather than the number of captures ≤ 45 mm because we had much larger sample sizes from observational data. We calculated the proportion of recruits for the same time windows as we calculated Lincoln-Peterson population estimates to allow for direct comparisons. Given the life-history of *A. stamineus* we expect that an increase in the proportion of recruits in the 0-3 month post removal window would reflect greater survival of individuals that had recruited prior to or immediately after the AIS removal events. On the other hand, an increase in the proportion of recruits 3-6 months post removal might indicate greater survival of larvae and recruits from a spawning event that happened following AIS removals.

We calculated the proportion of recruits of both control and removal reaches separately and used linear regression to determine if the reaches were demographically independent. We then calculated the proportion of recruits of the whole stream population (i.e., control and removal reaches). An ANOVA with a Tukey's post-hoc test was used to determine if the post-removal proportion of recruits was significantly different from estimates from prior to the removal.

If the proportion of recruits in the populations increases, then there should be a concomitant shift in size-frequency distributions toward a greater relative proportion of smaller individuals. To test this hypothesis, we conducted a Kolmogorov-Smirnov test (K-S test) to compare size-frequency distributions of all populations prior to AIS removal to size-frequency distributions of the populations in the post-AIS removal period(s) during which we detected a significant increase in recruitment.

We also tested the hypothesis that the magnitude of the response in the proportion of recruits was related to the effectiveness of AIS removals. From the ANOVAs, we identified time periods that differed from the pre-removal period. We calculated the difference in the proportion of recruits in each population between the pre-removal period and post-removal time periods that were found to be significantly different. We used regression analysis to determine if the pre- to post-removal change in recruitment was correlated with the percent density change (pre-post) in Poeciliids, Loricariids, and all other AIS. Furthermore, we tested the hypothesis that the magnitude of the response in the proportion of recruits was related to stream hydrology. We used regression analysis to determine if the pre- to post-removal change in the proportion of recruits was correlated with the mean and CV of stream discharge. We tested for the interactive effects of hydrology and removal efficacy on the response in the proportion of recruits if the main effects were significant.

Statistical analysis of demographic responses: did the growth rate or condition of *A. stamineus* increase following AIS removals?

Competition with AIS for habitat space or resources is expected to cause a depression in growth rates and body condition of *A. stamineus*. To assess whether AIS removal resulted in an increase in growth and body condition, we calculated growth rates and changes in body condition of recaptured individuals. For each recaptured *A. stamineus,* we took the total length (mm) and weight (g) at the time of recapture and subtracted the length and weight from the previous capture event. We divided the difference by the number of days between capture events to calculate growth rates in length and weight. With body condition calculated as follows:

$$Condition\ (C) = \frac{Weight\ (g)}{Standard\ Length\ (mm)^\wedge 3}$$

change in body condition was calculated as the condition at recapture minus the condition at previous capture divided by the number of days between capture events. An ANOVA with a sequential contrasts post-hoc test was used to determine if the growth rates and change in body condition varied significantly between time periods. Two streams, Hālawa and Kāwā , were removed from growth rate and condition analysis because of a lack of recaptured individuals either during pre-removal, post-removal, or both periods.

We then tested the hypothesis that the magnitude of the response in growth and change in condition factor was related to the effectiveness of AIS removals. From the ANOVAs, we first identified time periods that differed from the pre-removal period. We calculated the difference in growth (mm/d and g/d) and condition (g/mm3·d) between the pre-removal period and post-removal time periods that were found to be significantly different. We used regression analysis to determine if pre- to post-removal change in growth and/or condition was correlated with the percent density change in Poeciliids, Loricariids, and all other AIS. Furthermore, we tested the hypothesis that the magnitude of the response in growth and condition was related to stream hydrology. We used regression analysis to determine if the pre- to post-removal change in growth and condition was correlated with the mean and CV of stream discharge. We tested for the interactive effects of hydrology and removal effectiveness on the response in the growth and condition if the main effects were significant.

Statistical analysis of demographic responses: did immigration or emigration of A. stamineus into and out of removal reaches increase following AIS removals?

Competition with AIS for habitat space or resources is expected to displace native *A. stamineus*. AIS removals may lead to a higher degree of residency of fish within the treatment reach, and/or may cause more fish to move into the treatment reach following removal if niche space has opened up. We also calculated movement rates of recaptured individuals by dividing the number of individuals that moved between reaches by the number of individuals that remained in the same reach between capture events. We used a t-test to determine whether movement rates differed between pre- and post-removal periods. Furthermore, we used locally estimated scatterplot smoothing (LOESS) to determine whether there were trends in the time-series of the number of marked individuals that moved between reaches, the number of marked individuals that stayed resident in their previous reach, and whether movements into or out of the removal reach changed over time.

5.3 LIFE HISTORY VARIATION IN RELATION TO AIS, DISCHARGE, AND NUTRIENTS IN HAWAIIAN STREAMS

Study species and study design.

This study focused on *A. stamineus* (Fig. 4) because it is the most abundant and widespread native fish on Oʻahu, and because (to date) its life history has been more extensively studied than the life history of the three other facultative migratory native fishes in Hawaiian streams (Hogan et al. 2014, Heim-Ballew et al. 2020). We evaluated life history variation in *A. stamineus* captured in 12 of the 13 study watersheds (Fig. 2, Table 1), as one watershed (Kīpapa stream) did not contain native fishes and so was removed from analyses in this study. All *A. stamineus* were collected by snorkelers using hand nets and were anesthetized using MS-222. Whole fish samples were stored in polypropylene vials filled with 95% ethanol.

AIS densities and hydrology.

All AIS densities and native species densities were characterized in the study watersheds according to snorkel surveys (described above) conducted prior to AIS removals. For this study, daily stream discharge was characterized between October 2015 and October 2016. For 8 watersheds, discharge was characterized from USGS gauging station records downloaded with R package “dataRetrieval” (Hirsh and De Cicco 2014). For Waianu Stream, discharge records were gathered for the station closer to the river mouth on Waiahole Stream (16294100) rather than the station in the Waianu sub-watershed. For streams without stations (Keaʻahala, Kāwā, and Kīpapa streams), discharge was monitored using HOBO U20 water level data loggers in removal reaches corrected for atmospheric pressure monitored by a Hobo U20L data logger in the adjacent riparian zone (described above).

Nutrients.

We characterized average inorganic nitrogen and soluble reactive phosphorus (SRP) concentrations for each stream as a proxy for anthropogenic nutrient pollution (Lisi et al. 2018). Dissolved inorganic nutrients were quantified at approximately monthly intervals from May 2015 through January 2018 in each stream. Samples were taken at or near base flows from streams using syringes, the water was filtered immediately (Whatman GFX; 0.45 µm), and then stored frozen until ready to be analyzed. Thawed samples were simultaneously run for SRP (µg/L) and nitrate (NO_3, µg/L) using standard colorimetric methods on an Astoria-Pacific Autoanalyzer II (Clackamas, Oregon, USA). We calculated mean SRP and NO_3 concentrations across the full study period to capture nutrient variation among streams.

Collection of *A. stamineus* otoliths.

Otolith material is laid down continuously as fish grow, and as otoliths are formed, their calcium-carbonate matrix passively incorporates trace elements from the water, thereby creating a detailed, permanent record of the chemical environment an individual has experienced until the time of sampling. Therefore otolith microchemistry data can be used to quantify the frequency and spatial distribution of migratory and resident larvae recruiting to *A. stamineus* populations (Hogan et al. 2014). Accordingly, **s**agittal otoliths were extracted from each individual using forceps and a fine-bristled paintbrush and distilled water were used to remove tissue from the calcareous structure. Otoliths were then mounted on a petrographic slide using Crystalbond adhesive resin and stored until further analysis. This sampling was conducted under the TAMUCC IACUC protocol AUP# 11-15 TAMUCC.

Characterization of migratory life history.

Extracted and mounted otoliths were first finely polished with fine-grit diamond coated lapping paper of varying grit sizes (range 60 µm - 0.5 µm) to reveal the growth rings from the primordium to the edge. Otolith trace element chemistry was then analyzed to determine migration history (i.e., migrant or resident) using Laser Ablation Inductively Coupled Plasma Mass-Spectrometry (LA-ICP-MS) at the Jackson School of Geosciences, University of Texas-Austin. A New Wave Research UP193-FX fast excimer (193 nm wavelength, 4-6 ns pulse width) laser coupled to Agilent 7500ce inductively coupled plasma mass spectrometry (ICP-MS) was used according to Seeley et al. (2015). A laser transect was set to run from the otolith edge (adult stage) to the opposite otolith edge and by bisecting the primordium to establish a palindromic chemical signature (Hogan et al., 2014). Otoliths were pre-ablated to remove any external contaminants prior to data collection (40% power, 10 Hz, 50 µm spot size, 40-50 (µm/s) scan rate, He cell flow of 300 mL/min). Ablations were operated at RF Power 1600 W with an average Ar carrier flow of 1 L/min. Samples were analyzed at 5 µm/s line traverses, using a 25 µm spot at 60% power, 10 Hz repetition rate, and He cell flow of 700 mL/min Laser energy (fluence) during analytical sessions averaged 3.57 J/cm^2 with < 3.0% variance. Ten analytes ($^{24\text{-}25}$Mg, $^{42\text{-}43}$Ca, ^{55}Mn, ^{66}Zn, ^{68}Zn, ^{88}Sr, $^{137\text{-}138}$Ba, ^{7}Li, ^{208}Pb) were measured using a 25-millisecond integration time. Magnesium, zinc, and manganese were used to help identify areas where the laser crossed the epoxy prior to the otolith material being analyzed. Calcium was measured to identify variations in otolith calcium carbonate topography and for standardizing otolith strontium concentrations. Barium has a higher concentration in freshwater than the ocean and strontium shows the opposite pattern, and both elements can be used to verify which aquatic environment an individual experienced at a given time in the life-history. The analytical sampling period was 0.6512 seconds, which is equivalent to a reading every 7.678 µm and corresponds to 85% measurement time. Intensities of isotopes (counts) were converted to

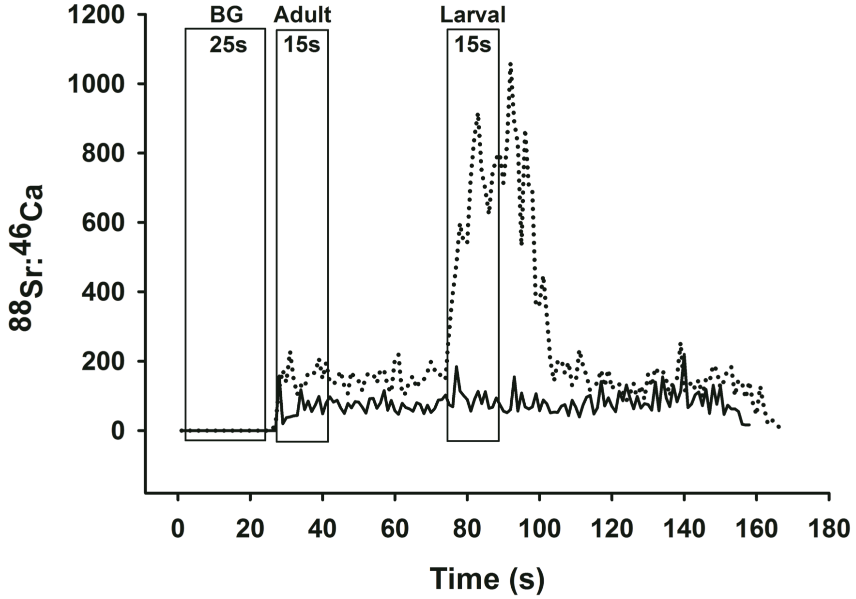


Figure 5: Example of migrant (dashed line) and resident (solid line) life history otolith profiles; high Sr:Ca ratios in the middle of the profile indicate larval residence in saltwater. Boxes indicate the method of signal selection

for each sample; a 25 s window selected for background (BG) signal, and 15 s windows for both the post-metamorphic (Adult) and Larval signals.

concentration (ppm) using Ioline software using ^{43}Ca as the internal standard. Baseline gas concentrations of the analytes was determined prior to the analysis of each otolith by taking 30 second gas blank intervals that were measured while the laser was off. USGS MACS-3, a synthetic calcium carbonate standard, was used as the primary reference standard for all analytes. Recoveries (relative standard deviations versus GeoREM preferred values) among analytes for secondary standards (NIST612) were run as unknowns against the primary standard. Element concentrations were calculated assuming a concentration of 38.3% Ca (FEBS-1 otolith standard certificate) in the sampled outer calcium carbonate layer. Integrated Sr:Ca ratios were used to determine marine and freshwater habitat use by fish during the larval stage (pre-metamorphosis) and juvenile/adult stage (post-metamorphosis). Fish were considered migrant contingents if the Sr:Ca ratio prior to the metamorphosis mark was greater than 4 mmol/mol (Fig. 5); all other fish were considered residents (Hogan et al., 2014). The proportion of contingents for a stream was determined by evaluating the Sr:Ca signatures of all fish collected from a stream. Residents were assigned a value of 0, and migrant contingents were assigned a value of 1 for statistical analysis.

Statistical model selection and analysis.

Unless otherwise stated, all variables were not transformed and all statistical analyses were conducted in R (R Core Team 2013), with significance determined at $\alpha = 0.05$.

Dredge model selection was performed to identify factors that most strongly influence the proportion of residents in populations of *A. stamineus.* The best model was identified by the lowest model AIC. The factors identified in the best model were considered to have the largest effect on the ratio of migrants to residents within each stream. The independent variables included as main effects in the full model were Loricariid density (#/m^2), Poeciliid density (#/m^2), daily median discharge (m^3/s), discharge variability (m^3/s), *A. stamineus* population density for each stream (#/m^2), inorganic nitrate (µg/L), soluble reactive phosphorus (µg/L), and to reduce collinearity in the model two interaction terms were also included being *A. stamineus* density x Loricariid density and soluble reactive phosphorus x discharge variability. Each models AIC and AUC were compared to evaluate the goodness of fit and whether it should be considered for further analysis. One additional watershed (He‘eia) was excluded from the analysis due to low sample size (n = 1). The total sample size for the analysis was 495 *A. stamineus*.

The top three models identified by the dredge selection process were analyzed by a multiple logistic regression. The significance of model terms (p-value < 0.05 was considered significant), overall model significance, AIC, and AUC were compared to identify the best and final model.

5.4 GENOMIC AND LIFE HISTORY RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Study design and sample collection.

This study focused on *A. stamineus* (Fig. 4), the most abundant and widespread of the amphidromous Hawaiian goby species (Keith et al. 2003, Moody et al. 2017). Prior work on otolith microchemistry has demonstrated that *A. stamineus* exhibits partial migration with ocean-going larvae, (i.e., migrants) having an average larval duration time of 118 days exhibiting a bimodal distribution with modes around 75 days and 155 days (Hogan et al. 2014). Prior studies also have demonstrated that many *A. stamineus* larvae remain in freshwater (i.e., residents) and that local populations of A*. stamineus* are often a mix of life history variants (Hogan et al. 2014, Heim-Ballew 2019). It is not uncommon for residents to dominate local populations, especially on Oʻahu (Heim-Ballew 2019). Additionally, prior work has found that nuclear and mitochondrial genetic variation in *A. stamineus* are inversely related to AIS densities in streams across the Hawaiian archipelago (Blum et al. 2014). Consistent with this, populations of *A. stamineus* on the island of Oʻahu harbor lower levels of genetic diversity than populations on other islands in the archipelago (Blum et al. 2014).

We evaluated genomic and life history variation of *A. stamineus* across twelve watersheds on the island of Oʻahu where we conducted experimental AIS removals (Fig. 2, Table 1). Kīpapa watershed was excluded from further consideration because no native species (including *A. stamineus*) were found there during the study period. Located on the windward and leeward sides of the Koʻolau Range, the study watersheds were selected to circumscribe the range of surface flow conditions (i.e., mean discharge, discharge variability; Table 1) as well as variation in the distribution and abundance of AIS on the island. This approach enabled us to leverage prior characterizations of hydrology and biotic communities (described above and below) to identify abiotic and biotic factors that influence genomic and life history variation of *A. stamineus*. Likewise, it enabled us to assess genomic and life history variation of *A. stamineus* prior to and following AIS removals.

Genomic and life history variation were assayed from post-larval immigrating *A. stamineus* recruits (< 45 mm) collected with hand nets between May 2015 and February 2018. A total of 262 samples were used to concurrently study genomic and life history variation across the study sites, with sample sizes ranging from 1 to 44 individuals per watershed (Table #). An additional 234 samples (total n = 496) were used to gain further insight into life history variation (described above and below). All individuals were euthanized with MS-222 and measured for total body length. Whole specimens preserved in 95% ethanol until being transferred to long-term storage at 20 °C. All collections were conducted in accordance with Tulane University IACUC protocol #0463.

Otolith extraction and migratory type identification.

As done for our other work on life history variation (described above and below), sagittal otoliths were extracted from all individuals using forceps and a combination of distilled water and a fine-bristled paintbrush was used to remove tissue from the calcareous structure of each otolith. This sampling was conducted under the TAMUCC IACUC protocol AUP# 11-15 TAMUCC. Otoliths were mounted on a petrographic slide using Crystalbond adhesive resin and stored until further processing. As described above, otolith microchemistry was analyzed following Hogan et al. (2014) and Heim-Ballew et al. (2020) to characterize migratory life history, with each individual identified as a resident or migrant type.

DNA extraction, sequencing, and genotyping.

Genomic DNA was extracted from each individual using DNeasy Blood & Tissue spin-column kits (QIAGEN, Toronto, ON, Canada), quantified using the Quant-iT dsDNA pico-green assay kit (Life Technologies, Grand Island, NY, USA), and visualized on 1.0% agarose gel to assess degradation. Genotyping by sequencing libraries were prepared with the 6-base cutter restriction enzyme Sph1 and Tas1, targeting fragment sizes of 475-560 base pairs (bp) and sequenced on an Illumina HiSeq2000 at the Texas A&M University – Corpus Christi Genomic Core Laboratory. A total of six 182-plex libraries were constructed for pair-end sequencing (150 bp target length), each containing blank controls. Individuals within each library were tagged with unique barcodes (5-8 bp in length) for identification.

Raw data filtering and SNP identification.

The software package FASTQC (Andrews 2010) was used to identify Illumina or universal adapters from raw reads as well as to examine the quality of raw reads. We used the software package STACKS v.2.1.1 (Catchen et al., 2013) for processing all sequence data. We allocated reads to individual samples, removed adapters, trimmed the reads to 146 base pairs using the STACKS demultiplexing program PROCESS_RADTAGS, and filtered out low-quality reads. We used the STACKS DENOVO_MAP program on a subset of 60 samples (encompassing all populations) to determine optimal parameter settings (m=3, M=3, n=3) for *A. stamineus* (Rochette and Catchen 2017, Paris et al. 2017). We used the USTACKS program to identify putative loci and SNPs at each locus. A subset of individuals (100) encompassing all populations and with coverage > 30x were used in the CSTACKS program to create a catalog of loci. All samples were then aligned to the catalog with the program SSTACKS. The *populations* program was used to export population SNPs and haplotypes.

Analysis of genomic diversity and population structure.

Unless otherwise stated, all variables were not transformed and all statistical analyses were conducted in R (R Core Team 2013), with significance determined at $\alpha = 0.05$.

Nucleotide diversity (π) and expected heterozygosity (H_e) were calculated for each population and migratory type for the full study period as well as prior to and following AIS removals using STACKS v2.1.1 (Catchen et al. 2013). Using generalized linear models, we examined relationships between genomic variation and hydrology, focusing on mean discharge and discharge variability according to coefficients of variation, as well as the biomass of Poeciliids and the biomass of Loricariids removed from the study sites. Population genomic structure was assessed through discriminant analysis of principal components (DAPC) in the package adegenet with clusters defined a priori as being each of the watershed group or migratory ecotype, as well as through admixture coefficient estimation, measures of divergence between populations, and relative proportions of genetic variance across hierarchical levels of organization. Pairwise F_{ST} values were calculated by study site and migratory ecotype using the package BEDASSLE (Bradburd et al. 2013), which relies on Weir and Hill's θ to assess divergence (Weir et al. 2002). Finally, an analysis of molecular variance (AMOVA) was performed using the package poppr (Kamvar et al. 2019) to determine the hierarchical (e.g., individual, migratory ecotype, watershed) structure of observed genomic variation.

Analysis of genomic and life history variation.

We first conducted a Welch's t-test to determine if expected heterozygosity (i.e., genomic diversity) differed between residents and migrants, with individuals across all populations pooled according to migratory type. We also conducted an analysis of variance to determine whether genomic variation in *A. stamineus* differed between populations in combination with migratory type. We then conducted Welch's t-tests to determine if AIS removals elicited a change in *A. stamineus* genomic diversity, with all individuals pooled as residents and migrants, differentiated according to treatment (i.e., residents in removal reaches, residents in control reaches, etc.). We also conducted generalized linear modeling to assess whether genomic diversity varied according to discharge (mean and coefficient of variation) as well as the biomass of Poeciliids and Loricariids (respectively) taken out of removal reaches.

We undertook a similar set of analyses to assay life history responses to AIS removals using z-tests, supplemented by an additional set of analyses designed to detect short-lived responses. Like some demographic responses associated with migration (e.g., recruitment) AIS removals might elicit transient shifts in life history variation. We first compared the proportion of resident (i.e., non-migrants) recruits before and after AIS removals across all watersheds, with all individuals pooled as residents and migrants. We then determined whether the proportion of resident (i.e., non-migrants) recruits differed between the two months before and two months following AIS removals, with data combined across all watersheds. We also assayed how the proportion of resident (i.e., non-migrants) recruits differed across four time windows centered around the removal date. As was done for demographic analyses (described above), individuals were binned in “Prior”, “0-3 Month Post”, “3-6 Months Post”, and “>6 Months Post” categories. All data prior to AIS removal was first pooled into one time-window to capture a more thorough representation of baseline conditions. In a parallel analysis, we constrained the pre-removal time period to the three months prior to AIS removals. For both analyses, data collected >6 months following removals was pooled to examine longer-term responses and to detect possible effects from increased sample sizes. To further align analyses of life history and demographic responses to AIS removals, individuals were not differentiated according to treatment; rather, all individuals (i.e., across all populations) were pooled according to migratory type.

5.5 ECOSYSTEM RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Study Design.

We evaluated the ecosystem outcomes of AIS removal using a BACI study design in 13 watersheds on the island of Oʻahu (Fig. 2). Focal watersheds were selected to capture the range of surface flow on the windward and leeward sides of the Koʻolau Range. These 13 watersheds also capture the variation in the distribution and abundance of all AIS present on Oʻahu (Blum et al. 2014; Moody et al. 2017; Lisi et al. 2018). For 12 watersheds, the removals occurred in March 2016. The removal in the remaining watershed (Punaluʻu) occurred in July 2017. In each watershed, we first conducted an assessment of baseline biotic and hydrological conditions in a 100 m ‘removal’ reach, as well a 100 m ‘control’ reach located 25 m upstream of the removal reach. Each pair of removal and control reaches were located in the lower region of the study watersheds (Fig. 2), which is common practice for BACI-style experiments in aquatic systems (Rytwinski et al. 2019). As described above, we then removed AIS from each 100 m removal reach using three-pass electrofishing with block nets at the upper and lower boundaries to prevent exchange of fish and prawns during the manipulation. To ensure the safety of native amphidromous fishes and macroinvertebrates (e.g., *Macrobrachium* prawns), snorkelers netted them by hand before electrofishing began; they were counted and measured, then held in aerated, streamside tanks near their capture location until completion of AIS removal, after which they were released to their place of capture. All captured AIS were euthanized with MS-222 following Tulane University IACUC protocol #0463. Identification, length, and wet mass were recorded for every individual within 24 hours to calculate AIS biomass density based on the streambed area of the removal reach. On the same day that the removal was completed in each stream, two team members walked through the upstream control reach to mimic the substrate disturbance from netting and electrofishing in the removal reach. Analyses of ecosystem responses focused on comparing conditions measured in the months leading up to and the months immediately following manipulations, when the effects of AIS removals were expected to be strongest and most apparent.

Watershed land use, hydrology, sediment and nutrient conditions.

Land use in each watershed was assessed using USGS StreamStats data, which is based on the 2001 National Land Cover Dataset (the most recent available for Hawai'i; Homer et al. 2007). The catchment delineated upstream of the end of the removal reach was used to characterize watershed size, basin slope, and land use. We characterized land use in each watershed as percent area urban (high, moderate, low intensity, and developed open space), impervious surfaces, forest (tree canopy cover), and agriculture (crop + pasture).

Hourly discharge was used to characterize surface flow regimes for each study watershed. When possible, we used data from a USGS gage near the experimental reaches (Table 2). In watersheds without USGS gaging stations (Waianu, Keaʻahala, Kāwā, and Kīpapa), water level was monitored hourly using HOBO U20 water level data loggers (Onset Computer Corporation, Bourne, MA, USA) placed within the stream at the study site, corrected for fluctuations in atmospheric pressure recorded by a second logger in the adjacent riparian zone. Water level measurements were used to estimate discharge following standard methods based on a power function relationship between water level and instantaneous discharge calculated from velocity and depth measurements (Gore and Banning 2017).

TSS and dissolved nutrients were measured at 2-3 times in the two months before and after AIS removals in each reach. TSS was characterized from 1.0-3.0 L of stream water filtered onto pre-weighed, pre-combusted filters (ProWeigh), and stored frozen until being oven-dried (48 hr, 60°C) and re-weighed. For nutrient analyses, water samples were collected in the thalweg using syringes, filtered through a glass fiber filter (Whatman GFX; 0.45 µm), and stored frozen until analyzed. Ammonium (NH_4) was measured within 24 hours according to fluorometric measures (Holmes et al. 1999) taken with an AqualFluor Fluorometer (Turner Designs San Jose, CA). SRP and NO_3 were analyzed using standard colorimetric methods on an Astoria-Pacific Autoanalyzer II (Clackamas, Oregon USA).

Analysis of nutrient limitation.

AIS can mediate primary productivity by limiting nutrient availability to algae, especially those that are not heavily shaded and that do not receive abundant nutrients from upstream pollution. Considering this, we evaluated nutrient limitation of periphyton using nutrient diffusing substrates (NDS; Tank et al. 2007) deployed in both reaches of each stream for 15 days. Plastic containers were filled with agar mixed with dissolved salts of nitrogen (0.5 M NH_4Cl), phosphorus (0.5 M KH_2PO_4), nitrogen and phosphorus (0.5 M KH_2PO_4+0.5 M NH_4Cl) or no nutrients. A fritted glass disk was placed on top of the agar and served as a clean algal growth substrate. Randomized complete blocks of each treatment (Control, N, P, and NP) were anchored to the stream bottom (n = 3 per treatment) in each reach before and after AIS removals. After collection, the fritted discs were separated from the agar, wrapped in aluminum foil, and frozen for later analysis. Chlorophyll *a* was extracted in cold 90% buffered ethanol for 24 hours, with extracts then analyzed using fluorometry. Though nutrient limitation was assayed in all 13 watersheds prior to AIS removals, several post-removal NDS deployments were destroyed by high flow events, stream drying, or vandals. Thus, the full BACI design was not achieved for all 13 watersheds.

Leaf litter breakdown.

Leaf-litter breakdown rates integrate physical, chemical, and biological properties of streams (Benfield et al. 2017). AIS removal might alter leaf decomposition by altering nutrient availability to microbial biofilms, which constitute a large proportion of N content of submerged leaves (hence leaf litter C:N ratios reflect microbial growth). We assessed microbial decomposition of leaves from *Hibiscus tiliaceaus*, a naturalized riparian plant introduced to the Hawaiian archipelago by early Polynesian settlers. Senescent *H. tiliaceaus* leaves were collected while still attached to the tree, then air dried for several weeks. Material from whole leaves (stems removed) was weighed (2.00 ±0.05 g) and folded into envelopes of window screen to prevent access by shredders. Five leaf packs were anchored to the stream bottom at the downstream end of each reach. Control leaf packs (n = 6) were carried once into the field to quantify mass loss due to handling, but were never wetted. After 15 days, leaf packs were collected, frozen, and freeze dried. Inorganic sediment and invertebrates were removed before weighing on a microbalance. A subset of samples from each reach (n = 3) were ground into a fine powder and analyzed for organic content (via loss on ignition) and C:N ratio (via elemental analysis).

Whole stream metabolism.

To complement measures of leaf litter breakdown, we also assessed net ecosystem production (i.e., the total activities of autotrophs and heterotrophs) using the diel oxygen method (Odum 1956). Dissolved oxygen and temperature were recorded hourly using an automated logger (Hobo U26, Onset Corp., Bourne, MA, USA; 0.02 mg L^{-1} and 0.02°C sensor resolutions), with the Bayesian Metabolic Model (BAMM, (Holtgrieve et al. 2010a) used to simulate diel dissolved oxygen dynamics as a function of water temperature and light (PAR). The model estimates gross primary productivity (GPP), ecosystem respiration (ER), and gas exchange with the atmosphere (K) over a 24 hour period. Average stream width, depth, and canopy cover (via conical densitometer) were calculated from measurements every 10 m in each reach. O_2 measurements were calibrated in municipal water that had been circulated with a minipump and aerated to reach oxygen saturation. We confirmed that sensor drift did not exceed 0.05 mg/L per hour by recording changes in saturated water held in darkness. Oxygen dynamics were recorded at the bottom of each reach for 4-5 days just before AIS removals, and again 10-14 days afterwards. The timing of post-removal metabolism measurements was designed allow the ecosystem to respond to reduced consumer densities, while minimizing potentially confounding effects from community reassembly (i.e., recolonization). We avoided sampling during large rain events to minimize possible effects of hydrological disturbance on stream metabolism. Nonetheless, cascades in some streams created high gas transfer velocities that precluded quantification of GPP and ER (Holtgrieve et al. 2010).

Statistical analyses.

Unless otherwise stated, all variables were not transformed and all statistical analyses were conducted in R (R Core Team 2013), with significance determined at $\alpha = 0.05$.

Algal nutrient limitation was tested in each NDS assay using one-way ANOVA followed by Tukey's HSD pairwise post-hoc comparisons. The identity of the limiting nutrient was inferred as N (N > C or N = N+P > C), N and P co-limitation (N+P > C), primarily N and secondarily P (N+P > N > C), or no limitation by N or P (N = P = N+P ≤ C) following Tank and Dodds (2003).

To quantify the overall impact of AIS removals on stream ecosystem functioning, we used permutational multivariate analysis of variance (perMANOVA), which treats all streams as comparable replicates, to test whether removal reaches differed from control reaches before and after removal (i.e., four groups reflecting before-after and control-removal combinations). We conducted separate perMANOVAs for water chemistry (nutrients, TSS), NDS experiments (P, C, NP, N), leaf litter breakdown (rate, %N, %C, C:N), and metabolic metrics (GPP, ER, GPP:ER) (Table 1). We also performed a multivariate homogeneity of group dispersions test to assess pair-wise differences in variance among groups (Anderson 2006). Ecosystem metrics were log standardized using the biostats code compilation (McGarigal 2009), and analyses were conducted using the Vegan (Oksanen et al. 2016) and MASS packages.

Because we purposefully selected watersheds to represent broad gradients in biotic and abiotic characteristics, we also explored the influence of individual stream characteristics on BACI experimental outcomes. We did so by assessing the correlation between each characteristic and the effect size of removing AIS. We used log response ratios to quantify effect size (Hedges et al. 1999):

$$Effect\ size = \log\left(\frac{\frac{R_{after}}{R_{before}}}{\frac{C_{after}}{C_{before}}}\right)$$

where *R* and *C* are the response variable in the removal and control reaches, respectively, as measured before and after AIS removal. Negative effect sizes indicate a greater decrease in the response variable following AIS removal in the removal reach than the control reach, while positive values indicate a greater increase in the removal reach. Effect sizes near zero indicate that there is no temporal difference between reaches. For each type of response variable (i.e., nutrients, leaf breakdown, metabolism), we then tested for a correlation between effect size and each contextual gradient (i.e., land use, discharge, light, background nutrients, pre-removal AIS densities). To account for potential functional differences among AIS, we tested for separate correlations focusing on the removal of Loricariid armored catfish, Poeciliid live-bearers, and all AIS, respectively.

5.6 COUPLED OCEAN-WATERSHED MODELING OF WHOLE-WATERSHED AIS REMOVAL

We developed a general analytic model of the ‘clean-slate’ approach that considers an arbitrary species subdivided into subpopulations, which may represent spatial units or life history stages (Appendix B), that are variably connected via two-way propagule exchange. Invasive species extermination is assumed to entirely remove all invaders from the habitat of one subpopulation of the target native species. This depiction of a metapopulation yields recovery of native species at the restoration site so long as there is at least one connected subpopulation as a source of recruits. We then compared the dynamics of native species demography, including the pace and degree of subpopulation growth, between clean-slate and unrestored sites (Appendix B). The model is designed to be general and phenomenological; it depends only on differences between native and invasive species life history, and the strength of negative effects of invasive species on native species. It can also be extended to consider the implications for subpopulation recovery of deviations from the most general clean-slate approach, including differences in metapopulation structure and spillover, and pitfalls such as Allee effects and temporal variation in connectivity among subpopulations. To illustrate this, we undertook ancillary analyses that address the consequences of density-dependence, transient effects following extermination, and the temporal dynamics of subpopulation recovery (Appendix B).

Simulation model structure and dynamics.

The general model was tailored to investigate the application of ‘clean-slate’ restoration to Hawaiian stream gobies, capturing the general properties of a metapopulation in species with complex life cycles involving obligate transitions between two distinct habitats (Fig. 1, Fig. 6). The structure of the model features a group of individual watersheds connected by marine dispersal. Local dynamics within each watershed are a function of competition and watershed conditions, including the impact of invaders (see Appendix B for details of model structure and parameterization). These parameters determine the maximum density of native species in a watershed, production of propagules, and the ability of incoming larvae to colonize. Larvae at sea disperse according to a three-dimensional Lagrangian ocean circulation model developed for the Hawai‘i region by the (Hybrid Coordinate Ocean Model (HYCOM) consortium (Jia et al. 2011, http://hycom.org/hycom/overview). The HYCOM covers the full Hawaiian archipelago (16° N – 26° N and 166° W to 150° W) at a resolution of 1/25th or a degree latitude and longitude (~4 km resolution). Following Moody et al. (2019), the 3-dimensional hydrodynamics of open-ocean and coastal environments (Fig. 7) captured by the HYCOM was coupled with a particle tracking model to simulate dispersal of larval fish as a passive tracer. Combined with empirical observations about life history such as the frequency distribution of larval period duration, we calculated average larval import and export between each of 51 subpopulations (Fig. 7) across a four-year period (2007-2011) (Moody et al. 2019). The resulting connectivity matrix leads to dynamic interactions between subpopulations, allowing us to calculate a minimum larval death rate during marine dispersal.

To maximize the realism of the model, we ran simulations for just one of the five endemic gobies of the Hawaiian archipelago (Kido 2008). *Awaous stamineus* is the best-studied species, reflecting the fact that it is the most abundant and widespread at present (Lisi et al. 2018). The only facet of its biology that we excluded from the model for simplicity is facultative amphidromy (Hogan et al. 2014), which was beyond the scope of this study. Because the model is structured to capture general properties of amphidromous life history, the qualitative patterns in our results are expected to apply to species other than *A. stamineus*, particularly given our approach of permuting stream conditions across watersheds. Although competition between gobies probably plays a role in determining the structure of their communities (Kido 2008), they all share an amphidromous life history that differs fundamentally from that of most invasive species. Given that gobies coexist readily within watersheds (Kido 2008), our results would be unchanged so long as intraspecific competition coefficients (Appendix B) exceed interspecific competition coefficients.

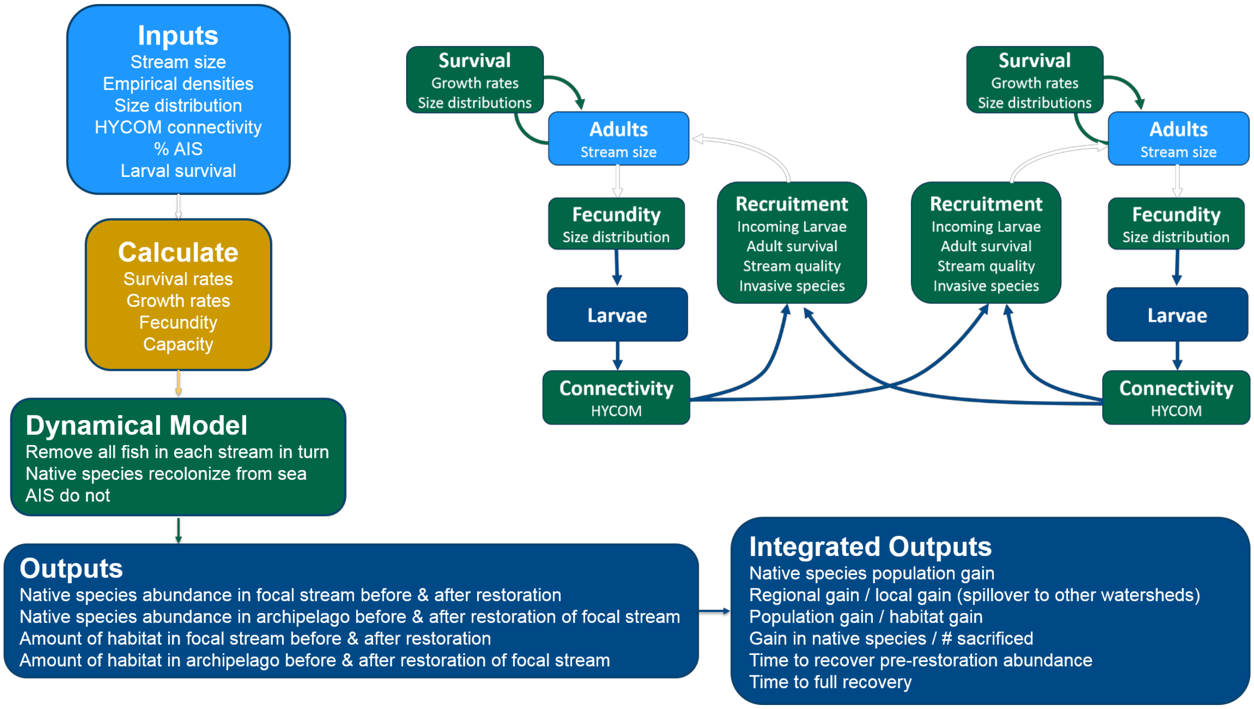


Figure 6: Conceptual framework and structure (inset) of the coupled ocean-watershed model implemented to explore local and regional outcomes of whole-watershed AIS removals across the Hawaiian Islands. Model inputs include empirically derived or estimates of watershed geomorphology, ocean circulation, stream biotic communities, and native species demography. Inputs are used to calculate demographic rates that serve as parameters for dynamical modeling, which outputs estimates of local and regional native species demography and habitat availability before and after AIS removal.

Although we parameterized the model using empirical data for our focal goby species (*A. stamineus*) from across the Hawaiian archipelago, we do not claim that the resulting parameter values match specific streams in Hawaiʻi. Rather, our goal was to capture the range of goby abundances, invadedness, and connectivity patterns that are present across the archipelago. As such, we do not report results for specific streams, but instead independently permute watershed carrying capacities and invadedness across locations in the HYCOM connectivity matrix to return probability distributions of outcomes at each of 51 stream locations (n = 1000 permutations).

We specified the dynamical model for subpopulations in each stream using Beverton-Holt competition (Beverton and Holt 1957), and simulated the resulting dynamics as they arise from in-stream interactions and dispersal between watersheds. We examined the resulting subpopulation sizes and dynamics to obtain local- and archipelago-scale outcomes of exterminating all invasive species from a single watershed. For simplicity, we assumed that the focal native species is also eliminated by incidental mortality during extermination, such that the new population of native species initially arises solely through propagule import from other subpopulations. However, the ideal implementation of clean-slate restoration includes sequestering many-to-all individuals of native species before extermination occurs (Fig. 1).

Modeling questions.

We applied the model to each of 51 Hawaiian streams to investigate the range of outcomes that occur when all fish, both native and introduced, are locally exterminated at one time. The system was then allowed to recover with no further intervention. We addressed five related questions to explore this phenomenon:

1. What range of changes occur in local subpopulation size?
2. What range of changes occur in aggregate metapopulation size across the archipelago?
3. How do archipelago- and local-scale changes compare?
4. What is the payoff in terms of native subpopulation size after *versus* before ‘clean slate’ eradication?
5. How long does it take each native species subpopulation to recover to its pre-eradication size?

For each of these questions, we were interested in determining how they depend on model parameters, such as local carrying capacity, invadedness, and connectivity. Thus we compared outcomes across sites that differ in these parameters to identify the kinds of restoration sites that provide the highest return on investment from eradicating invaders.

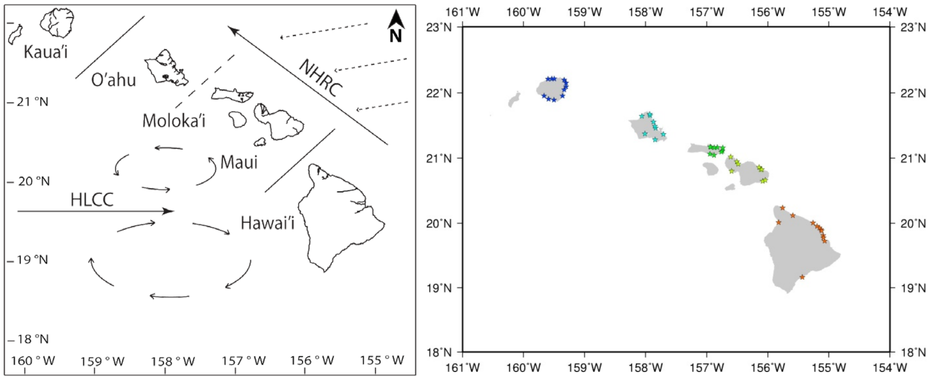


Figure 7: (Left) Map of the Hawaiian archipelago, with the streams (black lines) included in the study of ocean-stream connectivity. Solid arrows indicate major surface currents within the archipelago (the Hawaiian Lee Countercurrent and the North Hawaiian Ridge

Current including two circulations). Dashed arrows indicate the direction of prevailing winds. Lines separating the islands indicate the position of consensus (solid line) and putative (dashed line) genetic breaks for marine species within the archipelago (redrawn from Toonen et al. 2011). (Right) Stream mouth locations (stars) for release and settlement of virtual larvae across the watersheds included in the study of ocean-stream connectivity.

Implementing clean-slate restoration in the model.

Each simulation was run for 1000 generations to evaluate the effect on one native species of exterminating all invasive species from a single focal stream. After 500 generations at baseline conditions, both native and invasive species were extirpated by setting native subpopulation size to 0 and setting invader impact (I_x) to 0. Dynamics then proceeded for another 500 generations to observe subsequent outcomes.

The change in focal native subpopulation size ("local gain") resulting from local eradication of invasive species was calculated as the difference between means of pre-and post-eradication equilibria. During both time periods, subpopulation size was found to stabilize after 100 generations, so comparisons were between generations 101-500 versus 601-1000. Our simulations, and the resulting burn-in period before achieving equilibrium, did not account for temporal or spatiotemporal variability, which were beyond the scope of our analyses. The change in archipelago-wide metapopulation size ("archipelago gain") resulting from invasive species eradication in the focal stream was similarly calculated, based on the sum of subpopulation sizes across all 51 watersheds.

In some contexts (e.g. Fig. 1, Fig. 6), it is useful to evaluate the multiplicative change in the archipelago-wide metapopulation. We use "metapopulation multiplier" to refer to how many times larger the native metapopulation becomes post-eradication compared to pre-eradication.

We also calculate a "payoff ratio" for each simulation that reflects the archipelago gain by native species relative to the pre-eradication size of the local subpopulation. In the scenario where incidental mortality of native species arises from eradication efforts, the payoff ratio captures the large-scale demographic benefit versus the worst-case local cost of cleaning the slate. This multiplier deliberately considers the worst-case scenario should the local subpopulation in the focal site be extirpated, even though we advocate for avoiding that outcome during implementation of the 'clean-slate' strategy (Fig. 1). Provided that the payoff ratio is ≥1, 'clean-slate' restoration is assured of yielding a net increase in the native metapopulation.

Recovery time ("time lag before local gain") addresses how long it takes the subpopulation in the focal stream to rebound to its pre-eradication size. When 'clean-slate' restoration is accompanied by incidental mortality, recovery time quantifies a worst-case period of transient negative effects on the subpopulation. This is distinct from the time to achieving full restoration potential (i.e., the period between eradication and achieving maximum local subpopulation and archipelago metapopulation sizes).

Permuting stream conditions, connectivity, and recruitment limitation.

We were interested in exploring the full range of outcomes of eradications in order to understand what conditions make 'clean-slate' restoration most successful. We also wished to clearly determine what conditions could yield long-term detrimental impacts on native species. To do this, we simulated 'clean-slate' restoration in every watershed, respectively. Moreover, because we were interested in a range of realistic streams rather than sites selected *a priori*, we permuted stream carrying capacity and invadedness (see Appendix B for definitions) independently with respect to the connectivity matrix. In practice, this means that we established a permuted set of carrying capacity, invadedness, and connectivity values for every stream, then ran 51 individual simulations of exterminating invaders from a single stream. These sets of simulations were repeated for 1000 permutations of carrying capacity, invadedness, and connectivity. Furthermore, we ran each set of simulations with low and high levels of propagule survival (i.e., the probability that a goby larva would die at sea versus recruit to a stream). The high propagule survival rate was used to evaluate the implications of recruitment limitations for restoration success. For comparison, the low propagule survival rate was used to eliminate recruitment limitation. Each of the outcome variables from our simulations was calculated for every 'clean-slate' simulation (n = 102,000 simulations = simulations for 51 watersheds, two levels of propagule survival rates, and 1000 permutations of watershed attributes).

The resulting probability distributions of outcomes over all permutations allowed us to evaluate the robustness of outcomes from the 'clean-slate' approach. That is, we were able to evaluate the predictions from using estimates for each variable derived from observations at the 51 sites, but separately to permute those variable values to identify what general attributes of a restoration site (e.g., river size, invadedness, connectivity) are salient to achieve the maximum return on exterminating invasive species.

6 Results and Discussion

6.0 AIS REMOVAL FROM HAWAIIAN STREAMS

Stream hydrology.

Mean stream discharge spanned an order of magnitude, ranging from 0.084 m^3s^{-1} in Heʻeia stream to 0.785 m^3s^{-1} in Punaluʻu stream (Fig. 1). Streams also ranged in discharge variation. Stream discharge was less variable in watersheds known to receive more rainfall on the windward side of the island (e.g., Waianu, CV = 1.06; Punaluʻu, CV = 0.91) and in groundwater dominated streams (e.g., Keaʻahala, CV = 0.13). Conversely, leeward watersheds had more variable flows (e.g., Kīpapa, CV= 2.47; Waimea, CV = 4.29). Two leeward streams (Hālawa and Kīpapa) were ephemeral with stream channels that had intermittent dry channels for at least ~10% of the study period (Table 2, Q90).

AIS removals.

Prior to electrofishing, 283 native fish were hand netted (252 *A. stamineus*, 5 *E. sandwicensis*, and 26 *S. hawaiiensis*), safely sequestered, and returned to the stream after completion of all electrofishing passes. In comparison, 331 native fish (210 *A. stamineus*, 35 *E. sandwicensis*, 50 *S. hawaiiensis*, 33 *S. stimpsoni*, 3 *Kuhlia sp.*) were captured during electrofishing. Exposure to electrical current resulted in a ~2% mortality rate (i.e., 7 total mortalities consisting of 3 *E. sandwicensis,* 2 *A. stamineus*, and 2 *S. hawaiiensis*) (Table 4). All native fish mortality occurred during the second or third pass with pulsed current of ≥175 V at 60 Hz and 12% duty cycle (Table 4). Exposure to electrical current also resulted in a <2% non-lethal injury rate (i.e., 5 injuries, with injuries sustained by 3 *E. sandwicensis* and 2 *A. stamineus*). The only injury sustained from DC current occurred at 200 V; all other injuries occurred with pulsed current at 200 V, 60 Hz, and a duty cycle of 12% (Table 4).

Table 4. Mortality and injury records of native species with their recorded total length (TL) and mass during electrofishing with reference to the date and site where electrofishing was conducted, as well as the electrofishing pass and the corresponding instrument settings (DC = direct current, SP = standard pulse). ES = *Eleotris sandwicensis*, AS = *Awaous stamineus*, SH = *Stenogobius hawaiiensis*

Record	Site	Pass	Waveform	Volt (V)	Freq (Hz)	Duty Cycle	Species	TL (mm)	Mass (g)
Mortality	Waimea	3	SP	275	60	12%	*ES*	96	8.8
Mortality	Hālawa	3	SP	275	60	12%	*AS*	114	11.31
Mortality	Hālawa	3	SP	275	60	12%	*AS*	81	3.27
Mortality	Kāwā	2	SP	200	60	12%	*SH*	35	0.31
Mortality	Waimānalo	3	SP	200	60	12%	*ES*	99	9.17
Mortality	Waimānalo	2	SP	175	60	12%	*ES*	97	9.32
Mortality	Waimānalo	2	SP	175	60	12%	*SH*	72	2.69
Injury	Kāwā	2	SP	200	60	12%	*ES*	134	37.07
Injury	Kāwā	2	SP	200	60	12%	*ES*	106	13.55
Injury	Keaʻahala	2	DC	200	N/A	N/A	*AS*	145	25.38
Injury	Keaʻahala	3	SP	200	60	12%	*AS*	68	2.76
Injury	Waimānalo	3	SP	200	60	12%	*ES*	100	10.48

We captured a total of 25,782 AIS individuals across all electrofishing passes (Table 5), with ~66% of individuals being members of the two target groups (Loricariids *n* = 4,281; Poeciliids *n* = 12,797; other AIS *n* = 8,704). This is consistent with observations of greater pre-removal densities of Poeciliids compared to loricariid densities (mean ± SD: loricariids = 2.6 ± 3.3 m^{-2}, poeciliids = 3.6 ± 5.1 m^{-2}). Loricariids captured during electrofishing passes included *Ancistrus temmincki* and *Hypostomus watwata*. Poeciliids captured included *Poecilia sphenops*, *Xiphophorus helleri*, *Poecilia reticulata*, and *Gambusia affinis.* As expected, for all AIS and our target groups (Loricariids and Poeciliids), catch increased from the first to the second pass, then decreased in the third pass (Fig. 8). Size, measured as total length, of captured organisms was inversely related to the voltage ($F_{1,3482}$ = 8.632, P = 0.00332).

Discussion.

Despite AIS being consistently identified as a primary threat to native aquatic biodiversity in Hawaiian streams (e.g., Brasher 2003, Walter et al. 2012, Gagne et al. 2015, Gagne et al. 2018, Hain et al. 2019), to date there have been sparingly few efforts to develop methods to control AIS and to evaluate native species responses to management interventions (Nico and Walsh 2011). Past efforts to study mitigation and control have sometimes been constrained or denied approval in Hawaiʻi, despite State prioritizations to eliminate threats stemming from AIS (see Hawaiʻi Interagency Biosecurity Plan 2017-2027, https://dlnr.Hawaiʻi .gov/hisc/files/2017/02/Hawaiʻi -Interagency-Biosecurity-Plan.pdf). These restrictions partly stem from concerns that prospective methods to control AIS can potentially result in more harm than good. In some instances, these concerns have been codified, such as the moratorium that has been placed on electrofishing (Hawaiʻi Revised Statutes Section 118-23 and Hawaiʻi Administrative Rules Sections 13-75-6 and 187A-6), which is widely used for controlling AIS in continental streams (Murphy and Willis 1996). We were granted agency permission because, similar to other AIS control efforts in systems with species of ecological and cultural significance (Nico et al. 2015, McEwan and Crisp 2019), we proposed to assess an approach that prioritized the safety and well-being of native species. This study also warranted special consideration because it was intended to provide a thoroughly documented case-study illustrating how native species can be protected during efforts to control AIS in Hawaiian streams.

Most attempts of AIS removal in oceanic island freshwater ecosystems have focused on the use of chemical toxicants (e.g., rotenone) (Lintermans 2000; Pham et al. 2013; Nico et al. 2015) and have produced mixed outcomes (Nico and Walsh 2011). Even though rotenoid compounds are active ingredients in plants traditionally used by indigenous Hawaiian s and other Pacific Islanders for fishing (Barrau 1955, Rickard and Cox 1986, Armstrong et al. 2011), concerns have been raised about the possibility that rotenone use in streams could be detrimental to the biota in downstream coastal waterbodies (e.g., near-shore coral reefs). This has led to broad restrictions of its use, including in Hawaiʻi, where unpermitted release of chemical toxicants can be a criminal offense (Hawaiʻi Administrative Rules Sections 13-75-7).

While our study and those on other oceanic islands (Kwak and Cooney 2008) bypassed concerns about toxicants, we addressed similarly-minded concerns that electrofishing could result in widespread harm to native species. Our findings indicate that collateral harm can be contained by modifying use of electrofishing for stream specific conditions (e.g., pH, conductivity, depth, presence of sensitive species). Adjustments were made to the current form (unpulsed direct versus pulsed) as well as voltage and amperage. These considerations alongside logistical adjustments, such as completing a first pass with the instrument set to unpulsed direct current, enabled us to effectively remove AIS with significantly reduced exposure of native species to potentially harmful conditions (Fig. 8) (Whaley et al. 1978, Sharber et al. 1994, McMichael et al. 1998). Notably, we also reduced the potential for exposure by

Table 5. The species and number of individuals removed from each watershed during electrofishing (summed across all passes).

Species	Hālawa	He‘eia	Kahalu‘u	Kalihi	Kāwā	Kea‘ahala	Kīpapa	Mānoa	Punalu‘u	Waianu	Waikāne	Waimānalo	Waimea
Ancistrus temmincki		1571	825	42	57	821	3	303					
Hypostomus watwata					69			273			73	244	
Clarias fuscus	27		1				9	2	1		1	23	1
Gambusia affinus					5	219						19	
Poecilia sphenops	3927		40			80		19	1180	710	9	835	
Poecilia reticulata	1837	589	320	171		247	99	12		226	173	31	
Poecilia sp	213	24	58			1		1		153	11	558	
Xiphophorus helleri		268	106	1	1	265				168	209	12	
Micropterus dolomieui				14			1	20					
Archocentrus nigrofasciatus					29	18		21					
Hemichromis elongatus					4				1				2
Oreochromis sp												87	
Monopterus albus					2								
Misgurnus anguillicaudatus		16					4						
Macrobrachium lar	169	19		14			6		248	225	173	101	125
Neocaridina denticulata		5308	497	110		143	29	61	124				
Procambarus clarkii			32	8		99		21				9	
Bufo marinus	9				2	1	2					23	14
Rana catesbeiana	2	3	1									1	
Rana rugosa											1		
Unidentified tadpoles	5	4					191	15		4		481	

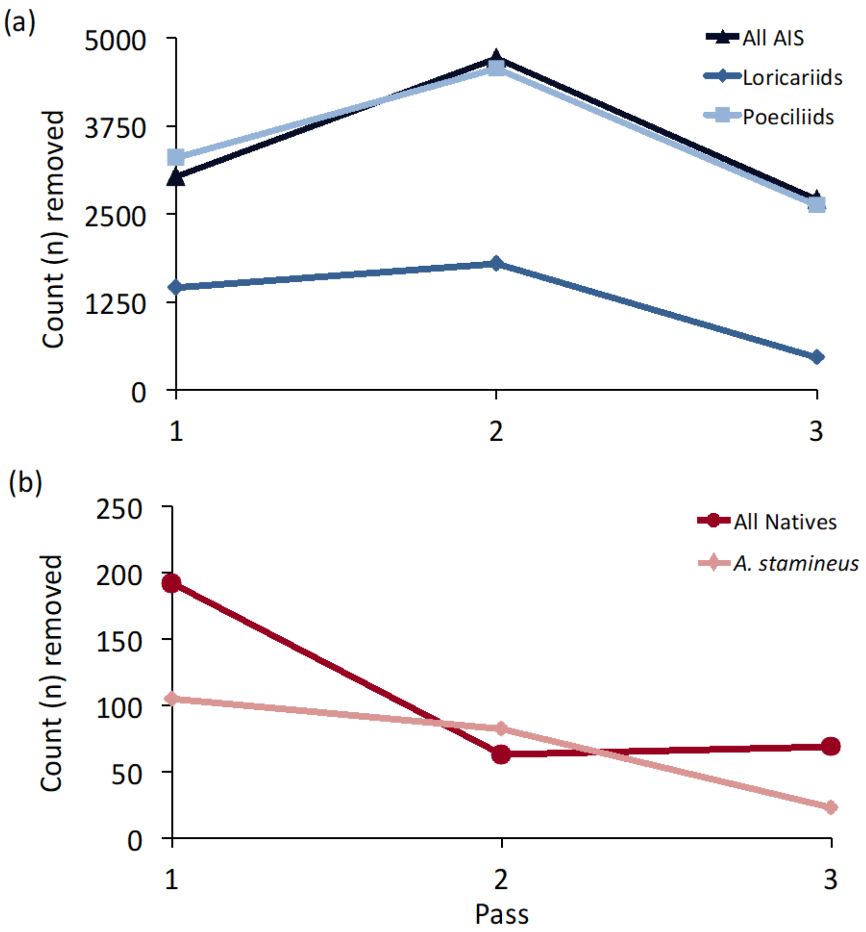


Figure 8. Number of (A) AIS and (B) native species removed from removal reaches during each electrofishing pass summed across all streams.

taking additional steps to safeguard species of concern. Akin to work done on AIS control in temperate oceanic island streams (Buktenica et al. 2013, Pham et al. 2013), we combined hand-netting and stream-side sequestration of native species with electrofishing tailored to reduce collateral harm. Our results indicate that incorporating protective controls can be implemented to reduce exposure risks. For example, hand-netting and sequestration of *A. stamineus* reduced exposure risk by >50%. Lower reductions were achieved for other species, however, with differences reflecting ease of capture, habitat use and densities. For instance, *E. sandwicensis* is a sit-and-wait ambush predator with cryptic coloration (Kido 1996), thus they are hard to detect and less likely to be captured (i.e., ~12.5% capture rate), particularly compared to *A. stamineus* (i.e., ~54% capture rate). It is notable that several quick and elusive native species (e.g., *S. stimpsoni* and *K. sandvicensis*) that were only caught during electrofishing (n = 33 and 3, respectively), suffered no harm from exposure to electrical current (Table 4). Nonetheless, taking steps to address potential differences in pre-removal capture rates will be key to achieving further risk reduction, especially for elusive species like *E. sandwicensis* that are highly sensitive to electrical current.

Contrary to reports that complex habitat characteristics limit the utility of electrofishing (Baker and Foster 1992, Nico and Walsh 2011), our findings demonstrate that it can be effective for controlling AIS in Hawaiian streams. We had little trouble executing electrofishing passes, encountering conditions

Table 6. Multiple linear regressions of immediate pre- to post-removal percent density changes (i.e., the day before and the day after AIS removal) with backward model selection for all AIS, Loricariids, and Poeciliids, respectively. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$

Response variable: % density change of taxonomic group	Full Model: ~ Mean discharge + (Mean discharge)^2 + Discharge variation + (Discharge variation)^2 + Pre-removal AIS density + + Invasive abundance removed + Invasive biomass removed										
	AIC	R^{2}adj	F	*P*	Reverse selected best model	AIC	R^{2}adj	F	*P*	Best model parameters	Coefficients
All AIS	78.57	0.51	2.19	0.14	Mean discharge + (Mean discharge)^2 + Pre-removal AIS density + Invasive abundance removed	74.74	0.64	6.36	0.01	Mean discharge	-316***
										(Mean discharge)^2	372***
										Pre-removal AIS density	-3.97**
										Invasive count removed	-0.01
Loricariids	90.63	0.44	2.33	0.18	Full Model	90.63	0.44	2.33	0.18	Mean discharge	213
										(Mean discharge)^2	-189
										Discharge variability	-6.97
										(Discharge variability)^2	1.70
										Pre-removal AIS density	-5.81
										Invasive abundance removed	0.019*
										Invasive biomass removed	-0.003
Poeciliids	88.54	0.33	1.854	0.28	Mean discharge + (Mean discharge)^2 + Discharge variability + Invasive abundance removed + Invasive biomass removed	86.43	0.45	2.95	0.09	Mean discharge	-423***
										(Mean discharge)^2	526***
										Discharge variability	1.70*
										Invasive abundance removed	-0.01
										Invasive biomass removed	0.01*

Table 7. Multiple linear regression models of the effects of mean discharge rate, discharge variability, and pre-removal AIS densities on the count and biomass removed during electrofishing for each invasive species group (All AIS, Loricariids, Poecillids) summed across all watersheds (n=13). * *p* < 0.05, ** *p* < 0.01, * *p* < 0.001**

Taxonomic group	Response variable	df	Model F	R^2adj	Model parameters	Parameter coefficient	t
All AIS	Count removed	3,35	17.73	0.57***	Mean discharge	-192.7	-0.15
					Discharge variability	-204.3	-0.83
					Pre-removal AIS density	335.5	6.31***
	Biomass removed	3,35	1.40	0.03	Mean discharge	1005.1	0.39
					Discharge variability	-254.4	0.62
					Pre-removal AIS density	211.8	1.93*
Loricariids	Count removed	3,9	6.65	0.56**	Mean discharge	- 368.0	-0.83*
					Discharge variability	-169.8	-1.95*
					Pre-removal AIS density	58.3	3.09**
	Biomass removed	3,9	0.27	-0.22	Mean discharge	-920.4	0.21
					Discharge variability	-422.8	-0.48
					Pre-removal AIS density	102.3	0.54
Poeciliids	Count removed	3,9	0.47	-0.15	Mean discharge	206.3	0.09
					Discharge variability	139.9	0.30
					Pre-removal AIS density	104.8	0.34
	Biomass removed	3,9	0.66	-0.09	Mean discharge	1486.5	1.17
					Discharge variability	-237.7	-0.96
					Pre-removal AIS density	46.0	0.41

typical to wadable streams in the archipelago (and conditions akin to those commonly found in continental streams). We did find, however, that complex habitat characteristics like steep elevation changes, pools, and large substrate types (e.g., boulders) can present challenges like increased difficulty of netting fish (Baker and Foster 1992, Mollenhauer et al. 2017). Despite these considerations, our results demonstrate that electrofishing can be effective so long as approaches are tailored to optimize capture and removal success, like what has been done in New Zealand and Puerto Rico (Hicks et al. 2007, Kwak and Cooney 2008).

We also found that electrofishing was highly effective for capturing species thought to be recalcitrant to electrofishing, including benthic catfish with plated armor and more mobile lotic species like Poeciliids (Nico and Walsh 2011). We found that both Loricariids and Poeciliids were susceptible to electrical current, which rendered them immobile long enough for capture and removal. However, susceptibility to electrofishing did differ among AIS taxonomic and functional groups and varied according to instrument settings and use (Table 5, Table 6, Table 7). For example, capture rates of Loricariid catfish were high when direct and pulsed current was delivered, whereas Poeciliids were responsive to pulsed current. The difference is likely a result of more electric current passing through Loricariids due to their larger body size (Zalewski 1983, Zalewski and Cowx 1990) and because of their benthic habit, which reduces the likelihood that the delivery of direct current will instigate movement beyond the range of potential exposure. Thus, usage of direct current is likely sufficient for targeted control of Loricariids, thereby reducing the risk to native species that have evaded hand-netting.

6.1 COMMUNITY RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Removal efficacy: did removal efforts elicit an immediate change in AIS and native species densities?

Biotic surveys conducted the day before and the day after AIS removals in each stream revealed differences among streams in the effect size of the removals. Most streams with Poecillids present showed a decrease in Poeciliid density immediately after AIS removal, however, the percent difference varied by two orders of magnitude (Fig. 9). Hālawa stream showed the greatest before-after difference of -13.43 m^{-2} (59% decrease). Mānoa stream showed a slightly positive difference of 0.13 m^{-2} indicating a very small (4%) increase in density, the only stream to do so. Loricariid densities also typically declined after AIS removal with a similar degree of variation among streams as the Poecillids. The largest before-after difference was found in Kahaluʻu stream (-12.3 m^{-2}; 53%). Kalihi stream exhibited a 22% increase in density after removals (before-after difference = 0.26 m^{-2}).

Measures of percent density changes (((post-removal – pre-removal) / pre-removal) *100) demonstrated that electrofishing immediately reduced overall AIS densities (t = 2.71, *p* = 0.018), Loricariid densities (t = 2.55, *p* = 0.02), and Poeciliid densities (t = 2.17, *p* = 0.049), compared to the control reaches (Fig. 9). Removal events did not reduce total native species densities compared to control reaches (t = 1.63, *p* = 0.12) nor were densities of *A. stamineus* reduced in the removal reaches compared to the control reaches (t = 0.80, *p* = 0.44).

Removal efficacy: did AIS removal differ according to hydrology and pre-removal densities?

The number of individuals removed varied by more than an order of magnitude among streams from n = 142 in Waimea stream to n = 7,802 in Heʻeia stream (Table 5). Biomass removed also varied by an order of magnitude among streams from 1,286.4 g in Kīpapa stream to 13,347.4 g in Mānoa stream. AIS capture varied according to discharge and pre-removal AIS densities (Table 6, Table 7). The number of individuals or biomass of all AIS removed and Poeciliid individuals removed did not reflect mean discharge or discharge variation (Table 6, Table 7). The number of Loricariid individuals captured was greater in streams with lower mean discharge and less variation in discharge (Fig. 10, Table 6, Table 7), whereas percent density change of Loricariids did not reflect stream discharge (Fig. 10, Table 6, Table 7). In contrast, percent density change of all AIS and Poeciliids was greatest in streams with intermediate mean discharge (range 0.24-0.55; Fig. 10). We found that percent density change of Poeciliids reflected discharge variability, where the percent density change was greater in streams with greater discharge variability (Fig. 10, Table 6, Table 7). We also found that there was a smaller overall percent reduction in streams with greater pre-removal AIS densities (Fig. 10, Table 6, Table 7) despite a positive relationship between pre-removal AIS densities and the number of all AIS and Loricariids removed (Table 6, Table 7). Similarly, there were smaller percent density changes in both Loricariids and Poeciliids in streams where there were more AIS individuals or a greater biomass of AIS removed (Fig. 10, Table 6, Table 7).

Post-removal re-assembly: Were temporal responses to AIS removals ephemeral or persistent?

AIS removals resulted in long-lasting shifts in species densities, indicated by a significant BACI effect. We detected a significant period x reach interaction (F = 3.81, *P* = 0.02; Table 8, Table 9), reflecting a persistent reduction after removal events of AIS density in removal reaches (β = -4.07, t = 4.6, *p* < 0.0001) and downstream reaches (β = -2.13, t = 2.42; *p* = 0.02), but not upstream reaches (β = -0.66; t = 0.76; *p* = 0.45) (Fig. 11). We detected an overall pattern of persistent reduction in Loricariid densities (F = 12.00, *p* < 0.001; Fig. 11), yet we did not find a significant period x reach interaction for Loricariid densities (F = 2.42; *p* = 0.09). However, post-hoc tests showed that Loricariid density were significantly lower in removal reaches (β = -1.36; t = 3.73; *p* < 0.001), with no change in density detected in either control reach. An overall pattern of reduction was also found for Poeciliid densities (F = 19.22, *p* < 0.0001; Fig. 11). Similar to Loricariid densities, we did not find a significant period x reach interaction for Poeciliid densities (F = 1.33; *p* = 0.27), but post-hoc tests showed that Poeciliid densities were

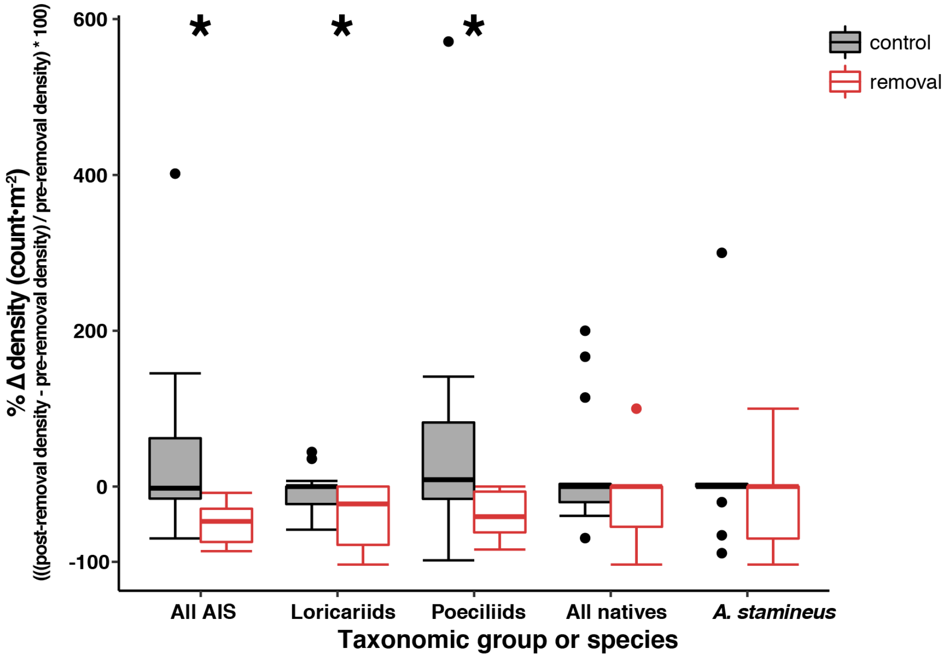


Figure 9: The immediate percent density change (((post-removal density – pre-removal density)/pre-removal density) *100) for each group of interest between removal reaches (red) and control reaches (grey). Whiskers are minimum and maximum values relative to the first and third quartiles, respectively, whereas median values are denoted with bars. * = significant difference in percent density changes between reaches.

significantly lower in removal reaches (β = -1.78; t = 3.74, *p* < 0.001), as well as in the downstream control reach (β = -1.16; t = 2.42; *p* = 0.02). Likewise, we found evidence of density reductions for individual species within our targeted taxonomic groups (e.g., *A. temminki*; *H. watawata*, *P. reticulata*, *P. sphenops*) as well as some non-target species (Table 8, Table 10, Table 11).

There were no significant changes in overall native species densities resulting from AIS removals (F = 1.43, *P* = 0.23; Fig. 11). There was no significant period x reach interaction and post-hoc tests showed that densities were not reduced in control or removal reaches (Table 8). Similarly, there were no significant changes to *A. stamineus* densities resulting from AIS removals (F = 1.03, *P* = 0.31; Fig. 11, Table 8); there was no significant period x reach interaction and post-hoc tests showed that densities were not reduced in control or removal reaches.

The GLS models of density changes across each cycle showed that AIS density reductions persisted and revealed that changes in density were related to environmental and biotic covariates (Fig. 11, Table 9). AIS densities remained lower over the duration of the post-removal study period (F = 3.58; *p* < 0.0001). We did not find a significant overall reach x cycle interaction, but post-hoc tests showed that AIS densities remained lower in removal reaches in later cycles (12-27) compared to early cycles (1-4). This pattern was not detected in either of the control reaches, though there were differences in AIS density in very early cycles (1-2) compared to very late cycles (21-27). Not surprisingly post-removal AIS densities were positively correlated with pre-removal AIS densities (β = 0.34; F = 25.71; *p* < 0.0001). Conversely, native species densities in the removal and control reaches were not affected by removal events (cycle: F = 1.44; *P* = 0.07; reach*cycle: F = 0.64; *P* = 0.98; Fig. 11).

While both Loricariid and Poeciliid densities remained lower over the duration of the post-removal study period (cycle: F = 2.31; *p* = 0.0002; F = 2.75; *p* < 0.0001, respectively; Fig. 11), constituent species did not respond the same way to removal events. For example, *A. temmincki* densities remained lower over time, but *H. watawata* densities did not (Fig. 11, Table 8). Likewise, both *P. reticulata* and *P. sphenops* densities remained lower, whereas *X. helleri* densities did not (Fig. 11, Table 8).

Among native species, only *A. stamineus* densities varied over time (Fig. 11, Table 9). Post-hoc tests revealed that *A. stamineus* densities initially increased in the removal reaches following AIS removal (from cycle 9 to 10: β = 0.18; t = 3.63; *p* = 0.05, and from 9 to 11: β = 0.29; t = 4.52; *p* = 0.002), decreased from cycle 11 to 12 (β = -0.18; t = -3.87; *p* = 0.02), and then stabilized around the average pre-removal density (Fig. 11). No significant changes in *A. stamineus* densities were found in either control reach.

We detected a persistent overall shift in community composition across time, which differed in magnitude according to reach and community type, as well as discharge and the number of AIS individuals removed by electrofishing (Fig. 12, Table 12). Changes in whole community dissimilarity also were largely reflective of changes in the AIS community (Fig. 12). Post-hoc analyses revealed a significant increase in dissimilarity for the whole community between early cycles (1-8) and later cycles (12 to 27) across all reaches. Despite no overall significant reach x cycle interaction (F = 0.95; *p* = 0.58), post-hoc tests also showed there was an increase in dissimilarity between cycles 11 and 12 for the removal reach (β = 0.23; t = 3.96; *p* = 0.03). This pattern was evident for the AIS community as well (Fig. 12, Table 12), with an increase in dissimilarity between cycles 11 and 12 in the removal reach (β = 0.24 t = 4.01, *p* = 0.02) in the absence of an overall significant reach x cycle interaction (F = 0.95, *p* = 0.57). The GLS models revealed that whole and AIS community dissimilarity increased with higher mean discharge (whole: β = 0.08, t = 3.65, *p* < 0.001; AIS: β = 0.08, t = 3.48, *p* < 0.001). There also was evidence that

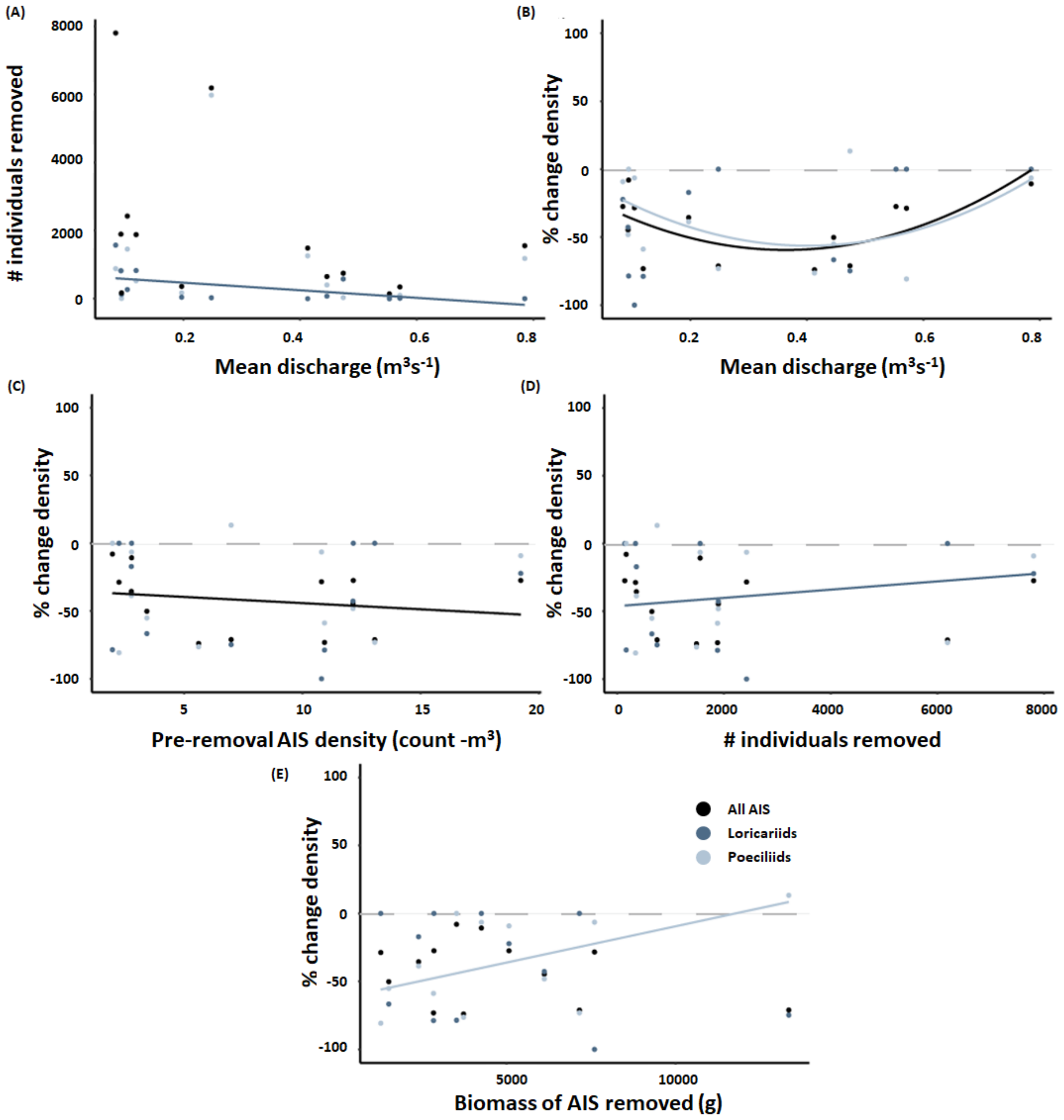


Figure 10: Metrics of removal efficacy (i.e., count, percent density change) versus hydrology and pre-removal densities. (a) Variation in the number of AIS (dark blue), loricariids (medium blue), and poeciliids (light blue) captured relative to mean discharge conditions (m^3s^{-1}); (b-e) Variation in percent density change (((post-removal density – pre-removal density)/pre-removal density) *100) of AIS, loricariids, and poeciliids versus mean discharge (m^3s^{-1}), pre-removal AIS density, the number of AIS removed, and the biomass of AIS removed. Only significant relationships are represented with linear or non-linear trend lines. The grey dashed line = no change in density.

Table 8. Results of generalized least squares models of pre- to post-removal changes in targeted taxonomic groups and constituent species densities over the full duration of the study.

Taxonomic group or species	Non-native or native	Parameter	df	F	*P*
All AIS	Non-native	Period	1, 788	20.45	< 0.0001
		Reach	2, 788	0.50	0.61
		Period*Reach	2, 788	3.81	0.02
Loricariids	Non-native	Period	1, 788	11.99	< 0.001
		Reach	2, 788	0.36	0.7
		Period*Reach	2, 788	2.42	0.09
Poeciliids	Non-native	Period	1, 788	19.22	< 0.0001
		Reach	2, 788	0.50	0.61
		Period*Reach	2, 788	1.33	0.27
A. temmincki	Non-native	Period	1, 662	7.65	0.01
		Reach	2, 662	0.09	0.92
		Period*Reach	2, 662	1.23	0.29
H. watwata	Non-native	Period	1, 486	7.51	0.01
		Reach	2, 486	1.64	0.19
		Period*Reach	2, 486	2.62	0.07
G. affinis	Non-native	Period	1, 144	1.83	0.18
		Reach	2, 144	0.67	0.50
		Period*Reach	2, 144	0.46	0.63
P. sphenops	Non-native	Period	1, 666	7.42	0.01
		Reach	2, 666	0.46	0.63
		Period*Reach	2, 666	0.09	0.91
P. reticulata	Non-native	Period	1, 644	12.78	< 0.001
		Reach	2, 644	0.23	0.80
		Period*Reach	2, 644	0.30	0.73
X. helleri	Non-native	Period	1, 603	5.82	0.02
		Reach	2, 603	1.67	0.19
		Period*Reach	2, 603	1.40	0.25
All native species	Native	Period	1, 788	1.43	0.23
		Reach	2, 788	0.99	0.37
		Period*Reach	2, 788	0.24	0.79
A. stamineus	Native	Period	1, 746	1.03	0.31
		Reach	2, 746	1.55	0.21
		Period*Reach	2, 746	0.42	0.65

Table 9. Results of generalized least squares models of cycle-to-cycle variation in targeted taxonomic groups and constituent species densities over the full duration of the study.

		Best forward selected model				
Taxonomic group or species	Non-native or native	AIC	Best model parameters	df	F	*P*
All AIS	Non-native	4087	Reach	2, 706	1.65	0.19
			Cycle	27, 706	3.58	< 0.0001
			Reach*Cycle	54, 706	0.72	0.94
			Mean discharge	1, 706	1.1	0.29
			Discharge variation	1, 706	3.68	0.06
			AIS abundance removed	1, 706	3.61	0.06
			Pre-removal AIS density	1, 706	25.71	< 0.0001
Loricariids	Non-native	2665	Reach	2, 708	0.12	0.89
			Cycle	27, 708	2.31	0.0002
			Reach*Cycle	54, 708	0.88	0.71
			AIS abundance removed	1, 708	4.02	0.45
			Pre-removal AIS density	1, 708	7.79	0.01
Poeciliids	Non-native	3160	Reach	2, 708	2.21	0.11
			Cycle	27, 708	2.75	< 0.0001
			Reach*Cycle	54, 708	0.64	0.98
			Discharge variation	1, 708	13.05	0.0003
			AIS abundance removed	1, 708	25.74	< 0.0001
A. temmencki	Non-native	2202	Reach	2, 582	0.01	0.99
			Cycle	27, 582	2.04	0.002
			Reach*Cycle	54, 582	0.81	0.84
			AIS abundance removed	1, 582	95.79	< 0.0001
			Pre-removal AIS density	1, 582	8.44	0.004
H. watawata	Non-native	-466	Reach	2, 408	0.38	0.68
			Cycle	27, 408	1.46	0.07
			Reach*Cycle	54, 408	0.78	0.86
			Mean discharge	1, 408	6.70	0.01
			AIS biomass removed	1, 408	105.34	< 0.0001
			Pre-removal AIS density	1, 408	24.11	< 0.0001
G. affinis	Non-native		Reach	2, 66	0.00	1.00
			Cycle	27, 66	4.35	< 0.0001
			Reach*Cycle	54, 66	4.77	< 0.0001
P. sphenops	Non-native		Reach	2, 586	1.65	0.19
			Cycle	27, 586	2.33	0.0002
			Reach*Cycle	54, 586	0.46	0.99
			AIS abundance removed	1, 586	14.98	< 0.0001
			Pre-removal AIS density	1, 586	8.76	0.003
P. reticulata	Non-native		Reach	2, 636	6.20	0.002
			Cycle	27, 636	2.07	0.001

			Reach*Cycle	54, 636	0.79	0.86
			AIS abundance removed	1, 636	43.22	< 0.0001
			Pre-removal AIS density	1, 636	39.34	39.34
X. helleri	Non-native		Reach	2, 524	14.91	< 0.0001
			Cycle	27, 524	1.97	0.003
			Reach*Cycle	54, 524	0.56	0.99
			Pre-removal AIS density	1, 524	5.47	0.02
All native species	Native	-389	Reach	2, 708	0.14	0.87
			Cycle	27, 708	1.44	0.07
			Reach*Cycle	54, 708	0.64	0.98
			Mean discharge	1, 708	3.88	0.05
			AIS biomass removed	1, 708	5.98	0.01
A. stamineus	Native	-596	Reach	2, 668	0.15	0.86
			Cycle	27, 668	1.90	0.004
			Reach*Cycle	54, 668	0.88	0.70

dissimilarity declined in streams where more AIS individuals were removed, but the effect sizes were small (whole: β = -0.00002, t = -3.4, $p < 0.001$; AIS: β = -0.00002, t = -3.38, $p < 0.001$).

Native species community dissimilarity also changed across time, but not between reaches. As with the whole and AIS communities, we did not find a significant reach x cycle interaction (Table 12). Unlike what was found for whole and AIS communities, however, post-hoc analyses did not detect differences between cycles across all reaches or for different reaches. Despite a seemingly stochastic pattern of variation, the GLS models (Table 12) revealed that native community dissimilarity was higher with greater discharge variability (β = 0.22, t = 4.68, $p < 0.001$) and in streams where more AIS biomass was removed, though the effect size was small (β = 0.00001, t = 2.89, $p = 0.004$).

Discussion.

We found that a combination of pre-removal trapping and sequestration of native species with a tailored 3-pass depletion electrofishing protocol resulted in immediate reductions in AIS biomass and abundance without disrupting population densities of native species. AIS removal also resulted in a sustained reduction in overall AIS density and a shift in community composition reflective of reduced densities of target AIS species, despite differences in removal efficacy across AIS taxonomic groups, AIS prevalence, and hydrological regimes. In contrast, overall native species densities did not decrease overtime. These findings indicate that electrofishing can be a valuable tool for AIS management in tropical and subtropical Pacific island streams, so long as steps are taken to reduce risk to native species. Our results also suggest that careful consideration of surface flow and pre-removal biotic conditions can help maximize desirable outcomes of removal efforts.

AIS removal efficacy. Notably, our results indicate that removal efficacy varied according to hydrology, with greater AIS density reductions achieved in streams exhibiting mid-range mean discharge conditions (Fig. 10). The observed variation in electrofishing efficiency is consistent with prior work showing that capture success (Hense et al. 2010, Mollenhauer et al. 2017) and consequently AIS reductions vary

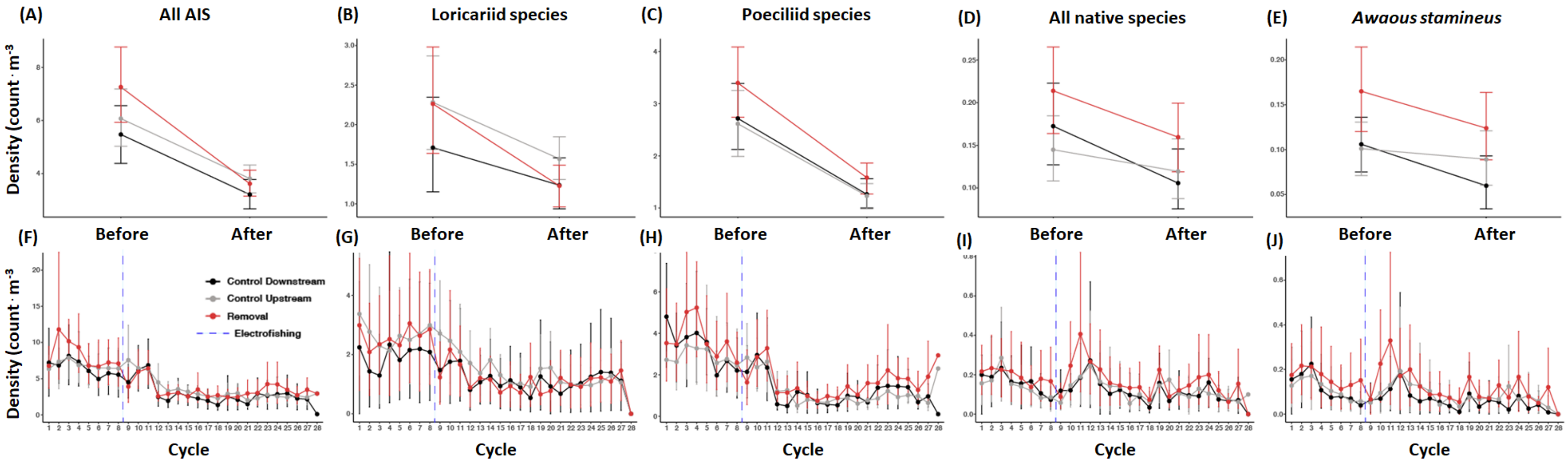
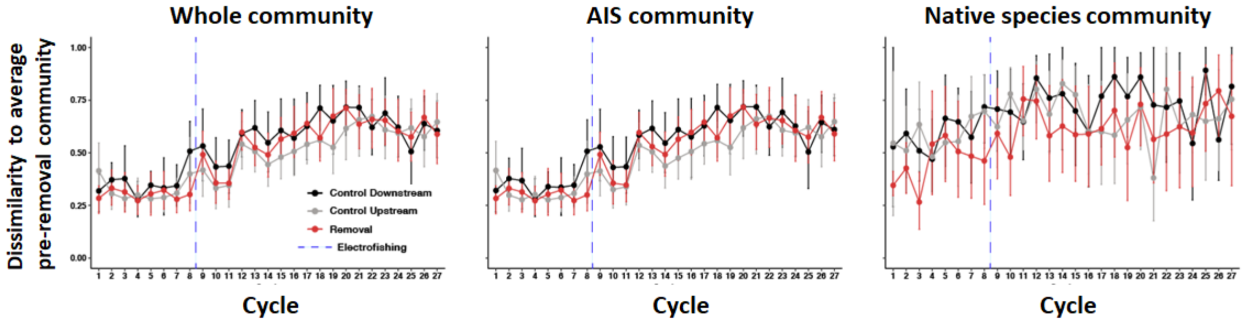


Figure 11: (Top) Reaction norm plots of changes in density for the full duration of the study between periods (pre-removal, post-removal) across each reach (red = removal, black = control downstream, grey = control upstream): (a) All AIS, (b) Loricariid species, (c) Poeciliid species, (d) All native species, and (e) *A. stamineus*. (Bottom) Mean and SE plots of long-term changes in density for each reach according to snorkel survey cycle: (f) All AIS, (g) Loricariid species, (h) Poeciliid species, (i) All native speices, and (j) *A. stamineus*. The whiskers are standard errors, and mean values are denoted with dots. The blue dashed line denotes when the AIS removal event occurred.



Whole community

AIS community

Native species community

Figure 12: Mean and SE of long-term changes in community dissimilarity across each post-removal snorkel survey cycle for each reach (red = removal, black = control downstream, grey = control upstream) for the whole community, AIS community, and native species community. The whiskers are standard errors, and mean values are denoted with dots. The blue dashed line denotes when the AIS removal event occurred.

among streams (Kolar et al. 2010). The observed pattern in efficacy reflects something of a "Goldie Locks" scenario for AIS removal. That is to say- electrofishing might be relatively easier to conduct in low discharge streams, but AIS might nonetheless evade capture as a result of being present in relatively high densities (Fig. 13), and conversely the physical challenges of electrofishing in high discharge streams reduce the likelihood of capturing AIS despite potentially lower densities or abundance. Accordingly, removal efforts might elect to first target streams with intermediate discharge as a foundation for broader campaigns to control AIS. Likewise, our results suggest that prioritization should consider AIS prevalence and densities relative to discharge conditions. For example, it may very well be possible to remove more Loricariids in low discharge streams (Fig. 10) without achieving a proportionate reduction in density relative to density reductions that can be achieved in mid-range discharge streams. Similarly, efforts might result in the removal of the same number of Poeciliids across streams with different discharge regimes (Fig. 10), with the greatest density reductions being achieved in mid-range discharge streams.

Table 10. Results of generalized least squares models for non-target species densities over the entire duration of the study categorized as densities before or after AIS removal (i.e., long-term, longitudinal response).

Taxonomic group or species	Non-native or native	Parameter	df	F	*P*
E. sandwicensis	Native	Period	1, 294	0.13	0.72
		Reach	2, 294	2.89	0.06
		Period*Reach	2, 294	0.28	0.76
S. hawaiiensis	Native	Period	1, 441	0.69	0.41
		Reach	2, 441	2.01	0.14
		Period*Reach	2, 441	0.31	0.73
S. stimpsoni	Native	Period	1, 414	2.42	0.12
		Reach	2, 414	0.32	0.73
		Period*Reach	2, 414	0.57	0.57
A. bisulcata	Native	Period	1, 336	3.46	0.06

		Reach	2, 336	1.16	0.32
		Period*Reach	2, 336	0.33	0.72
K. sandvicensis	Native	Period	1, 291	0.02	0.89
		Reach	2, 291	0.44	0.65
		Period*Reach	2, 291	0.40	0.67
M. grandimanus	Native	Period	1, 426	7.66	0.01
		Reach	2, 426	1.16	0.31
		Period*Reach	2, 426	0.59	0.55
Bufo marinus	Non-native	Period	1, 666	1.24	0.27
		Reach	2, 666	0.38	0.69
		Period*Reach	2, 666	0.29	0.75
Clarias fuscus	Non-native	Period	1, 165	0.89	0.35
		Reach	2, 165	2.27	1.01
		Period*Reach	2, 165	0.89	0.41
Corbicula fluminea	Non-native	Period	1, 225	4.99	0.03
		Reach	2, 225	0.17	0.85
		Period*Reach	2, 225	1.00	0.37
Macrobrachium lar	Non-native	Period	1, 629	2.17	0.14
		Reach	2, 629	1.59	0.21
		Period*Reach	2, 629	0.28	0.76
Melanoides tuberculatus	Non-native	Period	1, 788	1.92	0.17
		Reach	2, 788	0.06	0.94
		Period*Reach	2, 788	0.01	0.99
Neocaridina denticulata sinensis	Non-native	Period	1, 695	1.16	0.23
		Reach	2, 695	0.21	0.81
		Period*Reach	2, 695	5.58	0.01
Procambarus clarkii	Non-native	Period	1, 644	2.11	0.15
		Reach	2, 644	0.29	0.74
		Period*Reach	2, 644	0.01	0.99

Persistent outcomes of AIS removal. We expected rapid recolonization and return to pre-removal conditions following single time point removals of AIS from our 100 m study reaches. No barriers to movement were present to limit recolonization, thus we expected that AIS present outside of the removal reaches would readily move into areas with few if any fish present (Koehn et al. 2000). We instead found that removals resulted in long-standing reductions in AIS densities and concomitant shifts in community composition (Simberloff 2011). These results suggest that removals restructured local community dynamics by shifting AIS, Loricariid and Poeciliid species densities (Fig. 11, Fig. 12). However, it is possible that some of the observed shifts in community dynamics across reaches and watersheds could also potentially reflect seasonal (e.g., rainfall) and climatic variation (e.g., El Niño-Southern Oscillation cycle) not quantified in this study.

We detected temporal shifts in community dissimilarity corresponding to AIS removals in the study reaches (Fig. 12). Community dissimilarity increased from pre-removal to post-removal conditions across all reaches, however there was a shift of increased community dissimilarity between cycles 11-12 in the removal reaches with sustained dissimilarity thereafter. This pattern is consistent with the premise that AIS removals create ecological 'voids' (i.e., areas where resources become newly available), which are then 'filled' by individuals moving in from outside of removal reaches (Koehn et al. 2000), resulting in the establishment of an new equilibrium community structured by lower overall densities in the removal and control reaches.

Table 11. Results of generalized least squares models for non-target species densities over the entire duration of the study categorized by cycle.

Response Variable: density	Best forward selected model AIC	Best model parameters	df	F	*P*
E. sandwicensis	-1360	Reach	2, 214	3.87	0.02
		Cycle	27, 214	1.38	0.11
		Reach*Cycle	52, 214	1.05	0.39
		AIS biomass removed	1, 214	21.15	<0.0001
		Pre-removal AIS density	1, 214	9.56	0.002
S. hawaiiensis	-1307	Reach	1, 360	2.81	0.06
		Cycle	2, 361	1.01	0.46
		Reach*Cycle	54, 361	0.66	0.97
		Pre-removal AIS density	1, 361	29.2	<0.0001
S. stimpsoni	-1478	Reach	2, 335	0.07	0.93
		Cycle	27, 335	1.28	0.16
		Reach*Cycle	54, 225	0.54	0.99
		Pre-removal AIS density	1, 225	10.43	0.001
A. bisulcata	-1523	Reach	2, 257	0.78	0.46
		Cycle	27, 257	2.64	<0.0001
		Reach*Cycle	54, 257	0.94	0.59
		Pre-removal AIS density	1, 257	7.07	0.01
K. sandvicensis	-725	Reach	2, 211	0.64	0.53
		Cycle	27, 211	1.28	0.17
		Reach*Cycle	54, 211	0.94	0.6
		AIS biomass removed	1, 211	24.04	<0.0001
		Pre-removal AIS density	1, 211	13.82	0.0003
M. grandimanus	-1954	Reach	2, 347	0.78	0.46
		Cycle	27, 347	1.26	0.18
		Reach*Cycle	54, 347	0.39	1.00
		Pre-removal AIS density	1, 347	18.15	<0.0001

The observed pattern might also (i.e., instead of, or in combination with) be reflective of a demographic collapse perhaps driven by Allee effects, where the local populations were culled below a density threshold, thus resulting in sustained suppression of population size and growth (Taylor and Hastings 2005, Potapov and Lewis 2008, Tobin et al. 2011). An Allee effect is plausible, as it is likely that larger reproductive individuals were disproportionately removed from the local populations (i.e., due to greater sensitivity to current exposure and ease of capture), leaving smaller non-reproductive individuals within the removal reach and reducing overall potential reproduction across the removal and control reaches (Sabaj et al. 1999, Carvahlo et al. 2009) due to reduced interactions.

Notably, we did not find evidence of unanticipated and undesirable effects of AIS removals on at-risk native species (Fig. 11, Fig. 12). AIS control can result in unexpected or unwanted secondary outcomes, like ecological release of non-target invasive species, which can prevent recovery or trigger declines in native species of concern (Zavaleta 2002, Morrison 2007, Campbell et al. 2011). It can also shift trophic interactions that prove unfavorable to native species. For example, removing invasive prey species might increase consumption of native species by native and invasive predators (Zavaleta et al. 2001).

Table 12. Results of the generalized least squares models of cycle-to-cycle community dissimilarity for all species, all AIS, and all native species, respectively over the full duration of the study.

Response Variable: dissimilarity	AIC	Best model parameters	df	F	*P*
Whole community	-541	Reach	2, 683	2.10	0.12
		Cycle	26, 683	4.34	< 0.0001
		Reach*Cycle	52, 683	0.95	0.58
		Mean discharge	1, 683	13.34	0.0003
		AIS abundance removed	1, 683	11.55	0.001
AIS community	-513	Reach	2, 683	2.07	0.13
		Cycle	26, 683	4.28	< 0.0001
		Reach*Cycle	52, 683	0.95	0.57
		Mean discharge	1, 683	12.14	0.001
		AIS abundance removed	1, 683	11.44	0.001
Native community	292	Reach	2, 682	1.05	0.35
		Cycle	26, 682	1.55	0.04
		Reach*Cycle	50, 682	1.05	0.39
		Mean discharge	1, 682	24.09	<0.0001
		AIS abundance removed	1, 682	1.63	0.20
		AIS biomass removed	1, 682	6.34	0.01

Removing invasive species might also increase infection of native species by invasive parasites (Gagne et al. 2016). Yet we found that average densities of *A. stamineus* remained relatively unchanged throughout the study, with a few notable exceptions. For example, we found that densities temporarily increased in removal reaches during a ~3 month period immediately following AIS removals. It is possible that removals may have resulted in greater recruitment of juvenile *A. stamineus* into removal reaches, as prior work suggests that removals could have temporarily elevated survival of recruits by increasing resource availability (Lisi et al. 2018). Likewise, a pulse of recruitment might have occurred because AIS removals alleviated predation and competition pressures that manifest as "migratory gauntlets" (Hain et al. 2019). Careful demographic analyses (e.g., via mark-recapture) could shed further light on the processes underlying the observed increase in *A. stamineus* (Hain et al. 2016).

Adoption and broader applications. It is broadly acknowledged that AIS are a major and growing threat to the endemic biodiversity of oceanic islands across the Pacific (Molnar et al. 2008, Boykin et al. 2012, Moody et al. 2017, Champion 2018, Lisi et al. 2018). Increasingly difficult challenges lie ahead for prevention, management, and mitigation of AIS as global travel and transport of goods continue to escalate. While complete eradication of AIS is often the desired endpoint, it is rarely feasible because of cost and logistical constraints (for example, see Report to Twenty-Ninth Legislature, State of Hawaiʻi, 2018 Regular Session, Budgetary and Other Issues Regarding Invasive Species). A pragmatic compromise then is to execute well-conceived control and mitigation efforts to (re)establish ecological functionality in order to promote the recovery of native species or to boost valued ecosystem services (Walsh et al. 2016).

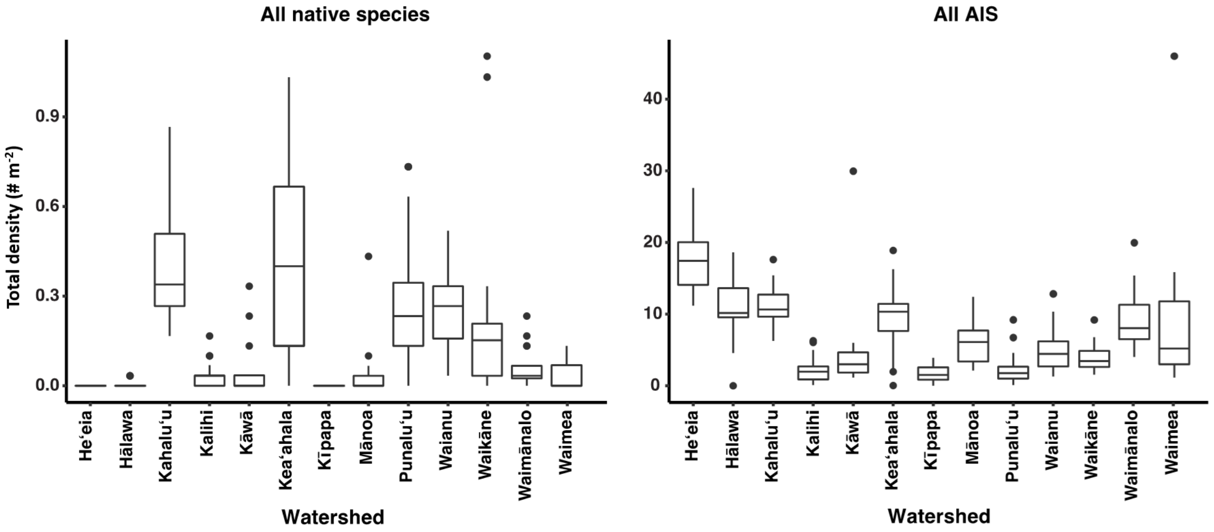


Figure 13. Whisker box plot of variation in pre-removal native and AIS species abundances in the removal reach of each study watershed.

Our findings demonstrate that localized and targeted AIS removals can establish a beachhead for the pursuit of more ambitious control efforts. Our work showed that AIS can be controlled with little collateral harm to native species of concern, illustrating that localized AIS control (following protocols that prioritize the welfare of native species) could bolster larger-scale restoration efforts. Notably, this study demonstrates that even one removal event can trigger immediate and sustained suppression of targeted AIS, with little adverse consequences for native species and communities (i.e., low mortality and injury rates, stable or increasing post-removal densities). This suggests that greater benefits could be derived from a well-timed series of removals, provided no subsequent introductions occur (e.g., via aquarium releases, fish stocking, biological control, etc.) (Kulp and Moore 2000). Similarly, combining AIS removal with habitat restoration could ease the pressures imposed by migratory gauntlets, thus promoting emigration and immigration of native species (Hain et al. 2019). Our work also suggests that pursuing other aspirational goals, such as reducing AIS densities across entire streams (i.e., across lengths much greater than 100 m removal reaches) is feasible, particularly in streams with features that provide for favorable logistics (e.g., wadable depths) and that constrain potential recolonization (e.g., terminal waterfalls). Because the native fauna are amphidromous, achieving more aspirational goals like whole-watershed control could deliver benefits that not only bolster local populations but also elevate range-wide persistence by increasing the overall size of propagule pools and thus the potential for recruitment across the archipelago. Understanding the potential for local and range-wide outcomes might thus be a pertinent element of decision-making (e.g., determining the greatest return on investment) (Laplanche et al. 2018), which could be well supported with coupled ocean-watershed models that estimate demographic outcomes of connectivity and life history variation (e.g., Moody et al. 2019).

Despite the many challenges, controlling invasive species is vital to protecting the biological legacies that are unique to tropical and subtropical oceanic islands. In 2001, the Hawaiʻi legislature passed a resolution stating, “the silent invasion of Hawaiʻi by alien invasive species is the single greatest threat to Hawaiʻi’s economy, natural environment, and the health and lifestyles of Hawaiʻi’s people and visitors,” (Senate Concurrent Resolution 45, H.D.1 [2001]). It is thus prudent to remember that streams and other vulnerable ecosystems do not occur in isolation but rather fall within a larger socioeconomic context. Accordingly, real or perceived health risks to residents, their pets and livestock, and native species may lead to restricted use of some eradication tools. It is therefore important for researchers and practitioners to engage stakeholder communities through cooperative management and restoration efforts (e.g., by participating in stakeholder-led initiatives), with the aim of establishing meaningful partnerships that provide for the support and resources necessary to undertake AIS management (Duffy and Martin 2019) with the well-being of all in mind (McClelland et al. 2011).

6.2 DEMOGRAPHIC RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Mark-recapture summary statistics.

Across all streams, we captured and measured a total of 4,887 individuals over the course of the study, including recapture events. We marked a total of 4,099 fish with individual identifying tags. We had 1,238 recapture events, 788 individuals were recaptured and re-measured at least once for an overall recapture rate of 19.2%. We recaptured 276 individuals more than once; two individuals from Keaʻahala stream were re-captured eight times. Some streams had no or few recapture events (Hālawa, n = 0; Kāwā , n = 1). However, five streams had over 50 recapture events (Kahaluʻu, Keaʻahala, Punaluʻu, Waikāne, and Waianu) (Table 13). All streams except for Hālawa and Kāwā had at least one recapture event before and after the AIS removal.

Did *A. stamineus* population sizes increase following AIS removals?

Lincoln-Peterson estimates of population size were significantly correlated between control and removal reaches ($r^2 = 0.989$, $p < 0.001$) which indicates a lack of demographic independence between reaches within streams. Therefore, we proceeded with further analyses using whole stream estimates of population size. An ANOVA indicated there was a significant difference among time periods in estimated population size ($F = 7.362$, $p < 0.01$; Fig. 14). Tukey’s post hoc tests indicated that the >6 month post removal time period had significantly smaller population sizes than prior to AIS removals ($p = 0.05$), than 0-3 months post removal ($p < 0.01$), and 3-6 months post removal ($p = 0.01$) (Fig. 14).

The response of *A. stamineus* population sizes in the >6 month post removal time period was not significantly correlated with the effectiveness of the removal of Poeciliids ($r^2 = 0.24$, $p = 0.33$) or Loricariids ($r^2 = 0.36$, $p = 0.28$), although the response was positively correlated with the effectiveness of removals of all other AIS (slope = 9.78, $r^2 = 0.54$, $p = 0.06$) (Fig. 15). Population size responses to AIS removals were not correlated with mean stream discharge ($r^2 = 0.01$, $p = 0.80$; Fig. 15) or variance in discharge ($r^2 = 0.27$, $p = 0.24$) (Fig. 15).

Was there an increase in recruitment of *A. stamineus* following AIS removals?

Table 13. Summary statistics for mark-recapture study including the number of marked fishes (n), the number of recapture events pre-/post-AIS removal, the mean and standard deviation of length (mm), mass (g), and condition factor (*C*) for captured fishes in 12 Oʻahu streams.

Stream	n	N recapture events pre/post	Mean Length (mm)	s.d. Length (mm)	Mean Mass (g)	s.d. Mass (g)	Mean Condition (g/mm^3)	s.d. Condition (g/mm^3)
Heʻeia	53	1/19	101.0754717	50.38270806	17.48320755	23.28197188	7.88797E-06	1.50755E-06
Hālawa	47	0/0	79.53191489	42.57517702	8.994042553	14.56936999	7.54271E-06	1.52295E-06
Kahaluʻu	1632	46/380	54.67647059	18.44486002	1.757205882	3.835533888	7.0606E-06	1.15856E-05
Kalihi	124	1/12	69.19354839	42.01967743	7.479596774	15.80812498	6.91064E-06	2.01961E-06
Kāwā	15	0/1	69.2	40.87472149	6.487142857	10.39707905	7.05128E-06	1.71426E-06
Keaʻahala	1277	43/348	72.95458105	27.09546235	4.553766641	6.262315401	7.35484E-06	1.24077E-06
Mānoa	153	3/8	77.24183007	37.71482784	7.189542484	11.78601867	7.41961E-06	1.56855E-06
Punaluʻu	844	171/34	77.2014218	26.63642469	5.630592417	6.293422681	8.1674E-06	1.29945E-06
Waianu	199	40/11	80.73366834	27.93976967	6.218542714	5.922549307	8.16287E-06	1.18227E-06
Waikāne	238	26/48	96.94957983	34.99917992	11.27063025	13.28877208	8.09318E-06	1.41081E-06
Waimānalo	190	4/12	69.68947368	38.38972243	5.915263158	10.26971974	7.24503E-06	1.93208E-06
Waimea	115	9/10	58.46956522	37.62938422	3.734521739	6.007844615	6.33018E-06	1.71674E-06

The proportion of recruits was significantly correlated between control and treatment reaches ($r^2 = 0.775$, $p < 0.01$) indicating a lack of demographic independence between reaches within streams. Therefore, we proceeded with further analyses using whole stream estimates of the proportion of recruits. An ANOVA indicated there was a significant difference among time periods in the proportion of recruits (F = 4.998, $p < 0.01$) (Fig. 16). Tukey's post hoc tests indicated that the proportion of recruits in the populations was higher 3-6 months post AIS removal compared to before AIS removals ($p < 0.01$) but was not significantly different than 0-3 months post removal ($p > 0.05$). We also found that recruitment during the 0-3 months post removal period was also not significantly different than the period prior to AIS removals ($p > 0.05$) (Fig. 16). A K-S test indicated a significant difference in size-frequency distributions of the post-removal populations compared to before AIS removals and compared to the 3-6 months post removal period (D = 0.28934, $p < 0.01$). Size-frequency histograms confirmed that there was a significant shift 3-6 months post AIS removal toward smaller (≤ 45 mm) individuals (Fig. 17).

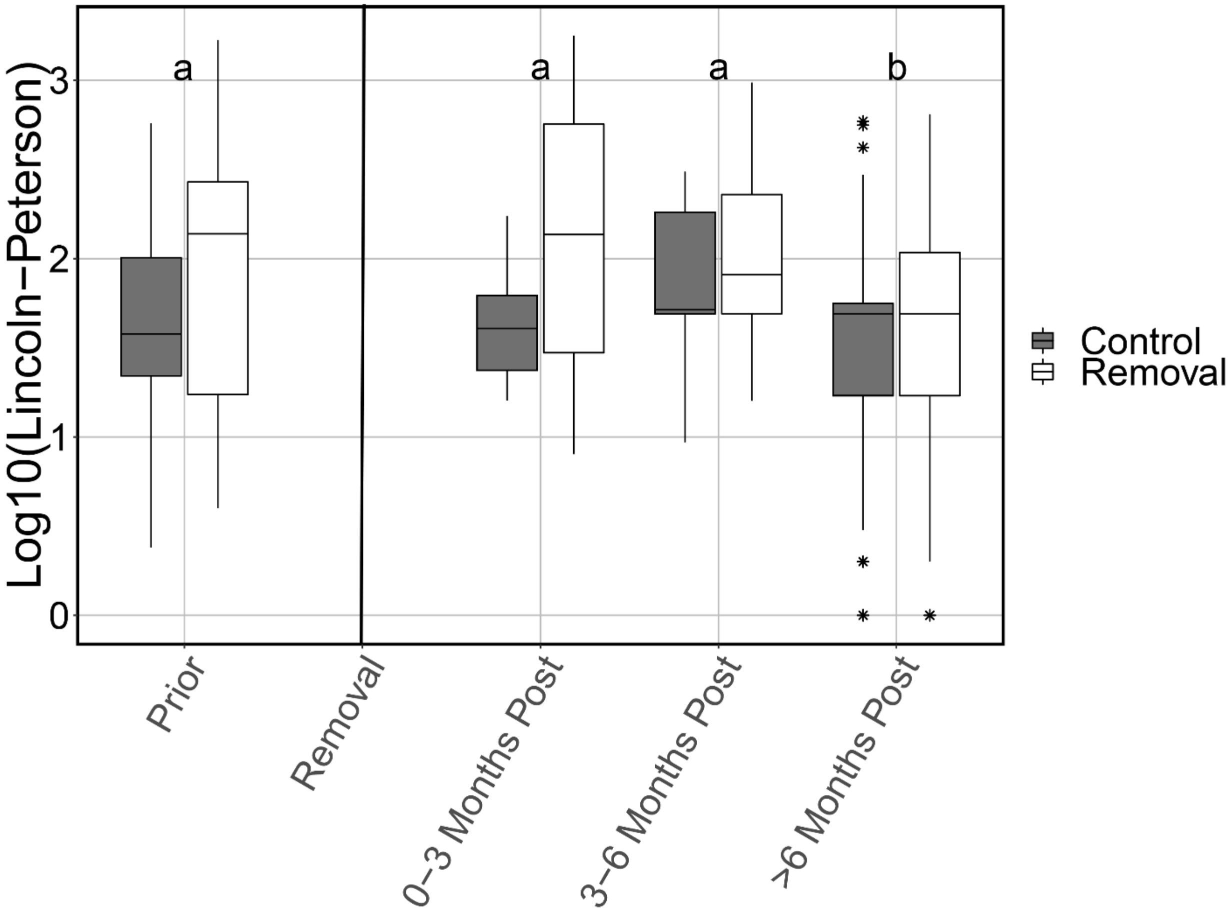


Figure 14. Lincoln-Peterson population estimates for ten streams prior to AIS removal, and 0-3, 3-6 and >6 months following AIS removals. Box plots show the mean (horizontal line), the first and third quartiles (box boundaries) and minimum and maximum data range (vertical lines) with outlier data points represented by asterisks. Heavy vertical black line indicates the AIS removal event. Population estimates for both control and removal reaches are depicted here to demonstrate the nature of the BACI design, however, due to strong demographic correlation between reaches within streams an ANOVA was performed on whole population estimates. Letters above box plots indicate significant differences among periods identified by a post-hoc test.

The increase in the proportion of recruits in *A. stamineus* populations in the 3-6 month post removal time period was significantly positively correlated with the effectiveness of the removals of Poeciliids (slope = 1.09, $r^2 = 0.78$, $p < 0.001$) (Fig. 18). However, the recruitment response was not correlated with the efficacy of removals of Loricariids ($r^2 = 0.24$, $p = 0.21$) or all other AIS ($r^2 = 0.18$, $p = 0.17$) (Fig. 18). The population responses to AIS removal was not correlated with mean stream discharge ($r^2 = 0.01$, $p = 0.89$) (Fig. 18) or the variance in discharge ($r^2 = 0.03$, $p = 0.59$) (Fig. 18).

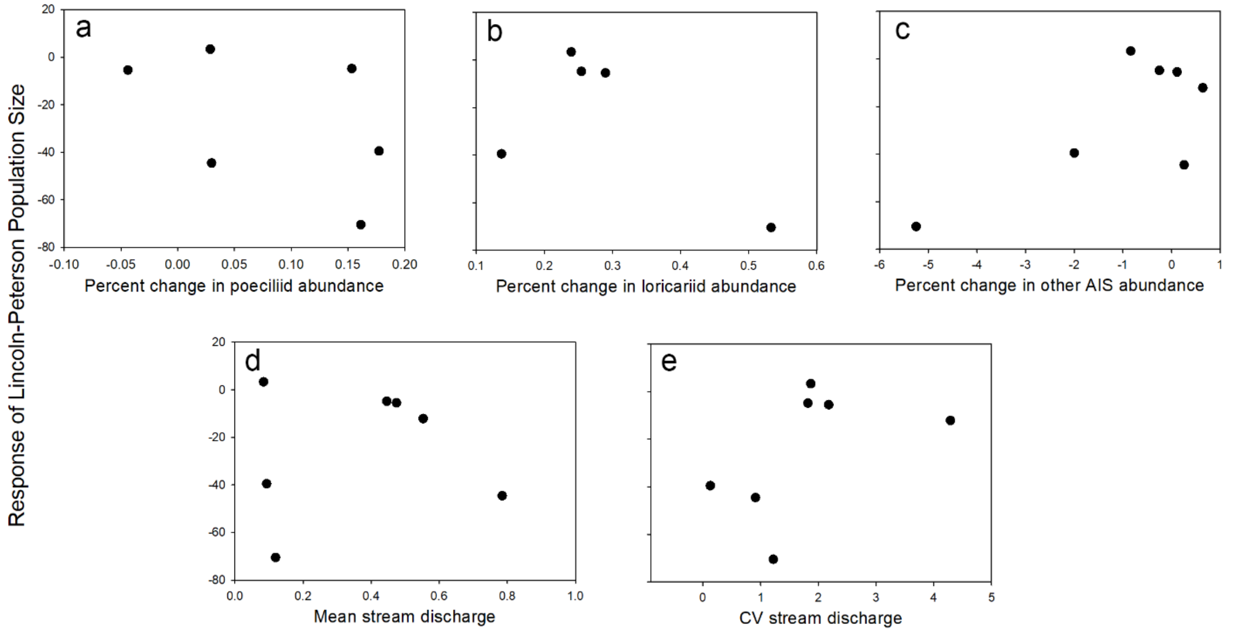


Figure 15. The magnitude of the response in the Lincoln-Peterson estimates of *A. stamineus* population sizes >6 months post AIS removal compared to the effectiveness of AIS removals (top row) and hydrological conditions (bottom row); specifically (a) the percent change in Poeciliid populations, (b) the percent change in Loricariid populations, (c) the percent change in all other AIS populations, (d) mean stream discharge and, (e) the coefficient of variation in stream discharge. Trend lines have been added to significant relationships.

Did the growth rate and/or condition of *A. stamineus* increase following AIS removals?

We had 1,268 recapture events throughout the course of the study from which growth rates and change in condition factor could be determined. The average rate of growth among all recaptured fish was 0.0013 mm/d (±0.0020 mm/d) and 0.027 g/d (±0.253 g/d). The average change in condition factor was -0.053 g/mm^3 (±2.122 g/mm^3). We found that 1,219 fish (96.2%) grew in size (mm) between capture events, while 1,139 fish (89.8%) gained weight between recapture events. A total of 824 fish (65.0%) had an increase in condition factor between recapture events. Rates of change in length ($CV_{mm/d}$ = 44.7%, $CV_{g/d}$ = 83.4%) and condition factor (CV = 358.3%) and means varied by an order or magnitude among streams.

The growth rate in body mass (g/d) of *A. stamineus* increased significantly following AIS removals conducted across the study watersheds (F = 3.38, $p = 0.018$). Sequential contrasts showed that the rate of change in body mass (g/d) increased in the 0-3 month post removal period compared to the period prior to removal (t-ratio = 2.356, *p* = 0.019). Growth rates remained higher than pre-removal and did not differ significantly from the 0-3 month post-removal period or in the 3-6 month (t-ratio = 0.059, *p* = 0.953) and >6 month (t-ratio = −1.395, *p* = 0.163) post-removal time periods (Fig. 19). Growth rates in body length (mm/d) however did not differ among time periods (F = 1.79, *p* = 0.15). The rate of change in body condition factor (*C*/d) also differed among time periods, albeit the effect was small and marginally insignificant (F = 2.61, *p* = 0.060) (Fig. 19). Sequential contrasts showed a similar pattern to that of mass growth rates, where condition factor increased on a per-day rate faster in the 0-3 month post-removal period compared to pre-removal (t-ratio = 1.906, *p* = 0.057). Rates of change in condition remained higher than the rate during the pre-removal period, but did not differ significantly from the 0-

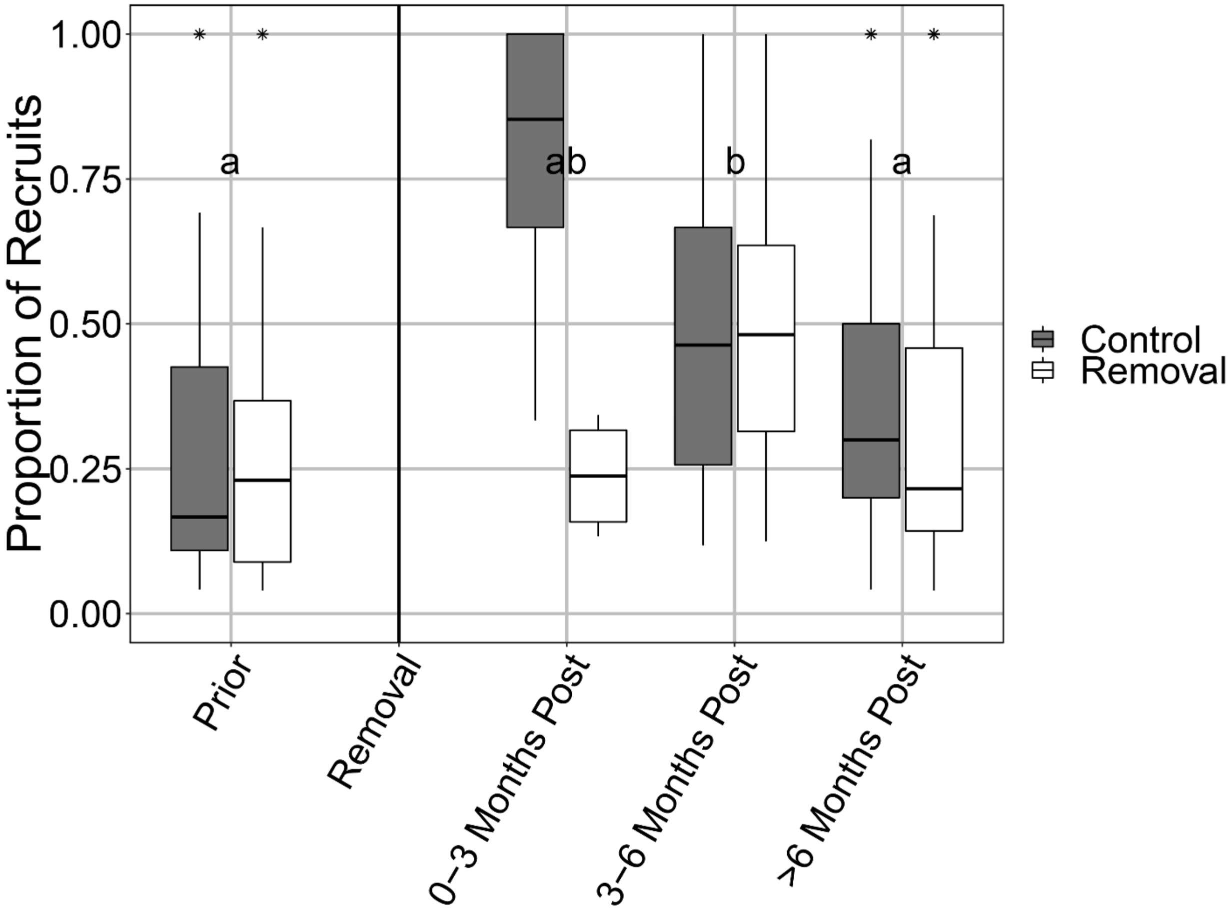


Figure 16. The mean proportion of recruits prior to AIS removal, and 0-3, 3-6 and >6 months post AIS removal. Box plots show the mean (horizontal line), the first and third quartiles (box boundaries) and minimum and maximum data range (vertical lines) with outlier data points represented by asterisks. Heavy vertical black line indicates the AIS removal event. Population estimates for both control and removal reaches are depicted here to demonstrate the nature of the BACI design, however, due to strong demographic correlation between reaches within streams an ANOVA was performed on whole population estimates. Letters above box plots indicate significant differences among periods identified by a Tukey's post-hoc test.

3 month post-removal period, the 3-6 month (t-ratio = -0.313, $p = 0.754$) and the >6 month (t-ratio = −0.303, $p = 0.762$) post-removal time periods (Fig. 19).

The increase in growth rates (g/d) of *A. stamineus* in the 0-3, 3-6, and >6 months post removal time periods were not significantly correlated with the effectiveness of removals of any AIS taxon group, or either of the stream hydrological parameters (all, $p > 0.05$). However, the change in growth rates in the >6 month post-removal period was correlated with mean stream discharge, albeit the effect was marginally insignificant ($r^2 = 0.38$, $p = 0.08$).

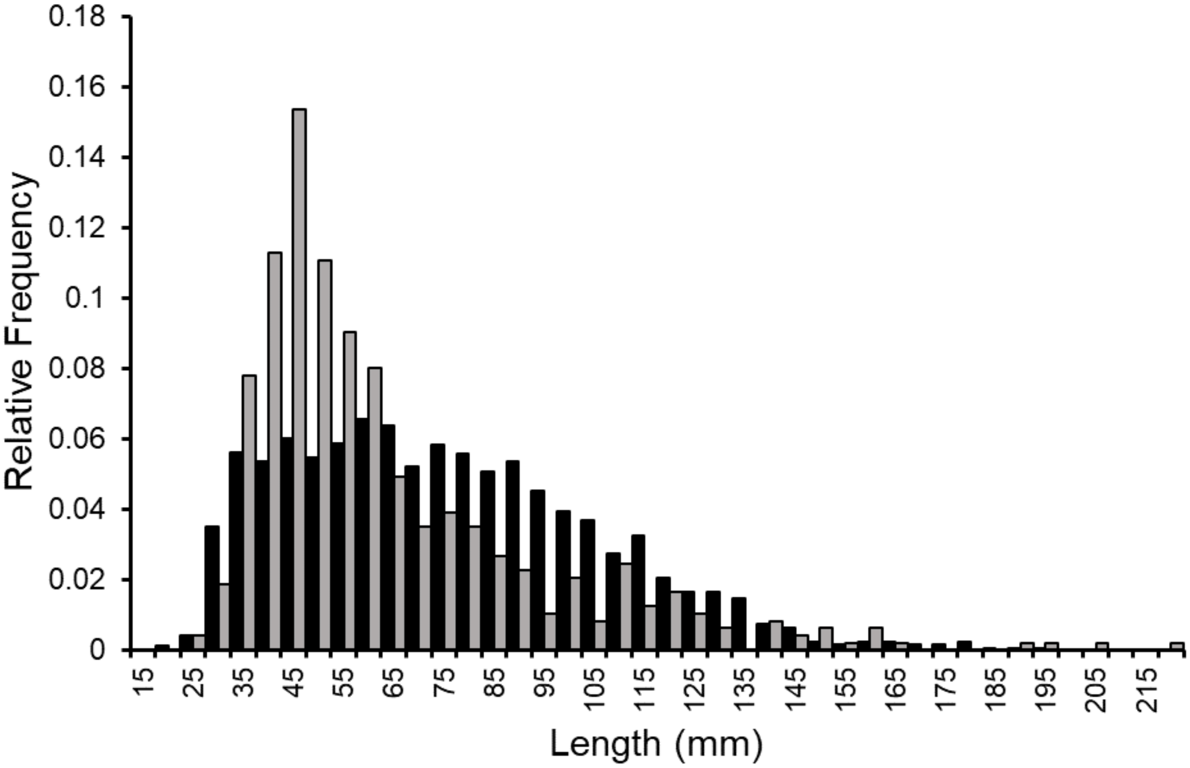


Figure 17. Relative size frequency distributions of *A. stamineus* populations in the period prior to AIS removal (black bars) and 3 – 6 months post AIS removal (grey bars).

The increase in the rate of change of body condition (C/d) in the 3-6 month post removal time period was significantly positively correlated with the effectiveness of the removals of Loricariids (slope = 0.09, $r^2 = 0.82$, $p = 0.03$) (Fig. 20) and negatively correlated with mean stream discharge (slope = -0.06, $r^2 = 0.60$, $p = 0.04$) (Fig. 20). The interaction between percent change in Loricariid densities and mean stream discharge however was not significant ($p = 0.33$). The increase in the rate of change of body condition (C/d) in the 0-3 month post-removal time period was also positively correlated with the effectiveness of Loricariid removals, however the effect was marginally insignificant ($r^2 = 0.79$, $p = 0.11$). There was no relationship between condition response and either metric of stream discharge in the 0-3 month post-removal period (both, $p > 0.6$).

Did movement of *A. stamineus* into and out of removal reaches increase following AIS removals?

We had 1,238 total recapture events throughout the course of the study from which we could determine movement rates and patterns of marked fish. Of those, we recorded 122 instances in which a fish was caught in a different reach upon its next recapture, indicating movement among reaches. We recorded 1,116 recapture events in the same reach in which the fish was previously caught. We observed 46 movements from the treatment reaches into the surrounding control reaches or the interstitial buffer areas between reaches. We observed 43 movements from the control reaches and buffer areas into the treatment reaches. The remainder of the movements occurred between control reaches (upstream and downstream) or between control and buffer reaches. The overall movement rate was 10.9%, but movement rates varied considerably among streams (CV = 114%). Mānoa and Kalihi streams had the highest movement rates (66.7% and 55.6% respectively). Heʻeia and Kahaluʻu streams had very low movement rates (5.9% and 5.7% respectively). One stream, Waimānalo, had no observed movements despite 15 recapture events. The average movement rate among streams differed between pre-removal and post-removal periods. The mean pre-removal rate was 11% while the mean post-removal rate was 20%, however, a t-test indicated this difference was not significant as among stream variation was very high (t = -1.71; d.f. = 9; *p* = 0.121). Variance in movement rates within streams was smaller over time (mean pre-post CV = 56%) than variance among streams. Trend analysis using LOESS confirmed that the number of individuals that moved (Fig. 21), the number of resident individuals (Fig. 21), the number of movement events into the treatment reach (Fig. 21), and movement out of the treatment reach (Fig. 21) did not change significantly over the course of the study. There was a subtle but non-significant trend of increasing numbers of resident individuals over time (Fig. 21).

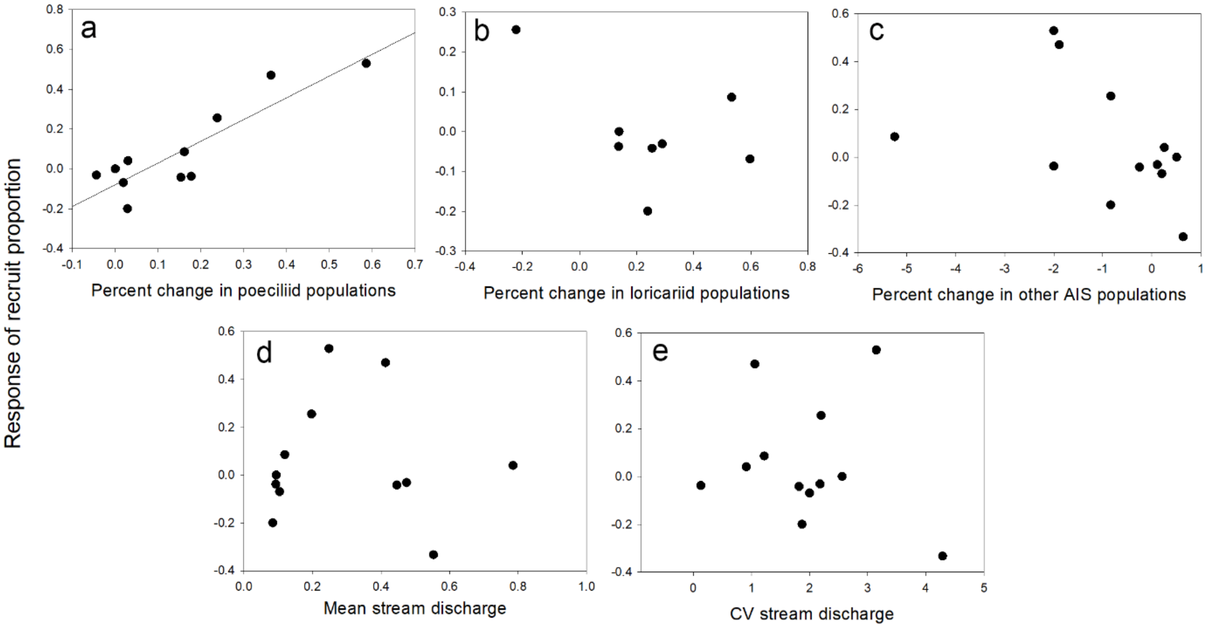


Figure 18. The magnitude of the response in the proportion of recruits in *A. stamineus* populations 3 – 6 months post AIS removal compared to the effectiveness of AIS removals (top row) and hydrological conditions (bottom row) specifically; (a) the percent change in Poeciliid populations, (b) the percent change in Loricariid populations, (c) the percent change in all other AIS populations, (d) mean stream discharge and, (e) the coefficient of variation in stream discharge. Trend lines have been added to significant relationships.

Discussion.

Here we assessed whether the removal of AIS from Hawaiian streams resulted in positive outcomes for the population demography of the at-risk endemic goby, *Awaous stamineus*. We found that AIS removals were followed by an increase in recruitment of juveniles and improved body growth and condition of *A. stamineus*. The increase in recruitment was short-lived, with recruit proportions returning to pre-removal levels starting >6 months after AIS removal. The increase in body growth and condition began during the first three months following AIS removals and persisted for the duration of the study. Streams that experienced more efficacious AIS removals saw greater increases in recruitment and growth, indicating that AIS removal was the driver of observed demographic responses. We did not find, however, that there was an increase of in-stream movement into the AIS removal reaches, or overall increases in population size caused by a pulse of recruitment. These results suggest that AIS removals can recover demographic processes in native Hawaiian fishes but that regular interventions will be required unless AIS can be wholly extirpated from the streams.

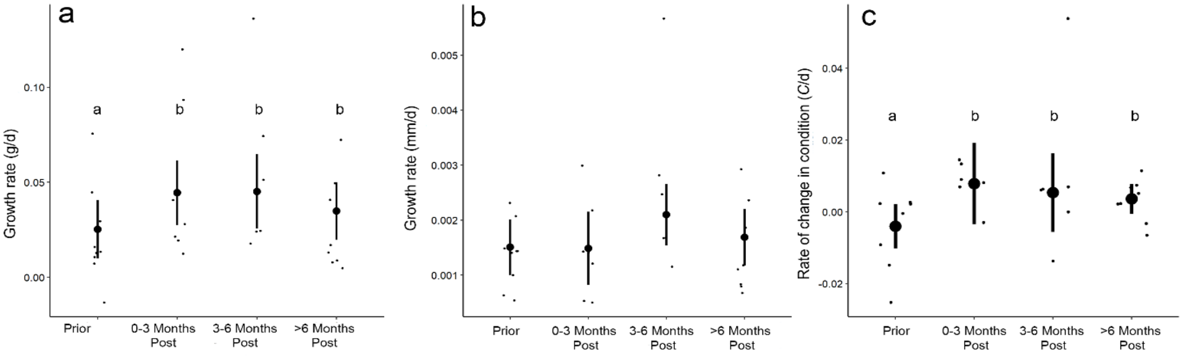


Figure 19. Mean growth rates (large black dots) in (a) grams per day, (b) mm per day, (c) condition factor (*C*) per day in ten stream populations (small black dots) prior to AIS removal, and 0-3, 3-6, and >6 months post AIS removal. Black vertical bars indicate 95% confidence intervals. Letters (a, b) indicate significant differences among periods identified by sequential contrasts post-hoc test.

The observed increase in recruitment in the 3-6 month post-removal time period aligns with our predictions. We hypothesized that Poeciliids depredate larvae and young juveniles in *A. stamineus* populations (Holitzki et al. 2013), and that the level of predation was high enough to reduce recruitment. Two lines of evidence support this inference. First, the timing of the recruitment increase (3-6 months following AIS removals), matches precisely with the early life-history ecology of *A. stamineus*. The range of larval durations for *A. stamineus* is 3-5 months (Hogan et al. 2014). Alleviation of Poeciliid predation on larvae via their removal would increase larval supply and ultimately recruitment 3-5 months later. The migratory life-history of *A. stamineus* would allow larval migrants to escape predation pressure by heading to sea, suggesting that local AIS removal may not have a strong impact on local recruitment. However, it has been shown that most of the larvae that recruited to our populations during the study period were non-migrants (mean = 87%, range: 44-98%; Heim-Ballew 2019), which might explain the significant increase in larval recruitment in the 3-6 month post-removal time period. The second line of evidence that supports this inference is that the magnitude of the recruitment response correlated with the efficacy of the removal of Poeciliids among streams. Streams that had a greater percent change in Poeciliid density had larger pulses in recruitment 3-6 months later. This, coupled with the fact that a great majority of larvae completed their life-cycle in-stream, indicates that removals of Poeciliids can elevate recruitment in *A. stamineus* populations.

Stream hydrology can also play a role in recruitment because sufficient surface flow is required for migrant larvae to egress and ingress, and thus highly variable flows can be a threat to survival of non-migrant larvae (e.g., due to stream drying) as well as to immigrants (e.g., due to limited ingress). Surface flow has also been shown to impact the effectiveness of AIS removals in Oʻahu streams (described above). For example, we have found that Poeciliid removal is more effective in streams with intermediate mean discharge and greater discharge variability. We might therefore expect that there is an interaction between stream hydrology and Poeciliid removals in structuring demographic responses, such as recruitment, but contrary to this expectation, we did not find evidence of a significant interaction. This may be because the hydrological conditions that are best for Poeciliid removals and what is best for recruitment do not match. For example, while removal of Poeciliids was most effective at intermediate mean flows, greater mean discharge would benefit passive downstream drift and attract more immigrant recruits (i.e., due to increased ocean-stream connectivity and stronger freshwater cues (McDowall 2007b)). Likewise, Poeciliid removals may be more readily captured in streams with greater discharge variability because when streams reach low flows, they become trapped in pools (i.e., which reduces their opportunity to escape capture). However, extreme low flows and drying events would likely increase mortality and reduce recruitment of native migratory fish. This is evidenced by the fact that we had low sample sizes in three of our most hydrologically variable streams (Hālawa, Kāwā , and Kīpapa). Accordingly, contrary as opposed to complementary effects of hydrology on AIS removals and native species demography could very well result in non-significant interactions.

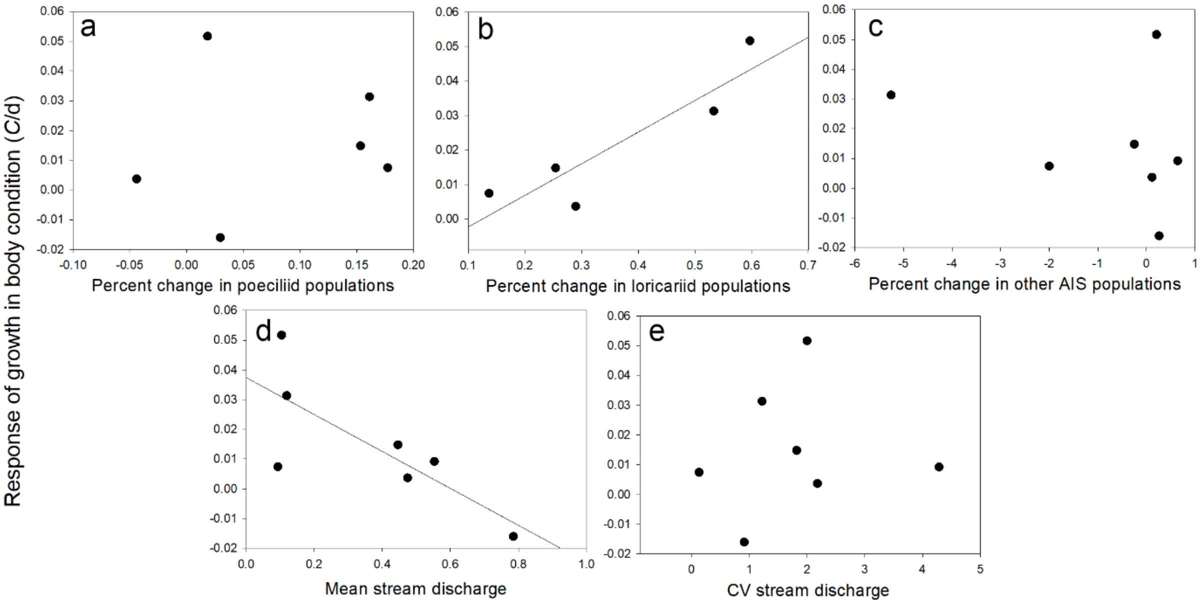


Figure 20. The magnitude of the response in growth of body condition in *A. stamineus* populations 3-6 months post AIS removal compared to the effectiveness of AIS removals (top row) and hydrological conditions (bottom row) specifically; (a) the percent change in Poeciliid populations, (b) the percent change in Loricariid populations, (c) the percent change in all other AIS populations, (d) mean stream discharge and, (e) the coefficient of variation in stream discharge. Trend lines have been added to significant relationships.

The observed increases in growth (g/d) and condition (*C*/d) following AIS removals were also consistent with our predictions. We hypothesized that AIS in general, but specifically Loricariids, compete with *A.*

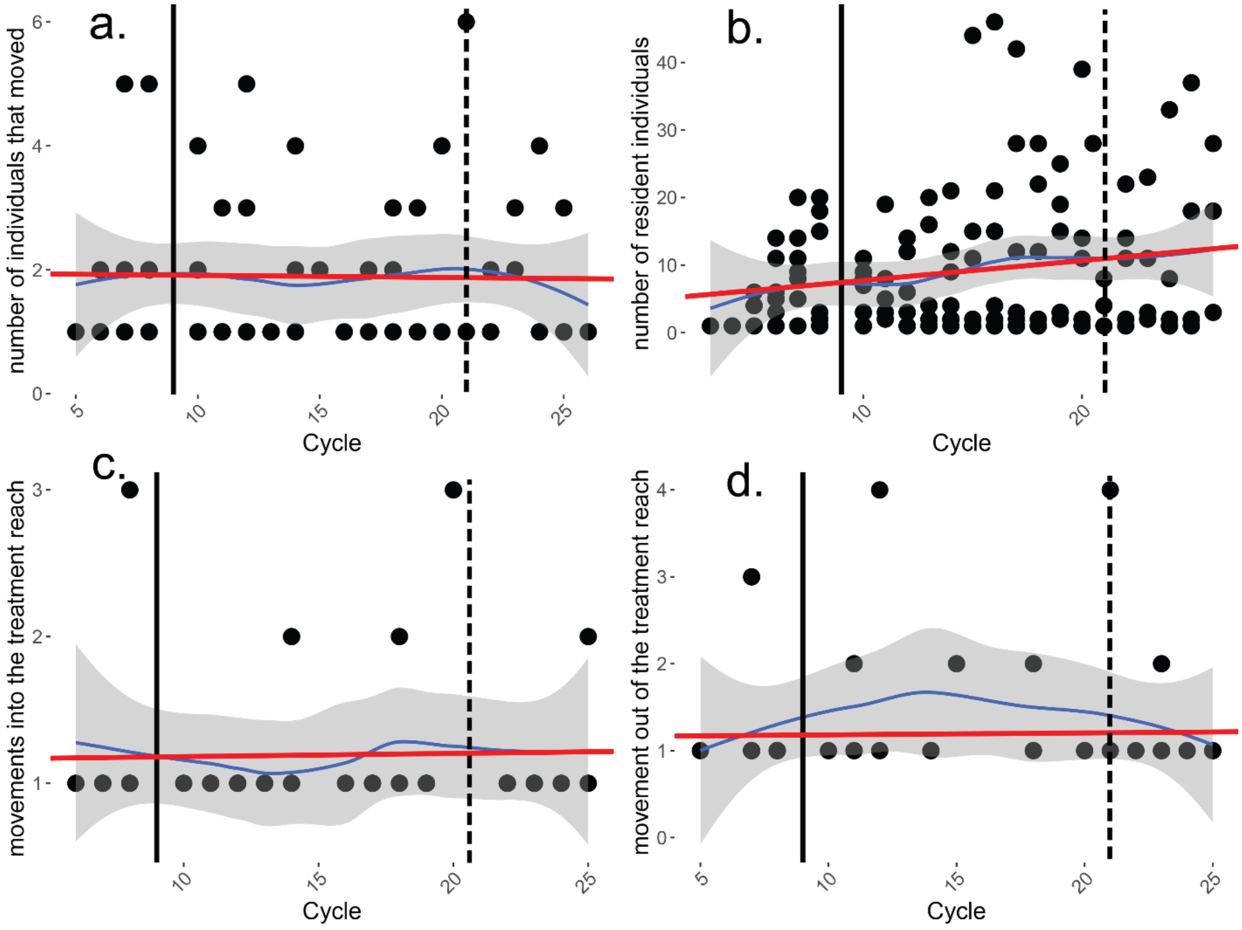


Figure 21. Summary of fish movement and residency events across the study timeline in 12 Oʻahu streams determined by mark-recapture events. (a) the number of recaptured individuals that moved between reaches within a stream; (b) the number of recaptured individuals that remained resident in the same reach as previously caught; (c) the number of movement events into the AIS removal reach; (d) the number of movement events out of the AIS removal reach. The solid black bar indicates the timing of the AIS removal event in 11 of the 12 streams. The dashed black bar indicates the timing of the AIS removal event in Punaluʻu stream. The red line is the linear trendline, the blue line is the LOESS trend line, the grey shaded area is the 95% confidence area in the LOESS estimates.

stamineus adults and juveniles for food and habitat space, and that the removal of these species could result in increases in growth rates and body condition. Loricariids are known to reduce phosphorus availability and the quality and quantity of algal food resources (Capps and Flecker 2015) and are known to compete for food resources with native species like *A. stamineus* that feed on algae and diatoms (Kido et al. 1993, Kido 1996). Our findings indicate that the removal of Loricariids elicited increases in growth and condition in *A. stamineus*. First, the removal of Loricariids was highly successful. The average percent density change among populations was a 26% reduction, with some streams seeing up to 60% decrease in density of Loricariids. The densities of Loricariid populations changed immediately, and low densities persisted for the duration of the study (Fig. 9, Fig. 11). This timeline matches with the observation that the rate of growth and increase in condition occurred in the first 3 months following AIS removals and persisted for the duration of the study. In addition, the magnitude of the rate of change of body condition (*C*/d) was correlated with the efficacy of Loricariid removals in the 0-3 months ($r^2 = 0.79$), and 3-6 months ($r^2 = 0.82$) post-removal periods.

As with Poeciliid removals, we might expect that there is an interaction between stream hydrology and Loricariid removals in structuring the demography of *A. stamineus*. The effectiveness of Loricariid removals in the study watersheds was correlated with hydrology (Fig. 10), where more Loricariids were removed from streams with lower mean discharge and lower variation in discharge. Additionally, it is known that the body shape of amphidromous Hawaiian gobies is locally adapted to the hydrological environment they recruit to, where fish that recruit to streams with steep slopes and faster moving waters (i.e., on Hawaiʻi) tend to have narrower bodies compared to fish recruiting to lower slope, slower flow streams (i.e., on Kauaʻi) presumably to reduce drag and improve swimming and climbing performance (Blob et al. 2010). Thus, we might expect that the change in body condition following AIS removal would be significantly higher in lower discharge environments, both due to the relaxation of selection for narrower bodies in lower flow environments and due to more effective removal of Loricariids. This could lead to an interaction between the efficacy of Loricariid removals and mean discharge conditions. Consistent with this, we found that the rate of change in body condition following AIS removals was greater in streams with lower mean discharge, possibly due to greater alleviation of resource competition and slower flows permitting individuals to attain greater girth. In this instance, the hydrological conditions that allow for greater girth and allow for more effective removals are identical, but we still did not detect a significant statistical interaction. This is most likely due to a lack of statistical power to detect an effect. Not only did the main terms have small effect sizes with marginally significant p-values, there likely were not enough degrees of freedom to detect an interactive effect given our relatively small sample sizes.

Though we detected significant post-removal increases in recruitment that were correlated with the effectiveness of AIS removals, we did not observe a significant increase in population sizes of *A. stamineus*. Recruitment can be an important driver of dynamics in size-structured populations of aquatic organisms with complex life-histories similar to those exhibited by Hawaiian migratory stream fishes (Victor 1983, Caley et al. 1996). Although the size of recruitment cohorts in some marine fishes has been shown to determine adult population sizes (Hjort 1914), recruitment does not always regulate population dynamics and increases in recruitment do not guarantee increases in population size. Competition and predation are known to impose significant mortality on recruitment size-classes (Hixon and Webster 2002; Hixon and Beets 1993; Carr et al. 2002). Populations at or near carrying capacity may experience density-dependent mortality from competition, which will dampen the size of recruitment cohorts (Hixon and Webster 2002). Even if populations are recruitment limited, density-independent mortality from predation can be significant enough to dampen the size of recruitment cohorts. Hixon and Beets (1993) showed, for example, that recruits in coral reef fish populations suffered >90% mortality in the first three months due primarily to density-independent predation. In *A. stamineus* populations, the pulse of elevated recruitment observed following AIS removals was short lived. We monitored post-removal conditions for a long enough time period (~2 years) in most of our streams that we should have detected the recruits from that cohort joining the adult population. Thus it is likely that mortality from competition and/or predation dampened the cohort enough to make its effects on adult population sizes indeterminable. While AIS removals resulted in a sustained reduction in predatory Poeciliids and competitors like Loricariids, neither group was extirpated from any of our study sites. Accordingly, it is likely that the effects of AIS on *A. stamineus* populations were alleviated but not eliminated. In addition, populations of *A. stamineus* are known to recruit year-round (Lindstrom 1998, Heim-Ballew 2019) and adult populations are composed of a number of different cohorts, so it is possible that a short-lived increase in recruitment is simply insufficient to significantly increase adult population sizes.

We observed immediate and sustained increases in growth rate that were correlated with the effectiveness of AIS removals. There can be several outcomes from an increased growth rate which can be an important mechanism of population regulation (Vincenzi et al. 2002). Increases in growth rate might be expected to reduce mortality as fish of larger size will be less susceptible to predation (Houde 1997, Hixon and Beets 1993). Fish with greater body condition might be expected to perform better (Vincenzi et al. 2002). Fish body mass and condition is also related to fecundity. For example, increased body mass and condition at maturity can increase fecundity for female fishes (Rideout and Morgan 2010, Barneche et al. 2018). This might be expected to manifest in greater reproductive output from the populations and greater survival to adult size classes, and thus it might moderate recruitment variation (Rickman et al. 2000). Increased body mass and condition as a result of AIS removal are expected to result in a hyperallometric increase in total reproductive energy indicating that for every additional gram of body mass growth added due to AIS removals, reproductive output is expected to increase 1.2-fold (Barneche et al. 2018). Unfortunately, we did not investigate reproductive output in *A. stamineus* populations, and we did not observe an overall increase in population sizes on average, although we did observe an increase in recruitment. The timing of the recruitment pulse matches with predictions that AIS removals led to improved in-stream conditions, which led to increased body growth, which led to a bout of spawning and increased recruitment 3-6 months following removals. Further work is thus warranted to definitively determine whether increases in body mass and condition of *A. stamineus* can lead to increases in recruitment.

Local population sizes of *A. stamineus* can increase not only due to increased production and recruitment, but also from in-stream movement of adults and juveniles. Adults of *A. stamineus* have been shown to be site-attached (Kinzie 1990), and males appear to defend territories in which females will nest (Ha and Kinzie 1996). Post-larvae and recruits are migratory and will move upstream, up to several kilometers, before settling on a territory (Smith and Smith 1998). Despite the relatively sedentary nature of adults and juveniles, in-stream movement of adult fishes is expected on small spatial scales (~100m; Rodriguez 2002). If AIS were affecting habitat use by *A. stamineus*, we might expect to see fish move into the remediated study reaches as more habitat becomes available. Contrary to this expectation, we found that movement rates were low throughout the study. Of all recapture events, we found that only ~10% of individuals exhibited movement between study reaches or buffer areas between study reaches. We also did not observe an appreciable increase in movement following AIS removals either into or out of the remediated reaches. It appears that fish of markable size in our study maintained a high level of residency, consistent with prior observations of territoriality. This suggests that AIS are not affecting *A. stamineus* habitat use, at least at the reach scale. However, there may be significant overlap in microhabitat use between AIS and *A. stamineus* within reaches that our study could not discern. Additional work will be necessary to determine whether microhabitat use of *A. stamineus* is affected by AIS and whether it changes following remediation.

Prior et al. (2018) hypothesized that small, simple native communities that are characterized by strong trophic links, like those on islands (Jones et al. 2016) and in aquatic ecosystems (Gallardo et al. 2016), may be more likely to recover after invasive species removal. Insular freshwater systems, like those in Hawaiʻi are characterized by depauperate communities and strong trophic linkages (Coat et al. 2009). Consistent with these expectations, our findings indicate that AIS removals can result in the demographic recovery of native Hawaiian migratory fishes through the alleviation of predation and competition. Suppression of AIS was enough to detect significant increases in some processes like recruitment and growth rates. This also aligns with previous studies showing that complete eradication is not required to reduce AIS impacts (Green et al. 2014). While the observed responses indicate that Hawaiian stream communities might be ideal systems to effect ecological recovery via AIS removal (Prior et al. 2018), our findings also indicate that experimental AIS removals were not adequate to effect long-term changes in *A. stamineus* demography. The observed post-removal increase in recruitment was short lived. Consistent with this, snorkel surveys found evidence of a brief increase in population densities during the first five months following AIS removal (Fig. 11), whereas mark-recapture assessments did not detect any increase in adult and juvenile population sizes. Thus, effecting long-term ecological recovery in these system might require undertaking more regular or more aggressive AIS removal than that done for this study.

Our findings suggest that regularly scheduled AIS removals, particularly during the peak breeding season for Hawaiian gobies, may lead to persistent increases in recruitment. The observed pulse of recruitment occurred 3-6 months following removals and declined thereafter. Thus, regular AIS removal timed every 3-6 months may be enough to sustain elevated recruitment, which could result in increased adult abundances particularly if recruit mortality is lower due to higher growth rates. It might be possible that less frequent, more aggressive removals might effect greater responses, as post-removal AIS densities remained depressed for the duration of our study (Fig. 11). This also highlights that a whole stream intervention, resulting in extirpation of AIS would likely be an effective strategy to achieve the recovery of *A. stamineus* populations (Fig. 1).

Table 14. Daily discharge (Q) of each study stream summarized by the mean, coefficient of variation (CV: ratio of the standard deviation relative to the mean), and the 95th, 90th, 50th 10th or 5th exceedance percentiles of discharge between October 2015 and October 2016. Stream discharge was monitored by USGS gages or with stationary HOBO U20 pressure loggers. Nitrate = inorganic nitrate, SRP = soluble reactive phosphorus.

Sites	USGS site	mean Q	CV	Q95	Q90	Q50	Q10	Q5	Nitrate (μg/L)	SRP (μg/L)
Hālawa	16226200	0.351	2.68	0.001	0.003	0.078	0.78	1.64	52.49	8.88
He‘eia	16275000	0.108	1.99	0.052	0.055	0.068	0.18	0.2	38.2	18.71
Kahalu‘u	16283200	0.133	1.23	0.066	0.082	0.101	0.16	0.27	89.39	21.1
Kalihi	16229000	0.262	2.05	0.022	0.026	0.136	0.54	0.99	38.7	9.64
Kea‘ahala	Hobo-U2	0.093	0.06	0.087	0.088	0.092	0.1	0.104	126.43	35.37
Mānoa	16241600	0.599	2.11	0.118	0.121	0.289	1.08	1.79	64.62	11.59
Punalu‘u	16301050	0.881	0.77	0.513	0.543	0.72	1.16	1.71	10.42	12.13
Waianu	16294100	1.286	0.94	0.659	0.715	1.125	1.64	2.14	41.3	16.36
Waihe‘e	16284200	0.258	1.04	0.173	0.177	0.206	0.32	0.43	58.68	23.99
Waikāne	16294900	0.464	1.69	0.063	0.07	0.305	0.72	1.41	60.33	12.69
Waimānalo	16249000	0.113	1.74	0.052	0.053	0.064	0.15	0.33	314.82	17.4
Waimea	16330000	0.738	1.97	0.026	0.041	0.331	1.53	2.53	9.21	4.86

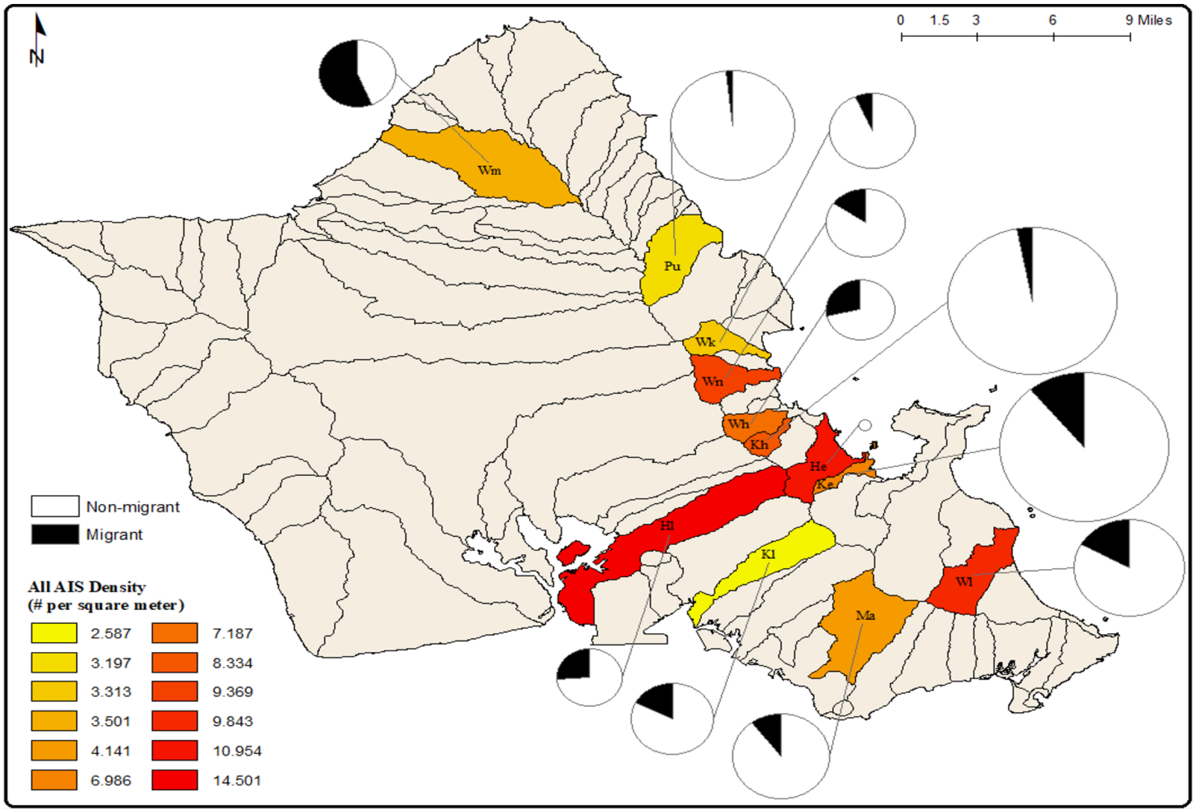
6.3 LIFE HISTORY VARIATION IN RELATION TO AIS, DISCHARGE, AND NUTRIENTS IN HAWAIIAN STREAMS

Sample collections.

A total of 496 *A. stamineus* were collected from the 12 Oʻahu watersheds. The largest sample size was from Keaʻahala (n=101) and the lowest sample size was from Heʻeia (n = 1) (Fig. 22). The sample from Heʻeia (and thus the watershed, also) was excluded from statistical analyses.

AIS Densities.

Poeciliid densities ranged from 0.001 m^{-2} in Waimea to 13.943 m^{-2} in Hālawa (Fig. 22). There were four streams that did not have Loricariids or had near zero Loricariid densities (Waimea, Waianu, and Hālawa: 0 m^{-2}; Punaluʻu: 0.001 m^{-2}). Waiheʻe watershed exhibited the highest density of Loricariids (3.628 m^{-2}) (Fig. 22).



Watershed	Kalihi (Kl)	Punaluu (Pu)	Waikane (Wk)	Waimea (Wm)	Manoa (Ma)	Keaahala (Ke)	Waihee (Wh)	Kahaluu (Kh)	Waianu (Wn)	Waimanalo (Wl)	Heeia (He)	Halawa (Hl)
N	28	59	30	25	38	101	21	100	26	48	1	19
Non-migrants	23	58	28	11	34	90	15	97	22	40	1	14
Migrants	5	1	2	14	4	11	6	3	4	8	0	5
Poecillid Density	1.428	2.95	2.63	0.001	1.225	4.268	3.406	4.604	9.114	7.153	3.66	13.943
Loricariid Denisty	0.883	0.001	0.0331	0	0.499	2.577	3.628	3.45	0	0.065	5.83	0

Figure 22. Proportions of migrant and non-migrant (i.e., resident) *Awaous stamineus* relative to overall invasive species density (# / m^2) in each study watershed, with reference to sample size (N), Poeciliid density, and Loricariid density.

Stream Flow.

Median daily discharge (Q) ranged from 0.064 to 0.331 m^3/sec. Median discharge was lowest in Waimānalo watershed and highest in Waimea watershed. Mean discharge ranged from 0.093 m^3/sec in Keaʻahala to 1.286 m^3/sec in Waianu (Waiahole) watershed, and discharge variability (CV) ranged from 0.06 to 2.68 m^3/sec in Keaʻahala and Hālawa, respectively (Table 14).

Nutrient Concentrations.

Nitrate concentrations were lowest in Waimea watershed (9.21 µg/L) and highest in Waimānalo watershed (314.82 µg/L) (Table 14), whereas soluble reactive phosphorus was lowest in Waimea watershed (4.86 µg/L and highest in Keaʻahala watershed (35.37 µg/L) (Table 14).

Migration Behavior.

Overall, the resident contingent (87%) was more common than the migrant contingent (13%) among the sampled individuals. Of the streams with robust representation, Punaluʻu harbored the highest proportion (98%; n = 59) of residents, whereas Waimea watershed harbored the lowest proportion (44%; n = 25) of residents (Fig. 22) among the study watersheds.

Model Selection.

There was no significant effect of Loricariid density on the proportion of resident *A. stamineus* in the study watersheds ($p = 0.57$, Table 15). However, there were fewer residents when Loricariid density was higher and *A. stamineus* population density was lower ($p = 0.0065$, Fig. 23). Additionally, there were fewer residents when discharge variability and Poeciliid density were higher ($p = 0.033$ and $p = 0.0029$, respectively) (Fig. 23). Also, there were fewer residents when discharge variability was higher and soluble reactive phosphorus was lower ($p = 6.64^{-07}$, Fig. 23) and at both extremes of the inorganic nitrate concentration spectrum ($p = 0.00018$, Fig. 22). The relationship between resident proportions and SRP concentrations was complex, however, as there were slightly more residents at higher concentrations ($p = 0.0028$, Fig. 23). Lastly, there were more residents when Loricariid and *A. stamineus* population density ($p = 6.64^{-07}$) and SRP were high at all levels of discharge variability (p = 0.0065) (Fig. 24, Fig. 25).

Table 15. Logistic regression output for best selected model explaining variation in the proportion of resident contingents in the study watersheds.

Coefficients	Estimate	Std. Error	Z value	p-value	Relative Contribution (%)	Significance
Intercept	10.248697	3.446396	2.974	0.002942	-	**
Discharge Variability	-2.634966	1.185061	-2.223	0.026183	2.896115	*
Poeciliid Density	0.128751	0.060225	2.138	0.032532	3.147698	*
Loricariid Density	0.295555	0.516758	0.572	0.567361	0	ns
Phosphorus	-0.242487	0.081197	-2.986	0.002823	0.381879	**
Nitrate	0.014077	0.003763	3.741	0.000183	10.67714	***
Awaous Density	-0.829836	0.270559	-3.067	0.002161	26.46525	**
Discharge Variability* Phosphorus	-0.186752	0.037565	-4.971	6.64e-07	40.76633	***

Loricariid Density * Awaous Density	0.215804	0.079351	2.720	0.006536	15.66559	**

Significance indicated: '.' *p*-value <0.1, '*' *p*-value <0.05, '**' *p*-value<0.01, '***' *p*-value<0.001

Discussion.

Impaired water quality, reduced water quantity, and AIS are widely recognized threats to populations of native Hawaiian migratory fishes (Brasher 2003, Lisi et al. 2018). We aimed to test how the quality and quantity of surface water and AIS densities affect life history variation in the native fish *A. stamineus*, particularly whether the proportion of residents changed in response to one or all of these factors. We found that variability in stream flow, the density of invasive Poeciliids, elevated nitrogen and phosphorus, as well as the density of *A. stamineus* all had significant direct effects on the proportion of

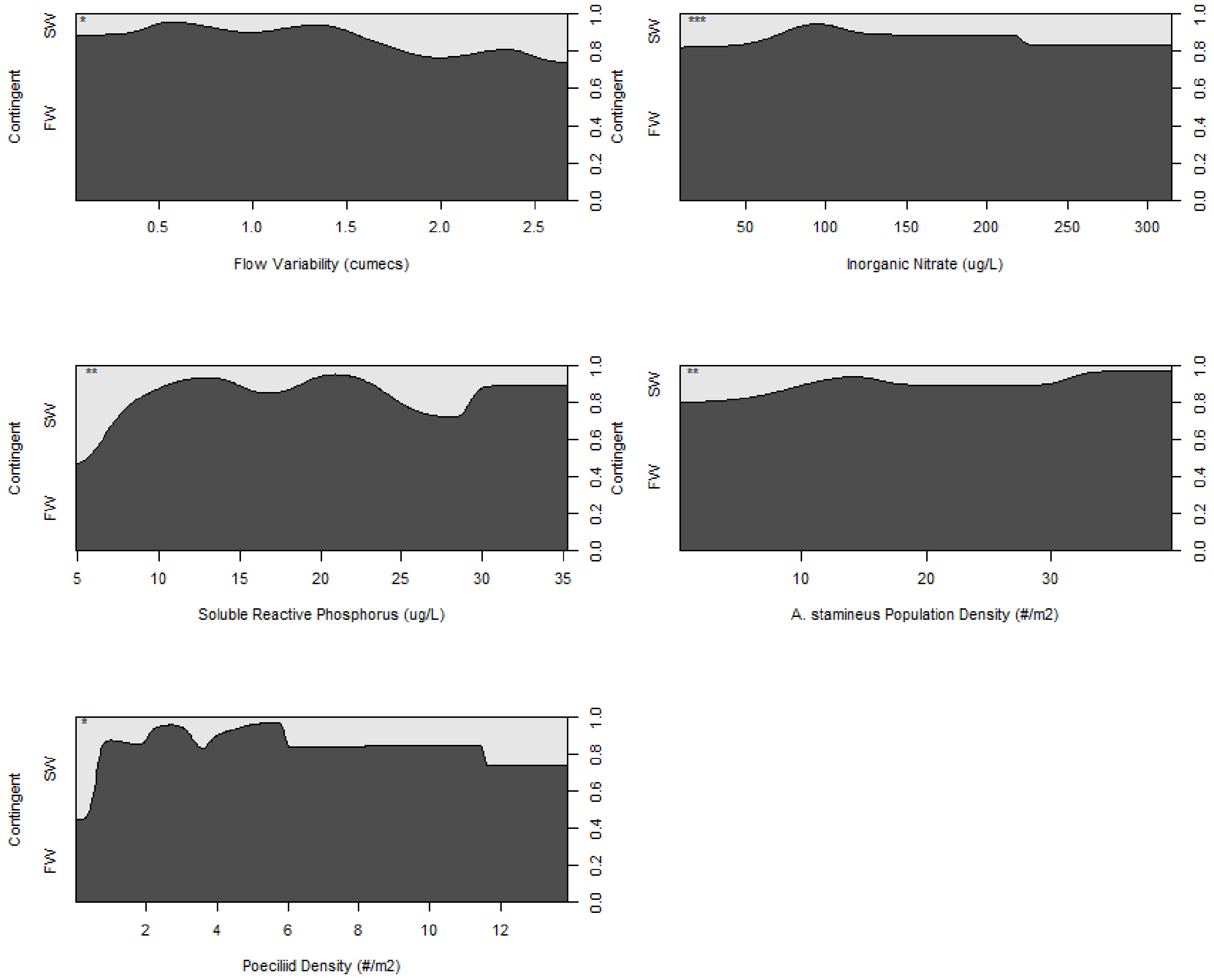


Figure 23. Main effects plots for significant variables; Significance indicated: '.' *p*-value <0.1, '*' *p*-value <0.05, '' *p*-value<0.01, '***' *p*-value<0.001**

residents in the studied populations. Overall, the proportion of residents was greater when phosphorus concentrations were higher and *A. stamineus* densities were higher. While there was no direct effect of Loricariid density on the proportion of residents, there was a significant interaction between *A. stamineus* population density and Loricariid density (i.e., there were more residents where there were higher Loricariid and *A. stamineus* population densities). The densities of invasive Loricariid catfishes also increased with higher phosphorus concentrations and thus trended with greater resident proportions. These findings suggest that populations of Loricariids and *A. stamineus* may be better supported in systems that are more enriched with phosphorus and other nutrients that may co-occur from land-based human activities. Notably, resident proportions were lower in streams with higher discharge variability and greater densities of Poeciliids. Streams with intermediate nitrogen concentrations tended to have a greater proportion of residents.

Phosphorus is typically a limiting nutrient in freshwater systems (Elser et al. 2007); low levels of phosphorus may indicate that a system is nutrient limited and thus may not be capable of supporting high densities of resident *A. stamineus*. Since Loricariids sequester phosphorus (Hood et al. 2005), one might expect that higher Loricariid densities would correspond to lower phosphorus concentrations, but the opposite was found. Loricariid density and phosphorus were positively collinear. Finding excess phosphorus in the study systems with higher densities of Loricariids indicates that phosphorus must be entering the system faster than it can be sequestered by the catfish (Carlson and Simpson 1996). Nitrate also had a significant effect on the proportion of residents; there were more residents at intermediate nitrate levels. Phosphorus may be entering streams from effluent and may be accompanied by other nutrients associated with land-based activities (Ellison and Brett 2006). Nitrate was positively correlated with phosphorus, which may indicate the two nutrients, and likely many others, are transported to streams by similar means. Thus nutrients like phosphorus and nitrate may act as a proxy for other terrestrial runoff that can either decrease water quality or stimulate food web production (Ellison and Brett 2006). Additionally, the high and positive correlation between Loricariid density and phosphorus may indicate that Loricariids are disturbing stream banks enough to cause phosphorus to be transported

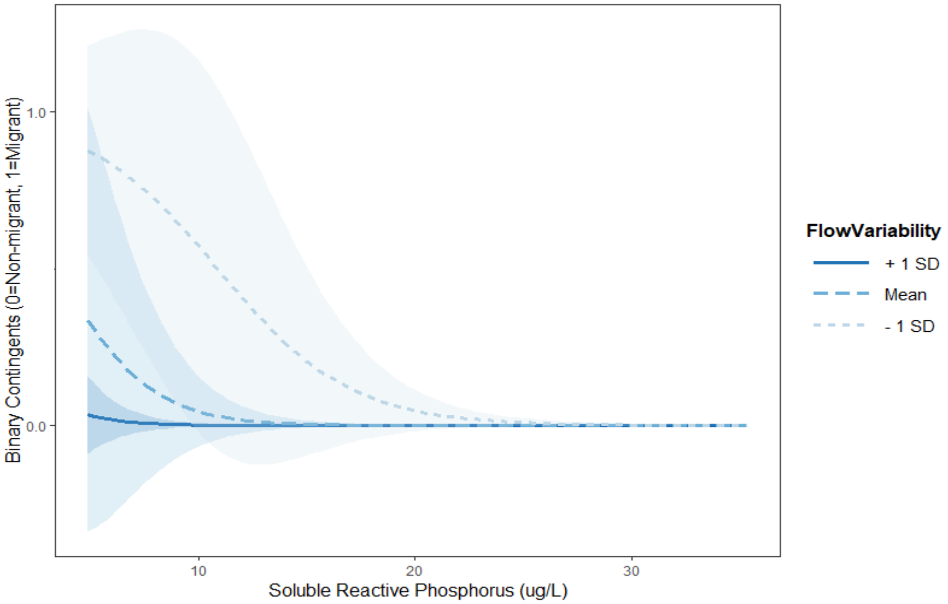


Figure 24. Interaction plot between flow variability and soluble reactive phosphorus.

from sediment to the water column; stream bank erosion is a primary and natural means of phosphorus entering rivers and streams (Ellison and Brett 2006).

Among our study watersheds, we detected more residents when discharge variability was lower, indicating stable flows likely improve in-stream conditions, thereby resulting in greater survival of resident larvae. This is consistent with evidence that more stable base flow can increase the feeding potential of larvae (Rothschild and Osborn 1988, Sundby and Fossum 1990, MacKenzie et al. 1994) by increasing encounters between larvae and prey. On the other hand, when flow variability is high, habitat may be lost from the drying up of streams, which may increase mortality of residents. There was also a clear relationship between SRP and discharge variability, where the concentration of SRP declined with increasing discharge variability. It is possible that greater discharge variability increases nutrient transport, which may promote primary and secondary producers that act as nutrition sources for larvae (Ellison and Brett 2006). Conversely, if a stream tends to dry up due to high discharge variability, then the transport or movement of phosphorus within a stream may be limited or reduced (Ellison and Brett 2006). More stable base flows may also facilitate the physical movement of larvae and interactions with prey sources may also increase. More residents may thus survive the larval stage because of greater access to more abundant prey (Houde 2008). Interestingly, we found that there were more residents, regardless of flow variability levels when phosphorus concentrations were intermediate to high, indicating that some phosphorus enrichment may increase overall stream productivity.

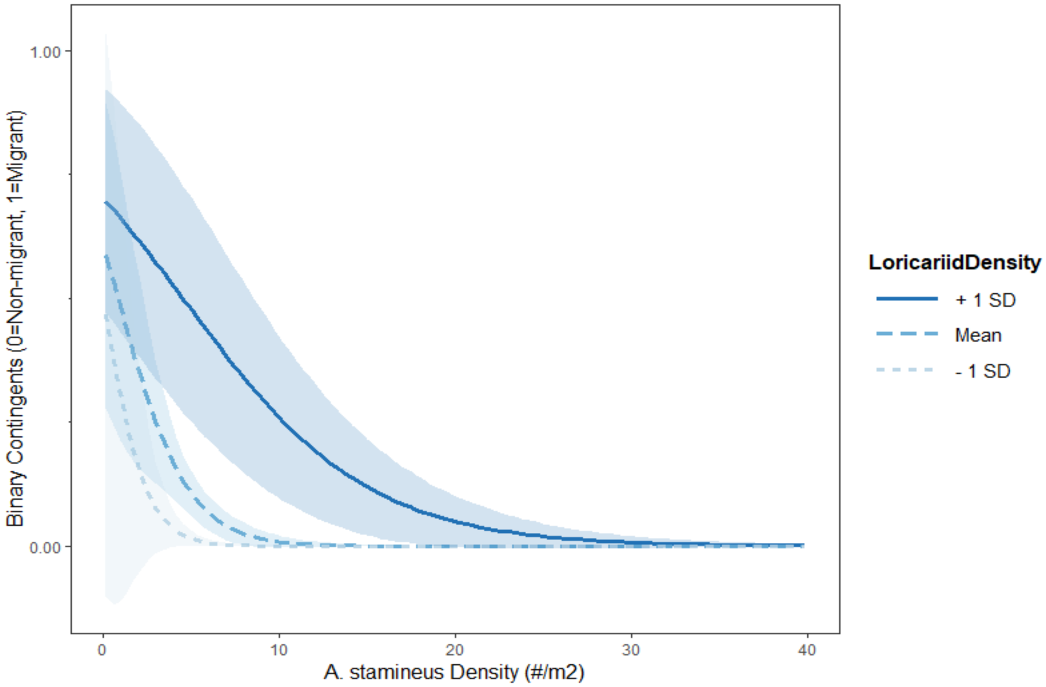


Figure 25. Interaction plot between Loricariid density and *Awaous stamineus* population density

Our results also indicate that the proportion of residents were lower with higher discharge variability and greater densities of Poeciliids. As predators of fish at early life stages, Poeciliids can directly elevate the mortality of resident larvae (Holitski et al. 2013, Hain et al. 2019). Migrant larvae would also be subject to mortality by Poeciliids, but only in the first few hours before they flush out to sea (Lindstrom

1998), so their relative abundance likely will be much less affected by in-stream predation. Notably, predation pressures can be elevated when discharge variability is high, as drying tends to cause water to pool, which concentrates in-stream residents and thus increases interactions between resident larval prey and predatory Poeciliids. Dewatering of streams would be expected to similarly elevate predation rates, though it might also lower the abundance of migrants by reducing ocean-stream connectivity.

Our findings demonstrate that interactions between physical and biological processes shape mortality and recruitment success in *A. stamineus* (Houde 2008), with higher quality habitats generally supporting higher population densities with higher proportions of resident contingents. We recovered relationships suggesting that several aspects of anthropogenic activity affect the proportion of residents in *A. stamineus* populations. Certain AIS like predatory Poeciliids appear to exert a negative pressure on resident larvae, too high and too low nutrient concentration may increase larval mortality, and likewise more variable discharge can elevate mortality by reducing stream habitat and nutrient or prey availability. Nonetheless, residents appear to be the dominant recruits to *A. stamineus* populations on Oʻahu, which may be due to relatively higher and more variable rates of mortality for ocean-going migrants (Heim-Ballew 2019).

Our findings also suggest that *A. stamineus* populations on Oʻahu are likely very responsive (i.e., susceptible) to shifts in conditions that can alter resident recruitment, including interventions intended to promote population stability and resilience. For example, restoration and management strategies might be more successful if interventions stabilize base flows, promote greater nutrient balance, and reduce AIS densities. Given evidence of complex interactions among stressors, more holistic management approaches will likely be necessary to protect or restore at-risk populations. Further work on other facultative migrants could offer additional insight on how to prioritize management efforts. Likewise, understanding how local interventions can yield 'spillover' benefits to other watersheds on an island- or other islands- would also offer valuable perspective on how to address broader geographies of anthropogenic pressures acting on native species of concern.

6.4 GENOMIC AND LIFE HISTORY RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Genomic variation.

Patterns of genomic diversity reflected abiotic and biotic variation across the study watersheds. Cleaning and trimming raw ddRAD sequence file yielded 35,001 SNPs that passed quality control for further analysis of the 262 *A. stamineus* juvenile samples that were analyzed for both life history and genomic variation (Table 16). Analysis of SNP variation among the sampled *A. stamineus* revealed the presence of genomic differentiation (Table 17, Table 18) and variable levels of expected heterozygosity within and among watersheds (Table 19). While we did not recover a relationship between expected heterozygosity and mean watershed discharge ($p = 0.37$) (Fig. 26), we did find that expected heterozygosity increased with increasing discharge variability ($p < 0.001$) (Fig. 26).

Comparisons according to migratory life history revealed that genomic diversity differed between resident and migrant types. Despite there being a larger number of residents ($n_{residents} = 219$, $n_{migrants} = 43$) among the sampled individuals, migrant *A. stamineus* were found to harbor higher expected heterozygosity ($t= 3.40$; $p = 0.001$) (Fig. 27), although the extent of differences varied among watersheds ($p < 0.001$) (Fig. 27).

Table 16. The number of individuals per migratory type in each watershed that were used for genomic analyses.

Watershed	Number of migrants	Number of residents
Heʻeia	0	1
Hālawa	5	11
Kahaluʻu	2	42
Kalihi	6	17
Keaʻahala	6	36
Mānoa	3	22
Punaluʻu	1	18
Waianu	3	19
Waihee	3	15
Waikāne	1	6
Waimānalo	1	23
Waimea	12	9
Total	**43**	**219**

Genomic differentiation.

Even though the overall estimate of genomic differentiation among the study watersheds was quite low (global F_{ST} = 0.009), estimates of differentiation were quite variable between watersheds with pairwise F_{ST} values ranging from 0.007 to 0.03 (Table 17). We also recovered a greater amount of pairwise differentiation between resident and migratory types (global F_{ST} = 0.012) (Table 18). Consistent with

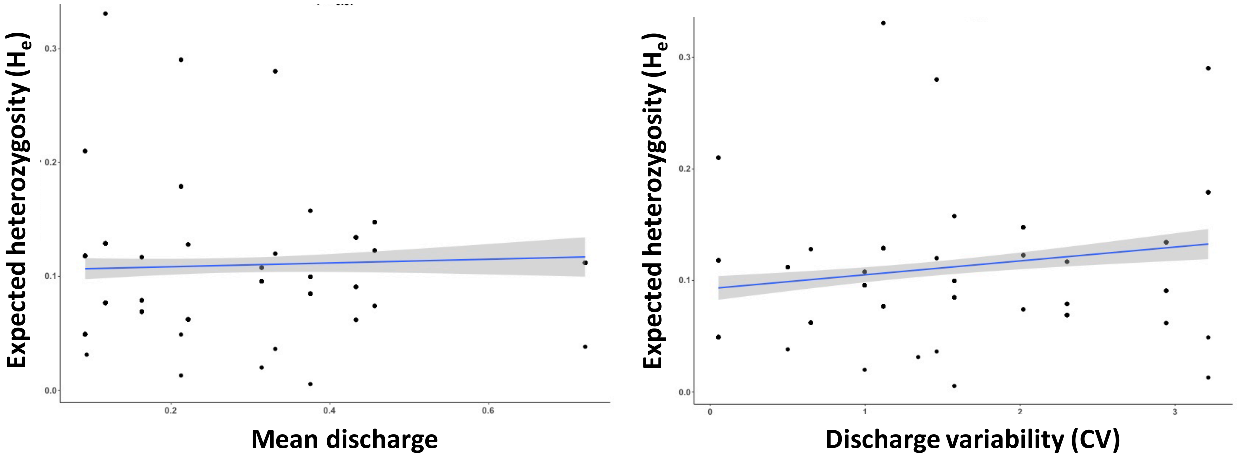


Figure 26. While there was no relationship between genomic diversity and (a) mean discharge, there was a significant positive relationship with (b) discharge variability.

.

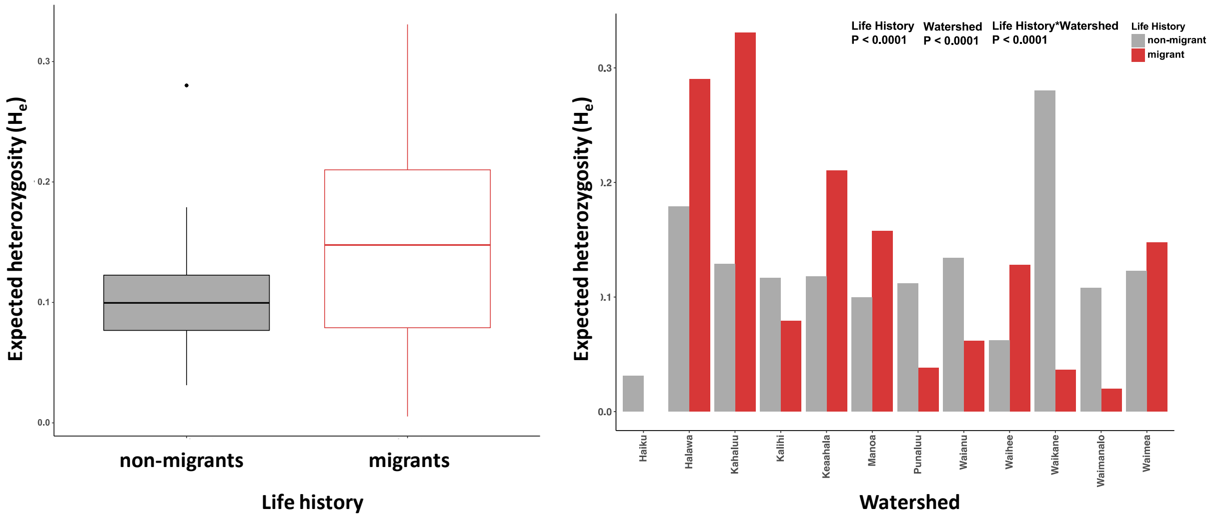


Figure 27. Genomic diversity differed according to migratory ecotype (left), with migrants exhibiting greater diversity with individuals pooled across all watersheds. Genomic diversity varied, however, by migratory type among watersheds (right).

this, an analysis of molecular variance (AMOVA) found that 86% of the total variation occurred within individuals, with the next largest component (11.6%) being among migratory types within watersheds (Table 19). Variation between watersheds constituted 2.4% of the total variation We recovered a pattern of significant genomic differentiation among *A. stamineus* structured by watershed (Fig. 28), despite patterns of genomic diversity differing among watersheds according to surface discharge and AIS removal.

Table 17. Pairwise F_{ST} estimates between watersheds with all individuals pooled by location.

	He'eia	Hālawa	Kahalu'u	Kalihi	Kea'ahala	Mānoa	Punalu'u	Waianu	Waihee	Waikāne	Waimānalo	Waimea
He'eia	0.000	0.021	0.003	0.019	0.021	0.007	0.007	0.009	0.011	0.031	0.009	0.017
Hālawa		0.000	0.012	0.007	0.008	0.011	0.014	0.010	0.010	0.012	0.013	0.009
Kahalu'u			0.000	0.005	0.004	0.006	0.006	0.004	0.005	0.015	0.005	0.005
Kalihi				0.000	0.006	0.008	0.009	0.007	0.007	0.011	0.010	0.007
Kea'ahala					0.000	0.006	0.006	0.005	0.005	0.010	0.008	0.005
Mānoa						0.000	0.009	0.006	0.007	0.011	0.007	0.006
Punalu'u							0.000	0.008	0.008	0.022	0.010	0.007
Waianu								0.000	0.009	0.011	0.010	0.007
Waihee									0.000	0.012	0.006	0.007
Waikāne										0.000	0.023	0.010
Waimānalo											0.000	0.014
Waimea												0.000

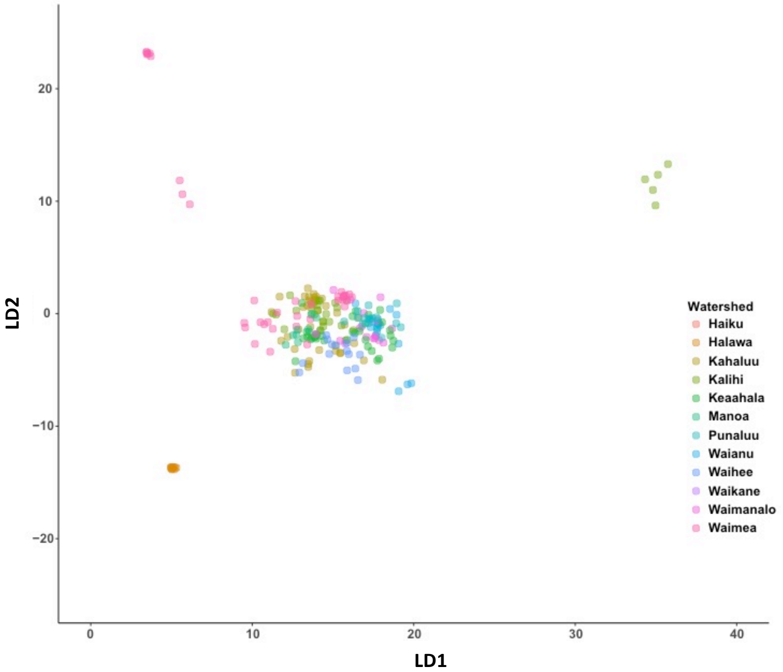


Figure 28. Discriminant analysis of principal components illustrating genomic variation between *A. stamineus* within watersheds and differentiation between watersheds.

Notably, DAPC-based assignment analysis revealed evidence of genetic divergence according to migratory life history (Fig. 29), despite contextual patterns of genomic diversity and differentiation across watersheds. Strong clustering by migratory life history is consistent with the higher estimates of pairwise genomic differentiation recovered in comparison of residents and migrants.

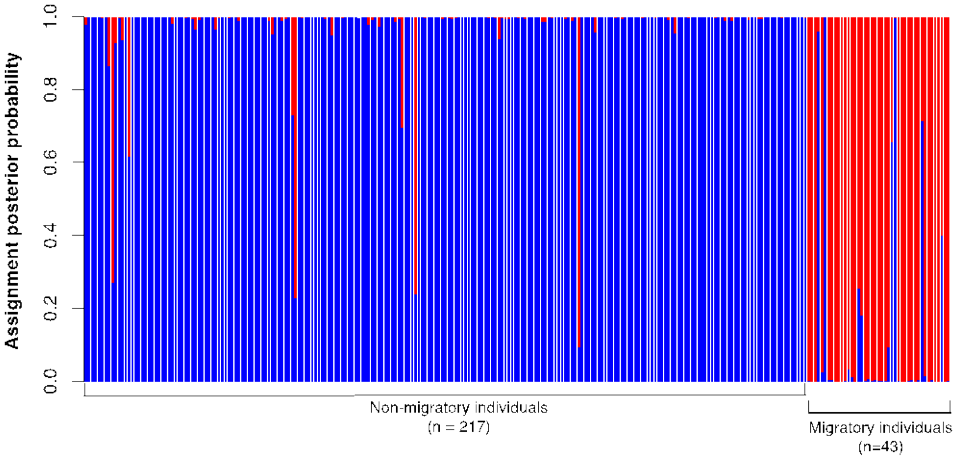


Figure 29. Bar plot from DAPC of genetic differentiation between migratory types (non-migratory residents = blue, versus migrants = red) across all watersheds, indicating that genetic variation in A. stamineus is structured according to migratory life history.

Table 18. Pairwise F_{ST} estimates within and among watersheds with individuals categorized according to migratory life history (R = resident; M = migrant).

	Heeia (R)	Halawa (R)	Kahaluu (R)	Kalihi (R)	Keaahala (R)	Manoa (R)	Punaluu (R	Waianu (R)	Waihee (R)	Waikane (R)	Waimanalo (R)	Waimea (R)	Halawa (M)	Kahaluu (M)	Kalihi (M)	Keaahala (M)	Manoa (M	Punaluu (M)	Waianu (M)	Waihee (M)	Waikane (M	Waimanalo (M	Waimea (M)
Heeia (R)	0	0.018	0.006	0.011	0.007	0.01	0.013	0.012	0.014	0.031	0.009	0.019	0.024	0	0.027	0.034	0.004	0	0.006	0.008	0	0	0.015
Halawa (R)		0	0.005	0.007	0.005	0.004	0.007	0.007	0.007	0.011	0.006	0.009	0.01	0.019	0.008	0.011	0.015	0.022	0.012	0.013	0.019	0.02	0.009
Kahaluu (R)			0	0.005	0.004	0.004	0.004	0.004	0.004	0.006	0.004	0.005	0.006	0.006	0.005	0.005	0.008	0.009	0.004	0.005	0.009	0.007	0.005
Kalihi (R)				0	0.005	0.005	0.007	0.005	0.005	0.008	0.005	0.007	0.008	0.011	0.006	0.007	0.011	0.012	0.008	0.01	0.023	0.015	0.006
Keaahala (R)					0	0.004	0.004	0.005	0.004	0.006	0.004	0.006	0.007	0.008	0.006	0.006	0.007	0.007	0.005	0.006	0.009	0.008	0.005
Manoa (R)						0	0.005	0.005	0.006	0.008	0.004	0.007	0.008	0.01	0.007	0.007	0.01	0.012	0.006	0.008	0.02	0.01	0.006
Punaluu (R)							0	0.006	0.006	0.01	0.004	0.008	0.011	0.013	0.008	0.008	0.013	0.015	0.009	0.011	0.017	0.016	0.006
Waianu (R)								0	0.006	0.009	0.005	0.008	0.009	0.013	0.009	0.008	0.012	0.013	0.01	0.012	0.015	0.014	0.006
Waihee (R)									0	0.009	0.004	0.007	0.009	0.014	0.007	0.007	0.011	0.019	0.009	0.009	0.021	0.015	0.007
Waikane (R)										0	0.008	0.01	0.014	0.025	0.013	0.014	0.014	0.034	0.013	0.016	0.039	0.037	0.01
Waimanalo (R)											0	0.007	0.01	0.011	0.007	0.008	0.011	0.013	0.008	0.007	0.011	0.012	0.007
Waimea (R)												0	0.01	0.015	0.011	0.012	0.014	0.02	0.011	0.011	0.034	0.016	0.006
Halawa (M)													0	0.019	0.008	0.011	0.007	0.007	0.005	0.013	0.019	0.02	0.009
Kahaluu (M)														0	0.028	0.029	0.015	0	0.009	0.014	0	0	0.017
Kalihi (M)															0	0.014	0.015	0.031	0.012	0.012	0.045	0.038	0.008
Keaahala (M)																0	0.015	0.04	0.015	0.007	0.028	0.041	0.01
Manoa (M)																	0	0.011	0.021	0.021	0.015	0.007	0.014
Punaluu (M)																		0	0.006	0.009	0.003	0	0.02
Waianu (M)																			0	0.013	0.013	0.011	0.009
Waihee (M)																				0	0.011	0.013	0.012
Waikane (M)																					0	0.016	0.015
Waimanalo (M)																						0	0.016
Waimea (M)																							0

Genomic responses to AIS removals.

Overall, we found that *A. stamineus* genomic diversity increased (Fig. 30) compared to pre-removal genomic diversity (t = 4.11; *p* < 0.001) (Fig. 30). We also detected a reach effect, where shifts in genomic diversity were larger for removal reaches than control reaches (t = -2.11, *p* = 0.04) following AIS removals. Though residents appear to exhibit a larger increase in genomic diversity compared to migrants, the difference was not significant (*p* = 0.82; Fig. 30).

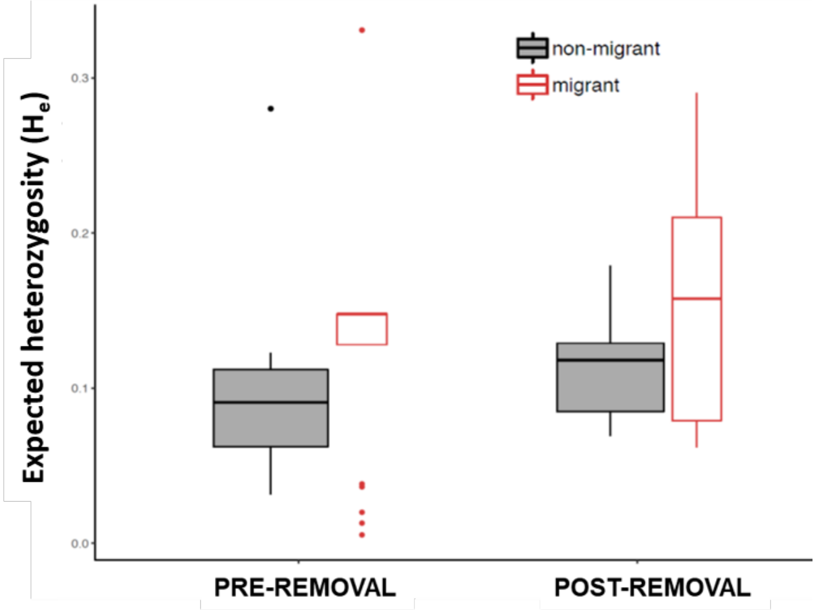


Figure 30. Genomic diversity before and after AIS removals with individuals pooled across all watersheds and binned according to migratory life history.

Consistent with these results, we found that the genomic diversity of *A. stamineus* following AIS removals was positively correlated with the amount of Poeciliid biomass (*p* < 0.001; Fig. 31) taken out of removal reaches. In contrast, no correlation was recovered between the genomic diversity of A. stamineus before AIS removals and the amount of Poeciliid biomass (*p* > 0.05; Fig. 31) taken out of removal reaches. No correlations were recovered with the amount of Loricariid biomass removed from the study watersheds for either time period (both, *p* > 0.05; Fig. 31).

Life history responses to AIS removals.

Despite evidence indicating that life history variation is influenced by AIS densities across our study watersheds (Fig. 22), we did not find a significant shift in the proportion of *A. stamineus* residents following AIS removals (*p* > 0.05) when pooling all individuals across all watersheds according to time period. Consistent with this, we also did not detect a significant difference between control and removal reaches across all watersheds.

Likewise, comparisons of pre-removal and post-removal proportions in individual watersheds suggest that AIS removal did not elicit a consistently strong life history response. Responses varied among individual watersheds, though some variation is attributable to low samples sizes (before or after removals, or during both time periods) in some of the study watersheds. Nonetheless, comparisons limited to the four watersheds that had the greatest pre- and post-removal sample sizes (Keaʻahala,

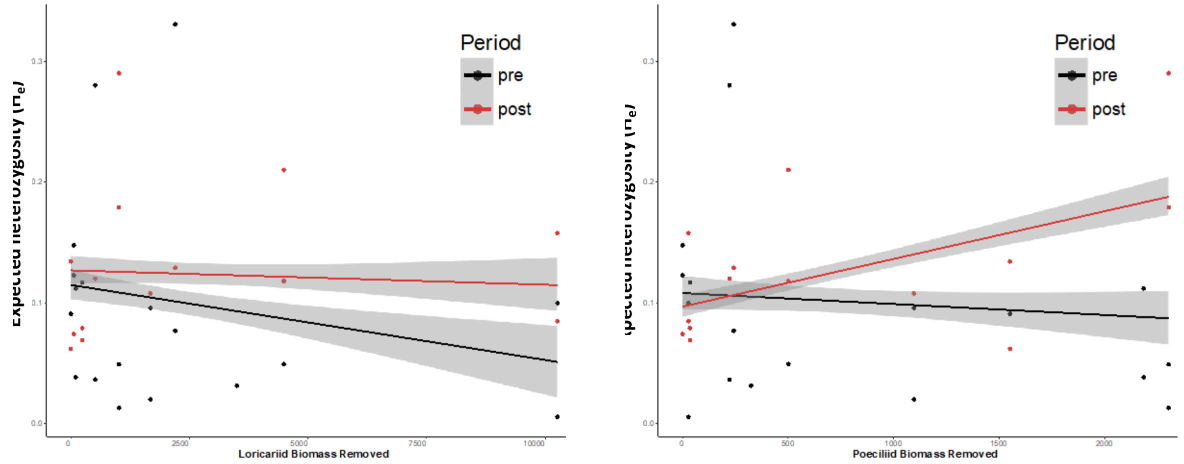


Figure 31. Genomic diversity pre- and post-removal in relation to AIS biomass (g) removed of (left) Loricariids and (right) Poeciliids.

Kahaluʻu, Mānoa, Waimānalo) did not recover statistically significant differences in the proportion of residents by time period or reach (both, *p* > 0.05).

Comparisons designed to explore the possibility of a short-lived response found stronger evidence of a shift in life history centered around AIS removals. Overall, a statistically significant shift was not detected (F = 2.09; d.f. = 3; *p* = 0.12), but there was a provocative trend in life history variation over the course of the study period (Fig. 32). The trend persisted when watersheds with low sample sizes (n < 3) that had a fixed (i.e., 0 or 1) proportion were removed from consideration, and strengthened further when pre-removal data was constrained to the three month window immediately preceding AIS removals (F = 2.91; d.f. = 3, *p* = 0.058). Post hoc Tukey tests revealed that the largest difference occurred between the 0-3 months pre-removal period relative to the >6 months post-removal period (*p* = 0.056), suggestive of a progressive rise in the proportion of residents following AIS removals. Similarly-minded comparisons between the 2-month pre-removal to 2-month post-removal time periods detected a statistically non-significant 12% increase (*p* = 0.24) in the proportion of residents when all individuals were pooled across all watersheds.

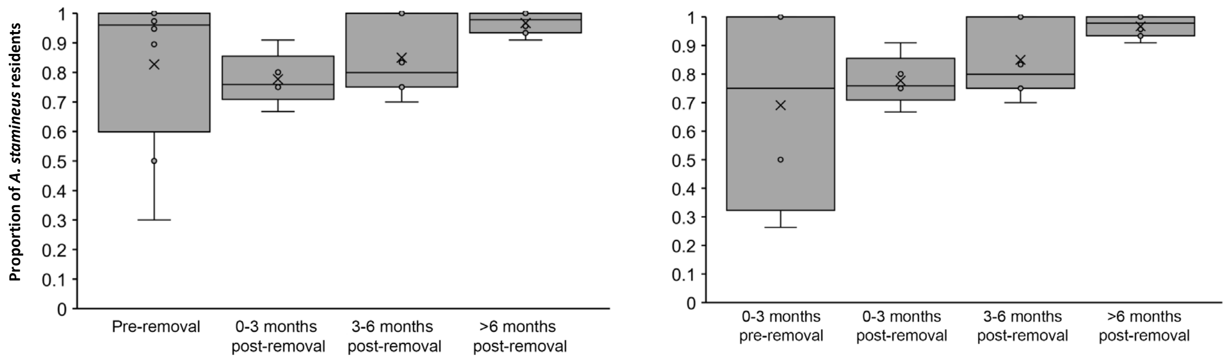


Figure 32. Box-whisker plots of the proportion of residents in populations of *Awaous stamineus* across 12 study watersheds on Oʻahu prior to AIS removals (left) and during a 0-3 month pre-removal period (right) relative to 0-3 months post-removal, 3-6 months post-removal, and > 6 months post-removal. X indicates the mean, horizontal line indicates the median, boxes delineate the 1st and 3rd quartiles, whiskers indicate the minimum and maximum range of the data. Data points outside the whiskers are outliers.

Discussion.
In this study, we examined whether AIS removals elicited genetic and demographic rescue of *A. stamineus* populations across 12 watersheds on the island of Oʻahu. Baseline surveys revealed that the proportions of resident and migrant life history types varied across watersheds and that migrants exhibit greater genomic diversity compared to residents. We also found evidence of weak but significant genomic differentiation between watersheds and unexpectedly strong differentiation according to migratory life history. AIS removals elicited an increase in genomic diversity, but the shift in diversity was not associated with an increase in immigration, as theory would predict for genetic rescue. Rather, we detected an increase in the proportion of residents concurrent with a brief rise in recruitment, which suggests that the observed increase in genomic diversity is due to localized demographic rescue (e.g., greater survival and reproduction of local residents). These findings illustrate that conservation practices like AIS removal can promote recovery through localized responses, even in migratory species with the potential for high vagility. Our results also suggest that conservation practices should not solely focus on increasing population connectivity or be solely based on the expectation that recovery will result from immigration. Rather, greater consideration should be given to how practices can concurrently promote local demographic and genomic rescue to foster the recovery of threatened species like *A. stamineus*.

Populations of *A. stamineus* are at risk across the Hawaiian archipelago, particularly on Oʻahu (Brasher et al. 2003, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). It appears, however, that *A. stamineus* are more resilient to watershed impairment than most, if not all, of the other native amphidromous fishes in Hawaiian streams (Lisi et al. 2018). Some evidence suggests that the resilience of *A. stamineus* may in part be due to its facultative migratory life history (Hogan et al. 2014, Heim-Ballew 2019). Yet, three of the four other native amphidromous fishes also exhibit evidence of facultative migration (Heim-Ballew et al. 2020), and it appears that residents dominate *A. stamineus* populations across Oʻahu, even in highly impaired watersheds (Heim-Ballew 2019) (Fig. 22). This runs contrary to the expectation that local populations should be sustained by immigration because impairment constrains survival and reproduction of residents (Waits et al. 2008, Blum et al. 2014). Evidence of dietary flexibility (Lisi et al. 2018) suggests that *A. stamineus* are more capable of coping with impairment, raising the possibility that trophic plasticity also confers greater capacity for local demographic recovery. This, along with the relatively high proportion of residents in local populations (Fig. 22), may facilitate demographic rescue across Oʻahu rather than genetic rescue via immigration from populations elsewhere in the archipelago.

We predicted but did not find evidence that AIS removal elicits a demographically cryptic rise in genomic variation driven by replacement of residents by migrants. Rather, we found evidence of elevated genomic diversity as well as a brief rise in population densities (Fig. 11), recruitment (Fig. 16) and the proportion of resident *A. stamineus* (Fig. 32) following AIS removals. In aggregate, these findings are more consistent with predicted outcomes of demographic rescue, including a concomitant rise in genomic diversity. It is possible that the observed shifts reflect direct and indirect release from pressures imposed by AIS. AIS removals likely reduced competition, allowing *A. stamineus* to gain greater access to habitat and resources. Consistent with this, we found evidence of greater growth and elevated body condition following AIS removals (Fig. 19). The timing of the observed rise in population densities (Fig. 11) and recruitment (Fig. 16) suggests that reduced competition may have indirectly promoted higher reproductive output (i.e., which would increase larval supply and ultimately recruitment 3-5 months later). Removing AIS may also have concurrently reduced predation of early life stages, particularly resident larvae that experience greater exposure to predators like Poeciliid live-bearers (Hain et al. 2019). While plausible, this hypothesis warrants further study (e.g., of depredation and reproduction according to AIS presence and densities) to gain greater perspective on the factors and pathways that lead to population recovery following AIS removal.

Genetic rescue via immigration may be more likely to occur in other native migratory gobies under particular sets of conditions. It might be especially likely for *S. stimpsoni*, which is the only obligate amphidromous goby in Hawaiian streams (Heim-Ballew et al. 2020). Gene flow is high between populations of *S. stimpsoni* (Moody et al. 2015), which might suggest that genetic rescue via immigration is a viable conservation strategy. Yet in-stream post-settlement selection is strong enough to overcome influences from immigration (Moody et al. 2015, Moody et al. 2019). Evidence that selection produces divergent morphologies across streams reflecting highly localized pressures (for waterfall climbing or predator evasion; Blob et al. 2008, Blob et al. 2010, Moody et al. 2017) indicates that local production and contribution of larvae to offshore pools is likely key to persistence. Consequently, 'immigrants' might effect genetic and demographic rescue by sustaining locally adapted populations. If so, then conservation efforts might fall short if only focused on improving connectivity. It might instead be necessary to increase local reproductive output, reduce anthropogenic pressures across migratory gauntlets (Hain et al. 2019), and maintain ocean-stream connectivity. Genetic rescue that involves immigration of recruits drawn from well-mixed offshore pools is more likely to occur where either obligate or facultative amphidromous species are exceedingly rare. Several native migratory species- including *S. stimpsoni, L. concolor,* and the snail *Neritina granosa*- are thought to be extirpated or rare across Oʻahu (Devick et al. 1995, Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). Processes acting at very low population densities, like Allee effects, might prohibit contributions of residents to recruitment, particularly where impairment is constraining survival and reproduction (Taylor and Hastings 2005, Potapov and Lewis 2008, Tobin et al. 2011). Accordingly, a 'clean-slate' strategy (Fig. 1) that relies on immigration from non-local sources might be the most effective approach for restoring genetic and demographic viability of at-risk populations of native migratory species.

The unexpected finding of genomic differentiation among resident and migrant *A. stamineus* indicates that the spatial and temporal scale of management interventions can modify the composition and thus evolutionary trajectory of local populations. Evidence that AIS removals elevated genomic diversity likely through a proportional rise in residents- in populations already dominated by residents (Fig. 22)- suggests that our actions did little to change the composition or trajectory of *A. stamineus* in the study sites across Oʻahu. More aggressive measures, however, like 'clean-slate' approaches to restoration, may affect this outcome. To some, this is an acceptable risk, particularly when extirpation or extinction is imminent. To others, it reinforces long-standing concerns about the risks involved with managing for genetic rescue. The theory and concept of genetic rescue focus on the premise of increasing gene flow, via immigration, between small isolated populations (Ingvarsson 2001; Whiteley et al. 2015; Hufbauer et al. 2015). This has proven to be remarkably controversial in part because genetic rescue can occur without changes in demography (i.e., increasing population size), and thus populations can still be lost (Bell et al. 2019). Additionally, while immigration can be beneficial, it can also potentially result in the introduction of maladaptive alleles that can hasten demographic decline (Keller et al. 2001, Tallmon et al. 2004). Despite good evidence to the contrary (e.g., Fitzpatrick et al. 2020), this concern has been cited in objections to management efforts that promote dispersal, like assisted migration and other forms of transplantation (Hedrick 1995, Mills et al. 1996, Pimm 2006, Edmands 2007, Bell et al. 2019). Concerns about promoting maladaption are certainly relevant to native Hawaiian gobies, given evidence of local adaptation in *S. stimpsoni* (Moody et al. 2015, Moody et al. 2019). It is possible that actions taken to increase gene flow might overwhelm selective pressures that otherwise keep the influence of maladaptive alleles in check. Likewise, actions that alter the proportional representation of migratory phenotypes might similarly erode local adaptation, and potentially shift local populations to become more homogeneous. It might also cause populations to follow novel evolutionary pathways (Garant et al. 2007). Accordingly, consideration should be given to possible trade-offs between demographic and evolutionary recovery before interventions are undertaken.

Importantly, possible trade-offs might not always be evident, as evolutionary differentiation can go undetected (Wu et al. 2016; Dahruddin et al. 2017; Watanabe et al. 2017). This is certainly well illustrated by our finding that *A. stamineus* harbors highly divergent migratory phenotypes, as all prior studies of genetic variation in *A. stamineus* have found little evidence of intraspecific evolutionary divergence (e.g., Lindstrom et al. 2012, Blum et al. 2014, Alda et al. 2016). Similarly, recent studies of Atlantic cod have revealed that the largest component of genetic divergence corresponds to two migratory phenotypes that coexist in varying proportions rather than hierarchical population structure or differentiation among multiple, local populations with partial isolation. As it has long been thought that Atlantic cod exhibit fixed migratory strategies, the observed pattern of differentiation could have easily been misinterpreted as evidence of high gene flow and population connectivity. Instead, it served as the basis for determining that two migratory phenotypes are widely sympatric in Icelandic waters (Thorsteinsson et al. 2012), in the North Sea (Barth et al. 2017; Sodeland et al. 2016) and in the western Atlantic (Barney et al. 2017). Though more geographically extensive comparisons will be necessary to confirm prevalence and distributions, it likewise appears that there are two widely sympatric migratory phenotypes of *A. stamineus* in the Hawaiian Islands. The novelty of this finding raises the prospects that ongoing or planned management efforts may (though well intended) be ineffective or perhaps even cause more harm than good.

Efforts to conserve and manage native fauna in Hawaiian streams have long relied on the assumption that immigration can sustain or replenish threatened populations. Progressive study continues to reveal, however, that this is not a safe or simple assumption. Some work supports the premise that amphidromous species can readily (re)colonize streams, even if watersheds are impaired or disturbed (McDowall 2007a, McDowall 2007b). For example, studies of recent, volcanically formed freshwater habitat illustrate that some amphidromous species are capable of pioneering colonization (e.g., Schoenfuss et al. 2004). But, dispersal-driven (re)colonization is not a sure thing. The highly dynamic nature of oceanographic currents can result in few to no post-larvae recruiting back to freshwater habitats (Cook et al. 2009, Alda et al. 2016, Riginos et al. 2016, Moody et al. 2019). And, even if individuals reach available habitat, there may be little to no survival and establishment (Moody et al. 2019), in part because in-stream degradation can limit recruitment (Blum et al. 2014, Hain et al. 2019). Evidence that populations draw from highly localized pools of recruits either due to natal retention (Hogan et al. 2014, Heim-Ballew et al. 2020) or selection-driven adaptive constraints (Moody et al. 2015, Moody et al. 2019) also suggests that dispersal-driven immigration exerts little influence on local demography. Greater understanding of this paired dynamic can help clarify longstanding incongruencies in species distributions and abundance, including the rarity of several species across Oʻahu and low densities of native fauna in remote and relatively undisturbed watersheds on Oʻahu that should be capable of harboring verdant populations (Blum et al. 2014, Moody et al. 2017). Better understanding of how life history moderates genomic and demographic variation will likewise help ground management expectations and deliberations about executing interventions like AIS removal. It is imperative, however, to develop and expand pipelines that hasten translation of research into practice. As threats continue to build, collaborations that foster communication and build trust (i.e., among communities with sometimes disparate priorities and values) may very well prove vital to securing the biological legacies of the Hawaiian Islands and other oceanic archipelagos.

Table 19. Estimates of expected heterozygosity for *A. stamineus* populations across hierarchical levels (i.e., watershed; watershed by migratory ecotype; watershed by period (pre-/post-removal); watershed by migratory ecotype by period (pre-/post-removal)).

Spatial scale	Population	Migratory ecotype	Time period	Expected heterozygosity (He)
Watershed	Haiku	All	Both	0.34
	Hālawa	All	Both	0.34
	Kahaluʻu	All	Both	0.25
	Kalihi	All	Both	0.25
	Kāwā	All	Both	0.34
	Keaʻahala	All	Both	0.22
	Mānoa	All	Both	0.22
	Punaluʻu	All	Both	0.38
	Waianu	All	Both	0.22
	Waihee	All	Both	0.23
	Waikāne	All	Both	0.26
	Waimānalo	All	Both	0.23
	Waimea	All	Both	0.26
Watershed by migratory ecotype	Hālawa	Migrant	Both	0.23
	Kahaluʻu	Migrant	Both	0.15
	Kahlihi	Migrant	Both	0.11
	Kalihi	Migrant	Both	0.22
	Keaʻahala	Migrant	Both	0.23
	Mānoa	Migrant	Both	0.19
	Punaluʻu	Migrant	Both	0.09
	Waianu	Migrant	Both	0.17
	Waihee	Migrant	Both	0.21
	Waikāne	Migrant	Both	0.10
	Waimānalo	Migrant	Both	0.08
	Waimea	Migrant	Both	0.25
	Haiku	Resident	Both	0.09
	Hālawa	Resident	Both	0.25
	Kahaluʻu	Resident	Both	0.26
	Kalihi	Resident	Both	0.26
	Keaʻahala	Resident	Both	0.26
	Mānoa	Resident	Both	0.26
	Punaluʻu	Resident	Both	0.25
	Waianu	Resident	Both	0.26
	Waihee	Resident	Both	0.25
	Waikāne	Resident	Both	0.23
	Waimānalo	Resident	Both	0.26
	Waimea	Resident	Both	0.24
	Haiku	Both	Pre	0.34

Watershed by period	Hālawa	Both	Pre	0.21
	Kahaluʻu	Both	Pre	0.12
	Kalihi	Both	Pre	0.15
	Kāwā	Both	Pre	0.33
	Keaʻahala	Both	Pre	0.10
	Mānoa	Both	Pre	0.11
	Punaluʻu	Both	Pre	0.21
	Waianu	Both	Pre	0.11
	Waihee	Both	Pre	0.12
	Waikāne	Both	Pre	0.16
	Waimea	Both	Pre	0.19
	Wamanalo	Both	Pre	0.12
	Haiku	Both	Post	0.02
	Hālawa	Both	Post	0.22
	Kahaluʻu	Both	Post	0.19
	Kalihi	Both	Post	0.12
	Kāwā	Both	Post	0.18
	Keaʻahala	Both	Post	0.19
	Mānoa	Both	Post	0.18
	Waianu	Both	Post	0.21
	Waikāne	Both	Post	0.09
	Waimānalo	Both	Post	0.12
	Waimea	Both	Post	0.11
Watershed by migratory ecotype by period	Hālawa	Migrant	Pre	0.01
	Kahaluʻu	Migrant	Pre	0.33
	Mānoa	Migrant	Pre	0.01
	Punaluʻu	Migrant	Pre	0.04
	Waihee	Migrant	Pre	0.13
	Waikāne	Migrant	Pre	0.04
	Waimānalo	Migrant	Pre	0.02
	Waimea	Migrant	Pre	0.15
	Haiku	Resident	Pre	0.03
	Hālawa	Resident	Pre	0.05
	Kahaluʻu	Resident	Pre	0.08
	Kalihi	Resident	Pre	0.12
	Keaʻahala	Resident	Pre	0.05
	Mānoa	Resident	Pre	0.10
	Punaluʻu	Resident	Pre	0.11
	Waianu	Resident	Pre	0.09
	Waihee	Resident	Pre	0.06
	Waikāne	Resident	Pre	0.28
	Waimānalo	Resident	Pre	0.10

Waimea	Resident	Pre	0.12
Hālawa	Migrant	Post	0.29
Kalihi	Migrant	Post	0.08
Keaʻahala	Migrant	Post	0.21
Mānoa	Migrant	Post	0.16
Waianu	Migrant	Post	0.06
Hālawa	Resident	Post	0.18
Kahaluʻu	Resident	Post	0.13
Kalihi	Resident	Post	0.07
Keaʻahala	Resident	Post	0.12
Mānoa	Resident	Post	0.08
Waianu	Resident	Post	0.13
Waikāne	Resident	Post	0.12
Waimānalo	Resident	Post	0.11
Waimea	Resident	Post	0.07

6.5 ECOSYSTEM RESPONSES TO AIS REMOVAL FROM HAWAIIAN STREAMS

Survey of ecosystem conditions.

Baseline nutrient concentrations varied widely among our focal watersheds (Table 20). Across the spectrum of nitrate (2.7 to 640.1 µg L^{-1}), ammonium (0.62 to 61.5 ug L^{-1}), and soluble reactive phosphorus (2.6 to 58.0 µg L^{-1}), several streams showed consistently high dissolved nutrients (Keaʻahala, Waimānalo, Kāwā) while others were nutrient-poor (Waimea, Kīpapa). Watershed land use appears to explain some of this heterogeneity. For instance, percent forest cover was negatively correlated with SRP (slope=-0.385, $p = 0.002$, $r^2 = 0.53$) and NO_3 (slope=-3.84, $p = 0.036$, $r^2 = 0.34$) concentrations.

Table 20. Ecosystem variables measured in each stream prior to AIS removals. Streams are ordered by increasing pre-removal biomass density of armored catfish (g m^{-2}). Urban land use is represented by the percent of watershed area covered by impervious surfaces (Imperv) and forest (Forest). The median stream discharge (Q50) is shown as an index of stream size. Water chemistry metrics include concentrations of total suspended solids (TSS, mg L^{-1}), nitrate (NO_3, µg N L^{-1}), and soluble reactive phosphorus (SRP, µg P L^{-1}). As a proxy for light availability, we measured canopy closure using a conical densitometer in the stream. We measured periphyton accrual using Chlorophyll a (Chla, µg cm^{-2} day^{-1}) extracted from the control treatments of nutrient diffusing substrates (NDS). Whole-stream metabolism was modeled with the Bayesian Metabolism Model to estimate gross primary production (GPP, mg O_2 m^{-2} d^{-1}) and ecosystem respiration (ER, mg O_2 m^{-2} d^{-1}), and GPP:ER (P:R) was used to infer ecosystem-level heterotrophy when the ratio is <1.0. Leaf litter mass loss rate (Leaf Decay, g day^{-1}) was measured from *Hibiscus tiliaceaus*.

Stream	Catfish	Guppies	total AIS	Forest	Imperv	Q50	TSS	NO3	SRP	Light	Chla	GPP	ER	P:R	Leaf Decay
Waimea	0.0	0.0	4.7	90	0.58	0.14	0.0005	4.3	3.4	9.23	0.85	2.28	3.38	0.67	0.036
Waianu	0.0	3.1	7.5	62	1.3	0.28	0.0034	27.7	19.3	30	5.37	6.60	8.55	0.78	0.045
Punalu’u	0.0	2.6	4.8	76	1.53	0.62	0.0035	3.4	9.6	41.29	3.36	8.84	7.75	1.19	0.023
Halawa	0.0	8.6	26.6	86	31.67	0.04	0.0015	69.2	8.5	6.13	0.63				0.034
Kīpapa	0.0	0.0	1.9	70	12.92	0.07	0.0008	28.8	3.7	10.89	0.30	1.32	3.01	0.44	0.035
Kalihi	0.5	0.1	5.1	89	23.37	0.06	0.0012	28.2	9.3	11.41	0.18	1.16	8.84	0.13	0.039
Waikāne	0.7	0.3	2.0	79	1.11	0.23	0.0049	71.0	8.5	3.01	0.86	0.42	2.38	0.17	0.048
Waimānalo	1.5	2.0	13.8	60	11.12	0.26	0.0101	245.4	13.1	44.57	4.25	2.31	2.12	1.09	0.046
Kahalu’u	4.3	0.5	5.8	30	10.89	0.10	0.0006	75.5	21.6	8.11	0.86	0.65	1.15	0.57	0.058
He’eia	7.1	0.7	10.3	34	16.01	0.07	0.0011	49.3	20.9	33.22	2.14	0.68	1.03	0.66	0.050
Kāwā	9.2	0.0	12.7	23	29.78	0.04	0.0027	570.6	25.7	48.27	0.82	1.35	7.04	0.23	0.053
Keaʻahala	13.9	1.5	18.9	35	46.12	0.09	0.0024	166.6	48.1	35.22	3.11	0.70	7.19	0.09	0.038
Mānoa	23.0	0.1	31.4	60	35.86	0.16	0.0034	21.5	9.8	25.14	2.57	1.52	1.61	0.97	0.044

Leaf litter loss rates were also highly variable before removals (0.10 to 0.58 g day^{-1}) (Fig. 33). Leaf decomposition was slowest in Punalauʻu and Kīpapa watersheds, and most rapid in Kāwā and Waimānalo watersheds. Loss rates were positively correlated with dissolved nitrate (log$_{10}$-transformed) concentrations (β_0 = 0.018, slope = -0.012, p <0.001, r^2=0.36) but not ammonium or SRP, suggesting N-limitation of microbial activity. Although the C:N of leaf packs exhibited only modest variation, C:N of remaining leaf material was negatively related to leaf loss rates (slope=-227.20, p = 0.0017, r^2=0.18), reflecting the association between N-rich microbial biomass and organic decomposition rates.

NDS assays revealed the presence of a wide range of algal accrual rates in the absence of supplemental nutrients, ranging from nearly zero (e.g., in Kīpapa watershed) to rapid biofilm growth (0.1 to 7.6 mg Chl a d^{-1}) (Fig. 33). Before removals, NDS assays indicated N-limitation in two streams (Waimea, Kalihi

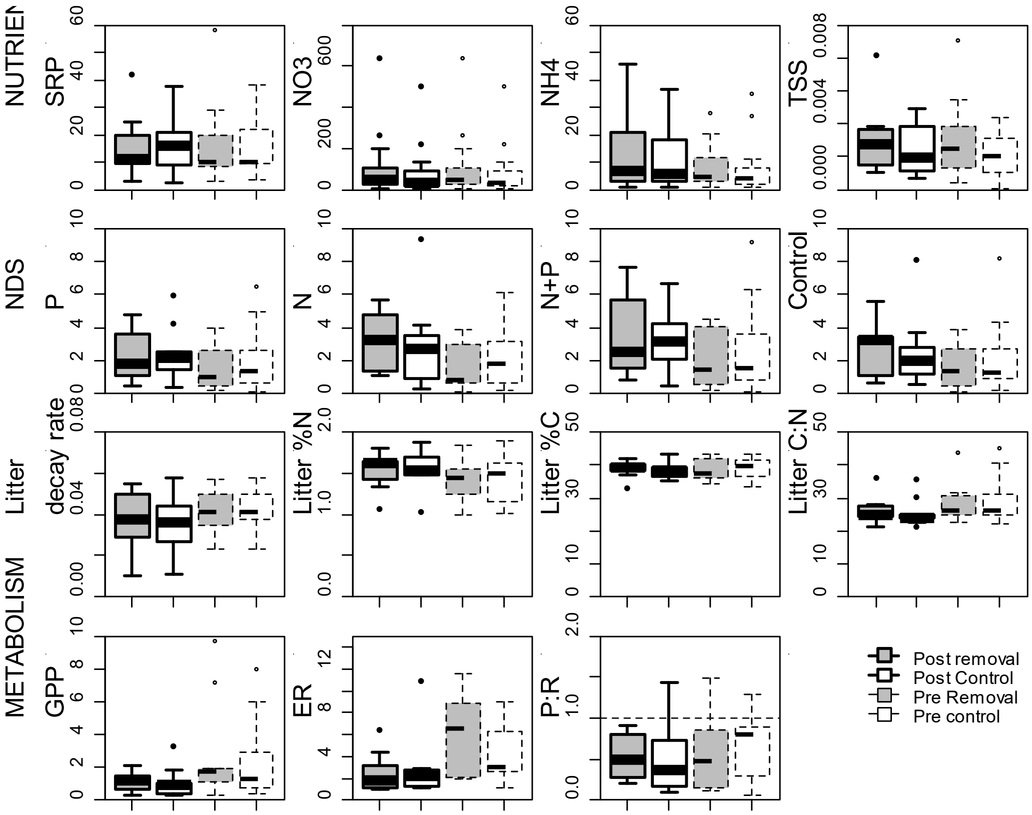


Figure 33. Summary statistics for ecosystem processes measured across 13 streams on Oʻahu in control (white) and removal (grey) reaches both before (dashed outline) and after (dark outline) experimental removal of aquatic invasive species. Top row indicates water chemistry variables: soluble reactive phosphorus (SRP, µg P L^{-1}), nitrate (NO_3, µg N L^{-1}), ammonium (NH_4, ug N L^{-1}), and total suspended solids (TSS, mg L^{-1}). Second row indicates results of algal accrual on nutrient diffusing substrates (NDS, µg cm^{-2} d^{-1}) supplemented with phosphorus (P), nitrogen (N), both (N+P), or neither (Control). Third row shows *Hibiscus tiliaceaus* leaf litter mass loss rate (g d^{-1}), %N, %C, and C:N ratio after incubation in streams. Last row shows estimates of whole-stream gross primary production (GPP, mg O_2 m^{-2} d^{-1}), ecosystem respiration (ER, mg O_2 m^{-2} d^{-1}), and GPP:ER (P:R, dashed line =1.0).

watersheds), P-limitation in three streams (Waimānalo, Heʻeia, Kāwā watersheds), and no evidence that nutrients limit algal growth in 8 streams (Hālawa, Kīpapa, Waianu, Waikāne, Kahaluʻu, Keaʻahala, Mānoa, Punaluʻu watersheds).

Most stream ecosystems were heterotrophic (GPP:ER ratio: median = 0.46, range = 0.07-1.48) (Fig. 33), but Punaluʻu and Waimānalo streams were net autotrophic, featuring obvious algal mats (Table 20). Both ER (1.00 to 11.51 mg O_2 m^{-2} d^{-1}) and GPP (0.27 to 9.82 mg O_2 m^{-2} d^{-1}) varied widely across streams (Table 20). NDS assays indicated that nutrients were not limiting in most streams prior to AIS removals, so we assessed the possible influence of light and grazers on stream GPP. Armored catfish presence, light (measured according to canopy closure), and their interaction were included in the best model ($F_{3,8}$= 72.11, $p < 0.001$, $r^2 = 0.951$). However, using catfish biomass rather than catfish presence as a predictor yielded a comparable model (ΔAICc < 2). High biomass of armored catfish, which are specialized algae- grazers, was clearly associated with low primary productivity (Fig. 34). In streams that had not been invaded by Loricariids, GPP was significantly positively associated with light availability (slope = 0.23) (Fig. 34). Algal productivity responses to light were weakly positive in streams containing armored catfish (slope = 0.024) (Fig. 34).

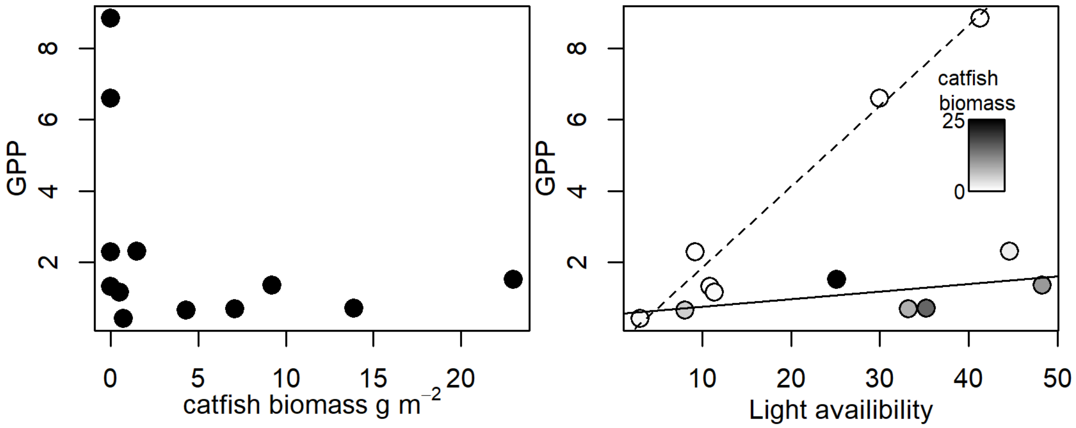


Figure 34. (Left) Average gross primary production (GPP, mg O_2 m^{-2} d^{-1}) in the removal reach measured before AIS removals shows strong suppression by armored catfish. (Right) Gross primary productivity as a function of percent light availability reach (inverse percent canopy cover). Gross primary productivity rises with light availability (% canopy cover) in the absence of armored catfish, but it is strongly suppressed in streams containing catfish (shaded markers, solid line) compared to streams lacking catfish (open markers, dashed line).

AIS densities, AIS removals and responses to AIS removal.

Of the 25,782 individual non-native fish and invertebrates removed from the study sites, ~66% were either Loricariid armored catfish (n = 4,281) or Poeciliid live-bearers (n = 12,797). Two species of Loricariids were captured: *Ancistrus temmincki* and *Hypostomus watwata*. At least four species of Poeciliids were captured: *Poecilia sphenops*, *Xiphophorus helleri*, *Poecilia reticulata*, and *Gambusia affinis.*

There was 11-fold variation in the density of AIS biomass removed from the study sites (1.89 to 31.41 g m^{-2}). AIS biomass density was dominated by armored catfishes at most sites. Mānoa Stream, near the University of Hawaiʻi campus, featured the highest AIS biomass density due to the abundance and size of

Loricariids at the site (Table 20). The biomass density of AIS was positively associated with road density (slope = 0.46, p = 0.003, r^2 = 0.53) and negatively related to forest cover in the watershed.

AIS removals shifted algal growth in three streams (Kīpapa, Waianu, Mānoa watersheds) from no apparent nutrient limitation to N+P co-limitation. In addition, Waikāne became P-limited, and Kahaluʻu and Keaʻahala became N-limited compared to their pre-removal state of no limitation. The opposite direction of change in nutrient limitation occurred in Kāwā watershed (P-limited to none) and Kalihi and Waimea watersheds (N-limited to none), while Heʻeia Stream switched from P- to N-limitation. The NDS assays put in after AIS removals were vandalized in Waimānalo Stream and desiccated in Hālawa Stream.

Table 21. Statistical tests of reach and time effects on different types of ecosystem processes using a test for homogeneity of multivariate dispersion to determine whether pair-wise difference among group variances are different (Anderson 2006). Above the diagonal, we show observed p-values for each pairwise comparison of upstream control (UpCon) and removal reaches both before and after removals. Below the diagonal, we show permuted p-values for pairwise tests of dispersion. Separate tests were conducted for metrics of nutrients (NH_4, SRP, NO_3, and TSS), algal growth in four nutrient supplementation treatments of NDS experiments (N, P, N+P, control), leaf litter decomposition (decay rate, %N, %C, C:N), and ecosystem metabolism (GPP, ER, P:R).

Nutrients	Post Removal	Post UpCon	Pre Removal	Pre UpCon
Post Removal		0.81	0.86	0.85
Post UpCon	0.79		0.95	0.92
Pre Removal	0.84	0.95		0.98
Pre UpCon	0.87	0.93	0.98	

NDS	Post Removal	Post UpCon	Pre Removal	Pre UpCon
Post Removal		0.99	0.15	0.32
Post UpCon	0.99		0.13	0.31
Pre Removal	0.14	0.14		0.88
Pre UpCon	0.30	0.30	0.88	

Leaf Litter	Post Removal	Post UpCon	Pre Removal	Pre UpCon
Post Removal		0.93	0.85	0.95
Post UpCon	0.92		0.73	0.84
Pre Removal	0.84	0.75		0.86
Pre UpCon	0.94	0.94	0.88	

Metabolic model	Post Removal	Post UpCon	Pre Removal	Pre UpCon
Post Removal		0.42	0.42	0.27
Post UpCon	0.44		0.93	0.65
Pre Removal	0.42	0.94		0.72
Pre UpCon	0.28	0.67	0.72	

Statistical comparison of reaches and time periods across all streams did not detect overarching effects of AIS removals on ecosystem processes. Water chemistry (nutrients, TSS) showed no overall differences (perMANOVA, p = 0.91) among reach-time groups or distinctions among pairwise comparisons of dispersion (p = 0.997) (Table 21). Periphyton accrual on NDS treatments were comparable overall (perMANOVA, *p* = 0.37), but exhibited a marginal difference in dispersion before versus after removals (*p* = 0.066). Leaf litter breakdown rates, %N, %C, and C:N ratios were also statistically comparable between reaches and time periods (perMANOVA, *p* = 0.88), as were ecosystem metabolism metrics (GPP, ER, GPP:ER; perMANOVA, *p* = 0.97; dispersion, *p* = 0.72) (Table 21).

While multivariate analyses treating the 13 study sites as comparable replicates suggested no consistent ecosystem responses to AIS removals, the effect sizes of removals for individual metrics indicate that heterogeneity across stream ecosystems likely masked the responses. In many cases, effect sizes were correlated with ecosystem conditions before AIS removals (Fig. 35). For instance, decreases in TSS concentrations were greatest in streams with high biomass of armored catfish (r = -0.53) and other AIS (r = -0.65), though there was no relationship with Poeciliid biomass. Reductions in ammonium concentrations were positively associated with Poeciliid biomass (r = -0.53) and light availability (r = -0.71). Removal effects on nitrate and SRP concentrations were positively associated with background nitrate (r = 0.8 and r = 0.5, respectively) and SRP (r = 0.83 and r = 0.95, respectively), yet were unrelated to AIS biomass density (r = -0.16 and r = -0.32, respectively). Effect sizes for leaf loss rates decreased with armored catfish biomass (r = -0.62).

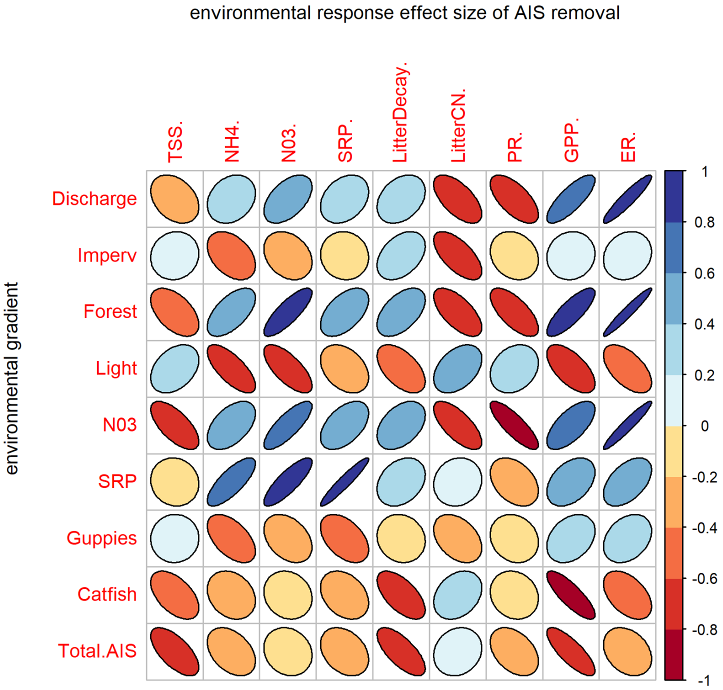


Figure 35: Pairwise correlation coefficients between the effect size (log response ratios,) of removing AIS on ecosystem processes (x-axis) and pre-removal conditions (y-axis) across 13 streams in O'ahu. Ellipse width, orientation, and color indicates the magnitude of positive (blue) or negative (red) correlation coefficients. Effect sizes were determined for concentration of total suspended solids (TSS), ammonia (NH_4), nitrate (NO_3), soluble reactive phosphorus (SRP); leaf litter breakdown rates of *Hibiscus tiliaceaus* leaves (Litter Decay) and C:N ratios of the leaves (Litter C:N); and ecosystem metabolism based on gross primary production (GPP), ecosystem respiration (ER), and the production:respiration ratio (PR). Stream conditions were described using pre-removal measurements of median discharge (Q50); percent of watershed are covered by impervious surfaces (Imperv) and forest cover (Forest); percent of canopy cover over the stream (Light), dissolved concentrations of NO_3 and SRP; and biomass density of invasive Poecilliids (Guppies), Loricariids (Catfish), or total AIS removed (g m^{-2}).

Although metabolism data were only obtained for 12 streams before removals and 8 streams after removals (due to high flow conditions and equipment failures), we found that ecosystem metabolism was particularly responsive to AIS removals when environmental conditions were taken into account. GPP and ER effect sizes following removals generally echoed each other, and varied systematically with stream discharge (positive), watershed forest cover (positive), nitrate concentrations (positive), and AIS biomass densities (negative). Because ER was more sensitive than GPP, the relationship of discharge, forest, and nitrate with effect size of removals on P:R ratios exhibited the opposite pattern (Fig. 35). The identity of AIS also mattered; GPP effect sizes responded far more to Loricariids than to Poeciliids. ER effect sizes were not consistently correlated with the amount of AIS removed, hence GPP:ER effect sizes were not either.

Discussion.

Our surveys of streams across the island of O'ahu demonstrated that non-native species have dramatically altered macrofaunal community structure with accompanying effects on ecosystem processes. In particular, armored catfish appear to limit GPP by grazing on periphyton, thereby mediating the influence of light and potentially nutrients. We also found that some components of ecosystems responded rapidly to reach-scale removals of AIS, while others did not. Importantly, AIS removals did not result in consistent, sweeping changes in any ecosystem metrics across all 13 streams. Rather, we found that some streams were far more responsive to AIS removals than others, suggesting that baseline assessments can help prioritize candidate sites to maximize the benefits of laborious interventions. Our results also indicate that a combination of interventions, such as improving water quality and habitat alongside AIS control, will likely be necessary to achieve ecosystem conditions that favor native species.

Pre-removal assessments of streams according to contextual biotic and abiotic gradients provided three insights into the effects of AIS on ecosystem attributes and processes. First, we found that Loricariid armored catfish exert stronger effects than Poeciliid live-bearers on ecosystem process rates. Poeciliids are now widespread across the Hawaiian archipelago (Moody et al. 2017). Prior studies indicate that Poeciliids can alter trophic structure and nutrient cycling (Holitzki et al. 2013) and prey upon early life stages of native migratory gobies (Hain et al. 2019). As omnivores, live-bearers may act as both competitors and predators of native gobies and prawns. Nonetheless, we did not find clear associations between variation in live-bearer densities and any of our ecosystem metrics, whereas we found clear evidence that armored catfish suppress GPP across a gradient of light availability (Fig. 34). This suggests that efforts to exterminate established invaders and discourage further introductions would likely be more profitable if focused on armored catfish.

Observed patterns of land use, nutrients, leaf-litter breakdown, and armored catfish distributions support a second key insight: that land use and other anthropogenic controls on nutrients have strong bottom-up effects on ecosystem process rates and perhaps even the invasibility of Hawaiian streams. We documented a frequently found pattern of land use transformation from forest to urban leading to higher nitrogen and phosphorus concentrations. Nitrogen was a strong predictor of microbial biomass on leaf litter, thereby elevating decomposition rates (Fig. 33, Table 20). Though phosphorus had no apparent effect on leaf litter mass loss, it appears that it might affect the likelihood that a stream is invaded by armored catfish. Because of their dermal armor, Loricariid catfish require more phosphorus to grow than most fishes (Hood et al. 2005). Consistent with this, sites with more substantial catfish biomass also featured moderate to high phosphorus concentration. While evidence of moderate phosphorus but high densities of small catfish suggest that Mānoa is an exception, anecdotal reports of high densities of large armored catfish in reaches close to our survey site align with this inference.

Further evaluation is thus warranted to evaluate the intriguing possibility that phosphorus pollution is requisite to armored catfish invasion success.

Finally, the strong pattern of pre-removal GPP with catfish presence and light availability strongly suggests that these grazers can overwhelm the benefits of light and nutrients for supporting algal growth. In systems lacking armored catfish, GPP was mediated primarily by light, as expected. This kind of interplay between physics and biology is well documented in rivers where salmon spawn; salmonids can re-engineer the stream bottom via nest digging (Moore and Schindler 2008), but only in streams with sufficiently small substrate (e.g., Holtgrieve et al. 2010b). Likewise, some Hawaiian streams are narrow enough that human activities determine whether the canopy is closed, while others are wide enough to always experience high light levels. Where light is sufficient to support high algal growth, armored catfish appear to be the primary factor mediating algal accumulation and GPP (Fig. 34). This suggests that contextual variation in biogeophysical conditions determines whether Loricariid invasions, and perhaps other species invasions, regulate ecosystem characteristics and processes in Hawaiian streams.

Experimental AIS removals also revealed heterogeneity in controls on ecosystem processes, with the added benefit of enabling us to separate cause and effect across contextual biotic and abiotic gradients. Ecosystem responses to removals depended on several aspects of background conditions, including the biomass and composition of the AIS that were removed from experimental reaches (Fig. 35). Little or no response occurred when background nutrients were high, light was low, and AIS were not abundant. Conversely, strong ecosystem responses were elicited by removals when a large amount of armored catfish biomass was removed, background nutrient levels were low to moderate, and a relatively open canopy allowed sufficient light to reach the streambed to fuel algal growth. For example, after a large number of armored catfish were removed from Heʻeia Stream (a stream with high light), we observed a clear decline in dissolved nutrients as GPP:ER increased (Fig. 35). Another general pattern was that sites where many catfish were removed experienced a drop in total suspended solids, presumably reflecting elimination of their role in resuspending detritus as they feed (Capps et al. 2015). Interestingly, leaf litter decay rates also declined after AIS removal in many streams, perhaps reflecting lower dissolved nutrients after eliminating recycling fluxes from non-native fishes and prawns. This aligns with the observation that the size of the downward shift in ammonium concentrations within removal reaches increased with the biomass of live-bearers removed across sites ($r = -0.53$), presumably reflecting the loss of excreted nitrogen waste from these small fishes. This interpretation is consistent with observations elsewhere in the archipelago (Holitzki et. al. 2013) that Hawaiian streams invaded by Poecillids have higher dissolved nitrogen and thicker algal biofilms compared to streams without Poeciliids.

Some ecosystem responses to AIS removals were less intuitive. For example, we observed the greatest rises in dissolved nutrients, leaf decomposition, and ER (as well as declining GPP:ER) in streams with high baseline nutrient concentrations. Also, in certain streams where armored catfish were removed in large numbers (e.g., Mānoa, Kahaluʻu, Heʻeia) and primary productivity rose rapidly, we observed an unanticipated shift toward NP- or N-limitation. The basis for this shift in nutrient limitation remains unclear.

Our study constitutes a notable experiment in invasive species control. Experimental removals were replicated across an unusually large number of streams, and despite being conducted at a small spatial scale, removals resulted in the sustained depression of AIS densities. Additionally, there was limited collateral harm to native species (described above). Yet the observed heterogeneity of ecosystem responses may in part be a consequence of incomplete eradication and recolonization. While removals achieved acute reductions in AIS densities and biomass (described above), some AIS nonetheless remained in experimental reaches following control efforts. Removal reaches also were left as open systems, thus AIS could recolonize soon after the end of control efforts. Similarly, this might have allowed AIS in neighboring reaches to move in and out of removal reaches (e.g., to feed). It is also possible that native gobies and prawns rapidly resumed their functional role as omnivores after AIS removals. However, AIS comprised at least 90% of all macrofaunal biomass in every stream, so compensatory responses by native species are unlikely to have offset the sudden loss of non-native taxa that previously dominated stream communities.

Our comparative analysis of ecosystem response to AIS removals revealed several important strategic dimensions of designing future restoration efforts. The most responsive sites were ones where nutrient pollution did not create highly enriched background conditions. For instance, high nutrient loads in urbanized reaches of Waimānalo, Kāwā, and Keaʻahala streams reduced ecosystem responses despite the removal of relatively large quantities of catfish and minimal canopy cover. This observation echoes earlier work on common carp showing that carp removals generally achieved water quality goals only after nutrient inputs had been reduced by 85-95% (Beklioglu et al. 2003, Schrage and Downing 2004). This highlights the concern that watershed development, agriculture, and septic system inputs (Whittier and El-Kadi 2014, Lisi et al. 2018) will boost nutrients enough to limit potential ecosystem response to AIS removals across the Hawaiian archipelago.

A second key factor regulating the outcomes of AIS removals in Hawai'i is spatial scale. We prioritized replication over scale, limiting the footprint of removals to 100 m experimental reaches. Recovery of some AIS within our removal reaches over the course of weeks to months suggest that a more comprehensive approach is needed to achieve lasting benefits. Watershed-scale ‘clean-slate’ eradication, for example, (described above and below) could potentially achieve lasting local and regional benefits from spillover benefits (Fig. 1, Fig. 6). Nonetheless, our experimental results show that even limited-scale AIS removals can produce ecosystem benefits when focused on sites with few other constraints. Even greater responses could presumably be achieved from complete eradication throughout a watershed.

At least two of our study watersheds-- Punaluʻu and Waianu—stand out as key places to protect by preventing species introductions and high-value targets for restoration through AIS removals. Both streams have moderate levels of nutrients, probably derived from traditional taro agriculture in the watershed, and sufficient light levels to support benthic algal growth. Notably, both also sustain greater diversity and biomass of native gobies and prawns than most other streams on Oʻahu. Most importantly, we found no armored catfish in these streams, though both harbor substantial biomass of live-bearers (mainly mollies). If introduced to these watersheds, we expect that armored catfish would affect dramatic changes by greatly reducing periphyton biomass and competing with endemic gobies and invertebrates that are obligate grazers (Lisi et al. 2018). Thus, at the very least, these watersheds should be prioritized for signage and educational campaigns to minimize the risk of catfish introductions.

This study places among the best-replicated BACI assessments of ecosystem-scale outcomes of AIS control. It provides a rigorous basis for inferring a range of ecosystem outcomes that arise from AIS removals conducted at a reach scale (and beyond). Our work illustrates that AIS removals may be effective in altering a wide range of ecosystem attributes and processes, particularly in streams that are minimally burdened by other stressors. Accordingly, we advocate prioritizing streams with lower nutrients, open canopies, and high existing biomass of non-native species when determining how best to maximize the return on major investments required to exterminate AIS from a watershed. Indeed, consideration should be given to undertake concurrent interventions to improve other in-stream conditions. For example, we recommend reducing nutrient loading in concert with AIS eradication campaigns. Similarly, concurrent habitat restoration (e.g., reducing canopy cover) could temporarily enhance GPP, thereby boosting conditions favorable to recovering native species. Accordingly, given the high cost of stream restoration (i.e., in terms of time, money, and political capital), our results can serve as a guide to selecting target watersheds whose characteristics maximize the odds of restoring ecosystem functioning and enhancing carrying capacity for native species through a portfolio of management strategies.

6.6 COUPLED OCEAN-WATERSHED MODELING OF WHOLE-WATERSHED AIS REMOVAL

Remarkably, we found that a 'clean-slate' restoration approach is virtually assured of success in boosting local populations of native gobies in Hawaiian streams (Fig. 36). Net increases in population size arise quickly after eradicating exotics; even if native adults also perished during extermination efforts, all local populations exceed their pre-extermination size within 10 years (Fig. 37). The most invaded streams show much shorter recovery times (mean ~1 year when invadedness > 75%). Local increases in native populations vary widely, however, and the largest gains arise in watersheds that are both large and highly invaded because these factors govern the additional carrying capacity for native species. When recruitment is limited, connectivity to other watersheds that supply incoming larvae also enhances the growth rate and population size of the native population following extermination of invaders.

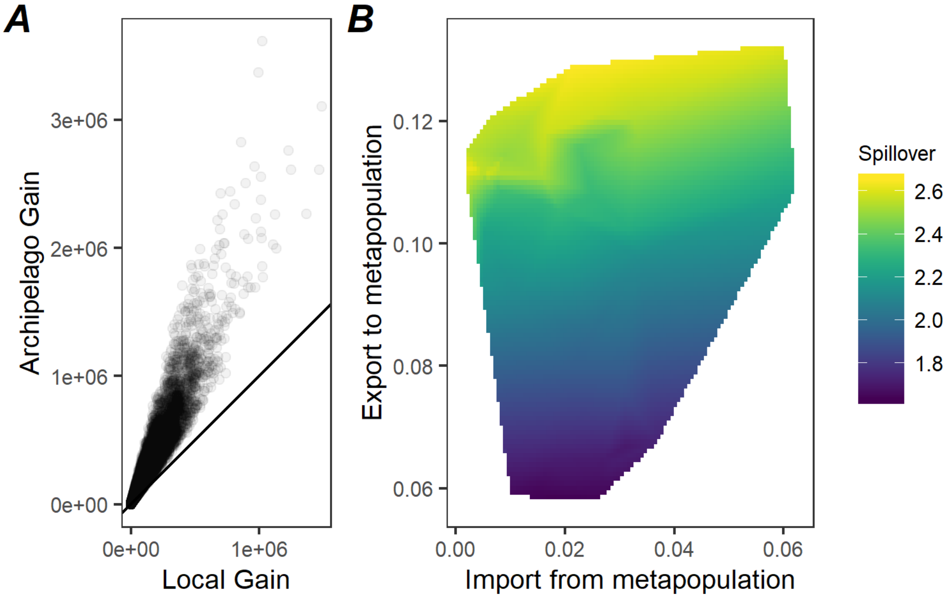


Figure 36. (A) Following extermination of invasive species from a single watershed, growth of the metapopulation across the Hawaiian archipelago greatly exceeds the local gains alone. In no case does the population decline. Spillover from 'clean-slate' restoration is a robust outcome when populations are recruitment limited: it occurs in each of 51 study streams, even when carrying capacities and invader densities were randomized across the archipelago (n=1000 permutations per watershed). If recruitment is not limited, no spillover occurs, and all points fall on the black line, indicating that gains are solely local. Gains measured as difference in population size 100 generations post-extirpation and population size pre-extirpation. (B) Connectivity via larval dispersal varies widely among watersheds, and strongly affects spillover (ratio of archipelago:local population gains). The color surface is fitted to simulation results depicted in (A), each of

which has a distinct combination of the import (proportion of all successfully dispersing larvae arriving at a given watershed) and export (proportion of all larvae from a given watershed that successfully disperse to any watershed) values estimated from a passive dispersal model applied to observed ocean currents from 2007-2011. Differences in spillover among streams depend primarily on export, so long as import is sufficient to ensure growth in the local population following eradication of invasive species.

Connectivity among watersheds strongly amplifies the metapopulation benefits of 'clean-slate' restoration. Demographic spillover occurs as the restored stream produces more larvae that boost populations elsewhere. This increase in propagule export is most dramatic in highly invaded streams because of their low production of larvae before restoration. When recipient watersheds are recruitment limited, not only do their populations grow but secondary spillover also ensues. Thus, eradication of invasive species in certain watersheds yields dramatic increases in metapopulation size via both direct and indirect connections (Fig. 37). For any given increase in the local population of the native species, there is a range of potential spillover, which we represent as the ratio of archipelago:local gains following extermination of invasive species. The variation in spillover ratio primarily reflects success in exporting larvae that reach other watersheds (Fig. 37). Additional small gains arise from greater receipt of larvae from elsewhere; these imported recruits support faster increases in the local native population following extermination of invaders.

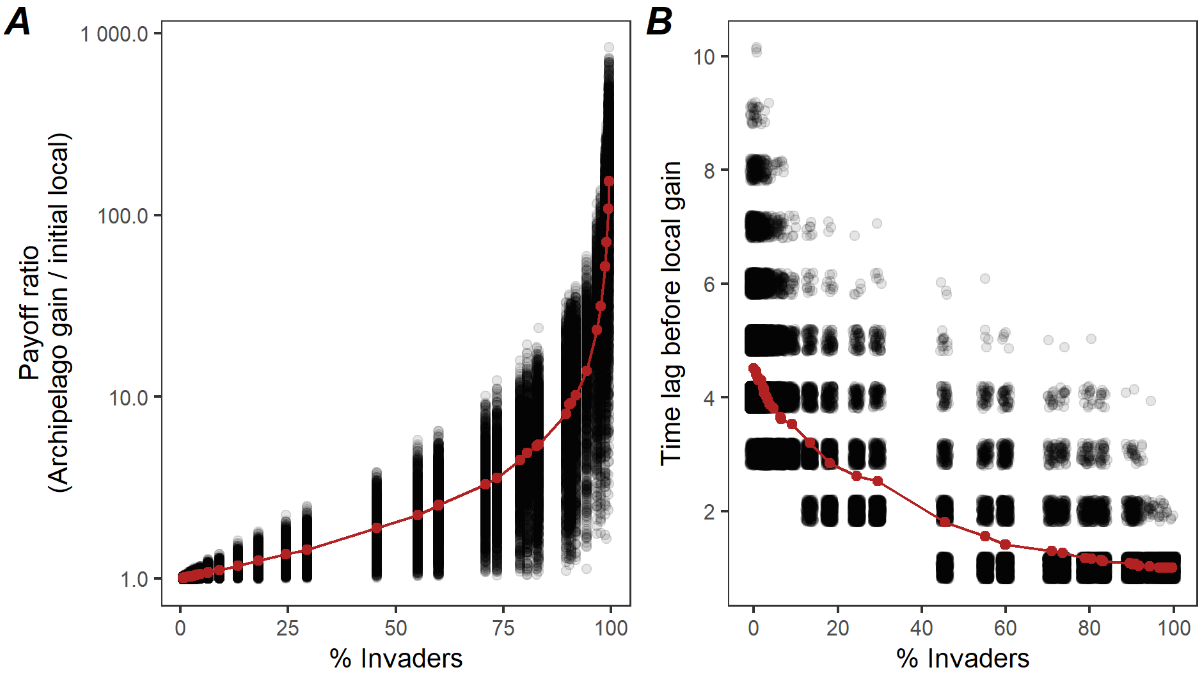


Figure 37. (A) Metapopulation payoff (ratio of archipelago gain to local population, pre-eradication) from eradication increases with invadedness of the local community. Variation among simulations within each invadedness level reflects the range of outcomes from permuting native species carrying capacity and metapopulation connectivity across 51 sites in the Hawaiian archipelago. Payoff is never less than 1, and so local populations are never harmed by eradication. (B) Years from eradication to reestablishment of pre-eradication population size decreases strongly with invadedness. Variation among simulations arises from permuting local carrying capacity and metapopulation connectivity. Results are illustrated under recruitment limitation; the same patterns hold when recruitment is not limited, but variance among simulations is far lower because payoff reflects only local gains.

The variation in metapopulation benefits across 51 single-watershed extermination simulations underscores the potential to maximize restoration benefits through strategic prioritization of restoration sites. Differences among streams in current fish communities and habitat size, as well as modeled larval exchange via ocean currents, indicate that there are only four watersheds where recovery time is rapid (< 2 years) and the archipelago metapopulation expands by 10-50% after eradicating invaders (Fig. 38). These high-return watersheds offer unique combinations of network connectivity, local carrying capacity, and local invadedness (Fig. 38). Importantly, none of these individual axes is enough to ensure a strong response to restoration, yet each is necessary for extraordinary success in metapopulation growth.

In the Hawaiian archipelago, every island except Molokaʻi has at least one watershed that, if restored, could yield at least a 4% increase in the total population of native gobies (Fig. 38). At their maximum, spillover benefits effectively double the local gains for native species of eliminating competition and predation from invaders. The geographic centrality of Oʻahu (Moody et al. 2019) and high human impacts (Moody et al. 2017) give rise to the widest range of compelling targets for 'clean-slate' restoration. This finding has a critical practical implication: Oʻahu also has the highest proportion of watersheds under government management, where restoration efforts and subsequent prevention of re-introduction of invasive species may be more feasible than in areas with complex mosaics of private ownership and public access.

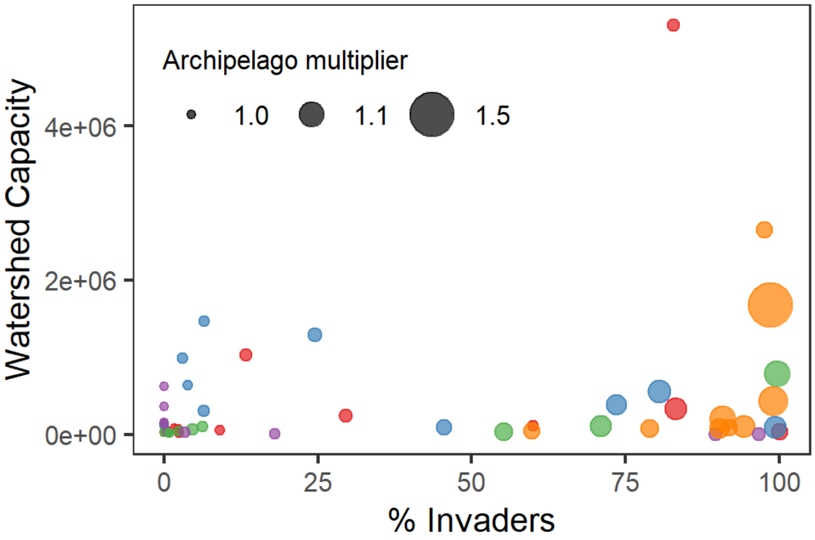


Figure 38. Archipelago-wide population increases with amount of local carrying capacity made available by eradicating invaders, which in turn reflects both invadedness and habitat area and quality. Multiplier is the ratio of archipelago population size post:pre eradication. Differences among the 51 sites in this study also arise from metapopulation connectivity, which differs among islands (Moody 2019); Red: Hawaiʻi, Green: Mauʻi, Purple: Molokaʻi, Orange: Oʻahu, Blue: Kauaʻi. There are streams on every island save Molokaʻi where 'clean-slate' restoration is predicted to substantially increase metapopulation size via the joint contributions of local gains and spillover. However, streams on Oʻahu tend to be especially favorable because they combine high invadedness and geographic centrality in the larval dispersal network.

As the impacts of biological invasions on native species and society continue to increase worldwide, our ‘clean-slate’ concept is distinct and important as a restoration strategy for priority ecosystems. Its hallmark feature is leveraging fundamental disparities in life history between invasive and native species that provide an opportunity to vigorously eradicate invaders while ensuring that native species can persist or recolonize from stored propagules. Unlike the predictable suite of life history traits that elevate invasion potential (Kleunen et al. 2015, McKnight et al. 2017), we expect that differences in storage life history between native and invasive species will be idiosyncratic and system-specific. Thus, a ‘clean-slate’ approach is not a universal prescription for restoration, but rather a strategy whose success depends upon the details of particular species and ecosystems. With the example of amphidromy in Hawaiian fishes (Alda et al. 2016), it is the combination of chemical options for eradicating all invaders throughout a watershed, propagule storage and transport via ocean currents for native species, and a distributed metapopulation structure that make us confident that a ‘clean-slate’ approach will achieve remarkable results.

Our inferences of near-zero risk of failure and impressive potential for local- and archipelago-scale demographic benefits rest on assumptions that invasive species have no storage or marine dispersal potential, and that the pre-extermination abundance of invaders is a proxy for the habitat carrying capacity for native species. We regard these as safe assumptions for Hawaiian streams. Importantly, additional simulations and our general model demonstrate that our results are robust to complicating factors such as self-recruitment or a series of poor years for marine larval dispersal (Appendix B). Dynamic exchanges across the metapopulation appear to be essential to the persistence of many populations of our focal species (and likely other endemic species that have more restricted current distributions and lower local abundances; Lisi et al. 2018), suggesting that the population rescue processes underlying our ‘clean-slate’ approach are already in operation. Indeed, the profoundly depleted state of most native populations in streams on Oʻahu makes it easy to understand why our simulations indicate that native species will never be worse off than before restoration efforts.

Notably, invasive-native differences in life history responses to extreme events are already used in certain control efforts but have not been recognized as a generalizable approach. Fire management of prairies relies on native species being better able to recover after frequent burning (Brockway et al. 2002; Kral et al. 2018). Further afield, human health depends on the recovery of helpful ‘native’ microbiomes after antibiotics clear infection by harmful ‘invasive’ counterparts (Iizumi et al. 2017). In aquatic ecosystems, consideration has been given to deliberate warming of lakes to eliminate cold-water refuges for invasive fishes (Lawson et al. 2015), and taxon-specific pesticides are widely used to control Sea Lamprey invasions with minimal harm to native species in Great Lakes tributaries (Siefkes 2017). Yet further opportunities to expand the use of a ‘clean slate’ approach surely abound. For example, native annual plants in deserts are well known to have persistent seed banks while some invaders do not (Li and Chesson 2018), hence wholesale removal of established plants could create the space needed for natives to thrive.

A notable outcome in our modeling of endemic species in Hawaiian streams is that the magnitude and timing of local population recovery is surprisingly insensitive to collateral mortality during eradication efforts. Rare-species management often focuses on preventing any loss of reproductive adults, yet we find little risk to local populations as long as invasive species eradication is successful. Since such ‘incidental take’ is likely during ecosystem-wide eradication efforts, relaxing no-take rules may be essential for restoration feasibility. Sequestering as many adults of native species as possible before exterminating invaders is still advisable, especially to safeguard local adaptations whose loss could slow subsequent population expansion. In Hawai’i, such local adaptation has strong fitness consequences

(Moody et al. 2015, 2019), although rapid colonization of multiple goby species following natural creation of streams (Schoenfuss et al. 2004) suggests that dispersal can still fuel successful recolonization. A conservative application of the ‘clean-slate’ approach would be to perform invasive species eradication in watersheds where natives have already been locally extirpated; newly rewatered streams in East Maui may soon provide such an opportunity if they become invaded before (re)colonization by native species.

Islands have been the scene of both profound ecosystem domination by invasive species (Sax and Gaines 2008, Glen et al. 2013) and spectacularly successful invader eradication efforts (Simberloff 2009), hence they are an ideal proving ground for ‘clean-slate’ restoration approaches. Indeed, the isolation and limited human populations that characterize many island ecosystems suit a key premise: we must be able to prevent re-introduction of the same invaders that have been laboriously eradicated. This requirement underscores the human dimensions of restoration efforts, which are only successful when they align with the values held by local communities (Glen et al. 2013, Russell et al. 2015, Crowley et al. 2017). In Hawaiʻi, stream gobies have deep cultural significance that engenders grassroots support for their protection. Given the precarious state of metapopulations of many endemic stream species in Hawaiʻi, communicating the value of experimenting with novel restoration strategies—as a complement to strict protections for most populations—will require sustained engagement with local communities (Russell et al. 2015). Moreover, if dramatic interventions simply reset the clock for the same invaders, ecosystem-scale extermination will not yield recovery of native populations. Community support and vigilance will be essential for avoiding anthropogenic reintroductions (Mack et al. 2000, Nico and Walsh 2011, Harris et al. 2012, Walsh et al. 2016), which is requisite to seizing the ecological opportunities provided by the storage effect and metapopulation biology. As society strives to counter biological invasions, the ‘clean-slate’ restoration approach affords great promise in rescuing native species of concern.

7 Conclusions and Implications for Future Research and Implementation

Conclusions.

Ensuring the continuity and long-term sustainability of DoD activities on Pacific islands requires effective stewardship of ecosystems that have been altered by the introduction of non-native species. This project was undertaken to provide DoD resource managers and their partners with knowledge and tools to recover key ecological processes and TER-S in Pacific island streams affected by AIS. Our primary research objectives were to (1) advance basic understanding of how ecological processes have been altered by AIS; (2) concurrently advance understanding of ecosystem and TER-S responses to AIS removal from Pacific island streams; (3) develop and validate restoration approaches that foster the recovery of TER-S; and (4) develop model-based decision tools to help DoD managers to design interventions that minimize resource expenditures and maximize in-stream conditions favorable to native species. By undertaking a set of complementary studies to meet these objectives, we have:

- Empirically demonstrated that (when modified to address prevailing physiographic and sociocultural concerns) electrofishing can be a valuable tool for AIS control in Hawaiian streams, illustrating how managers can maximize benefits of AIS control through careful consideration of target species densities, climate-driven hydrology, and surface flow management.
- Developed and demonstrated a model-based decision support tool that can serve as a defensible and transparent analytical framework for prioritizing interventions according to local, regional and archipelago-scale gains.

With an expanded knowledge base and toolkit, DoD resource managers and partner organizations should be able to achieve more effective watershed management and conservation of at-risk aquatic species to ensure the sustainability of military operations in the Hawaiian Islands and other oceanic islands in the Pacific.

Development of innovative approaches and actionable information for managing AIS has yielded several key findings and important conclusions, including:

- **AIS control is feasible and effective** with a tailored electrofishing protocol that minimizes collateral harm to TER-S
- **Control efforts can result in sustained reductions** in overall AIS densities and densities of targeted AIS (where removal efficacy is mediated by AIS densities and hydrology)
- **AIS removal can increase recruitment**, growth, and body condition of TER-S
- **AIS removal can increase genomic diversity** and composition of local TER-S populations (where gains are mediated by migratory life history)
- **Whole watershed 'clean-slate' restoration can lead to demographic spillover** that benefits populations elsewhere in the archipelago (leveraging life history differences between AIS and TER-S)

Transforming novel research findings into routine practice.

The studies described herein have yielded ground-breaking knowledge, innovative management protocols, and new decision-making tools to characterize and recover key ecological processes and at-risk native species in Pacific island streams impacted by AIS. This project has provided the first ever quantitative tests demonstrating how a host of key ecosystem and demographic processes respond to AIS control over management-relevant time frames. The project also has illustrated how surface flow conditions- and by extension flow modification, which is arguably the only other realistic approach to

AIS manipulation that can be widely applied- could dovetail with AIS removal to promote recovery of ecological processes and species of concern. *Our findings strongly indicate that these are effective approaches for improving the outlook of native gobies and other aquatic species of concern in Hawaiʻi and elsewhere in the Pacific.* Accordingly, the information gained from this project can provide immediate and lasting guidance for advancing adaptive watershed management on Pacific islands. Our findings demonstrate, for example, that electrofishing can be a useful tool for AIS management in the region so long as modifications are made to conventional methods to greatly reduce potential risks to native species (while ensuring capture efficacy of targeted AIS species). Likewise, better understanding of how life history moderates genomic and demographic variation can help ground management expectations and deliberations about executing AIS removals. However, specific guidance is required to promote incorporation of new knowledge and adoption of new methods and tools (i.e., so that novel advance soon become routine understanding and practice). Below we outline some specific recommendations that can spur applications of our findings. We also have appended a Fact Sheet and data collection sheets to facilitate the adoption of baseline survey and AIS control methods (Appendix C).

AIS control. Electrofishing has long been considered unacceptable in Hawaiʻi and other Pacific island archipelagos because of collateral risks to ecologically and culturally important native species. Our results demonstrate, however, that electrofishing can be highly effective for AIS control and that it can be conducted so that potential risks to native species are greatly reduced. Achieving high capture efficacy of all or targeted AIS with minimal collateral harm to native species of concern will require that prospective practitioners account for (1) the electro-sensitivity of native species; and (2) prevailing in-stream and watershed characteristics, including hydrology and existing AIS communities, when assessing the potential benefit of electrofishing for AIS management in the region. Further, while Pacific island streams provide excellent opportunities to attempt full eradication of AIS from watersheds connected only by marine dispersal pathways, practitioners must be cognizant that reintroductions might curtail potential benefits to native species.

Our findings from experimental AIS removals are consistent with expectations that AIS reductions will vary according to locality characteristics (i.e., specific to particular watersheds or sites). In part, this is a result of differences in complex habitat characteristics, such as steep elevational changes, plunge pools, and boulders, which can make electrofishing in Pacific island streams more challenging than in most continental streams. It is also important to consider that hydro-geomorphological (e.g., discharge) and the physiological differences among target groups can influence removal efficacy in Pacific island streams. We found, for example, that stream hydrology differentially influenced capture efficacy (i.e., the number of individuals that were removed and the relative change in densities) within and across species. As the smaller and more mobile of our target taxa, Poeciliids were typically less likely to be caught compared to armored catfish. We also found that electrofishing is not appropriate in saline or estuarine habitats or in freshwater habitats subject to tidal influence where risk of injury or mortality for native species can be much greater, due to the need for pulsed current run at higher voltages. It is worthwhile to note, however, that the results presented here are based on work conducted on Oʻahu; outcomes may be different elsewhere. Therefore, potential users must consider watershed and regional conditions to determine whether and where to employ electrofishing for AIS management on other Pacific islands.

With this in mind, we recommend practitioners employ a generalized strategy (summarized in the Fact Sheet flow chart included in Appendix C) to conduct modified electrofishing for AIS control in Pacific island streams, as follows:

- Hand collect native species prior to electrofishing and sequester all captured individuals streamside in well-aerated and covered tanks (or similar containment units), taking care to separate large predators from potential prey species.
- Conduct initial electrofishing passes using unpulsed DC current at the minimum voltage required to produce an effect, but to safely capture remaining individuals of native species from the removal reach. Be alert to ensure that electrodes do not come in to contact with native species.
- Consider conducting multiple passes at these settings to further reduce chances of collateral harm to electro-sensitive native species.
- Otherwise, conduct an additional pass using pulsed current at an increased voltage for the targeted removal of AIS, while still accounting for potential risk to larger, vulnerable native species.
- Conduct a final pass, increasing voltage if necessary, to achieve depletion of AIS, remaining alert to ensure the well-being of any native species remaining in the exposure area.
- Sequestered individuals should then be returned to their site of capture immediately following the final pass.

Although the methods highlighted here reduce the risk of adverse effects, electrofishing in Pacific island streams may still harm or kill native species of concern. Taking precautionary steps does not eliminate all risk of collateral injury and mortality. Therefore, all practitioners should seek prior agency approval, particularly in Hawaiʻi as **the use of electrofishing devices to take aquatic life is prohibited under Section 188-23, Hawaiʻi Revised Statutes (HRS) and Section 13-75-6, Hawaiʻi Administrative Rules (HAR), without a permit issued under Section 187 A-6, HAR**.

In addition, we recommend that (1) some or all electrofishing be done under the supervision of mission agencies, like the DAR in Hawaiʻi; and (2) in cases where native species are captured during electrofishing passes, practitioners cease electrofishing and tend to captured individuals to reduce further risk of stress-induced injury or mortality; and (3) if there is unexpectedly high mortality of native species, users terminate electrofishing and contact partners to improve their electrofishing protocol.

Identifying and prioritizing areas for AIS management. We developed and validated a coupled ocean-watershed model that combines information on climate, hydrology, and demography to help DoD managers establish and maintain best management practices. Our model (Fig. 1, Fig. 6, Appendix B) demonstrates the value of leveraging a distinct life history dichotomy between AIS and native migratory fauna. Model simulations focusing on *A. stamineus* showed, for example, that watershed-scale 'clean-slate' restoration can reliably convert demographic sinks into productive source populations that provide recruits to streams across the archipelago. Model simulations also showed that 'clean-slate' restoration is both low-risk (e.g., local increases in native species always occur quickly) and high-reward (e.g., spillover enhances all other populations), but that maximizing return on investment requires strategic prioritization and targeted execution of AIS eradication.

Variation in metapopulation benefits across the Hawaiian archipelago indicates that priority should be given to restoring large, highly-invaded watersheds that are well connected by ocean currents. An evaluation of network connectivity, local carrying capacity, and local invadedness in 51 watersheds (~15% of all watersheds on the five high islands with permanent streams) identified four where recovery time is particularly rapid (< 2 years) and where AIS eradication could expand the archipelago metapopulation by 10-50% (Fig. 38). While simulations also indicate that every island except Molokaʻi has at least one watershed that, if restored, could yield at least a 4% increase in the archipelago metapopulation of *A. stamineus* (Fig. 38), the geographic centrality and widespread degradation of streams on Oʻahu (Moody et al. 2017, Moody et al. 2019) give rise to the widest range of compelling targets for 'clean-slate' restoration including watersheds that are under government management. Thus it might very well be more feasible and more rewarding to eradicate AIS from streams on Oʻahu than elsewhere in the archipelago.

This guidance might very well suffice for a pilot or trial program, but further analyses will be required to build out a broader campaign or to develop management portfolios. Additional simulations tailored to address different questions and priorities should be undertaken, with consideration given to the distribution, demography and life history of other at-risk native species. Likewise, further modeling can help characterize the outcomes of ongoing and planned watershed management and restoration projects focusing on particular areas of interest (e.g., DoD bases and installations), thus serving as a valuable forecasting tool for assessing the potential benefits of individual or coordinated management actions within and among watersheds. For example, simulations might be useful for balancing trade-offs between regulating water use, constraining land use practices, or targeted control of invasive species across Hawaiʻi. This could demonstrate how to schedule water use (i.e., surface flow diversions, rewatering) to protect sensitive life history stages of native species with minimal disruption. It could also demonstrate when alternatives to modifying water use, including AIS removal, could be more cost-effective for sustaining at-risk populations. Accordingly, we recommend evaluating cost and benefit functions to determine where return on investments would be greatest under different stream flow conditions and climate scenarios so that DoD managers can act according to objective, defensible guidance for sustaining water availability and conserving at-risk native species. The current model configuration could be useful to begin this process. Further work to restructure the scope of the model (e.g., to encompass more or fewer watersheds) and to parameterize it for other species might be required, however, depending on the questions and objectives at hand.

Future research opportunities.

Novel perspectives and approaches for managing oceanic island stream ecosystems are more likely to be embraced if additional research were to be carried out to resolve some key remaining questions and uncertainties. There are three priority areas that warrant further consideration: (1) ecological and evolutionary drivers of life history variation; and (2) systematic conservation planning; and (3) development of environmental DNA (eDNA) based approaches for biotic monitoring and assessment.

Ecological and evolutionary drivers of life history variation. Progressive study continues to reveal that native Hawaiian fishes exhibit complex migratory life histories, and that accounting for life history variation is vital to successful watershed and TER-S management (especially when management efforts involve potentially contentious interventions). Efforts to protect and manage native stream fauna in the Hawaiian Islands ascribe to the increasingly out of date perspective that all migratory species exhibit fixed life histories, despite increasing evidence to the contrary. Our prior work completed for SERDP project RC-1646 (Blum et al. 2014) demonstrated that *A. stamineus* exhibit facultative amphidromy, and that life history variation results in lifelong consequences (Hogan et al. 2014). Building on this finding, work completed as part of this project has demonstrated that only one of the other four native fishes is obligately amphidromous; the three others exhibit some level of migratory flexibility (Heim-Ballew et al. 2020). We also have found clear evidence of genomic differentiation between *A. stamineus* resident and migrant contingents (Fig. 29), and that the two life history contingents exhibit distinct reproductive phenologies (Heim-Ballew 2019). These unexpected findings raise the possibility that residents and migrants are incipient species, and accordingly, that there is greater evolutionary diversity within native stream biota than is currently thought (Alda et al. 2016). Like-minded studies of other species could similarly reveal the presence of cryptic evolutionary diversity. Thus greater knowledge of genomic and phenological variation could offer further guidance for conservation and management, including when and where to undertake AIS removals in coordination with habitat and stream flow management (Brasher 2003, Walter et al. 2012). Notably, this project has also revealed that the proportional representation of life history contingents in local populations of *A. stamineus* varies according to in-stream conditions (Fig. 22). Evidence of genomic differentiation among residents and migrants suggests that life history is strongly heritable, and that proportional representation arises as a function of differential reproduction and survival. It is also possible, however, that some elements of migratory behavior are plastic, and accordingly, that proportional representation is in part environmentally determined. This uncertainty could be clarified by undertaking studies of heritability, plasticity, and heritable plasticity within and among populations. Not only would this help clarify the full range and drivers of facultative life history and proportional representation in local populations (and thus also shed further light on natal retention and dispersal), it would also reveal whether and how management interventions act as selective pressures (or ameliorate selective pressures) shaping the evolution of *A. stamineus* and other amphidromous species native to the Hawaiian Islands.

Systematic conservation planning. Though coupled ocean-watershed modeling has proven to be a highly productive and informative approach for forecasting outcomes of AIS removal and for identifying priority areas to execute interventions, undertaking complementary systematic conservation planning (SCP) could lead to further advances in conservation and management practices. SCP could be undertaken independently or in conjunction with developing model simulations that are tailored to specific questions and management objectives. SCP optimization analyses could help simultaneously maximize the benefits for native species and the relative costs of AIS removal and mitigation. Benefits could be characterized as expected increases in native species populations according to the number of species present and the deviation of observed population densities from those predicted by coupled ocean-watershed model simulations. Accordingly, expected benefits could be weighted by metrics of population connectivity derived from our advection-diffusion model of larval dispersal (Fig.1, Fig. 6, Fig. 7, Appendix B) and estimates of gene flow across watersheds (e.g., Fig. 28) and across the archipelago (Blum et al. 2014, Alda et al. 2016). For the Hawaiian Islands, SCP models could be parameterized using data on the density and diversity of AIS and native species from the Hawaiʻi Watershed Atlas (www.hawaiiwatershedatlas.com) and archipelago-wide surveys that were completed in 2009 and 2011 as part of SERDP project RC-1646 (Blum et al. 2014). AIS management costs could be based on habitat area and AIS richness, under the assumption that the challenge of successful management is proportional to the number and distribution of AIS to remove or mitigate. To bracket the range of interests expressed by DoD managers and the broader conservation community, SCP models could be developed and executed at multiple spatial scale (e.g., all watersheds on Oʻahu, all watersheds containing DoD assets across the archipelago, and all watersheds across the archipelago). Additional model scenarios could be constructed to evaluate outcomes of combining AIS eradication with other management interventions, such as complete or partial closing of water diversions. Likewise, model scenarios could also account for projected shifts in surface flow under near-future climate change scenarios (Oki 2004, Diaz et al. 2005, Chu and Chen 2005, Safeeq and Fares 2011). Changes in both diversions and climate are expected to affect surface flow conditions, but each will likely exert influence on a distinct set of watersheds, thus it would be informative to compare optimal restoration plans derived from SCP models of both scenarios.

eDNA monitoring and assessment. Better understanding of AIS distributions could improve control efforts aiming to sustain TER-S or curtail the loss of valued ecosystem services. Accordingly, tools that offer more thorough and accurate estimates of biodiversity could streamline operational decision-making, and thus improve core mission outcomes. Recent innovations in high-throughput sequencing and bioinformatic infrastructure have elevated eDNA as a potentially transformative platform for surveying biodiversity. An increasing body of evidence indicates that analysis of DNA captured from an environmental sample (i.e., without first isolating target organisms) can be a powerful, non-invasive, and cost-effective approach to survey biodiversity. Yet it is also clear that further work is necessary to fully realize eDNA as a management tool.

eDNA analysis can overcome limitations of conventional biodiversity survey methods by providing more detailed accounts of targeted species and whole communities via greater sampling diversity and taxonomic resolution. Studies have so far demonstrated the power of eDNA as a tool for delimiting species distributions (e.g., Laramie et al. 2015; Sigsgaard et al. 2015; Spear et al. 2015) and for determining community composition, especially in aquatic ecosystems (e.g., Miya et al. 2015; Dowle et al. 2016; Olds et al. 2016; Valentini et al. 2016). Some evidence also suggests that eDNA can be a tool for assessing relative abundance (e.g., Evans et al. 2016; Thomson et al. 2016, though see Lim et al. 2016). A number of factors, however, can complicate use of eDNA to estimate species occurrence and abundance. Bench-scale technical and procedural complications like primer mismatches can skew data outputs (e.g., relative read counts) and thus preclude or bias species detection (Deiner et al. 2017). Abiotic and biotic factors that govern the origin, fate and transport of DNA (i.e., the ‘ecology of eDNA’) can also influence species detection and abundance estimation (Barnes et al. 2014; Barnes & Turner 2016). For instance, detection probability can depend on source distance and transport (Deiner et al. 2017). Inferences may also vary depending on the sample taken; eDNA longevity is shorter in water, for example, than it is in benthic sediment (Turner et al. 2015). Though these and other concerns are well-recognized, they remain unresolved. Realizing eDNA as a management tool will require methodological optimization, more extensive calibration (e.g., via comparisons to conventional survey results), full-scale applications (i.e., definitive tests), and use in downstream conservation tools like species distribution modeling.

Hawaiian Island streams are an especially compelling ‘natural laboratory’ to advance eDNA tools for DoD resource management. Prevailing conditions on the islands are emblematic of the challenges facing DoD installations worldwide. Streams that cross the dense array of DoD assets and installations in the archipelago harbor endemic, culturally important species that are under state or federal protection, as well as a proliferation of aquatic invasive species (AIS). SERDP-funded studies also provide a strong foundation to build on. Not only have we demonstrated the value of genetic methods for monitoring native species in Hawaiian streams (RC-1646, Blum et al. 2014), we also have uncovered evidence of widespread decline on Oʻahu, where the majority of DoD installations are located (Blum et al. 2014, Moody et al. 2017, Lisi et al. 2018). This involved conducting cross-archipelago surveys of genetic diversity, species diversity, and population densities that can inform the development of eDNA survey approaches. Additionally, findings reported herein on outcomes of AIS control also illustrate how complementary efforts can enhance the value of eDNA tools. For example, preliminary coupled ocean-watershed modeling has identified watersheds on Oʻahu under DoD stewardship as promising candidates for restoration (Fig. 1, Fig. 6, Fig. 38, Appendix B); by improving knowledge of species distributions and relative abundance, eDNA tools could impart greater certainty in recommendations for taking action. Our field-based assessments (Fig. 9, Fig. 10, Fig. 11, Fig. 12) additionally suggest that eDNA tools could be extremely useful for assessing outcomes of stream restoration (e.g., native species responses to AIS control) across Hawaiʻi.

There would be great value in undertaking a portfolio of vertically-integrated studies to strengthen understanding and use of eDNA for DoD resource management on Pacific islands. Work focusing on Oʻahu could leverage the natural features of Hawaiian streams, 50+ years of archival monitoring data (Moody et al. 2017), and SERDP-funded studies to: optimize and calibrate laboratory methods for concurrent species detection and abundance estimation; in order to (2) assess how spatial-temporal variation in eDNA production and transport influences species detection and estimates of species abundance; to develop (3) eDNA-informed models of current TER-S and AIS occupancy and abundance, which could be compiled into an 'Atlas' to support DoD operations and land stewardship across the region.

Transfer and outreach.

As threats continue to build, collaborations that foster communication and build trust (i.e., among communities with sometimes disparate priorities and values) will likely be increasingly vital to securing the biological legacies of the Hawaiian Islands and other islands elsewhere in the Pacific. It is thus imperative that pipelines be developed and expanded to hasten translation of research into practice. Accordingly, knowledge gained from this work is being conveyed to practitioner and stakeholder communities through a step-wise process starting with (1) topical tutorials, demonstrations, and execution of new protocols in partnership with potential end users; (2) development and dissemination of fact sheets and technical guidance documents; and (3) broader outreach and public engagement. Building on existing partnerships, we have worked closely with a core group of Oʻahu-based resource managers to develop and validate new methods for AIS control. Efforts were made to coordinate with ongoing and planned watershed management programs (e.g., we conducted experimental AIS removals in study reaches that fall within areas that resource managers have been restoring for the benefit of at-risk populations of native species). To support future, independent endeavors, we have developed a technical 'user' guide for resource managers and research scientists to undertake AIS removals in Hawaiian streams. The guide offers detailed technical overviews on AIS control methods development and demonstration of capture efficacy as well as collateral risks of a modified electrofishing protocol. In addition to offering a one-page Fact Sheet (Appendix C) summary alongside detailed explanations of protocols, it also provides supporting technical documents, such as example templates for data collection sheets (Appendix C). To engender greater community awareness as well as consideration of new knowledge and adoption of new tools (and to encourage further third-party advancement), we also have disseminated project findings to scientific, practitioner and stakeholder communities as well as the general public through publication of peer-review papers, by presenting seminars at regional workshops and national conferences, and by conveying key information about our work through popular press articles and community outreach.

Continuation of these efforts would help expand and support end user communities working to address cross-boundary concerns through cooperative partnerships. Many, if not all, DoD installations in the Pacific face cross-boundary management concerns that are best addressed by cooperative, watershed-scale partnerships. Cooperative management of invasive species to benefit at-risk native species can promote the success of the military mission by enabling better use of limited resources and assuring mission flexibility. An effort focusing on Waimānalo Stream could serve as a model to guide the development of partnerships across the Hawaiian Islands. Waimānalo Stream crosses land that is controlled by the Air Force and Marine Corps (Bellows Air Force Station and the Marine Corps Training Area Bellow, hereafter jointly referred to as "Bellows"). As a multi-use site located within a densely urbanized landscape, Bellows is an excellent setting for demonstrating how watershed partnerships can address cross-boundary management. Development of a watershed partnership might begin by (1) identifying and contacting key federal, state and private stakeholders in the Waimānalo watershed (i.e., landowners and stewards of large properties or properties adjacent to the stream); (2) preparing a draft restoration plan emphasizing cross-boundary management, cost-sharing and collaboration; (3) hosting outreach and joint field exercise events to demonstrate restoration methods, including those outlined here for AIS control; and (4) initiating coordinated monitoring, emphasizing pooling of resources and community involvement. In addition to providing targeted training and outreach, this effort would deliver a generalizable template for coordinated watershed management of AIS and native species that would help promote cost savings and mission readiness by helping maintain access and greater use of Pacific island installations.

Summary.

In summary, the research presented here has expanded fundamental knowledge of insular stream ecology, responses of native amphidromous fishes to AIS, as well as outcomes of AIS control and mitigation. Meeting our research objectives has produced innovative approaches and actionable information for managing AIS that can substantively improve DoD stewardship of stream ecosystems that cross DoD lands, especially on islands like Oʻahu where installations harbor native species under federal or state protection. This project not only empirically demonstrated that modified electrofishing can be a valuable tool for AIS control in Hawaiian streams, it also illustrated how managers can maximize benefits of AIS control through careful consideration of target species and prevailing hydrological conditions. Likewise, the project delivered a model-based decision support tool that can serve as a defensible and transparent analytical framework for prioritizing interventions according to local, regional and archipelago-scale gains. With an expanded knowledge base and toolkit, DoD resource managers and partner organizations should better positioned to sustain the rich biological and cultural legacies found on Pacific islands, which in turn will help ensure the sustainability of military operations across the region.

8 Literature Cited

Ainslie, B.J, Post, J.R., and Paul, A.J. 1998. Effects of pulsed and continuous DC electrofishing on juvenile rainbow trout. Am. J. Fish. Manag. **18**: 905-918.

Albins, M. A. 2015. Invasive Pacific lionfish Pterois volitans reduce abundance and species richness of native Bahamian coral-reef fishes. Mar. Ecol. Prog. Ser. **522**: 231-243.

Albins, M. A., and Hixon, M. A. 2008. Invasive Indo-Pacific lionfish Pterois volitans reduce recruitment of Atlantic coral-reef fishes. Mar. Ecol. Prog. Ser. **367:** 233-238.

Alda, F., Gagne, R.B., Walter, R. P., Hogan, J. D., Moody, K. N., Zink, F., McIntyre, P. B., Gilliam, J. F., and Blum, M. J. 2016. Colonization and demographic expansion of freshwater fauna across the Hawaiian archipelago. J. Evol. Biol. **29**: 2054-2069.

Allendorf, F.W., Luikart, G.H., and Aitken, S.N. 2013. Conservation and the genetics of populations. 2nd edition. Wiley-Blackwell, Hoboken, New Jersey.

Andrews, S., 2010. FastQC: a quality control tool for high throughput sequence data. http://www.bioinformatics.babraham.ac.uk/projects/fastqc/

Anderson, M.J. 2006. Distance-based tests for homogeneity of multivariate dispersions. Biometrics **62**: 245-253.

Anton, A., Cure, K., Layman, C. A., Puntila, R., Simpson, M. S., and Bruno, J. F. 2016. Prey naiveté to invasive lionfish Pterois volitans on Caribbean coral reefs. Mar. Ecol. Prog. Ser. **544**: 257-269.

Armstrong K., Herdrich D., and Levine A. 2011. Historic fishing methods in American Samoa. U.S. Department of Commerce, National Oceanic and Atmospheric Administration (NOAA) Technical Memorandum, NMFS-PIFSAC-24, Honolulu, Hawaii.

Arostegui, M.C., Quinn, T.P., Seeb, L.W., Seeb, J.E., and McKinney, G.J. 2019. Retention of a chromosomal inversion from an anadromous ancestor provides the genetic basis for alternative freshwater ecotypes in rainbow trout. Mol. Ecol. **28**: 142-1427

Baker, J.A., and Foster, S.A. 1992. Estimating density and abundance of endemic fishes in Hawaiian streams. State of Hawaii, Department of Land and Natural Resources, Division of Aquatic Resources. 50 p.

Bailey, K.M., and Houde, E.D. 1989. Predation on eggs and larvae of marine fishes and the recruitment problem. *In*: Advances in Marine Biology. Academic Press **25**:1-83.

Baltz, D.M., and Moyle, P.B. 1993. Invasion resistance to introduced species by a native assemblage of California stream fishes. Ecol. Appl. 3:246-255.

Barber, R.T., and Chavez, F.P. 1983. Biological consequences of El Nino. Science **222**: 1203-1210.

Barneche, D.R., Robertson, D.R., White, C.R. and Marshall, D.J. 2018. Fish reproductive-energy output increases disproportionately with body size. Science. **360**: 642-645.

Barnes, M.A., Turner, C.R., Jerde, C.L., Renshaw, M.A., Chadderton, W.L., and Lodge, D.M. 2014. Environmental conditions influence eDNA persistence in aquatic systems. Environ. Sci. Technol. **48**: 1819-1827.

Barnes, M.A., and Turner, C.R. 2016. The ecology of environmental DNA and implications for conservation genetics. Conserv. Genet. **17**: 1-17.

Barney, B.T., Munkholm, C., Walt, D.R., and Palumbi, S R. 2017. Highly localized divergence within supergenes in Atlantic cod (*Gadus morhua*) within the Gulf of Maine. BMC Genomics **18**: 271.

Barrau, J. 1955. Fishing poisons of the South Pacific. South Pacific Commission Quarterly Bulletin **5**: 7-8.

Barth, J.M.I., Berg, P.R., Jonsson, P.R., Bonanomi, S., Corell, H., Hemmer-Hansen, J., Jakocbsen, K.S., Johannesson, K., Jorde, P.E., Knutsen, H., Moksnes, P.-O., Star, B., Stenseth, N.C., Svedäng, H., Jentoft, S., and André, C. 2017. Genome architecture enables local adaptation of Atlantic cod despite high connectivity. Mol. Ecol. **26**: 4452-4466.

Beard, K.H., Vogt, K.A. and Kulmatiski, A., 2002. Top-down effects of a terrestrial frog on forest nutrient dynamics. Oecologia **133**: 583-593.

Beklioglu, M., Ince, O., and Tuzun, I. 2003. Restoration of the eutrophic Lake Eymir, Turkey, by biomanipulation after a major external nutrient control I. Hydrobiologia **490**: 93-105.

Bell, D.A., Robinson, Z.L., Funk, W.C., Fitzpatrik, S.W., Allendorf, F.W., Tallmon, D.A., and Whiteley, A.R. 2019. The exciting potential and remaining uncertainties of genetic rescue. Trends Ecol. Evol. **34**: 1070-1079.

Bell, G., and Gonzalez, A. 2011 Adaptation and evolutionary rescue in metapopulations experiencing environmental deterioration. Science **332**: 1327-1330.

Bell, J. D., Craik, G. J. S., Pollard, D. A., and Russell, B. C. 1985. Estimating length frequency distributions of large reef fish underwater. Coral Reefs **4:** 41-44.

Bell, K.N.I. 1994. Life cycle, early life history, fisheries and recruitment dynamics of diadromous gobies of Dominica, W.I., emphasising *Sicydium punctatum* Perugia. Ph.D. thesis, Memorial University, St. John’s, Nfld. 275 p.

Benfield, E.F., Fritz, K.M., and Tiegs, S.D. 2017. Leaf-Litter Breakdown. *In* Methods in Stream Ecology: Third Edition. Elsevier. pp. 71–82.

Benstead, J.P., March, J.G., Pringle, C.M. and Scatena, F.N. 1999. Effects of a low-head dam and water abstraction on migratory tropical stream biota. Ecol. Appl. **9**: 656-668.

Bergstrom, M. A., and Mensinger, A. F. 2009. Interspecific resource competition between the invasive round goby and three native species: logperch, slimy sculpin, and spoonhead sculpin. Trans. Am. Fish. Soc. **138:** 1009-1017.

Beverton, R.J.H., and Holt, S.J. 1957. On the dynamics of exploited fish populations. *In* Volume 19 of Fishery investigations (Great Britain, Ministry of Agriculture, Fisheries, and Food). London: H. M. Stationery Office.

Bilotta, G.S., and Brazier, R.E. 2008. Understanding the influence of suspended solids on water quality and aquatic biota. Wat. Res. **42**: 2849-2861.

Blackburn, T.M., Cassey, P., and Lockwood, J.L. 2009. The role of species traits in the establishment success of exotic birds. Glob. Change Biol. **15**: 2852-2860.

Blob, R.W., Bridges, W.C., Ptacek, M.B., Maie, T., Cediel, R.A., Bertolas, M.M., Julius, M.L., and Schoenfuss, H.L. 2008. Morphological selection in an extreme flow environment: body shape and waterfall-climbing success in the Hawaiian stream fish *Sicyopterus stimpsoni*. Int. Comp. Biol. **48**: 734-49. doi:10.1093/icb/icn086. (Erratum) **49**: 732-734(2009).

Blob, R.W., Kawano, S.M., Moody, K.N., Bridges, W.C., Maie, T., Ptacek, M.B., Julius, M.L., and Schoenfuss, H.L. 2010. Morphological selection and the evaluation of potential tradeoffs between escape from predators and the climbing of waterfalls in the Hawaiian stream goby *Sicyopterus stimpsoni*. Int.Comp. Biol. **50**: 1185- 99.

Blum, M.J., Gilliam, J.F., and McIntyre, P.B. 2014. Development and use of genetic methods for assessing aquatic environmental condition and recruitment dynamics of native stream fishes on Pacific islands. Final report for the United State Department of Defense, SERDP RC-1646.

Booth, D.B., Roy, A.H., Smith, B., and Capps, K.A. 2016. Global perspectives on the urban stream syndrome. Freshw. Sci. **35**: 412-420.

Boykin, L.M., Armstrong, K.F., Kubatko, L., and De Barro, P. 2012. Species delimitation and global biosecurity. Evol. Bioinform. **8**: 1-37.

Bradburd, G.S., Ralph, P.L., and Coop, G.M. 2013. Disentangling the effects of geographic and ecological isolation on genetic differentiation. Evolution **67**: 3258-3273.

Brasher, A.M.D. 2003. Impacts of human disturbances on biotic communities in Hawaiian streams. BioScience **53**: 1052-1060.

Brasher, A.M.D., Luton, C.D., Goodbred, S.L., and Wolff, R.H. 2006. Invasion patterns along elevation and urbanization gradients in Hawaiian streams. Trans. Am. Fish. Soc. **135**: 1109-1129.

Britton, J.R., Brazier, M., Davies, G.D., and Chare, S.I. 2008 Case studies on eradicating the Asiatic cyprinid *Pseudorasbora parva* from fishing lakes in England to prevent their riverine dispersal. Aquat. Conserv. **18**: 867-876.

Britton, J. R., Davies, G. D., and Brazier, M. 2009. Eradication of the invasive Pseudorasbora parva results in increased growth and production of native fishes. Ecol. Freshwat. Fish. **18:** 8-14.

Brockway, D.G., Gatewood, R.G., and Paris, R.B. 2002. Restoring fire as an ecological process in shortgrass prairie ecosystems: initial effects of prescribed burning during the dormant and growing seasons. J. Environ. Manage. **65**: 135-152.

Brown, J.H., and Kodric-Brown, A. 1977. Turnover rates in insular biogeography: effect of immigration on extinction. Ecology **58**: 445-449.

Bryan, W.A. 1915. Natural History of Hawaii: Being an Account of the Hawaiian People, the Geology and Geography of the Islands, and the Native and Introduced Plants and Animals of the Group. The Hawaiian Gazette Co., Ltd., Honolulu, HI.

Buktenica, M.W., Hering, D.K., Girdner, S.F., Mahoney, B.D., and Rosenlund, B.D. 2013. Eradication of nonnative brook trout with electrofishing and antimycin-A and the response of a remnant bull trout population. N. Am. J. Fish. Manage. **33**: 117-129.

Byström, P., and García-Berthou, E. 1999. Density dependent growth and size specific competitive interactions in young fish. Oikos **1999**: 217-232.

Cahn, A.R. 1929. The effect of carp on a small lake: the carp as a dominant. Ecology **10**: 271-274.

Caley, M.J., Carr, M.H., Hixon, M.A., Hughes, T.P., Jones, G.P. and Menge, B.A. 1996. Recruitment and the local dynamics of open marine populations. Annu. Rev. Ecol. Syst. **27**: 477-500.

Capps, K.A., and Flecker, A.S. 2013. Invasive aquarium fish transform ecosystem nutrient dynamics. Proc. R. Soc. Lond. B **280**: 20131520.

Capps, K.A., and Flecker, A.S. 2015. High impact of low-trophic-position invaders: nonnative grazers alter the quality and quantity of basal food resources. Freshwat. Sci. **34**: 784-796.

Capps, K.A., Ulseth, A., and Flecker, A.S. 2015. Quantifying the top-down and bottom-up effects of a non-native grazer in freshwaters. Biol. Inv. **17:** 1253-1266.

Carlson, R.E., and Simpson, J. 1996. A coordinator's guide to volunteer lake monitoring methods. North American Lake Management Society **96:** 305.

Carlson, S.M., and Seamons, T.R. 2008. A review of quantitative genetic components of fitness in salmonids: implications for adaptation to future change. Evol. Apps. **1**: 222-238.

Carr, M.H., Anderson, T.W. and Hixon, M.A. 2002. Biodiversity, population regulation, and the stability of coral-reef fish communities. Proc. Natl. Acad. Sci. U.S.A **99**: 11241-11245.

Carson, H.L., and Clague, D.A. 1995. Geology and biogeography of the Hawaiian Islands. *In* Hawaiian biogeography: Evolution on a hotspot archipelago. *Edited by* W.L. Wagner and V.A. Funk. Smithsonian Institution Press, Washington, DC. pp. 14-29.

Carvalho, L.N., Zuanon J., and Sazima, I. 2009. Natural history of Amazon fishes. *In* Tropical biology and conservation management: case studies. Vol. XI. *Edited by* K. Del-Claro, O.S. Oliveria, and V. Rico-Gray. Eolss Publishers Co. Ltd., Oxford. pp.133-144.

Catchen, J., Amores, A., Hohenlohe, P., Cresko, W., and Postlethwait J. 2011. *Stacks: building and genotyping loci de novo from short-read sequences*. G3: Genes, Genomes, Genetics **1**: 171-182.

Caves, E.M., Jennings, S.B., HilleRisLambers, J., Tewksbury, J.J. and Rogers, H.S. 2013. Natural experiment demonstrates that bird loss leads to cessation of dispersal of native seeds from intact to degraded forests. PLoS One **8**(5).

Chadderton, W.L., Kelleher, S., Brow, A., Shaw, T., Studholme, B., and Barrier R. 2003. Testing the efficacy of rotenone as a piscicide for New Zealand pest fish species. In: Managing Invasive Freshwater Fish in New Zealand. pp. 113-130. DOC workshop, May 2001, Hamilton.

Champion, P.D. 2018. Knowledge to action on aquatic invasive species: Island biosecurity - the New Zealand and South Pacific story. Manag. Biol. Invasion. **9**: 383-394.

Chapin III, F.S., Zavaleta, E.S., Eviner, V.T., Naylor, R.L., Vitousek, P.M., Reynolds, H.L., Hooper, D.U., Lavorel, S., Sala, O.E., Hobbie, S.E., Mack, M.C., and Diaz, S. 2000. Consequences of changing biodiversity. Nature **405**: 234-242.

Chapman, B.B., Hulthén, K., Blomqvist, D.R., Hansson, L.-A., Nilsson, J.-Å., Brodersen, J., Nilsson, P.A., Skov, C., and Brönmark, C.. 2011a. To boldly go: individual differences in boldness influence migratory tendency. Ecol. Lett. **14**: 871-876.

Chapman, B.B., Bronmark, C., Nilsson, J.A., and Hansson, L.A. 2011b. The ecology and evolution of partial migration. Oikos **120**: 1764-1775.

Chapman, B.B., Hulthén, K., Brodersen, J., Nilsson, P.A., Skov, C., Hansson, L.A., and Brönmark, C. 2012. Partial migration in fishes: causes and consequences. J. Fish Biol. **81:** 456-478.

Chervinski, J. 1984. Salinity tolerance of the guppy, *Poecilia reticulata* Peters. J. Fish Biol., **24**: 449-452.

Chesson, P. 1994. Multispecies competition in variable environments. Theor. Popul. Biol. **45**: 227-276.

Chesson, P. 2000. Mechanisms of maintenance of species diversity. Annu. Rev. Ecol. Evol Syst. **31**: 343-366.

Cho, G.K., Heath, J.W., and Heath, D.D. 2002. Electroshocking influences Chinook salmon egg survival and juvenile physiology and immunity. T. Am. Fish. Soc. **131**: 224-233.

Chu, P.S., Chen, H. 2005. Interannual and interdecadal rainfall variations in the Hawaiian Islands. J Climate **8**:4796-4813.

Coat, S., Monti, D., Bouchon, C., and Lepoint, G. 2009. Trophic relationships in a tropical stream food web assessed by stable isotope analysis. Freshwat. Biol. **54**: 1028-1041.

Cole, J.E., Overpeck, J.T., and Cook, E.R. 2002. Multiyear La Niña events and persistent drought in the contiguous United States. Geophys. Res. Lett. **29:** 25-31.

Cole, M.L., Valiela, I., Kroeger, K.D., Tomasky, G.L., Cebrian, J., Wigand, C., McKinney, R.A., Grady, S.P., and Carvalho da Silva, M.H. 2004. Assessment of a δ 15 N isotopic method to indicate anthropogenic eutrophication in aquatic ecosystems. J. Environ. Qual. **33:** 124-132

Cook, B.D., Bernays, S., Pringle, C.M, and Hughes, J.M. 2009. Marine dispersal determines the genetic structure of migratory stream fauna of Puerto Rico: evidence for island-scale recovery processes. J. N. Am. Benthol. Soc. **28**: 709-718.

Cottenie, K. 2005. Integrating environmental and spatial processes in ecological community dynamics. Ecol. Lett. **8**: 1175-1182.

Covich, A., Crowl, T., and Scatena, F. 2003. Effects of extreme low flows on freshwater shrimps in a perennial tropical stream. Freshwater Biol*.* **48**: 1199-1206.

Crecco, V.A., and Savoy, T. 1987. Review of recruitment mechanisms of the American shad: the critical period and match-mismatch hypotheses reexamined. *In:* American Fisheries Society Symposium **1:** 455-468.

Crowley, S.L., Hinchliffe, S., and McDonald, R.A. 2017. Conflict in invasive species management. Front. Ecol. Environ. **15**: 133-141.

Cucherousset, J., and Olden, J.D. 2011. Ecological impacts of non-native freshwater fishes. Fisheries **36**: 215-230.

Cushing, D.H. 1973. The natural regulation of fish populations. Sea Fish. Res. **1973**.

Cushing, D.H. 1974. The possible density-dependence of larval mortality and adult mortality in fishes. *In*: The Early Life History of Fish. Springer, Berlin, Heidelberg, 103-111.

Cushing, D.H., and Harris, J.G.K. 1973. Stock and recruitment and the problem of density dependence. Rapp. P.-v. Reun. Cons. Int. Explor. Mer. **164**: 142-155.

Dahruddin, H., Hutama, A., Busson, F., Sauri, S., Hanner, R., Keith, P., Hadiaty, R., and Hubert, N. 2017. Revisiting the ichthyodiversity of Java and Bali through DNA barcodes: taxonomic coverage, identification accuracy, cryptic diversity and identification of exoctic species. Mol. Ecol. **17**: 288-299.

Dalbey, S.R., McMahon, T.E., and Fredenberg, W. 1996. Effect of electrofishing pulse shape and electrofishing-induced spinal injury on long-term growth and survival of wild rainbow trout. N. Am. J. Fish. Manage. **16**: 560-569.

D'antonio, C.M., and Dudley, T.L. 1995. Biological invasions as agents of change on islands versus mainlands. *In*: Islands, Springer, Berlin, Heidelberg, 103-121.

Deiner, K., Bik, H.M., Mächler, E., Seymour, M., Lacoursière-Roussel, A., Altermatt, F., Creer, S., Bista, I., Lodge, D.M., de Vere, N., Pfrender, M.E., and Bernatchez, L. 2017. Environmental DNA metabarcoding: Transforming how we survey animal and plant communities. Mol. Ecol. **21**: 5872-5895.

Devick, W.S., Fitzsimons, J.M., and Nishimoto, R.T. 1995. Threatened fishes of the world: *Lentipes concolor* Gill, 1860 (Gobiidae). Environ. Biol. Fish. **44**: 325-326.

Di, H.J., and Cameron, K.C. 2002. Nitrate leaching in temperate agroecosystems: sources, factors and mitigating strategies. Nutr. Cycling Agroecosyst. **46:** 237-256.

Diaz, H.F., Chu, P.S., and Eischeid, J.K. 2005. Rainfall changes in Hawaii during the last century. 16th Conference on Climate Variability and Change, Am Met Soc, Boston, MA (Available at http://ams.confex.com/ams/Annual2005/techprogram/paper_84210.htm)

Dinger, E.C., and Marks, J.C. 2007. Effects of high levels of Antimycin A on aquatic invertebrates in a warmwater Arizona stream. N. Am. J. Fish. Manage. **27**: 1243-1256.

Dodson, J.J., Aubin-Horth, N., Thériault, V., and Páez, D.J. 2013. The evolutionary ecology of alternative migratory tactics in salmonid fishes. Biol. Rev. **88**: 602-625.

Doherty, T.S., Glen, A.S., Nimmo, D.G., Ritchie, E.G., and Dickman, C.R. 2016. Invasive predators and global biodiversity loss. , M.A. 2002. Biodiversity, population regulation, and the stability of coral-reef fish communities. Proc. Natl. Acad. Sci. U.S.A **113**: 11261-11265.

Donlan, C.J., and Wilcox, C. 2007. Complexities of costing eradications. Anim. Conserv. **10**: 154-156.

Dowle, E.J., Pochon, S., Banks, J.C., Shearer, K., and Wood, S.A. 2016. Targeted gene enrichment and high-throughput sequencing for environmental biomonitoring: a case study using freshwater macroinvertebrates. Mol. Ecol. Res. **16**: 1240-1254.

Dudgeon, D., Arthington, A.H., Gessner, M.O., Kawabata, Z.-I., Knowler, D.J., Lévêque, C., Naiman, R.J., Prieur-Richard, A.-H., Soto, D., Stiassny, M.L.J., and Sullivan, C.A. 2006. Freshwater biodiversity: importance, threats, status and conservation challenges. Biol. Rev. **81**: 163-182.

Duffy, D.C., and Martin. C. 2019. Cooperative naturel resource and invasive species management in Hawaii. In Island invasives: scaling up to meet the challenge. Edited by: Veitch, C.R., Clout, M.N., Martin, A.R., Russell, J.C., and West, C.J. Occasional Paper SSC no. 62. Gland, Switzerland:IUCN.

Edmands, S. 2007. Between a rock and a hard place: evaluating the relative risks of inbreeding and outbreeding for conservation and management. Mol. Ecol. **16**: 463-475.

Ebner, B.C., Fulton, C.J., Cousins, S., Donaldson, J.A., Kennard, M.J., Meynecke, J.-O., and Schaffer J. 2015. Filming and snorkeling as visual techniques to survey fauna in difficult to access tropical rainforest streams. Mar. Freshwater Res. **66**: 120-126.

Ego, K. (1956). Life history of freshwater gobies. Freshwater Game Fish Management Research, Project **4**:1-23.

Egerer, M.H., Fricke, E.C. and Rogers, H.S. 2018. Seed dispersal as an ecosystem service: frugivore loss leads to decline of a socially valued plant, *Capsicum frutescens*. Ecol. Appl. **28**: 655-667.

Eldredge, L.G. 2000. Non-Indigenous Freshwater Fishes, Amphibians and Crustaceans of the Pacific and Hawaiian Islands. Invasive Species in the Pacific: A Technical Review and Draft Regional Strategy. 197 p. South Pacific Regional Environment Programme.

Ellison, M.E., and Brett, M.T. 2006. Particulate phosphorus bioavailability as a function of stream flow and land cover. Wat. Res. **40**: 1258-1268.

El-Sabaawi, R.W., Frauendo, R.T.C., Marques, P.S., Mackenzie, R.A., Manna, L.R., Mazzoni, R., Phillip, D.A., Warbanski, M.L., and Zandonà E. 2016. Biodiversity and ecosystem risks arising from using guppies to control mosquitoes. Biol. Lett. **12**: 20160590.

Elser, J. J., Marzolf, E. R., and Goldman, C. R. 1990. Phosphorus and nitrogen limitation of phytoplankton growth in the freshwaters of North America: a review and critique of experimental enrichments. Can. J. Fish. Aquat. Sci. **47**: 1468-1477.

Elser, J. J., Hayakawa, K., and Urabe, J. 2001. Nutrient limitation reduces food quality for zooplankton: Daphnia response to seston phosphorus enrichment. Ecology **82:** 898-903.

Elser, J. J., Bracken, M. E. S., Cleland, E. E., Gruner, D. S., Harpole, W. S., Hillebrand, H., Ngai, J. T., Seabloom, E. W., Shurin, J. B., and Smith, J. E. 2007. Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. Ecol. Lett. **10:** 1135-1142.

Elton, C.S. 1958. The Ecology of Invasions by Animals and Plants. University of Chicago Press, Methuen, London.

Elwood, J. W., Newbold, J. D., Trimble, A. F., and Stark, R. W. 1981. The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary producers. Ecology **62:** 146-158.

Englund, R.A. 1999. The impacts of introduced poeciliid fish and Odonata on the endemic Megalagrion (Odonata) damselflies of Oahu Island, Hawaii. J. Insect Conserv. **3**: 225-243.

Englund, R.A. 2001. Long-term monitoring of one of the most restricted insect populations in the United States, *Megalagrion xanthomelas* (Selys-Longchamps), at Tripler Army Medical Center Oʻahu, Hawaii (Zygoptera: Coenagrionidae). Odonatologica **30**: 255-263.

Englund, R.A., and Filbert, R.B. 1999. Flow restoration and persistence of introduced species in Waikele Stream, Oʻahu. Micronesica **31**: 143-154.

Engman, A. C., Kwak, T. J., and Fischer, J. R. 2017. Recruitment phenology and pelagic larval duration in Caribbean amphidromous fishes. Freshwat. Sci. **36:** 851-865.

Evans, N.T., Olds, B.P., Renshaw, M.A., Turner, C.R., Li, Y., Jerde, C.L., Mahon, A.R., Pfrender, M.E., Lamberti, G.A., and Lodge, D.M. 2016. Quantification of mesocosm fish and amphibian species diversity via environmental DNA metabarcoding. Mol. Ecol. Res. **16**: 29-41.

Fauvergue, X., Vercken, E., Malausa, T., and Hufbauer, R.A. 2012. The biology of small, introduced populations, with special reference to biological control. Evol. Appl. **5**: 424-443.

Finlayson, B.J., Schnick, R.A., Cailteux, R.L., Demong, L., Horton, W.D., McClay, W., and Thompson, C.W. 2002. Assessment of Antimycin A use in fisheries and its potential for reregistration. Fisheries **27**: 10-18.

Fisher, T.R., Peele, E.R., Ammerman, J.W., and Harding, Jr, L.W. 1992. Nutrient limitation in Chesapeake Bay. Mar. Ecol. Prog. Ser. **82:** 51-63.

Fitzsimons, J.M., McRae, M.G., and Nishimoto, R.T. 2007. Behavioral Ecology of Indigenous Stream Fishes in Hawaiʻi. Biology of Hawaiian Streams and Estuaries **3**:11-21.

Fitzpatrick, S.W., Bradburd, G.S., Kremer, C.T., Salerno, P.E., Angeloni, L.M. and Funk, W.C. 2020. Genomic and fitness consequences of genetic rescue in wild populations. Current Biology **30**: 517-522.e5.

Font, W.F. 2003. The global spread of parasites: what do Hawaiian streams tell us? *BioScience* **53**: 10611067.

Frankham, R. 2016. Genetic rescue benefits persist to at least the F3 generation, based on a meta-analysis. Biol. Conserv. **195**: 33-36.

Frankham, R., Ballou, J.D., Eldridge, M.D.B., Lacy, R.C., Ralls, K., Dudash, M.R., and Fenster, C.B. 2011.

Predicting the probability of outbreeding depression. Conserv. Biol. **25**: 465-475.

Frankham, R., Ballou, J.D., Ralls, K., Eldridge, M.D.B., Dudash, M.R., Fenster, C.B., Lacy, R.C., and Sunnucks, P. 2017. Genetic Management of Fragmented Animal and Plant Populations, Oxford University Press.

Fulton, W., Pendall, R., Nguyen, M., & Harrison, A. 2001. Who sprawls most? How growth patterns differ across the U.S. Center on Urban and Metropolitan Policy (24 p). Washington, DC.

Gaeta, J.W., Hrabik, T.R., Sass, G.G., Roth, B.M., Gilbert, S.J., and Vander Zanden, M.J. 2014. A whole-lake experiment to control invasive rainbow smelt (Actinoperygii, Osmeridae) via overharvest and a food web manipulation. Hydrobiologia **746**: 433-444.

Gagne, R.B., Hogan, J.D., Pracheil, B.M., McIntyre, P.B., Hain, E.F., Gilliam, J.F., and Blum, M.J. 2015. Spread of an introduced parasite across the Hawaiian archipelago independent of its introduce host. Freshwater Biol. **60**: 311-322.

Gagne, R.B., and Blum, M.J. 2016. Parasitism of a native Hawaiian stream fish by an introduced nematode increases with declining precipitation across a natural rainfall gradient. Ecol. Freshwat. Fish **25:** 476-486.

Gagne, R.B., Heins, D.C., McIntyre, P.B., Gilliam, J.F., and Blum, M.J. 2016. Mutual dilution of infection by an introduced parasite in native and non-native stream fishes across Hawaii. Parasitology **143**: 1605-1614.

Gagne, R.B., Sprehn, C.G., Alda, F., McIntyre, P.B., Gilliam, J.F., and Blum, M.J. 2018. Invasion of the Hawaiian Islands by a parasite of imperiled stream fishes. Ecography **41**: 528-539.

Gallardo, B., Clavero, M., Sánchez, M. I., and Vilà, M. 2016. Global ecological impacts of invasive species in aquatic ecosystems. Glob. Change. Biol. **22**: 151-163.

Garant, D., Forde, S.E., and Hendry, W.P. 2007. The multifarious effects of dispersal and gene flow on contemporary adaptation. Funct. Ecol. **21**: 434-443.

Giambelluca, T.W., Nullet, M.A., and Schroeder, T.A. 1986. Hawaii rainfall atlas. Hawaii Department of Land and Natural Resources Division of Water and Land Development Report, R76, 267.

Gido, K.B., and Franssen, N.R. 2007. Invasion of stream fishes into low trophic positions. Ecol. Freshw. Fish **16**: 457-464.

Gillespie, R. 2004. Community assembly through adaptive radiation in Hawaiian spiders. Science **303**: 356-359.

Glen, A.S., Atkinson, R., Campbell, K.J., Hagen, E., Holmes, N.D., Keitt, B.S., Parkes, J.P., Saunders, A., Sawyer, J., and Torres, H. 2013. Eradicating multiple invasive species on inhabited islands: the next big step in island restoration? Biol. Invasions **15**: 2589-2603.

Gore, J.A., and Banning, J. 2017. Discharge measurements and streamflow analysis. *In* Methods in stream ecology, vol 1, 3rd edn. *Edited by* F. R. Hauer and G.A. Lamberti. Academic Press, Boston, MA. pp 49-70.

Gopalakrishnan, C. 2005. Water allocation and management in Hawaii: A case of institutional entropy. *In*: Water institutions: Policies, Performance and Prospects. Springer, Berlin, Heidelberg, 1-23.

Green, S.J., Dulvy, N.K., Brooks, A.M., Akins, J.L., Cooper, A.B., Miller, S., and Côté, I.M. 2014. Linking removal targets to the ecological effects of invaders: a predictive model and field test. Ecol. Appl. **24**: 1311-1322.

Green, S.J., Underwood, E.B., and Akins, J.L. 2017. Mobilizing volunteers to sustain local suppression of a global marine invasion. Conserv. Lett. **10**: 726-735.

Gruner, D.S. 2007. Geological age, ecosystem development, and local resource constraints on arthropod community structure in the Hawaiian Islands. Biol. J. Linn. Soc. **90**: 551-570.

Ha, P.Y. and Kinzie, R.A. 1996. Reproductive biology of *Awaous guamensis*, an amphidromous Hawaiian goby. Environ. Biol. Fish. **45**: 383-396.

Haddad, N.M., Lars, A.B., Clobert, J., Davies, K.F., Gonzalez, A., Holt, R.D., Lovejoy, T.E., Sexton, J.O., Austin, M.P., Collines, C.D., Cook, W.M., Damschen, E.I., Ewers, R.M., Foster, B.L., Jenkins, C.N., King, A.J., Laurance, W.F., Levey, D.J., Margules, C.R., Melbournes, B.A., Nicholls, A.O., Orrock, J.L., Song, D.-X., and Townshend, J.R. 2015. Habitat fragmentation and its lasting impact on Earth's ecosystems. Sci. Adv. **1**: e1500052.

Hain, E.F., Lamphere, B.A., Blum, M.J., McIntyre, P.B., Nelson, S.A.C., and Gilliam, J.F. 2016. Comparison of visual survey and mark-recapture population estimates of a benthic fish in Hawaii. T. Am. Fish. Soc. **145**: 878-887.

Hain, E.F., Moody, K.N., Lamphere, B.A., Nelson, S.A.C., McIntyre, P.B., Gilliam, J.F., and Blum, M.J. 2019. Migratory gauntlets on oceanic islands: Watershed disturbance increases the cost of amphidromy. Ecol. Freshw. Fish. **28**: 446-458.

Hale, M.C., Thrower, F.P., Berntson, E.A., Miller, M.R., and Nichols, K.M. 2013. Evaluating adaptive divergence between migratory and nonmigratory ecotypes of a salmonid fish, *Oncorhynchus mykiss*. G3:Genes, Genomes, Genetics **3**: 1273-1285.

Hanski, I. 1998. Metapopulation dynamics. Nature, **396:** 41-49.

Harris, D.B., Gregory, S.D., Bull, L.S., and Courchamp, F. 2012. Island prioritization for invasive rodent eradications with an emphasis on reinvasion risk. Biol. Invasions **14**: 1251-1263.

Hauer, F.R., and Lamberti, G. eds. 2011. Methods in stream ecology. Academic Press.

Hecht, B.C., Hard, J.J., Thrower, F.P., Nichols, K.M. 2015. Quantitative genetics of migration-related traits in rainbow and steelhead trout. G3:Genes, Genomes, Genetics **5**: 873-889.

Hedges, L. V., Gurevitch, J., and Curtis, P.S. 1999. The meta-analysis of response ratios in experimental ecology. Ecology **80**: 1150.

Hedrick, P.W. 1995. Gene flow and genetic restoration – the Florida Panther as a case study. *Con. Biol.* **9:** 996-1007.

Heim-Ballew, H. 2019. Identifying Migration Flexibility and the Environmental Factors that Influence Variation in Recruitment Success in Partially Migratory Hawaiian Fishes Doctoral dissertation, Texas A&M University-Corpus Christi.

Heim-Ballew, H., Moody, K. N., Blum, M. J., McIntyre, P. B., and Hogan, J. D. 2020. Migratory flexibility in native Hawai'ian amphidromous fishes. J. Fish Biol., **962**: 456-468.

Hein, C.L., Vander Zanden, M.J., and Magnuson, J.J. 2007. Intensive trapping and increased fish predation cause massive population decline of an invasive crayfish. Freshw. Biol. **52**: 1134-1146.

Hense, Z., Martin, R.W., and Petty J.T. 2010. Electrofishing capture efficiencies for common stream fish species to support watershed-scale studies in the central Appalachians. N. Am. J. Fish. Manage. **30**: 1041-1050.

Hicks, B.J., Hamilton, D.P., Ling, N., and Wood, S. 2007. Top down or bottom up? Feasibility of water clarity restoration in the lower Karori reservoir by fish removal. Report prepared for the Karori Wildlife Sanctuary Trust. University of Waikato, Hamilton New Zealand, CBER Contract Report 70.

Higashi, G.R., and Nishimoto, R.T. 2007. The point quadrat method: A rapid assessment of Hawaiian streams. Bishop Mus. Bull. Cult. Environ. Stud. **3**: 305-312.

Hirsch, R.M., and De Cicco, L.A. 2015. User Guide to Exploration and Graphics for River Trends EGRET and dataRetrieval: R packages for hydrologic data version 2.0, February 2015: U.S. Geological Survey Techniques and Methods Book 4, A10, 93.

Hixon, M.A. and Beets, J.P. 1993. Predation, prey refuges, and the structure of coral-reef fish assemblages. Ecol. Monogr. **63**: 77-101.

Hixon, M.A. and Webster, M.S. 2002. Density dependence in reef fish populations. *In:* Coral reef fishes: dynamics and diversity in a complex ecosystem (ed: P.F. Sale). Academic Press, San Diego, California, USA, 303-325.

Hjort, J. 1914. Fluctuations in the great fisheries of northern Europe viewed in the light of biological research. ICES J. Mar. Sci. **1914:** 11 - 60.

Hjort, J. 1926. Fluctuations in the year classes of important food fishes. ICES J. Mar. Sci. **1:** 5-38.

Hogan, J.D., Blum, M.J., Walter, R.P. 2011. Characterization of ten novel microsatellite markers in *Awaous guamensis* with comments on cross amplification in congeners and other amphidromous fish native to Hawai'i. Conserv. Genet. Resour**. 3**: 275-277

Hogan, J.D., Blum, M.J., Gilliam, J.F., Bickford, N., and McIntyre, P.B. 2014. Consequences of alternative dispersal strategies in a putatively amphidromous fish. Ecology **95**: 2397-2408.

Hoese, D.F., and Gill, A. 1993. Phylogenetic relationships of Eleotridid fishes (Perciformes: Gobioidei). B. Mar. Sci. **52**: 415-440.

Holitzki, T.M., Mackenzie, R.A., Weigner, T.N., and McDermid, K.J. 2013. Differences in ecological structure, function, and native species abundance between native and invaded Hawaiian streams. Ecol. Appl. **23**: 1367-1383.

Holliman, F.M., Reynolds, J.B., and Kwak, T.J. 2003. Electroshock-induced injury and mortality in the spotfin chub, a threatened minnow. N. Am. J. Fish. Manage. **23**: 962-966.

Holtgrieve, G.W., Schindler, D.E., Branch, T.A., and A'mar, Z.T. 2010a. Simultaneous quantification of aquatic ecosystem metabolism and reaeration using a Bayesian statistical model of oxygen dynamics. Limnol. Oceanogr. **55**: 1047-1063.

Holtgrieve, G.W., Schindler, D.E., Gowell, C.P., Ruff, C.P., and Lisi, P.J. 2010b. Stream geomorphology regulates the effects on periphyton of ecosystem engineering and nutrient enrichment by Pacific salmon. Freshw. Biol. **55**: 2598-2611.

Homer, C., Dewitz, J., Fry, J., Coan, M., Hossain, N., Larson, C., Herold, N., McKerrow, A., VanDriel, J.N., and Wickham, J. 2007. Completion of the 2001 National Land Cover Database for the conterminous United States. Photogramm. Eng. Remote Sens. **73**: 337-341.

Horak, D.L., and Klein, W.D. 1967. Influence of capture methods on fishing success, stamina, and mortality of rainbow trout *(Salmo gairdneri)* in Colorado. T. Am. Fish. Soc. **96**: 220-222.

Hood, J.M., Vanni, M.J., and Flecker, A. S. 2005. Nutrient recycling by two phosphorus-rich grazing catfish: the potential for phosphorus-limitation of fish growth. Oecologia **146:** 247-257.

Houde, E.D. 1997. Patterns and trends in larval-stage growth and mortality of teleost fish. J. Fish Biol. **51**: 52-83.

Houde, E.D. 2008. Emerging from Hjort's shadow. J. N.W. Atl. Fish. Sci. **41**: 53-70.

Houde, E.D., and Hoyt, R.D. 1987. Fish early life dynamics and recruitment variability. Trans. Am. Fish. Soc. **1987.**

Huey, J.A., Crook, D.A., Macdonald, J.I., Schmidt, D.J., Marshall, J.C., Balcombe, S.R., Woods, R.J., and Hughes, J.M. 2014. Is variable connectivity among populations of a continental gobiid fish driven by local adaptation or passive dispersal? Freshwat. Biol. **59:** 1672-1686.

Hufbauer, R.A., Szucs, M., Kasyon, E., Youngberg, C., Koontz, M.J., Richards, C., Tuff, T., and Melbourne, B.A. 2015. Three types of rescue can avert extinction in a changing environment. P. Natl. Acad. Sci. USA **112**: 10557-10562.

Hothorn T., Bretz, F., and Westfall, P. 2008. Simultaneous Inference in General Parametric Models. Biometrical J. **50**: 346-363.

Iguchi, K., and Mizuno, N. 1991. Mechanisms of embryonic drift in the amphidromous goby, Rhino-gobius brunneus. Environ. Biol. Fish. **31:** 295-300.

Iizumi, T., Battaglia, T., Ruiz, V., and Perez Perez, G.I. 2017. Gut microbiome and antibiotics. Arch. Med. Res. **48**: 727-734.

Ingvarsson, P.K. 2001. Restoration of genetic variation lost – the genetic rescue hypothesis. Trends Ecol. Evol. 16:62-63.

Jenkins, A.P., Jupiter, S.D., Qauqau, I., and Atherton, J. 2010. The importance of ecosystem-based management for conserving aquatic migratory pathways on tropical high islands: a case study from Fiji. Aquat. Conserv. Mar. Freshwat. Ecosyst. **20:** 224-238.

Jia, Y., Calil, P.H.R., Chassignet, E.P., Metzger, E.J., Potemra, J.T., Richards, K.J., and Wallcraft, A.J. 2011. Generation of mesoscale eddies in the lee of the Hawaiian Islands. J Geophys. Res. Oceans **116**(C11)

Jombart, T. 2008. adegenet: a R package for the multivariate analysis of genetic markers. Bioinformatic*,* **24**: 1403-1405.

Jombart, T., Devillard, S., and Balloux, F. 2010. Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. BMC Gen. **11**: 94.

Jombart, T., and Ahmed, I. 2011. adegenet 1.3-1: new tools for the analysis of genome-wide SNP data. Bioinformatics **27**: 3700-3701.

Jones, H.P., Holmes, N.D., Butchart, S.H., Tershy, B.R., Kappes, P.J., Corkery, I., Aguirre-Muñoz, A., Armstrong, D.P., Bonnaud, E., Burbidge, A.A. and Campbell, K. 2016. Invasive mammal eradication on islands results in substantial conservation gains. Proc. Natl. Acad. Sci. USA, **113**: 4033-4038.

Jonsson, B., and Jonsson, N. 1993. Partial migration: niche shift versus sexual maturation in fishes. Rev. Fish Biol. Fisher **3**: 348-365.

Kamvar, Z.N., Tabima, J.F., Everhart, S.E., Brooks, J.C., Krueger-Hadfield, S.A., and Sotka, E. 2019. Package 'poppr'.

Karlsen, B. O., Klingan, K., Emblem, A., Jorgensen, T. E., Jueterbock, A., Furmanek, T.,Hoarau, G., Johansen, S. D., Steinar, D., Nordeide, J. T. &Moum, T. 2013. Genomic divergence between the migratory and stationary ecotypes of Atlantic cod. Mol. Ecol. **22**: 5098–5111.

Keith, P. 2003. Biology and ecology of amphidromous Gobiidae of the Indo-Pacific and the Caribbean regions. J. Fish Biol. **63**: 831–847.

Keller, L.F., Jeffery, K.J., Arcese, P., Beaumont, M.A., Hochachka, W.M., Smith, J.N.M, and Bruford, M.W. 2001. Immigration and the ephemerality of a natural population bottleneck: evidence from molecular markers. Proc. Roy. Soc. B **268:** 1387-1394.

Kerr, L.A., and Secor, D.H. 2010. Latent effects of early life history on partial migration for an estuarine-dependent fish. Environ. Biol. Fish. **89**: 479-492.

Kido, M.H. 1996. Morphological variation in feeding traits of native Hawaiian stream fishes. Pac. Sci. **50**: 184-193.

Kido, M. H. 1997. Food relations between coexisting native Hawaiian stream fishes. Environ. Biol. Fish. **49:** 481-494.

Kido, M. H., Ha, P., and Kinzie III, R. A. 1993. Insect introductions and diet changes in an endemic Hawaiian amphidromous goby, Awaous stamineus Pisces: Gobiidae. Pac. Sci. **471:** 43-50.

Kido, M.H. 2008. A persistent species assemblage structure along a Hawaiian stream from catchment-to-sea. Environ. Biol. Fish. **82**: 223-235.

Kier, G., Kreft, H., Lee, T.M., Jetz, W., Ibisch, P.L., Nowicki, C., Mutke, J., and Barthlott, W. 2009. A global assessment of endemism and species richness across island and mainland regions. P. Natl. Acad. Sci. USA **106**: 9322-9327.

Kiernan, J.D., Moyle, P.B. and Crain, P.K. (2012) Restoring native fish assemblages to a regulated California stream using the natural flow regime concept. Ecol. Appl. 22: 1472-1482.

King, A.J., Tonkin, Z. and Mahoney, J., 2009. Environmental flow enhances native fish spawning and recruitment in the Murray River, Australia. River Res. Appl. **25**: 1205-1218.

Kinzie III, R.A. 1988. Habitat utilization by Hawaiian stream fishes with reference to community structure in oceanic island streams. Environ. Biol. Fish. **22**: 179-192.

Kinzie III, R.A. 1990. Species Profiles: Life Histories and Environmental Requirements of Coastal Vertebrates and Invertebrates Pacific Ocean Region. Report 3. Amphidromous Macrofauna of Hawaiian Island Streams (No. 3). Hawaii Institute of Marine Biology, Kaneohe.

Kinzie, R.A., Chong, C., Devrell, J., Lindstrom, D. and Wolff, R. (2006) Effects of Water Removal on a Hawaiian Stream Ecosystem. Pac. Sci. **60**: 1-47.

Kirs, M., Kisand, V., Wong, M., Caffaro-Filho, R.A., Moravcik, P., Harwood, V.J., Yoneyama, B., and Fujioka, R.S. 2017. Multiple lines of evidence to identify sewage as the cause of water quality impairment in an urbanized tropical watershed. Water Res. **116**: 23-33.

Kleunen, M. van, Dawson, W., and Maurel, N. 2015. Characteristics of successful alien plants. Mol. Ecol. **24**: 1954-1968.

Koehn, J., Brumley, A., and Gehrke, P. 2000. Managing the Impacts of Carp. Bureau of Rural Sciences (Department of Agriculture, Fisheries and Forestry - Australia), Canberra. Available at: http://www.daff.gov.au/brs/land/ feral-animals/apamp/managing_the_impacts_of_carp

Kolar, C.S., Courtenay, W. Jr., and Nico, L. 2010. Managing undesired and invading fishes. *In* Inland fisheries management in North America. *Edited by* W.A. Hubert and M.C. Quist. American Fisheries Society, Bethesda, MD. pp. 213-259.

Kral, K., Limb, R., Ganguli, A., Hovick, T., and Sedivec, K. 2018. Seasonal prescribed fire variation decreases inhibitory ability of *Poa pratensis L*. and promotes native plant diversity. J. Environ. Manage. **223**: 908-916.

Kuang, T., Tornabene, L., Li, J., Jiang, J., Chakrabarty, P., Sparks, J. S., Naylor, G.J.P., and Li, C. 2018. Phylogenomic analysis on the exceptionally diverse fish clade Gobioidei (Actinopterygii: Gobiiformes) and data-filtering based on molecular clocklikeness. Mol. Phylo. Evol. **128**: 192-202.

Kulp, M.A., and Moore, S.E. 2000. Multiple electrofishing removals for eliminating rainbow trout in a small southern Appalachian stream. N. Am. J. Fish. Manage **20**: 259-266.

Kwak, T.J., Cooney, P.B., and Brown, C.H. 2007. Fishery population and habitat assessment in Puerto Rico streams: phase 1 final report. Federal Aid in Sport Fish Restoration Project F-50 Final Report. Submitted to Marine Resources Division, Puerto Rico Department of Natural and Environmental Resources, San Juan, Puerto Rico.

Kwak, T.J., Cooney, P. 2008. Fishery population and habitat assessment in Puerto Rico streams. Report to Puerto Rico Department of Natural and Environmental Resources.

Laplanche, C., Elger, A., Santoul, F., Thiede, G. P., and Budy, P. 2018. Modeling the fish community population dynamics and forecasting the eradication success of an exotic fish from an alpine stream. Biol. Conserv. **223**: 34-46.

Lathrop, R.C. 2007. Perspectives on the eutrophication of the Yahara lakes. Lake Reserv. Manag. **23**(4): 345-365.

Lathrop, R.C., Liebl, D.S., and Welke, K. 2013. Carp removal to increase water clarity in shallow eutrophic Lake Wingra. Lakeline Mag. North Am. Lake Manag. Soc. Madison WI **33**: 23-30.

Lande, R. 1988. Genetics and demography in biological conservation. Science **241**: 1455-1460.

Laramie, M.B., Pilliod, D.S., Goldberg, C.S. 2015. Characterizing the distribution of an endangered salmonid using environmental DNA analysis. Biol. Conserv. **183**: 29-37.

Lau, L.S., and Mink, J.F. 2006. Hydrology of the Hawaiian Islands. University of Hawai'i Press.

Lawrie, A.H. 1970. The sea lamprey in the Great Lakes. Trans. Am. Fish. Soc. **99**: 766-775.

Lawson, Z.J., Vander Zanden, M.J., Smith, C.A., Heald, E., Hrabik, T.R., and Carpenter, S.R. 2015. Experimental mixing of a north-temperate lake: testing the thermal limits of a cold-water invasive fish. Can. J. Fish. Aquat. Sci. **72**: 926-937.

Lemopoulos, A., Uusi-Heikkilä, S., Vasemägi, A., Huusko, A., Kokko, H., and Vainikka, A. 2018. Genome-wide divergence patterns support fine-scaled genetic structuring associated with migration tendency in brown trout. Can. J. Fish. Aquat. Sci. **75**: 1680-1692.

Lenthe, R., Singmann, H., Love, J., Buerkner, P, and Herve, M. 2020. emmeans: Estimated Marginal Means, aka Least-Squares Means, R package version 1.4.4, https://cran.r-project.org/web/packages/emmeans/index.html.

Lepak, J.M., Kraft, C.E. and Weidel, B.C. 2006. Rapid food web recovery in response to removal of an introduced apex predator. Can. J. Fish. Aquat. Sci. **63**: 569-575.

Lerner, H.R.L., Meyer, M., James, H.F., Hofreiter, M., and Fleischer, R.C. 2011. Multilocus resolution of phylogeny and timescale in the extant adaptive radiation of Hawaiian honeycreepers. Curr. Biol. **21**: 1838-1844.

Li, Y.M., and Chesson, P. 2018. Seed demographic comparisons reveal spatial and temporal niche differentiation between native and invasive species in a community of desert winter annual plants. Evol. Ecol. Res. 19: 71–84.

Liedvogel, M., Åkesson, S., and Bensch, S. 2011. The genetics of migration on the move. Trends Ecol. Evol. **26**: 561-569.

Lim, N.K., Tay, Y.C., Srivathsan, A., Tan, J.W., Kwik, J.T.B., Baloğlu, B., Meier, R., and Yeo, D.C. 2016. Next-generation freshwater bioassessment: eDNA metabarcoding with a conserved metazoan primer reveals species-rich and reservoir-specific communities. R. Soc. Open Sci. **3**: e160635.

Lindstrom, D. P. 1998. Reproduction, early development and larval transport dynamics of amphidromous Hawaiian gobioids Doctoral dissertation, University of Hawaii at Manoa.

Lindstrom, D.P., Blum, M.J., Walter, R.P., Gagne, R.B., and Gilliam, J.F. 2012. Molecular and morphological evidence of distinct evolutionary lineages of *Awaous guamensis* in Hawaiʻi and Guam. Copeia **2012**: 293-300.

Lintermans, M. 2000. Recolonization by the mountain galaxias *Galaxias olidus* of a montane stream after the eradication of rainbow trout *Oncorhynchus mykiss*. Mar. Freshwater. Res. **51**: 799-804.

Lisi, P.J., Childress, E.S., Gagne, R.B., Hain, E.F., Lamphere, B.A., Walter, R.P., Hogan, J.D., Gilliam, J.F., Blum, M.J., and McIntyre, P.B. 2018. Overcoming urban stream syndrome: Trophic flexibility confers resilience in a Hawaiian stream fish. Freshwater Biol. **63**: 492-502.

Lundberg, J. and Moberg, F. 2003. Mobile link organisms and ecosystem functioning: implications for ecosystem resilience and management. Ecosyst. **6:** 87-98.

Luton, C. D., Brasher, A.M.D, Durkin, C.D., and Little, P. 2005. Larval drift of amphidromous shrimp and gobies on the island of Oʻahu, Hawaiʻi. Micronesica **38**: 1-16.

Mack, R.N., Simberloff, D., Lonsdale, W.M., Evans, H., Clout, M., and Bazzaz, F.A. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. Ecol. Appl. **10**: 689-710.

MacKenzie, B.R., Miller, T.J., Cyr, S., and Leggett, W.C. 1994. Evidence for a dome-shaped relationship between turbulence and larval fish ingestion rates. Limnol. Oceanogr. **39:** 1790-1799.

Maciolek, J.A., and Timbol, A.S. 1980. Electroshocking in tropical insular streams. Prog. Fish-Cult. **42**: 57 58.

Mallin, M.A., Johnson, V.L., Ensign, S.H., and MacPherson, T.A. 2006. Factors contributing to hypoxia in rivers, lakes, and streams. Limnol. Oceanogr. **51:** 690-701.

Marchetti, M.P., and Moyle, P.B. 2001. Effects of flow regime on fish assemblages in a regulated California stream. Ecol. Appl. **11**: 530-53.

Marco-Rius F, Caballero P, Moran P, Garcia de Leaniz, C. 2013. Mixed-effects modelling of scale growth profiles predicts the occurrence of early and late fish migrants. PLoS ONE 8: e61744.

Marks, J.C., Haden, G.A., O'Neill, M. and Pace, C. 2010. Effects of flow restoration and exotic species removal on recovery of native fish: lessons from a dam decommissioning. Restor. Ecol. **18**: 934-943.

McCrimmon, H.R., and Bidgood, B. 1965. Abnormal vertebrae in the rainbow trout with particular reference to electrofishing. T. Am. Fish. Soc. **94**: 84-8.

McDowall, R.M. 2003a. Hawaiian biogeography and the islands' freshwater fish fauna. J. Biogeogr. **30**: 703- 10.

McDowall, R.M. 2003b. Shads and diadromy: implications for ecology, evolution, and biogeography. Biodiversity, Status, and Conservation of the World's Shads, 35.

McDowall, R. M. 2007a. On amphidromy, a distinct form of diadromy in aquatic organisms. Fish Fish. **81**: 1-13.

McDowall, R. M. 2007b. Hawaiian stream fishes: the role of amphidromy in history, ecology and conservation biology. Bishop Museum Bulletin in Cultural and Environmental Studies, **3**: 3-9.

McDowall, R.M. 2010. Why be amphidromous: expatrial dispersal and the place of source and sink population dynamics? Rev. Fish. Biol. Fisheries **20**: 87-100.

McEwan, A.J., and Crisp, P. 2019. Restoration of a New Zealand lagoon: Evaluation of two years of introduced fish control trials. Ecol. Rest. **37**: 90-100.

McGarigal, K. 2009. BIOSTATS R code compilation. Available from http://www.umass.edu/landeco/.

McGurk, M.D. 1986. Natural mortality of marine pelagic fish eggs and larvae: role of spatial patchiness. Mar. Ecol. Prog. Ser. **34:** 227-242.

McKnight, E., García-Berthou, E., Srean, P., and Rius, M. 2017. Global meta-analysis of native and nonindigenous trophic traits in aquatic ecosystems. Glob. Change Biol. **23**: 1861-1870.

McIntyre, P.B., Riedy Liermann, C., Childress, E., Hamann, E.J., Hogan, J.D., Januchowski-Hartley, S.R., Koning, A.A., Neeson, T.M., Oele, D.L., and Pracheil, B.M. 2016. Conservation of migratory fishes in freshwater ecosystems. Cambridge University Press: Cambridge, 324-360.

McMahon, R.F. 2002. Evolutionary and physiological adaptations of aquatic invasive animals: r selection versus resistance. Can. J. Fish. Aquat. Sci. **59**: 1235-1244.

McMichael, G A. 1993. Examination of electrofishing injury and short-term mortality in hatchery rainbow trout. N. Am. J. Fish. Manageme. **13**: 229-233.

McMichael, G.A., Fritts, A.L., and Pearsons T.N. 1998. Electrofishing injury to stream salmonids; injury assessment at the sample, reach, and stream scales. N. Am. J. Fish. Manage. **18**: 894-904.

Meffe, G.K. and Snelson, F.F. 1989. An ecological overview of Poeciliid fishes. In: Meffe, G.K. and Snelson F.F. (eds.), Ecology and Evolution of Livebearing Fishes (Poeciliidae). Prentice Hall, Englewood Cliffs, NJ, pp. 13-31.

Mesa, M.G., and Shreck, C.B. 1989. Electro-fishing mark recapture and depletion methodologies evoke behavioural and physiological changes in cutthroat trout. T. Am. Fish. Soc. **118**: 644-658.

Mills, L.S., and Allendorf, F.W. 1996. The one-migrant-per-generation rule in conservation and management. Conserv. Biol. **10**: 1509-1518.

Minckley, W.L., and Meffe, G.K. 1987. Differential selection by flooding in stream fish communities of the arid American Southwest. In: Matthews, W. J. and Heins D. C. (eds). Community and evolutionary ecology of North American stream fishes. pp 93-104. University of Oklahoma Press, Norman, Oklahoma, USA.

Mitton, C.J.A., and McDonald, D.G. 1994. Effects of electroshock, air exposure, and forced exercise on swim performance in rainbow trout (*Oncorhynchus mykiss*). Can. J. Fish. Aquat. Sci. **51**: 1799-1803.

Miya, M., Sato, Y., Fukunaga, T., Sado, T., Poulsen, J.Y., Sato, K., Minamoto, T., Yamamoto, S., Yamanaka, H., Araki, H., Kondoh, M., and Iwasaki, W. 2015. MiFish, a set of universal PCR primers for metabarcoding environmental DNA from fishes: detection of more than 23- subtropical marine species. Open Science **2**: e150088.

Molnar, J.L., Gamboa, R.L., Revenga, C., and Spalding, M.D. 2008. Assessing the global threat of invasive species to marine biodiversity. Front. Ecol. **6**: 485-492.

Mollenhauer, R., Mouser, J.B., and Brewer, S.K. 2017. Sampling the stream landscape: improving the applicability of an ecoregion-level capture probability model for stream fishes. Can. J. Fish. Aquat. Sci. **75**: 1614-1625.

Moody, K.N., Hunter, S.N., Childress, M.J., Blob, R.W., Schoenfuss, H.L., Blum, M.J., and Ptacek, M.B. 2015. Local adaptation despite high gene flow in the waterfall-climbing Hawaiian goby, Sicyopterus stimpsoni. Mol. Ecol. **24**: 545-563.

Moody, K.N., Gagne, R.B., Heim-Ballew, H., Alda, F., Hain, E.F., Lisi, P.J., Walter, R.P., Higashi, G.R., Hogan, J.D., McIntyre, P.B., Gilliam, J.F., and Blum, M.J. 2017. Invasion hotspots and ecological saturation of streams across the Hawaiian archipelago. Cybium **41**: 127-56.

Moody, K.N., Kawano, S.K., Bridges, W.C., Blob, R.W., Heiko, H.L., and Ptacek, M.B. 2017. Contrasting post-settlement selection results in many-to-one mapping of high performance phenotypes in the Hawaiian waterfall-climbing goby *Sicyopterus stimpsoni*. Evol. Eco. **31**: 489-516.

Moody, K.N., Wren, J.L.K., Kobayashi, D.R., Blum, M.J., Ptacek, M.B., Blob, R.W., Toonen, R.J., Schoenfuss, H.L., and Childress, M.J. 2019. Evidence of local adaptation in a waterfall-climbing Hawaiian goby fish derived from coupled biophysical modeling of larval dispersal and post-settlement selection. BMC Evol. Biol. **19**: 88.

Moore, J.W., and Schindler, D.E. 2008. Biotic disturbance and benthic community dynamics in salmon-bearing streams. J. Anim. Ecol. **77**: 275-284.

Moore, S.E., Kulp, M., Rosenlund, B., Brooks, J., and Propst, D. 2008. A field manual for the use of Antimycin A for restoration of native fish populations. National Resource Report NPS/NRPC/NRR-2008/033, U.S. National Park Service, Fort Collins, Colorado, USA. 149 p.

Morrison, S. 2016. Life history variation and diet preference in non-diadromous Otago galaxiids Doctoral dissertation, University of Otago.

Mosley, M. P. 2000. Regional differences in the effects of El Nino and La Nina on low flows and floods. Hydrolog. Sci. J. **45:** 249-267.

Mullner, S.A., Hubert, W.A., and Wesche, T.A. 1998. Snorkeling as an alternative to depletion electrofishing for estimating abundance and length-class frequencies of trout in small streams. N. Am. J. Fish. Manage. **18**: 947-953.

Murphy, B.R., and Willis, D.W. 1996. Fisheries Techniques, 2nd edn. American Fisheries Society. Bethesda, MD.

Neeson, T.M., Smith, S.D.P., Allan, J.D., and McIntyre, P.B. 2016. Prioritizing ecological restoration among sites in multi-stressor landscapes. Ecol. Appl. **26**: 1785-1796.

Nelson, S.G., and Eldredge, L.G. 1991. Distribution and status of introduced cichlid fishes of the genera *Oreochromis* and *Tilapia* in the islands of the South Pacific and Micronesia. Asian Fish. Sci. **4**: 11-22.

Nico, L.G., Jelks, H.L., and Tuten, T. 2009. Non-native suckermouth armored catfishes in Florida: description of nest borrows and burrow colonies with assessment of shoreline conditions. Engineer Research and Development Center, Vicksburg, MS, Environmental Lab.

Nico, L.G., and Walsh, S.J. 2011. Non-indigenous freshwater fishes on tropical Pacific islands: a review of eradication efforts. In: Island Invasives: Eradication and Management. Proc. of the Int. Conf. on Island Invasives (Veitch C.R., Clout M.N. & Towns D.R., eds), pp. 97-107. Switzerland: IUCN.

Nico, L.G., Englund, R.A., and Jelks, H.L. 2015. Evaluating the piscicide rotenone as an option for eradication of invasive Mozambique tilapia in a Hawaiian brackish-water wetland complex. Manag. Biol. Invasions **6**: 83-104.

O’Dowd, D.J., Green P.T., and Lake, P.S. 2003. Invasional ‘meltdown’ on an oceanic island. Ecol. Lett. **6**: 812-817.

Odum, H.T. 1956. Primary production in flowing waters. Limnol. Oceanogr. **1**: 102-117.

Olds, B.P., Jerde, C.L., Renshaw, M.A., Li, Y., Evans, N.T., Turner, C.R., Deiner, K., Mahon, A.R., Brueseke, M.A., Shirey, P.D., Pfrender, M.E., Lodge, D.M., and Lamberti, G.A. 2016. Estimating species richness using environmental DNA. Ecol. Evol. **12**: 4214-4226.

Oki, D.S. 2004. Trends in Streamflow Characteristics at Long-Term Gaging Stations, Hawaii. USGS Scientific Investigations Report 2004-5080, USGS, Denver, CO.

Oki, D.S., and Brasher, A.M.D. 2003. Environmental setting and implications for water quality and Aquatic Biota, Oahu, Hawaii. 98 p. U.S. Geological Survey, Water Resources Investigations, Report 03-4156, Honolulu, Hawaii.

Oksanen, J., Blachet, F.G., Kindt, R., Legendre, P., O'Hara, R.B., Simpson, G.L., Solymos, P., Stevens, M.H.H., and Wagner, H. 2016. vegan: Community Ecology Package. R package version 2.3-4.

Olsson, I.C., Greenburg, L.A., Bergman, E. and Wysujack, K. 2006. Environmentally induced migration: the importance of food. Ecol. Lett. **9**: 645-651.

Osenberg, C.W., Schmitt, R.J., Holbrook, S.J., Abusaba, K.E., and Flegal A.R. 1994. Detection of environmental impacts - natural variability, effect size, and power analysis. Ecol. Appl. **4**: 16-30.

Palmer, G., Hogan, J. D., Sterba-Boatwright, B. D., and Overath, R. D. 2016. Invasive lionfish *Pterois volitans* reduce the density but not the genetic diversity of a native reef fish. Mar. Ecol. Prog. Ser. **558**: 223-234.

Paris, J.R., Stevens, J.R., and Catchen, J.M., 2017. Lost in parameter space: a road map for stacks. Methods Ecol. Evol. **8**: 1360-1373.

Parrish, J.D., Maciolek, J.A., Timbol, A.S., Hathaway, C.B. Jr, Norton, S.E. 1978. Stream Channel Modification in Hawaii, Part D: Summary Report Columbia (MO): US Fish and Wildlife Service, National Stream Alteration Team FWS/OBS-78/19.

Pearse, D.E., Miller, M.R., Abadía-Cardoso, A., and Garza, J.C. 2014. Rapid parallel evolution of standing variation in a single, complex, genomic region is associated with life history in steelhead/rainbow trout. Proc. R. Soc. B **281**: 20140012.

Peterson, I., and Wroblewski, J.S. 1984. Mortality rate of fishes in the pelagic ecosystem. Can. J. Fish. Aquat. Sci. **41:** 1117-1120.

Pham, L., West, D., and Closs, G.P. 2013. Reintroduction of a native galaxiid (*Galaxias fasciatus*) following piscicide treatment in two streams: response and recovery of the fish population. Ecol. Freshw.Fish. **22**: 361-373.

Pickup, M., Field, D.L., Rowell, D.M., and Young, A.G. 2013. Source population characteristics affect heterosis following genetic rescue of fragmented plant populations. Proc. R Soc. B. **280**: 20122058

Pimm, S.L., Dollar, L., and Bass Jr, O.L. 2006. The genetic rescue of the Florida panther. Animal Con. **9:** 115-122.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., and R Core Team. 2019. *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3.1-140, https://CRAN.R-project.org/package=nlme.

Poff, L.N., and Ward, J. V. 1989. Implications of streamflow variability and predictability for lotic community structure: A regional analysis of streamflow patterns. Can. J. Aquat. Fish. Sci. **56**: 1805-1818.

Polhemus, D.A. 1993. Damsels in distress: A review of the conservation status of Hawaiian Megalagrion damselflies (Odonata:Coenagrionidae). Aquat. Conserv. **3**: 343-349.

Polhemus, D.A., and Asquith, A. 1996. Hawaiian Damselflies: A Field Identification Guide. 122 p. Honolulu: Bishop Museum Press.

Potapov, A.B., and Lewis, M.A. 2008. Allee effect and control of lake system invasion. B. Math. Biol. **70**: 1371.

Power, M.E., Stout, R.J., Cushing, C.E., Harper, P.P., Hauer, F.R., Matthews, W.J., Moyle, P.B., and Statzner, B. 1988. Biotic and abiotic controls in river and stream communities. J. North Am. Benthol. Soc. **7**: 456-479.

Prior, K.M., Adams, D.C., Klepzig, K.D., and Hulcr, J. 2018. When does invasive species removal lead to ecological recovery? Implications for management success. Biol. Inv. **202**: 267-283.

Pulido, F. 2011. Evolutionary genetics of partial migration—the threshold model of migration revis(it)ed. Oikos **120**: 1776-1783.

Puritz, J.B., Keever, C.C., Addison, J.A., Byrne, M., Hart, M.W., Grosberg, R.K., and Toonen, R.J. 2012. Extraordinarily rapid life-history divergence between *Cryptasterina* sea star species. Proc. Roy. Soc. B **279**: 3914-3922.

R Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

Resh, V.H., and De Szalay, F.A. 1995. Streams and rivers of Oceania. *In* River and stream ecosystems. *Edited by* C. E. Cushing, K. W. Cummins, and G. W. Minshall. Elsevier, Amsterdam. pp. 717-736.

Ricciardi, A., Hoopes, M.F., Marchetti, M.P., and Lockwood, J.L. 2013. Progress toward understanding the ecological impacts of nonnative species. Ecol. Monogr. **83**: 263-282.

Rickard, P.P., and Cox, P.A. 1986. Use of Derris as a fish poison in Guadalcanal, Solomon Islands. Econ. Bot., **40**: 479-484.

Rickman, S.J., Dulvy, N.K., Jennings, S. and Reynolds, J.D. 2000. Recruitment variation related to fecundity in marine fishes. Can. J. Fish. Aquat. Sci. **57**: 116-124.

Riginos, C., Crandall, E.D., Liggins, L., Bongaerts, P., and Treml, E.A. 2016. Navigating the currents of seascape genomics: how spatial analyses can augment population genomics studies. Curr. Zool. **62**: 581-601.

Rideout, R.M. and Morgan, M.J. 2010. Relationships between maternal body size, condition and potential fecundity of four north-west Atlantic demersal fishes. J. Fish Biol. **76**: 1379-1395.

Rochette, N.C., and Catchen, J. 2017. Deriving genotypes from RAD-seq short-read data using Stacks. Nature Protocols 12: 2640-2659.

Rodríguez, M.A. 2002. Restricted movement in stream fish: the paradigm is incomplete, not lost. Ecology. **83**: 1-13.

Roth, B., Moeller, D., and Slinde, E. 2003. Ability of electric field strength, frequency, and current duration to stun farmed Atlantic salmon and Pollock and relations to observed injuries using sinusoidal and square wave alternating current. N. Am. J. Aquacult **66**: 208-216.

Rothschild, B.J. 1986. Dynamics of marine fish populations.

Rothschild, B.J., and Osborn, T.R. 1988. Small-scale turbulence and plankton contact rates. J. Plankton Res. **10:** 465-474.

Rowe, D.K. 2007. Exotic fish introductions and the decline of water clarity in small North Island, New Zealand lakes: a multi-species problem. Hydrobiologia **583**: 345-358.

Russell, J.C., Innes, J.G., Brown, P.H., and Byrom, A.E. 2015. Predator-free New Zealand: conservation country. BioScience **65**: 520-525.

Rytwinski, T., Taylor, J.J., Donaldson, L.A., Britton, J.R., Browne, D.R., Gresswell, R.E., Lintermans, M., Prior, K.A., Pellat, M.G., Vis, C., and Cooke, S.J. 2019. The effectiveness of non-native fish removal techniques in freshwater ecosystems: a systematic review. Environ. Rev. **27**: 71-94.

Sabaj, M.H., Armbruster, J.W., and Page L.M. 1999. Spawning in *Ancistrus* (Siluriformes: Loricariidae) with comments on the evolution of snout tentacles as a novel reproductive strategy: larval mimicry. Ichthyol. Explor. Freshwaters **10**: 217-229.

Safeeq, M., and Fares, A. 2011. Hydrologic response of a Hawaiian watershed to future climate change scenarios. Hydrologic Processes. Published online in Wiley Online Library (Available at wileyonlinelibrary.com)

Sagouis, A., Cucherousset, J., Villeger, S., Santoul, F., and Bouletreau, S. 2015. Non-native species modify the isotopic structure of freshwater fish communities across the globe. Ecography **38:** 979-985.

Satterthwaite, W.H., Beakes, M.P., Collins, E.M., Swank, D.R., Merz, J.E., Titus, R.G., Sogard, S.M., and

Mangel, M. 2010. State-dependent life history models in a changing (and regulated) environment: steelhead in the California Central Valley. Evol. Appl. **3**: 221-243.

Sax, D.F., Gaines, S.D. 2008. Species invasions and extinction: The future of native biodiversity on islands. Proc. Natl. Acad. Sci. U.S.A USA **105**: 11490-11497.

Schoenfuss, H.L., Julius, M.L., and Blob, R.W. 2004. Colonization of a recent, volcanically formed freshwater habitat: an example of primary succession. Ichthyol. Explor. Fres. **15**: 83-90.

Schofield, P.J., and Nico, L.G. 2007. Toxicity of 5% Rotenone to nonindigenous Asian swamp eels. N. Am. J. Fish. Manage. **27**: 453-459.

Schrage, L.J., and Downing, J.A. 2004. Pathways of increased water clarity after fish removal from Ventura Marsh; a shallow, eutrophic wetland. Hydrobiologia **511**: 215-231.

Schreck, C.B., Whaley, R.A., Bass, M.L., Maughan, O.E., and Solazzi, M. 1976. Physiological responses of rainbow trout (*Salmo gairdneri*) to electroshock. Fish. Res. Bd. Can. **33**: 76-84.

Schwarz, C.J. 2015. Analysis of BACI experiments. In: Course Notes for Beginning and Intermediate Statistics. Available at http://www.stat.sfu.ca/~cschwarz/CourseNotes.

Seddon, P.J., Griffiths, C.J., Soorae, P.S., and Armstrong, D.P. 2014. Reversing defaunation: restoring species in a changing world. Science **345**: 406-412.

Seeley, M., Miller, N., and Walther, B. 2015. High resolution profiles of elements in Atlantic tarpon Megalops atlanticus scales obtained via cross-sectioning and laser ablation ICP-MS: a literature survey and novel approach for scale analyses. Environ. Biol. Fish. **98:** 2223-2238.

Shackleton, R.T., Shackleton, C.M., and Kull, C.A. 2019. The role of invasive alien species in shaping local livelihoods and human well-being: A review. J. Environ. Manage. **229**: 145-157.

Sharber, N.G., and Carothers S.W. 1988. Influence of electrofishing pulse shape on spinal injuries in adult rainbow trout. N. Am. J. Fish. Manage. **8**: 117-122.

Sharber, N.G., Carothers, S.W., Sharber, J.P., de Vos, J.C. Jr., and House, D.A. 1994. Reducing electrofishing-induced injury for rainbow trout. N. Am. J. Fish. Manage. **14**: 340-346.

Sheldon, S.P., and Creed, R.P. 1995. Use of a native insect as a biological control for an introduced weed. Ecol. Appl. **5**: 1122-1132.

Sherwood, A.R., and Coontz, K.M. 2009. Spatial and temporal changes in macroalgal diversity of a Hawaiian stream with the return of previously diverted water. Algological Studies **131**: 115-134.

Shmida, A., and Wilson, M.V. 1985. Biological determinants of species diversity. J. Biogeogr. **12**: 1-20.

Siefkes, M.J. 2017. Use of physiological knowledge to control the invasive sea lamprey (*Petromyzon marinus*) in the Laurentian Great Lakes. Conserv. Physiol. **5**: pcox031.

Sigsgaard, E.E., Carl, H., Møller, P.R., and Thomsen, P.F. 2015. Monitoring the near-extinct European weather loach in Denmark based on environmental DNA from water samples. Biol. Conserv. **183**: 46-52.

Simberloff, D. 2009. We can eliminate invasions or live with them. Successful management projects. Biol. Invasions **11**: 149-157.

Simberloff, D. 2011. How common are invasion-induced ecosystem impacts? Biol. Invasions. 13: 1255-1268.

Simberloff, D., Martin, J.-L., Genovesi, P., Maris, V., Wardle, D.A., Aronson, J., Courchamp, F., Galil, B., García-Berthou, E., Pascal, M., Pyšek, P., Sousa, R., Tabacchi, E., and Vilà, M. 2013. Impacts of biological invasions: what's what and the way forward. Trends Ecol. Evol. **28**: 58-66.

Sin, H., Beard, K.H. and Pitt, W.C. 2008. An invasive frog, Eleutherodactylus coqui, increases new leaf production and leaf litter decomposition rates through nutrient cycling in Hawaii. Biol. Inv. **10:** 335-345.

Smith, G.C., Covich, A.P., and Brasher, A.M.D. 2003. An ecological perspective on the biodiversity of tropical island streams. BioScience **53**: 1048-1051.

Smith, J. E. 2007. Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems. Ecol. Lett. **10:** 1135-1142.

Smith, R.J.F. and Smith, M.J. 1998. Rapid acquisition of directional preferences by migratory juveniles of two amphidromous Hawaiian gobies, *Awaous guamensis* and *Sicyopterus stimpsoni*. Environ. Biol. Fish. **53**: 275-282.

Smith, V. H., Tilman, G. D., and Nekola, J. C. 1999. Eutrophication: impacts of excess nutrient inputs on freshwater, marine, and terrestrial ecosystems. Environ. Poll. **100:** 179-196.

Snyder, D.E. 2003. Invited overview: conclusions from a review of electrofishing and its harmful effects on fish. Rev. Fish Biol. Fisher. **13**: 445-453.

Sodeland, M., Jorde, P E., Lien, S., Jentoft, S., Berg, P.R., Grove, H., Kent, M.P., Arnyasi, M., Olsen, E.M., and Knutsen, H. 2016. "Islands of Divergence" in the Atlantic cod genome represent polymorphic chromosomal rearrangements. Genome Biology and Evolution 8: 1012-1022.

Sorenson, P.W., and Hobson, K.A. 2005. Stable isotope analysis of amphidromous Hawaiian gobies suggests their larvae spend a substantial period of time in freshwater river plumes. Environ. Biol. Fish **74**: 31-42.

Spennemann, D.H.R. 2002. Traditional milkfish aquaculture in Nauru. Aquacul. Int. **10**: 551-562.

Spear, S.F., Groves, J.D., Williams L.A., and Waits, L.P. 2015. Using environmental DNA methods to improve detectability in a hellbender (*Cryptobranchus alleganiensis*) monitoring program. Biol. Conserv. **183**: 38-45.

Sundby, S., and Fossum, P. 1990. Feeding conditions of Arcto-Norwegian cod larvae compared with the Rothschild-Osborn theory on small-scale turbulence and plankton contact rates. J. Plankton Res. **12:** 1153-1162.

Tank, J.L., and Dodds, W.K. 2003. Nutrient limitation of epilithic and epixylic biofilms in ten North American streams. Freshw. Biol. **48**: 1031-1049.

Taylor, C.M., and Hastings, A. 2005. Allee effects in biological invasions. Ecol. Lett. **8**: 895-908.

Taylor, C.M., and Norris, D.R. 2007. Predicting conditions for migration: effects of density dependence and habitat quality. Biol. Lett. **3:** 280-284.

Thomsen, P.F., Møller, P.R., Sigsgaard, E.E., Knudsen, S.W., Jørgensen, O.A., and Willerslev, E. 2016. Environmental DNA from Seawater Samples Correlate with Trawl Catches of Subarctic, Deepwater Fishes. PLoS One **11**: e0165252.

Thompson, K.G., Bergersen, E.P., Nehring, R.B., and Bowden, D.C. 1997. Long-term effects of electrofishing on growth and body condition of brown trout and rainbow trout. N. Am. J. Fish. Manage. **17**: 154-159.

Thorsteinsson, V., Pálsson, O.K., Tómasson, G.G. Jónsdóttir, I.G., and Pampoulie, C. 2012. Consistency in the behavior types of the Atlantic cod: repeatability, timing of migration and geo-location. Mar. Ecol. Prog. Ser. **462**: 251-260.

Tobin, P.C., Berec L., and Liebhold, A.M. 2011. Exploiting Allee effects for managing biological invasions. Ecol. Lett. **14**: 615-624.

Toonen, R.J., Andrews, K.R., Baums, I.B., Bird, C.E., Concepcion, G.T., Daly-Engel, T.S., Eble, J.A., Faucci, A., Gaither, M.R., Iacchei, M. and Puritz, J.B. 2011. Defining boundaries for ecosystem-based management: a multispecies case study of marine connectivity across the Hawaiian Archipelago. J. Mar. Biol. **2011**.

Townsend, C.R. 2003. Individual, population, community, and ecosystem consequences of a fish invader in New Zealand streams. Conserv. Biol. **17**:38-47

Turner, C.R., Uy, K.L., and Everhart, R.K. 2015. Fish environmental DNA is more concentrated in aquatic sediments than surface water. Biol. Conserv. **183**: 93-102.

Valentini, A., Taberlet, P., Miaud, C., Civade, R., Herder, J., Thomsen, P.F., Bellemain, E., Besnare, A., Coissac, E., Boyer, F. and Gaboriaud, C. 2016. Next-generation monitoring of aquatic biodiversity using environmental DNA metabarcoding. Mol. Ecol. **25**: 929-942.

Vander Zanden, M.J., Hansen, G.J., Higgins, S.N. and Kornis, M.S., 2010. A pound of prevention, plus a pound of cure: early detection and eradication of invasive species in the Laurentian Great Lakes. J. Great Lakes Res. **36**: 199-205.

Vanni, M.J., Flecker, A.S., Hood, J.M., and Headworth, J.L. 2002. Stoichiometry of nutrient recycling by vertebrates in a tropical stream: linking species identity and ecosystem processes. Ecol. Lett. **5**: 285-293.

Van Kessel, N., Dorenbosch, M., Boer, M. D., Leuven, R. S. E. W., and Velde, G. V. D. 2011. Competition for shelter between four invasive gobiids and two native benthic fish species. Curr. Zoo. **576**: 844-851.

Victor, B.C. 1983. Recruitment and population dynamics of a coral reef fish. Science. **219**: 419-420.

Vincenzi, S., Satterthwaite, W.H. and Mangel, M. 2012. Spatial and temporal scale of density-dependent body growth and its implications for recruitment, population dynamics and management of stream-dwelling salmonid populations. Rev. Fish Biol. Fish. **22**: 813-825.

Vitousek, P.M. 1988. Diversity and biological invasions of oceanic islands. Biodiversity **20:** 181-189.

Vitousek, P.M., Aber, J.D., Howarth, R.W., Likens, G.E., Matson, P.A., Schindler, D.W., Schlesinger, W.H., and Tilman, D.G. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecol. Appl. **7:** 737-750.

Vitousek, P.M. 2004. Nutrient Cycling and Limitation: Hawaii as a Model System. Princeton University Press, Princeton.

Waits, E.R., Bagley, M.J., Blum, M.J., McCormick, F.H. and Lazorchak, J.M. 2008. Source–sink dynamics sustain central stonerollers (*Campostoma anomalum*) in a heavily urbanize catchment. Freshwater Biol. **53**: 2061-2075.

Waller, D.M. 2015. Genetic rescue: a safe or risky bet? Mol. Ecol. **24**: 2595-2597.

Walsh, C.J., Roy, A.H., Feminella, J.W., Cottingham, P.D., Groffman, P.M., and II, R.P.M. 2005. The Urban Stream Syndrome: current knowledge and the search for a cure. J. North Am. Benthol. Soc. **24**: 706-723.

Walsh, J.R., Carpenter, S.R., and Vander Zanden, M.J. 2016. Invasive species triggers a massive loss of ecosystem services through a trophic cascade. P. Nat. Acad. Sci. USA **15**: 4081-4085.

Walter, R.P., Hogan, J.D., Blum, M.J., Gagne, R.B., Hain, E.F., Gilliam, J.F., and McIntyre, P.B. 2012. Climate change and conservation of endemic amphidromous fishes in Hawaiian streams. Endangered Species Res. **16**: 261-272.

Wandrag, E.M., Dunham, A.E., Duncan, R.P. and Rogers, H.S. 2017. Seed dispersal increases local species richness and reduces spatial turnover of tropical tree seedlings. Proc. Natl. Acad. Sci. U.S.A **114**: 10689-10694.

Watanabe, K., Tominaga, K., Nakajima, J., Kakioka, R., and Tabata, R. 2017 Japanese Freshwater Fishes: Biogeography and Cryptic Diversity. In: Motokawa M., Kajihara H. (eds) Species Diversity of Animals in Japan. Diversity and Commonality in Animals. Springer, Tokyo.

Watanabe, S., Iida, M., Lord, C., Keith, P., and Tsukamoto, K. 2014. Tropical and temperate freshwater amphidromy: a comparison between life history characteristics of Sicydiinae, ayu, sculpins and galaxiids. Rev. Fish Biol. Fish. **24:** 1-14.

Weir, B.S., Carolina, N., and Hill, W.G. 2002. Estimating F-S Tatistics. Annals of Human Gen. **36**: 721-750.

Welker, M. T., Pierce, C. L., and Wahl, D. H. 1994. Growth and survival of larval fishes: roles of competition and zooplankton abundance. Trans. Am. Fish. Soc. **123:** 703-717.

Whaley, R.A., Maughan, O.E., and Wiley, P.H. 1978. Lethality of electroshock to two freshwater fishes. Prog. Fish-Cult. **40**: 161-163.

Whiteley, A.R., Fitzpatrick, S.W., Funk, W.C., and Tallmon, D.A. 2015. Genetic rescue to the rescue.

Trends Ecol. Evol. **30**: 42-49.

Whittier, R.B., and El-Kadi, A.I. 2014. Human heath and environmental risk ranking of on-site sewage disposal systems for the Hawiaan islands of Kauai, Molokai,Maui, and Hawaii. Honolulu, Hawaii School of Ocean and Earth Sciences, University of Hawaii at Manoa.

Wiles, G.J., Bart, J., Beck Jr, R.E. and Aguon, C.F. 2003. Impacts of the brown tree snake: patterns of decline and species persistence in Guam's avifauna. Conserv. Biol. **17**: 1350-1360.

Willi, Y., and Fischer, M. 2005. Genetic rescue in interconnected populations of small and large size of the self-incompatible *Ranunculus reptans*. Heredity **95**: 437-443.

Winder, M., Jassby, A.D. and Mac Nally, R. 2011. Synergies between climate anomalies and hydrological modifications facilitate estuarine biotic invasions. Ecol. Lett. **14**: 749-757.

Witter, M.S., and Carr, G.D. 1988. Adaptive radiation and genetic differentiation in the Hawaiian silversword alliance (Compositae: Madiinae). Evolution **42**: 1278-1287.

Woodward, G. U. Y., and Hildrew, A. G. 2002. Food web structure in riverine landscapes. Freshwat. Biol. **47:** 777-798.

Wu, T.H., Tsang, L.M., Chen, I.-S., and Chu, K.H. 2016. Multilocus approach reveals cryptic lineages of the goby *Rhinogobius duospilua* in Hong Kong streams: Role of paleodrainage systems shaping marked populations differentiation in a city. Mol. Phylogen. Evol. **104**: 112-122.

Yamamoto, M.N., and Tagawa, A.W. 2000. Hawaiʻi's Native and Exotic Freshwater Animals. Mutual Publishing, Honolulu, Hawaiʻi.

Zalewski, M. 1983. The influence of fish community structure on the efficiency of electrofishing. Fish. Manage. **14**: 177-186.

Zalewski, M., and Cowx, I.G. 1990. Factors affecting the efficiency of electric fishing. *In* Fishing with Electricity: Applications in Freshwater Fisheries Management. Edited by I.G. Cowx and P. Lamarque. Fishing News Books: Oxford, UK. pp. 89-111.

Zavaleta, E.S. 2002. It's often better to eradicate, but can we eradicate better. *In* Turning the tide: The eradication of invasive species. *Edited by* C.R. Veitch and M.N. Clout. Gland: IUCN.

Zavaleta, E.S., Hobbs, R.J., and Mooney H.A. 2001. Viewing invasive species removal in a whole-ecosystem context. Trends Ecol. Evol. **16**: 454-459.

9 Appendix A. Scientific and Technical Publications

Published.

Alda, F., Gagne, R., Walter, R., Hogan, J.D., Moody, K., Zink, F., McIntyre, P.B., Gilliam, J.F., Blum, M.J. 2016. Colonization and demographic expansion of freshwater fauna across the Hawaiian archipelago. Journal of Evolutionary Biology **29**: 2054-2069.

Gagne, R.B., Sprehn, C.G., Alda, F., McIntyre, P.B., Gilliam, J.F., Blum, M.J. 2018. Invasion of the Hawaiian Islands by a parasite of imperiled stream fishes. Ecography **41**: 528-539.

Gagne, R., Blum, M.J. 2016. Parasitism of a native Hawaiian stream fish by an introduced nematode increases with declining precipitation across a natural rainfall gradient. Ecology of Freshwater Fishes **25**: 476-486.

Gagne, R.B., Heins, D.C., McIntyre P.B., Gilliam, J.F., Blum, M.J. 2016. Mutual dilution of infection by an introduced parasite in native and non-native stream fishes across Hawaiʻi . Parasitology **143**: 1605-1614.

Hain, E.F., Lamphere, B.A., Blum, M.J., Nelson, S.A.C., McIntyre, P.B., Gilliam, J.F. 2016. Visual survey and mark-recapture population estimates of stream fish in Hawaiʻi. Transactions of the American Fisheries Society **145**: 878-887.

Hain, E.F., Moody, K.N., Lamphere, B.A., Blum, M.J., Nelson, S.A.C., McIntyre, P.B., Gilliam, J.F. 2019. The migratory gauntlet: watershed disturbance increases the cost of diadromy. Ecology of Freshwater Fishes **28**: 446-458.

Heim-Ballew, H. 2019. Identifying migration flexibility and the environmental factors that influence variation in recruitment success in partially migratory Hawaiian fishes. Doctoral dissertation. Texas A&M University-Corpus Christi.

Heim-Ballew, H., Moody, K.N., Blum, M.J., McIntyre, P.M., Hogan, J.D. 2020. Migration flexibility in native Hawaiian amphidromous fishes. Journal of Fish Biology **96**: 456-468.

Hogan, J.D., Kozdon, R., Blum, M.J., Gilliam, J.F., Valley, J.W., McIntyre, P.B. 2017. Reconstructing larval performance and habitat use in a diadromous fish using otolith increments, trace elements and oxygen isotopes. Journal of Fish Biology **90**: 1338-1355.

Lisi, P.J., Childress, E., Gagne, R.B., Hain, E.F., Lamphere, B.A., Walter, R.P., Hogan, J.D., Gilliam, J.F., Blum, M.J., McIntyre, P.B. 2018. Overcoming urban stream syndrome: trophic flexibility confers resilience in a Hawaiian stream fish. Freshwater Biology **63**: 492-502.

Moody, K.N., Hunter, S.N., Childress, M.J., Blob, R.W., Schoenfuss, H.L., Blum, M.J., Ptacek, M.B. 2015. Local adaptation despite high gene flow in the waterfall climbing Hawaiian goby, *Sicyopterus stimpsoni*. Molecular Ecology **24**: 545-563.

Moody, K., Gagne, R.B., Heim-Ballew, H., Alda, F., Hain, E.F., Lisi, P., Walter, R.P., Higashi, G., Hogan, J.D., McIntyre, P.B., Gilliam, J.F., Blum, M.J. 2017. Invasion hotspots and ecological saturation of streams across the Hawaiian Islands. Cybium **41**: 127-156.

Moody, K.N., Childress, M.J., Wren, J.L.K., Kobayashi, D.R., Blum, M.J., Blob, R.W., Ptacek, M.B. 2019. Evidence of local adaptation in a waterfall-climbing Hawaiian goby from coupled biophysical modeling of larval dispersal and post-settlement selection. *BMC Evolutionary Biology* **19**: 88.

Rosenthal, W. 2018. Genomic insights into adaptation of invasive guppies across the Hawaiian archipelago. Undergraduate senior thesis. University of Wisconsin-Madison.

Whitt, J. 2019. Targeted invasive species removal cascades to reduce non-target invasive species through apparent facilitation. Masters thesis. Texas A&M University-Corpus Christi.

In review or revision.

Javaid, A., Lisi, P.J., Hogan, J.D., Blum, M.J., McIntyre, P.B. Contrasting diets of stream-captured Cane Toads and American Bullfrogs in Hawaiʻi . Herpetological Review. *In revision*

Lisi, P.J., Hogan, J.D., Holt, G., Moody, K.N., Blum, M.J., McIntyre, P.M. Dual-habitat mediation of migration in a facultatively amphidromous fish. Ecology. *In review*

Moody, K.N., Scherer, A.E., O'Connor, D., Heim-Ballew, H., Lisi, P.J. Hogan, J.D., McIntyre, P.B., Blum, M.J**.** Removal and recolonization of invasive species in Hawaiian streams. Can. J. Fish. Aquat. Sci. *Revised and resubmitted*

Rosenthal, W., McIntyre, P.B., Lisi, P.J., Prather Jr., R.B., Moody, K.N., Blum, M.J., Hogan, J.D., Schoville, S.B, Invasion and rapid adaptation of guppies (*Poecilia reticulata*) across the Hawaiian archipelago. Evolutionary Applications. *In review*

In preparation.

Childress, E.S., Kraemer, B.M., Hain, E., Gilliam, J.F., Blum, M.J., McIntyre, P.B. Stream nutrients integrate signals of current land use and 4-million years of ecosystem development across the Hawaiian archipelago. *In preparation for Ecosystems*

Heim-Ballew, H., Gilliam, J.F., Bickford, N., Blum, M.J., McIntyre, P.M., Hogan, J.D Variable lunar timing of migration within and among native Hawaiian amphidromous fishes. *In preparation for Animal Behaviour*.

Heim-Ballew, H., Lisi, P.J., Gilliam, J.F., Blum, M.J., McIntyre, P.M., Hogan, J.D. Factors that influence differential survival of residents of a migratory Hawaiian freshwater fish across environmental and biological gradients. *In preparation for Journal of Fish Biology*

Hogan, J.D., O'Connor, D., Moody, K.N., Scherer, A., Heim-Ballew, H., Lisi, P.J., McIntyre, P.B., Blum, M.J**.** Demographic responses of an at-risk native amphidromous goby to invasive species removal from Hawaiian streams. *In preparation for Can. J. Fish. Aquat. Sci.*

Holt, G., Moody, K.N., Lisi, P.J., Heim-Ballew, H., Hogan, J.D., Blum, M.J., McIntyre, P.B. For the greater good: taking advantage of life history to improve invasive species management. *In preparation for Nature.*

Janssen, S.E., Lepak, R.F., McIntyre, P.B., Krabbenhoft, D.P., Buck, D.G., Lisi, P.J., Koning, A., Stevens, A., Roegner, A.F., Weyl. O.L.F., and Hurley, J.P. Tracing global mercury contamination of freshwater fisheries using stable isotopes. *In preparation for Environmental Science & Technology*

Lisi, P.J., Hamman, E., Hogan, J.D., Blum, M.J., McIntyre, P.M. Ecosystem responses to removing invasive macrofauna from Hawaiian streams. *In preparation for Ecosystems*

Moody, K.N., Gagne, R., Alda, F., Hogan, J.D., Walter, R., McIntyre, P.B., Gilliam, J.F., Blum, M.J. Genetic diversity and species diversity of stream fauna across the Hawaiian archipelago. *In preparation for Molecular Ecology*

Moody, K.N., Heim-Ballew, H., Lisi, P.J., Hogan, J.D., McIntyre, P.B., Blum, M.J. Genetic and demographic rescue moderated by life history variation in an at-risk native Hawaiian migratory stream fish. *In preparation for Molecular Ecology*

Roach K., Lisi P.J., Mooney, R.J., Hogan, J.D., Blum, M.J., McIntyre, P.B. Invasive species take-over of tropical island stream nutrient pools. *In preparation for Ecology Letters*

10 Appendix B. Ocean-watershed Model Simulations

GENERAL MODEL OF CLEAN SLATE RESTORATION

Total population comprised of interacting subpopulations.

Species populations are often subdivided into subpopulations, which contribute additively to the total population. At a given time, the total population *N* of a species comprised of multiple subpopulations, *s* = 1…n is the sum

$$N(t) = \sum\nolimits_{s=1}^{n} N_s(t)\,. \tag{S.1}$$

These subpopulations may be structured in many different ways, representing, for example, different life stages or populations in different locations.

Subpopulations often interact dynamically, affecting each other through time. While it is possible for some subset to not interact (e.g. populations in different locations with no dispersal connections), we are interested here in the case where those subpopulations have dynamical interactions. Specifically, the value of *N* in a focal subpopulation *p* is a function of the values in other subpopulations at previous times,

$$N_p(t) = f\left(N_{s=1\ldots n}(t - 0\ldots m)\right). \tag{S.2}$$

Expression (S.2) is quite general, accounting for any number of subpopulations, which may affect each other at different timescales. A more concrete example is a species comprised of two subpopulations, *a* and *b*, that affect each other in discrete time at the subsequent timestep,

$$\begin{aligned} N_a(t) &= f(N_b(t-1)) \\ N_b(t) &= f(N_a(t-1)) \end{aligned}\,. \tag{S.3}$$

For example, if N_b are larvae and N_a are adults, some number of the N_b at time $t - 1$ join the adult population N_a at *t*, and the N_a at $t - 1$ produce offspring contributing to N_b at *t*. Similarly, N_a and N_b might represent subpopulations in different locations connected by dispersal, so that some number of N_a from time $t - 1$ have moved to N_b at *t*, and vice versa.

Subpopulations as buffered storage.

Frequently, subpopulations are differently affected by environmental or density-dependent interactions. These differences can provide population buffering against detrimental conditions, as the less affected subpopulation provides the storage from which the overall population can recover (Chesson 1990). This outcome does not necessarily require that the subpopulations are differently affected on average by mortality, but only that particular events do not affect them equally. For example, seeds may be less affected by drought than growing plants, but more affected by ant predation. Populations in different watersheds may be equally sensitive to floods and experience the same overall flood frequency but can still buffer each other if the floods do not occur at the same time.

In the extreme case of total mortality within a focal subpopulation *p*, $N_p(t) = 0$. Buffering would then be provided by storage in other subpopulations if they did not also all go to 0. Because $N_p(t + m)$ is a function of those other subpopulations according to (S.2), it will increase from 0 as the dynamical relationships play out. In the particular case of two subpopulations in (S.3), if a mortality event sets $N_a(t) = 0$, N_b provides storage, with individuals joining N_a at $t + 1$ from N_b, allowing the overall population to recover dynamically.

A particular sort of this buffering is central to the action of the storage effect coexistence mechanism, where subpopulations are differently sensitive to competition, and so species can avoid the doubly-bad impact of poor environments and high competition (Chesson and Warner 1981; Chesson 1990, 2000). However, although the existence of storage is a prerequisite, the primary focus of the storage effect as a coexistence mechanism is around the covariance between environment and competition. It is density dependence in this covariance that provides the stabilization (Chesson 2000). Here, our focus is different; we are interested not in stabilized multispecies coexistence, but in eliminating invasive species, and particularly in how differences in the way subpopulations buffer mortality events might allow that to happen.

Native-invader differences in buffering allow clean-slate restoration.
The life history leading to different subpopulation structure can make invader removal possible if all interacting subpopulations of the invasive can be killed, while the native has a different subpopulation structure that allows at least one subpopulation to survive. Here we provide a general explanation of this conclusion.

Different species typically have different subpopulation structure, which may be qualitative or quantitative. For example, annual plants may all have a seed bank, but the fractionation of the population between growing plants and seeds may be different, and the dynamical relationships between those stages depends on specific life history traits. Species may qualitatively differ in the structure of their subpopulations, if, for example, one disperses between locations while another does not. It is these qualitative differences in storage between native and invasive that we focus on here in developing our restoration approach.

Define a native population $N(t)=\sum_{s=1}^{n} N_s(t)$ as split over a series of subpopulations $s = 1 \ldots n$ as in (S.1), and consider an invader population $L(t)=\sum_{q=1}^{u} L_q(t)$ split over subpopulations $q = 1 \ldots u$. An event that causes all of the $L_q = 0$ will cause the total population $L(t)$ to fall to 0, and there will not be a subpopulation from which to recover. If that same event spares at least one of the native subpopulations, i.e. at least one $N_s > 0$, $N(t)$ will also remain positive, and the other N_s can recover from the buffering provided by the remaining subpopulation. Note that this does not require the native population to be unaffected, it just must have at least one subpopulation that does not go wholly extinct. Two end-members bear quickly addressing. First, if the mortality event kills all of the natives across all subpopulations, they will also fail to recover. Thus, any restoration action built on this clean-slate approach must understand the life history driving the subpopulation partitioning to ensure the mortality event is designed in such a way to ensure retention of at least one subpopulation. Second, this general development includes the potential to kill all of the invaders and leave all subpopulations of the native unaffected. While ideal, such events are rarely possible, and our focus here is on the opportunities that arise from the subpopulation buffering, which allow temporarily sacrificing some part of the native population.

As a concrete example relevant to amphidromous Hawaiian gobies, consider two subpopulations, freshwater adults N_f, and at-sea larvae, N_s. For the moment, consider a single watershed, ignoring the metapopulation structure of watersheds in the archipelago, and so $N(t) = N_f(t) + N_s(t)$. The invasives likely have some set of life history buffers, but for the purposes of this example, we leave them generic, $L(t) = \sum_{q=1}^{u} L_q(t)$. The key here is that whatever buffering their life history provides occurs entirely in freshwater, and so all subpopulations are susceptible to mortality events occurring there. A restoration action imposing 100% mortality on all fish in the watershed would then cause all $L_q = 0$ and so $L(t) = 0$. In the case of the natives, the mortality event in freshwater sets $N_f(t) = 0$, but $N_s(t)$ will be unaffected, because those larvae are already at sea. Thus, $N(t) = 0 + N_s(t)$. At the next timestep, $t + 1$, the invasives have no subpopulation to recover from, and so $L(t + 1) = 0$. However, the native larvae return from sea, initiating a new adult population, and so $N_f(t + 1) > 0$ and the overall population $N(t + 1) > 0$. From this, the population will dynamically recover as those adults grow and produce additional larvae.

These results demonstrate that it is possible to extirpate both natives and invasives in a way that allows native, but not invasive, reestablishment. This conclusion depends on only on structuring the extirpation to take advantage of life history differences in a way that preserves a buffering subpopulation of natives.

Dynamical outcomes of native recovery.

What can we say about the extent of native recovery, beyond that the extirpated subpopulation will be re-established by the preserved subpopulation(s)?

The extent and speed of native recovery from a clean-slate intervention depend on population growth rates, the way subpopulations affect each other, and the removed impact of invasives. In many cases, we would presume that recovery would not be instantaneous, as the remaining subpopulation(s) are unlikely to provide enough inputs to the removed subpopulation to immediately replace all lost individuals. In general, the speed of recovery will depend on the nature of the dynamical relationships between subpopulations (S.2). For example, if the remaining subpopulation is spatial, consisting of adults that must migrate, contributions to the extirpated subpopulation are likely to be slow. However, if the remaining subpopulation is a large propagule bank, recovery may occur quickly.

Most conservatively, the extirpation of one subpopulation will not have detrimental long-run impacts on dynamics; natives will recover to at least what they would have been without being extirpated. The sole condition is that invasives were not benefitting natives either directly (e.g. facilitation, mutualism, serving as prey), or indirectly (e.g. trophic cascades, reducing competitors, or beneficial ecosystem engineering). While this may seem at first glance to be defining the problem in a way that ensures success, this definition is central to any restoration plan to remove invasives; invasive removal with the intention to restore natives should be predicated invasives negatively impacting natives.

We develop these conclusions from comparisons of the abundances and dynamics between two alternatives: a sequence of population sizes for a scenario in which restoration by extirpation is carried out, and a matching sequence where extirpation did not occur (the status quo). Assume that the native population is governed by some unspecified dynamical interactions with the environment, $E(t)$, invasive impact , $I(t)$, and the densities of its own, $N(t)$, and other species, $D(t)$, then

$$N(t+1) = f(N(t), D(t), E(t), I(t)). \tag{S.4}$$

We assume that increasing the invasive impact $I(t)$ is never positive for the natives (always 0 or detrimental), which may be due to directly reducing $N(t)$, reducing fitness (per-capita contribution of

$N(t)$ to $N(t+1)$) and thereby reducing $N(t+1)$, or both. Any sequence of years, $t = 1 \ldots n$, will have some corresponding sequence of N, D, E, and I. The status quo situation can be represented by the sequence $\{N^{+I}\}$ for the given sequence of D, E, and I, while the clean-slate scenario is the sequence $\{N^{-I}\}$, with the sequence of invasive impacts removed, i.e.

$$\begin{aligned}\{N_j^{+I}\} &= f(\{N_j^{+I}\},\{D^{+I}\},\{E\},\{I\}) \\ \{N_j^{-I}\} &= f(\{N_j^{-I}\},\{D^{-I}\},\{E\})\end{aligned} \quad . \tag{S.5}$$

Most simply, if the initial value of N is the same and the sequences experience the same set of environmental perturbations E, the sequence $\{N^{-I}\}$ will always be at least as large as the sequence $\{N^{+I}\}$. In the special case of $I = 0$, the sequences will be the same, and provided I is detrimental, $\{N^{-I}\}$ will be larger than $\{N^{+I}\}$. In the simplest case of $D^{+I} = D^{-I}$, only the direct invader impact is removed, increasing the sequence $\{N^{-I}\}$ by that amount over $\{N^{-I}\}$.

The actual behavior is unlikely to be quite so simple, however. The sequence D is likely to be affected by I and so will differ between the $+I$ and $-I$ cases. However, because of the condition that invaders do not have net benefits, including indirect benefits through other species, the joint combination of removing $\{I\}$ and switching to $\{D^{-I}\}$ will benefit natives and increase $\{ N^{-I} \}$ relative to $\{N^{+I}\}$. These differences between $\{D^{-I}\}$ and $\{D^{+I}\}$ could increase the benefit to natives, if, for example, competitors were feeding on the invasives. Conversely, the changes from $\{D^{+I}\}$ to $\{D^{-I}\}$ may represent an increase of competition, dampening the total benefit of invasive removal. However, provided the net direct and indirect impacts of invasives are negative, natives will receive a net benefit.
The extirpation action itself means we cannot strictly say that the $\{ N^{-I} \}$ is always larger than $\{N^{+I}\}$, unless we avoid all impacts on the natives while cleaning the slate. Thus, if the extirpation occurs at time 0, the starting values of N are not the same for the sequences $\{N^{-I}\}$ and $\{N^{+I}\}$. Alternatively, we could say that the starting value is some arbitrary point in the past, and so is identical, but the extirpation sequence $\{N^{-I}\}$ will have some period of depressed N relative to $\{N^{+I}\}$ during and after the clean slate intervention. The relevant question then is not whether the extirpation sequence is always larger than the status quo, but whether after some time it recovers to at least as large as it would have been without the extirpation. The answer is yes, particularly if we make the reasonable assumption that the population is regulated in some way, i.e. it will not grow exponentially to infinity.

For any transition from t to $t+1$, there is a per-capita population-level fitness, $\lambda(t) = {}^{N(t+1)}\!\big/_{N(t)}$. For any given time transition, the fitness in the situation where invaders have been removed, $\lambda^{-I}(t)$, will be at least as large as the fitness for the situation where they haven't, $\lambda^{+I}(t)$. Consider the case immediately following extirpation. The invader-free situation has only the remaining subpopulation, so $N^{-I}(t)$ is very likely lower than $N^{+I}(t)$. Even in the absence of density-dependent population regulation, the impact of invaders has been removed, and so whatever the growth rate would have been that year, it will be at least that large, plus however much the invaders were depressing growth. This larger growth rate just from eliminating invader impact I is itself sufficient for the extirpated population to eventually grow larger than the status-quo comparison, even in the case of exponential population growth. For the simplest illustration, imagine growth rates do not fluctuate, and that we define $r = \ln(\lambda)$, i.e. r is the multiplicative growth rate. Then at time t, $N(t) = N_0 e^{rt}$. For $N_0^{-I} < N_0^{+I}$ and $r^{-I} > r^{+I}$ (the extirpation lowers the native density, but yields a higher growth rate), we can find that the extirpated population catches the still-invaded population, $N^{-I}(t) = N^{+I}(t)$, at

$$t = \frac{\ln\left(N^{+I} \middle/ N^{-I} \right)}{r^{-I} - r^{+I}} \ . \tag{S.6}$$

More realistic, however, is the situation of a population regulated by density-dependence, and so not growing exponentially, which yields faster recovery to the pre-extirpation population size.

When we consider density-dependent regulation, it will take far less time for the extirpated population to reach the level it would have been without extirpation. The sequence $\{N^{+I}\}$ likely fluctuates, but because it is regulated, the mean growth rate over time, $\overline{r}$, will be 0. In the clean-slate sequence, not only is the invader's impact on growth removed, but the density dependent reduction in growth reduced or removed as well near the clean slate event, because $N^{-I}(t)$ is lowered relative to $N^{+I}(t)$. In this case, $r^{+I}(t) >> r^{-I}(t)$ for t near the clean slate event, and so the extirpated sequence will rapidly grow, while the status-quo sequence will not.
The time it takes for the difference between $\{N^{-I}\}$ and $\{N^{+I}\}$ to fully close will depend on the extent to which invaders depressed native populations. If $I = 0$, (invaders have no effect), the approach of $N^{-I}(t)$ to $N^{+I}(t)$ will decelerate, because they will be regulated to the same level. In contrast, if I had a large impact on native abundances, the $\{N^{-I}\}$ will be regulated at a higher level than $\{N^{+I}\}$, and so density dependence will decrease r^{-I} more slowly, and r^{-I} will be positive when $N^{-I} = N^{+I}$. Thus, $\{N^{-I}\}$ will continue growing past $\{N^{+I}\}$. The larger the invasive impact, the lower $\{N^{+I}\}$ will be relative to the equilibrial value of $\{N^{-I}\}$, and so the faster the clean slate sequence will recover back to $\{N^{+I}\}$ and beyond. The invasive impact is therefore inversely related to the speed at which the population recovers to the pre-extirpation status quo (seen in figure 3). In contrast, full restoration to the final post-extirpation level is independent of the level of invasion pre-extirpation. Because the post-clean-slate system simply behaves like an uninvaded system, the time it takes to reach full restoration depends on growth rates and the size of the new (possibly fluctuating) equilibrium.

The analysis thus far has been of situations where both possibilities (clean slate and status quo) have positive expected population sizes (the population is viable and self-sustaining pre-extirpation). We have shown that given these conditions, the native population will return to at least its pre-extirpation abundance post-extirpation. It is also possible, and maybe even likely in highly invaded situations, that the natives are on a trajectory towards extinction. In that situation, the sequence $\{N^{+I}\}$ trends towards 0, and the average r^{+I} is negative. All of the above arguments still hold in this case, with $r^{-I} \geq r^{+I}$ because the detrimental impact of invaders has been removed. If the removal of invaders is sufficient to increase r^{-I} above 0, then the natives will recover post-extirpation, while they would have gone extinct had nothing been done. The trickier case is when the population would go extinct with or without natives, i.e. $r^{-I} < 0$ as well. In that case, the per-capita decline will happen slower in the extirpated system, because r^{-I} is still larger than r^{+I}. However, it is possible in this case that the extirpation will hasten the date of extinction, simply by lowering the population size, from which it will continue to decrease. While it is still true that the ultimate outcome is a population at least as large as it would have been without the extirpation, a population of 0 is cold comfort. Thus, work should be done to ensure the population is either currently viable pre-extirpation (ensuring its viability post-extirpation), or that the population would be viable if not for invasive species.

The results from this general model show that if native populations are present and viable before the slate is cleaned, they will recover to at least that level post-extirpation. If invasives harm natives, the final restored native abundances will be higher than they would be otherwise. The more invaders impact native abundances, the more quickly the native population will bounce back to its pre-extirpation level. The time to achieve the full extent of native recovery though will be determined by growth rates and population regulation, rather than the level of invadedness pre-extirpation.

Exploratory analysis of more complex scenarios: spatial structure and spillover.

We have established that after some initial recovery period, and provided extinction does not occur, $\{N^{-I}\}$ will always be at least as large as the sequence $\{N^{+I}\}$. In general, this means more total propagules will be produced, and so continued numerical population growth will accelerate until slowed by density dependence. These additional propagules may also contribute to other connected populations. We have so far developed the general conclusions in the most conservative case of a single, isolated population, or as the total over a system of connected subpopulations. However, multiple scales may be at work. For example, the general case works for a single isolated watershed with freshwater adult and larval at-sea populations, or an isolated patch of annual plants with growing adults and a seed bank. However, if there are other watersheds, or other locations with annual plants, the increased propagule supply from the target area may boost the native population in those other areas, even if no intervention occurs there. For this to happen, the native populations in other locations must be recruitment limited; that is, they can hold more natives, but simply do not receive sufficient propagules, and so some proportion of the newly-produced propagules can establish.

Exploratory analysis of more complex scenarios: Allee effects.

The development above assumes no Allee effects, that is, that the extirpation does not push the native population below some level where growth rates are lowered because of small population sizes (Stephens et al. 1999). It is likely that most real populations have some level where Allee effects are a concern. If this level is crossed, then the statement that $r^{-I} \geq r^{+I}$ may not hold for low N^{-I}. Fundamentally, this is an empirical question, dependent on the life history of the species in question, and requires careful empirical work to establish that any extirpation would not reduce populations to a level that causes Allee effects.

Exploratory analysis of more complex scenarios: Temporal fluctuations.

As demonstrated above, for any particular t, $r^{-I}(t) \geq r^{+I}(t)$ and so for any sequences $\{r^{-I}\}$ and $\{r^{+I}\}$, $\overline{r}^{-I} \geq \overline{r}^{+I}$. However, even when the overall trend is for growth, it is possible that any particular time might yield $r^{-I}(t) < 0$, and so the population may decline from t to $t + 1$. This decline will be relatively smaller for the clean slate than the invaded condition, because the growth rates are always higher in the clean slate scenario. Thus, if the populations start at the same level, the clean slate scenario can weather any sequence of bad times that could be withstood by the status quo. However, if the series of bad times occurs immediately following the extirpation event, the clean slate population will start from a lower population size. In the absence of Allee effects, the extirpated population will still recover from any series of years that the status quo can recover from, provided they are not so bad that all subpopulations get driven to 0. As with Allee effects, the sensitivity to bad conditions following extirpation is an empirical question. However, it seems likely that given the higher growth rates in the extirpated population and existence of buffered life history, there is a narrow range of “bad times” that would drive the clean slate population extinct but spare the invaded status quo.

For example, if the population of adult plants is destroyed for many years in a row, its seedbank may eventually disappear, and so not provide the opportunity for recovery once the mortality pressure is removed, but this loss would occur for the status-quo population as well. In the case of amphidromous fish in an isolated stream, mortality occurring multiple years in a row will kill the adults that grow from returning larvae, and so no new larvae will be produced, and the buffering stage will disappear.

However, it is unlikely that this would not also drive the status quo to extinction. For this sequence to yield full extinction in the clean slate scenario but not the status quo would require the string of bad years to occur immediately post-extirpation, before the recolonizing post-larvae had become reproductive adults. Moreover, spatial buffering of the sort provided by a metapopulation with larval exchange is very robust. Further, even if one stream is completely driven to zero over a series of bad years, the larval subpopulation will persist and allow recovery provided all streams are not destroyed simultaneously over multiple years. Parallel arguments apply if there is a series of particularly poor years for larval survival at sea. The buffering provided by the adult population and the metapopulation structure will tend to allow recovery. These rescue processes will tend to occur more rapidly in the clean slate scenario, provided the bad larval years do not occur immediately following extirpation. Two points are relevant here: first, restoration based on native storage life history must understand the nature of the native storage phase. If the buffered stage is short lived, the extirpation events must occur on a shorter timescale. Second, if a sequence of mortality events occur naturally causing failure of the restoration (i.e. bad luck), this sequence would have also caused extinction in the absence of the restoration.

Table S1. Model notation

Term	Meaning
$N(t)$	Total native population size at time t
$N_s(t)$	Population size of native subpopulation s at time t
n	Index of the total number of native subpopulations
N_a, N_b	Example native subpopulations a and b
p	Index of a focal subpopulation for illustration
$L(t)$	Total invasive population size at time t
q	Index of the total number of invasive subpopulations
N_f, N_s	Example of native subpopulations in freshwater and at-sea, for illustration
$D(t)$	Response of N to species other than the invasive at time t
$E(t)$	Response of N to environmental conditions at time t
$I(t)$	Response of N to invasive species at time t (invader impact)
$\{x\}$	Time sequence of the variable inside the braces, generically x
Superscript $+I$	Indicates the value of the term applying in the case with invaders (status quo)
Superscript $-I$	Indiciates the value of the term applying in the clean slate scenario
$\lambda(t)$	Per-capita population-level fitness, $N(t + 1) / N(t)$
$r(t)$	Per-capita population growth rate, $\ln(\lambda(t))$
$\overline{r}$	Mean population growth rate over time

SIMULATION MODEL OF HAWAIIAN FISH POPULATIONS

Analytical basis.

The simulation model of fish populations in 51 Hawaiian streams considers each watershed x as a unit, containing $N_{j,x}(t)$ adults of species j at time t. The number of adults at the next timestep, $N_{j,x}(t + 1)$ is then the production of those adults, minus death, plus any post-larvae that successfully join the adult population at time t. There are many possibly ways to model the competition among incoming post-larvae. The nature of competition among post-larvae for a place in the adult population, among already present adults, is likely to be well-approximated by contest competition: the adults present are likely to keep their place, and the post-larvae either gain a place as an adult or die. Thus, we choose Beverton-Holt competition (Beverton and Holt 1957) for our examples, leading to the dynamical equation

$$N_{j,x}(t+1) = N_{j,x}(t)(1-\delta_{j,x}(t)) + \frac{\left(R_{j,x}(t) + A_{j,x}(t)\right)}{1+C} \ . \tag{S.7}$$

The adults present in watershed *x* at time *t*, $N_{j,x}(t)$, lay $F_{j,x}(t)$ eggs, representing average per capita fecundity. Further, some proportion $\delta_{j,x}(t)$ of the adults in watershed *x*, $N_{j,x}(t)$, die at time *t*. New individuals join the adult population according to the second term of the equation. The total post-larvae competing to join the adult population is the sum of post-larvae arriving from sea, $A_{j,x}(t)$, and potentially any resident post-larvae, $R_{j,x}(t)$, allowing for facultative amphidromy.

The denominator, 1 + *C*, specifies Beverton-Holt contest competition, that is, how many of the competing post-larvae in the numerator join the adult population at *t* + 1. We assume that this competition reflects competitive effects from both other incoming larvae and already-present adults,

$$C_{j,x}(t) = \kappa_x \sum\nolimits_k \alpha_{j,k} \left(N_{k,x}(t)(1-\delta_{k,x}(t)) + \left(R_{k,x}(t) + A_{k,x}(t) \right) \right) \ . \tag{S.8}$$

The total response to competition, $C_{j,x}(t)$ is modelled as proportional to the total number of individuals, both adults and arriving juveniles in the stream, modified by two sorts of competition coefficient, κ_x, which accounts for watershed properties and invadedness, and $\alpha_{j,k}$, which captures intra- and interspecific competition. We do not distinguish here between the effects of juveniles and adults, because the outcome, namely joining the adult population, occurs at the adult stage, and so we must consider the effect of each individual as an adult. Note that equation (S.7) means that adults do not experience competition. Once adults establish, they contribute to competition, but are not affected by it. While unlikely to be exactly true, the basic structure that adults are reproductive, while younger fish need to disperse and then successfully settle to become adults is captured by this simple model. More complex emergent properties related to stage structure will not be captured here but are unlikely to alter the general conclusions. The key is the net outcome: how many disperse from *x* to *y* and successfully settle there.

The term κ_x modifies the usual competition coefficient $\alpha_{j,k}$ to yield watershed-specific properties. In effect, it is used to set a phenomenological carrying capacity dependent on invadedness, watershed size, and watershed quality. If $\alpha_{j,k}$ is the competitive effect of species *k* on species *j* in an uninvaded watershed of the mean size and quality, then

$$\frac{1}{\kappa_x} = \left({L_x}/{\bar{L}} \right) \left({E_x}/{\bar{E}} \right) (1 - I_x) \, , \tag{S.9}$$

Where we define on the reciprocal scale to make the connection to watershed capacity more intuitive. Although $1/\kappa_x$ is not exactly the carrying capacity of the watershed, it is directly related to it, with larger, better quality watersheds having higher capacity. The first two terms are defined relative to the mean watershed, which is the point at which $\alpha_{j,k}$ is defined. The overall size of the watershed determines how much space there is to fill and is accounted for by ${L_x}/{\bar{L}}$, which is the size of watershed *x* relative to the mean watershed size. Similarly, watersheds differ in quality, with some able to support more fish per unit size than others. These quality differences are captured by ${E_x}/{\bar{E}}$, the relative quality of watershed *x* compared with the mean watershed. Finally, the presence of invasive species reduces the available space for natives. We do not attempt to model the exact form of this reduction, but instead capture its phenomenological effect as the fractional reduction, I_x, in the watershed capacity of x for natives. Thus, an uninvaded stream with invader reduction $I_x = 0$ has the full capacity available to natives, while a fully invaded stream has $I_x = 1$, and no capacity available for natives.

The resident post-larvae are the product of the local adult population, fecundity, the residency (stay-home) rate, and the in-stream larval survival rate, h,

$$R_{j,x}(t) = N_{j,x}(t)F_{j,x}(t)s_{j,x}(t)h_{j,x}(t)\,. \tag{S.10}$$

The residency (stay-home) rate s allows for facultative amphidromy. The amphidromous fraction of the larvae produced from watershed x is then $(1-s_{j,x}(t))$, with $s = 0$ being complete amphidromy.

The post-larvae arriving from sea, $A_{j,x}(t)$ is the number of larvae arriving at watershed x from all watersheds in the archipelago (including x), and so depends on larval production from all watersheds at $t-1$, and the connectivity patterns among watersheds. Thus, $A_{j,x}(t)$ is not a function, but is the x'th item in the vector

$$\mathbf{A}_j(t) = \mathbf{M}_j(t-1)\mathbf{D}_j(t-1) \tag{S.11}$$

Where the vector **M** contains the total migratory larvae leaving each watershed; the x'th item in **M** is $N_{j,x}(t-1)F_{j,x}(t-1)(1-s_{j,x}(t-1))$, with $F_{j,x}$ being the mean per capita fecundity of species j in watershed x. The product of this production vector **M** and the connectivity matrix **D** then determines the vector of arrivals at each site **A**. In our modelling, we use the HYCOM model as analyzed by Moody at al. (2019) to reflect larval dispersal chaacteristics to define **D**. At-sea dispersal mortality $a_j(t)$ is contained in the matrix **D**. The $A_{j,x}(t)$ of each watershed x is thus the outcome of larvae arriving from all watersheds in the archipelago.

Table S2. Model notation

Term	Meaning	Value used
$N_{j,x}(t)$	Density or abundance of species j at site x and time t	Model outcome
$\delta_{j,x}(t)$	Adult death rate	0.4241
$R_{j,x}(t)$	Resident post-larvae	Model outcome
$A_{j,x}(t)$	Post-larvae arriving from sea	Model outcome
$s_{j,x}(t)$	Larval residency fraction (1-amphidromy rate)	Typically 0, sometimes varied from 0 to 1
$F_{j,x}(t)$	Fecundity (per capita egg production)	179,119
$C_{j,x}(t)$	Competition (reduction in population fitness due to density-dependence)	Model outcome
$\alpha_{(j,k)}$	Per capita competitive effect of species *k* on *j* in average stream	$\alpha_{(j,j)} = 0.00287$
κ_x	Stream-specific carrying capacity adjustment	Varies, dependent on L_x and E_x relative to mean
L_x	Watershed size	From Hawai'i DAR
E_x	Watershed quality	From empirical density
I_x	Invadedness of watershed (reduction in native capacity caused by invasives)	From 0 to 1, estimated from empirical surveys
$\mathbf{M}_j(t)$	Vector of migratory larvae of species *j* leaving each site and going to sea	Model outcome
$\mathbf{D}_j(t)$	Connectivity matrix for species *j*	See (Moody et al. 2019)
$a_j(t)$	At-sea larval survival	0.1 to 10^{-5}, including 0.0991 (HYCOM lost at sea)
$h_{j,x}(t)$	In-stream larval survival	When used, same range as *a*

Simulation and implementation.

We use the general model established in section *Analytical basis* to develop a simulation model capturing important processes for amphidromous Hawaiian gobies. We do not claim to be able to parameterize the model well enough to be explicit about absolute numbers or outcomes for particular watersheds. Instead, we parameterize the model to capture the salient characteristics of the system and put reasonable bounds on values and distributions of values over the watersheds. Then, because each simulated watershed does not accurately represent a real watershed, we independently permute watershed size and quality and invadedness relative to position in the connectivity matrix. This approach yields a probability distribution of outcomes for any given test, dependent on the specific arrangement of watershed characteristics in the archipelago.

The connectivity, and therefore dynamical links between watersheds, means that the population dynamics of neither single streams or the archipelago can be calculated analytically. Instead, we simulate local interactions within watersheds according to equation (S.7), with larval inputs at each time step determined by the connectivity matrix and outputs from all streams.

Specific parameterization.

Because we are not trying to exactly represent the system, but instead capture the important characteristics of it, we typically run the simulations for a single species. Most of the terms have been estimated from *Awaous stamineus* data, although we typically do not include facultative amphidromy. Moreover, the model is structured to capture general properties of goby life history, and so while estimates of parameter values from *A. stamineus* may differ in magnitude from those of other species, the qualitative results will not, particularly given our focus on permuting stream conditions among watersheds. Although competition between gobies may be quite important for their community structure, the questions here are around how goby life history compares with that of invasives. The interactions between gobies within watersheds will still occur but are not the focus of this work. All of our results would be unchanged if we simply make the intraspecific competition coefficients $\alpha_{j,j}$ larger than the interspecific competition coefficients $\alpha_{j,k}$, that is, if we assume that the gobies can coexist within watersheds as they seem to have done historically (Kido 2008).

We run all simulations with 51 streams, representing those streams in the HYCOM ocean circulation model (Moody et al. 2019). These streams are spread across the five main Hawaiian islands (Hawai'i, Mau'i, Moloka'i, O'ahu, and Kaua'i). HYCOM provides the connectivity matrix for four years, although unless specified otherwise, the results presented here use the mean connectivity over those four years. There is a distinct overall pattern of flow in HYCOM from southeast (Hawai'i) to northwest (Kaua'i) (Moody et al. 2019). Larvae were assumed to settle in the first stream within 5 km of their position after a marine phase of at least 50 days (Moody et al. 2019). Any larva that drifted for >150 days was considered lost at sea, thereby approximating the duration of marine larval dispersal for *A. stamineus* (mean 118 days, maximum 248 days; (Hogan et al. 2014). Thus, the outcome of HYCOM contains mortality from those lost at sea, averaging 0.9009 over all watersheds, and ranging from 0.8674 to 0.9422.

We expect significant additional larval mortality at sea, due to predation, starvation, and other factors, and so we include the term $a_j(t)$ in the model to account for this. There is little data about larval survival rates at sea, and so we explore outcomes for a range of values. Most relevant to the results presented here, this larval survival rate can be used to determine the extent of recruitment limitation. If survival is high, $A_{j,x}(t)$ is high and streams tend not to be recruitment limited, while low survival rates introduce recruitment limitation. Here, we use recruitment limitation to refer to the situation where the abundance of natives in the watershed does not reach the stream capacity because there are insufficient incoming post-larvae, $A_{j,x}(t)$. Adjusting recruitment limitation in this way operates on the overall limitation over the archipelago. The extent of recruitment limitation varies on a stream-by-stream basis, depending on capacity and position in the connectivity matrix relative to production from other streams, i.e. the larval supply.

Recruitment limitation also depends on adult death rates; as adult death rates increase, more post-larvae need to arrive every year to fill those spaces. Rather than adjust two parameters to control recruitment variation, however, the simulations presented here fix adult death rates and only vary larval mortality. Adult death rates are set at 0.4241, estimated from growth rates and size distributions according to (Van Sickle 1977). Growth rates during the adult stage were estimated from otoliths (Hogan et al. 2014), with size distributions from mark-recapture surveys in 2009 and 2011 (Hain et al. 2019). This is necessarily a crude estimate of the death rate, and requires assuming a stationary size distribution, but the goal here is to obtain reasonable values, which is supported by comparison with empirical age structure. The estimated death rate yields an estimated mean lifespan of 2.36 years, very close to field estimates of age structures, which have mean 2.4 (Hogan et al. 2014).

Facultative amphidromy is known to occur in some but not all of the native Hawaiian gobies, and so we include a term for the larval residency rate, $s_{j,x}(t)$. Unless specified otherwise, this term is set at 0 for all modelling presented here, representing purely amphidromous species for simplicity. In these cases, there are no resident post-larvae, $R_{j,x}(t) = 0$. It is, however, important to explore whether our conclusions hold for different levels of amphidromy, and so we consider cases of $s_{j,x}(t) \neq 0$ in supplementary information.

Density estimates of both natives and invasives were available for 32 watersheds from snorkel surveys (Lisi et al. 2018; Hain et al. 2019) and mark recapture studies (Hain et al. 2019), and form the estimates of watershed quality, E_x. The values of E_x for the remaining 19 streams were imputed from a regression of fish densities on the PC1 of land use (as in Lisi et al. 2018), island, and their interaction ($R^2 = 0.49$). Watershed size, L_x, was calculated two different ways, either stream length or watershed area, both obtained from the Hawaiʻi Division of Aquatic Resources' Atlas. In both cases, the percent of the watershed classified as headwaters was removed, as this area is unlikely to be a significant contributor to the amount of goby habitat, particularly *A. stamineus*. The results obtained using length and area are not qualitatively different, and so results reported here use watershed area, as it better captures the three dimensional size of the streams. These values (E_x and L_x) were multiplied to obtain total watershed capacity. The value of the competition coefficient $\alpha_{j,j}$ was then set to a value yielding the abundance of fish (both native and invasive) in the average watershed. The stream-specific native coefficient, κ_x, includes the relative values of E_x and L_x, but also a term accounting for the reduction of capacity available to natives, I_x. This invasive impact was calculated as the fraction of total fish contributing to E_x that were invasives. Strictly speaking, this estimation assumes that natives and invasives are directly 1 for 1 replacement for simplification sake. While this assumption is unlikely to be true, the approach used here captures the critical process of a detrimental impact of invasives on natives across a reasonable range, and it is not necessary to estimate the exact impact.

Per-capita fecundity, $F_{j,x}(t)$, is estimated as 179,119, using the regression equation of fecundity on standard length from Ha and Kinzie (Ha and Kinzie 1996) and the size distributions from snorkel and mark-recapture surveys (Lisi et al. 2018; Hain et al. 2019). This value falls within previously estimated ranges based on body size for *A. stamineus* (Ha and Kinzie 1996). Much like the values of adult death, the value of *F* matters primarily in relation to larval mortality, as they jointly determine larval supply. Since we vary larval mortality to achieve different levels of recruitment limitation, the value of *F* need not be exactly right. It is worth noting, however, that other goby species are likely to have much lower fecundity (Lindstrom 1998), and so may be more recruitment limited than *A. stamineus.*

LITERATURE CITED IN APPENDIX B

Beverton, R.J.H., and Holt, S.J. 1957. On the dynamics of exploited fish populations. *In* Volume 19 of Fishery investigations (Great Britain, Ministry of Agriculture, Fisheries, and Food). London: H. M. Stationery Office.

Chesson, P. 1990. Geometry, heterogeneity and competition in variable environments. Philosophical Transactions: Biological Sciences **330**: 165-173.

Chesson, P. 2000. Mechanisms of maintenance of species diversity. Annual Review of Ecology And Systematics **31**: 343-366.

Chesson, P., and Warner, R. 1981. Environmental variability promotes coexistence in lottery competitive systems. American Naturalist **117**: 923-943.

Ha, P.Y., and Kinzie, R.A. 1996. Reproductive biology of *Awaous guamensis*, an amphidromous Hawaiian goby. Environmental Biology of Fishes **45**: 383–396.

Hain, E.F., Moody, K.N., Lamphere, B.A., Nelson, S.A.C., McIntyre, P.B., Gilliam, J.F., and Blum, M.J. 2019. Migratory gauntlets on oceanic islands: Watershed disturbance increases the cost of amphidromy. Ecology of Freshwater Fish **28**: 446-458.

Hogan, J.D., Blum, M.J., Gilliam, J.F., Bickford, N., and McIntyre, P.B. 2014. Consequences of alternative dispersal strategies in a putatively amphidromous fish. Ecology **95**: 2397-2408.

Kido, M.H. 2008. A persistent species assemblage structure along a Hawaiian stream from catchment-to-sea. Environ Biol Fish **82**: 223-235.

Lindstrom, D.P. 1998. Reproduction, early development and larval transport dynamics of amphidromous Hawaiʻi n gobioids. Doctoral dissertation, University of Hawaiʻi at Mānoa.

Lisi, P.J., Childress, E.S., Gagne, R.B., Hain, E.F., Lamphere, B.A., Walter, R.P., Hogan, J.D., Gilliam, J.F., Blum, M.J., and McIntyre, P.B. 2018. Overcoming urban stream syndrome: Trophic flexibility confers resilience in a Hawaiian stream fish. Freshwater Biology **63**: 492-502.

Moody, K.N., Wren, J.L.K., Kobayashi, D.R., Blum, M.J., Ptacek, M.B., Blob, R.W., Toonen, R.J., Schoenfuss, H.L., and Childress, M.J. 2019. Evidence of local adaptation in a waterfall-climbing Hawaiian goby fish derived from coupled biophysical modeling of larval dispersal and post-settlement selection. BMC Evolutionary Biology **19**: 88.

Stephens, P., Sutherland, W., and Freckleton, R.P. 1999. What is the Allee effect? Oikos **87**: 185-190.

Van Sickle, J. 1977. Mortality rates from size distributions: The application of a conservation law. Oecologia **27**: 311-318.

11 Appendix C. Fact Sheet and Data Sheets

STOP Stop! Under the Hawaiʻi Revised Statutes Section 118-23 and Hawaiʻi Administrative Rules Sections 13-75-6 and 187A-6, it is illegal to conducting electrofishing in the State of Hawaiʻi. Protocol approval and permitting from the State of Hawaiʻi Division of Aquatic Resources is required for electrofishing. Do not proceed unless the proper permits have been aquired. **STOP**

Aquatic Invasive Species Removal via Electrofishing in Hawaiian Streams

Day 1

- Collect TSS and water chemistry data to access pre-removal stream conditions.
- Conduct point-quadrat snorkel surveys to quantify pre-revomal biotic composition.

Day 2

Pre-electrofishing:

- Collect native species via hand-netting and place in stream side sequestration tanks labelled with collection location.
  - Place weighted, labelled flags at collection location.

*Remember to place *Eleotris sandwicensis* in tanks separate from the other native species.


Awaous stamineus


Sicyopterus stimpsoni


Stenogobius hawaiiensis


Macrobracium grandimanus


Eleotris sandwicensis

Electrofishing:

- Measure water temperature and conductivity of stream.
- Program electrofishing unit to the appropriate conditions following the chart below.
- Place AIS in stream-side collection buckets (separate from the native species).
- Place any native species captured during electrofishing in individual holding tanks, photo document injury (if occurred), and monitor for recovery.

	Pass 1	Pass 2[a]	Pass 3
Waveform	Direct current	Standard pulse	Standard pulse
Voltage (V)	100	175-200 (decrease with higher conductivity)	175-200 (decrease with higher conductivity)
Frequency (Hz)	N/A	60	60
Duty Cycle	N/A	12%	12%

[a] If a high number of native species are still present in the removal area, repeated pass 1 conditions.

*If native faunal injures occurs, reduce voltage immediately and change to direct current.

**If native faunal mortality occurs, discontinue electrofishing and contact DAR immediately.

Post-electrofishing:

- Return native species back to collection locations.
- Euthanize AIS with MS-222, store and freeze for further analysis.

Day 3

- Collect TSS and water chemistry data to access post-removal stream conditions.
- Conduct point-quadrat snorkel surveys to quantify post-revomal biotic composition.

Date/ time	Stream	Reach	Nutr. Rep.	air (°C)	water (°C)	DO mg/L	DO %Sat	Cond. µs/cm	TSS mL	TSS filter numbr	TSS filter mass	dry mass (mg)	ashed mass (mg)

Stream Code: ____________________________ **Reach:** _________________ **Visit #:** _______

Observer: ______________________ **Date:** ____________________ **Time:** ______________

Weather: _________________________ **Reach-wide notes:** ______________________________

Nat Fish: Ag, Es, Lc, Ss, Sh, Ks, Mc **Nat Inverts** Ab, Mg, Ng, Nv	**NN Fish:** An, At, Ca, Cf, Ga, Hc, He, Koi, Lm, Md, Pr, Psp, Poec, Til, Xh, **NN Inverts:** Cor, Ml, Mt, Nds, Pc
Sub: sed, sand, grav, cob, rub, bould, bed, conc, veg **Vis:** clear, turbid, muddy, stained, (<0.1, <0.5, <1, >1m), veg **Flow:** slow, med, fast **Type:** run, riff, casc, chute, eddy, pool, spool, ppool	

X/Y Dist		Plot#	Obs.	Species and Size (mm)		Sub	Vis	Flow	Type	Dep (m)
1	5									
1	3									
1	3									
4	8									
4	3									
2	4									
3	8									
3	7									
1	8									
1	5									
1	5									
1	6									
1	1									

2	8									
2	5									
3	7									
1	2									
1	2									
1	2									
1	4									
1	2									
2	7									
2	2									
2	2									
4	1									
1	7									
4	1									
3	1									
2	1									
4	3									
1	1									
1	5									

4	4									
2	5									
3	5									
4	8									

Date of Use	**Stream Name**	**Date of Make** (on bottle)	**Volume Made -** Concentration: (on bottle) 20g MS2222 + 50gNaHCO3 / literH20	**Use:** Knock-out / Kill (#)	**Starting Concentration:** Stock / Stream H20	**Ending Concentration:** Stock / Stream H20	**Notes:**
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			
				( )			

DATASHEETS: MARK AND RECAPTURE, GENETIC SAMPLING

Stream: ______________________________ **Date:** _________________________ **Visit/Cycle:** _________ /_________

Observers: ____________________________ **Reaches Fished:** ____________________________ **Time Start/End:** __________ /_________

Reach	# (Taken #, Smple #)	Status (Recap?)	Size (mm)	Weight (g)	Sex (m, f, f(g), u)	Sample # (Genetics)	Tagged?	Tag ID / Color code	Photo	Recovery Notes	Additional Notes	Time In/Out	Tagger
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	
							Yes / No					___ /___	