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Review of Dredging Elutriate Application Factors: Relevance to Acute-to-Chronic Protection, Contaminant, and Endpoint Specificity

Alan J. Kennedy, Guilherme R. Lotufo, and Jeffery A. Steevens

July 2015

Environmental Laboratory

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Review of Dredging Elutriate Application Factors: Relevance to Acute-to-Chronic Protection, Contaminant, and Endpoint Specificity

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Final report

Approved for public release; distribution is unlimited.

Prepared for U.S. Army Corps of Engineers Washington, DC 20314-1000

Abstract

The purpose of this technical report is to review the use of application factors (AF) in determining the limiting permissible concentration (LPC) for water column (elutriate) toxicity evaluations for dredged material placement operations. Application factors are used as multipliers; AFs are applied to median effect toxicity endpoints in an effort to determine "safe" contaminant levels in the open water environment. While the default AF in the Marine Protection Research and Sanctuaries Act is 0.01. Federal regulations allow use of justifiable alternatives. An underprotective, high AF would underestimate the toxic effects of contaminants and potentially impact the organisms at the open-water, dredged material placement site. However, an overly conservative, low (excessively restrictive) AF would needlessly impose volume restrictions and increase dredged material management needs and costs. Herein the authors identify cases where use of the default AF is too conservative or even inappropriate. Examples include situations in which it is applied to short-exposure-duration dredging placement operations (e.g., < 24 hours), where the exposure concentrations are decreasing steadily over time, and instances in which it is applied to nonpersistent contaminants and to inappropriate toxicological endpoints. Particularly, use of the default AF is overly conservative when high levels of nonpersistent chemicals such as ammonia are present in elutriate water, and when toxicity is assessed using the very sensitive embryo/larval development tests. Since the default AF was not originally intended to be applied to nonpersistent contaminants or larval development endpoints, the authors propose alternatives to the default AFs that are specific to ammonia toxicity and the development toxicity test endpoint.

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Preface

This technical report reviews the use and applicability of application factors in dredging evaluations. This research was performed by Alan J. Kennedy and Dr. Guilherme R. Lotufo, both of the Environmental Risk Assessment Branch (CEERD-EPR), Environmental Processes and Engineering Division (CEERD-EP); and Dr. Jeffery A. Steevens, Senior Scientist, U.S. Army Engineer Research and Development Center (ERDC)-Environmental Laboratory (EL), Vicksburg, Mississippi. This research was funded by the USACE South Atlantic Division and Jacksonville District and by the Dredging Operations Technical Support Program, which is managed by Cynthia Banks.

At the time this report was published, Buddy Goatcher was Branch Chief, CEERD-EPR; Warren P. Lorentz was Division Chief, CEERD-EP; Dr. Jack Davis was Deputy Director, ERDC-EL; and Dr. Beth Fleming was Director, ERDC-EL.

LTC John T. Tucker III was Acting Commander of ERDC and Dr. Jeffery P. Holland was Director of ERDC.

1 Background

Dredged material evaluations

The U.S. Army Corps of Engineers (USACE) maintains navigable waterways through dredging operations. Federal navigation channels must be dredged to maintain project depth for vessel passage. An economically desirable option for managing relatively clean dredged material (DM) is to place it in open water at a designated site. However, prior to open water placement, the sediment must be evaluated to determine whether contaminants are present and whether the levels of contaminants may be expected to induce an adverse impact on the ecosystem at the placement site.

Placement of DM is regulated under the Clean Water Act (CWA) or the Marine Protection Research and Sanctuaries Act (MPRSA) for inland/coastal waters and open ocean (seaward of the national baseline), respectively. Technical guidance is available for conducting dredging evaluations under MPRSA (USEPA/USACE 1991) and CWA (USEPA/ USACE 1998); generally, these guidance documents recommend use of existing information previously generated at the project site, analytical screening for contaminants, and biological (toxicity) testing to inform management decisions.

Since it is often uncertain from analytical screening of contaminant concentrations whether adverse biological effects are expected, toxicity testing is generally required, especially under MPRSA. Toxicological testing of DM consists of three types of bioassays: (1) sediment elutriate tests that estimate the potential acute (or short-term) biological effects of DM on water column organisms during the short period DM is suspended in the water column after placement; (2) whole sediment toxicity tests that determine the potential for biological effects of the placed DM to benthic organisms; and (3) whole sediment bioaccumulation tests that determine the potential for contaminant uptake from the placed DM into the tissue of benthic organisms, including estimated implications to higher trophic levels. The points of interest at the DM placement site that these biological tests are intended to simulate are illustrated in Figure 1A. Figure 1. Representations of dredged material placement at a designated open-water site. Panel A illustrates the areas of interest simulated by sediment elutriate tests, whole sediment toxicity tests, and sediment bioaccumulation tests. Panel B illustrates the mixing zone concept in the water column evaluation. Panel C illustrates laboratory preparation of the sediment elutriate that is used to estimate water column exposure potential. Removal of sediment particles from elutriate water prior to bioassay testing is contingent on the applicable regulations, and can be determined on a case-specific basis.







Water column toxicity evaluations

The focus of this technical report is the sediment elutriate toxicity tests used during the water column evaluation. The water column evaluation determines the potential for contaminants released from DM to the water column during placement to have adverse impacts on the ecosystem at the placement site. In practice, a limiting permissible concentration (LPC) is established either by comparison to water quality screening values (Water Quality Criteria or Standards) or through biological testing, when the results of water quality screening are incomplete (not all contaminants present in the sediment have water quality screening values) or uncertain (potential for synergism among contaminants). Compliance is determined by comparing the LPC with the concentrations of contaminants in the water column inside and outside the mixing zone (see Figure 1B) at the placement site, predicted by mathematical models that determine the effects of mixing the DM releases with the water (STFATE; see USEPA/USACE 1998). Contaminant concentrations in the DM plume must be lower than the LPC at all times after placement outside of the mixing zone or placement site. In addition, under the MPRSA, contaminant concentrations in the DM plume must be lower than the LPC within the placement site mixing zone, after allowing four hours mixing. The process for determining the LPC from the results of biological testing is further described below.

Elutriate toxicity tests are performed to estimate the potential for shortterm toxicity of DM to pelagic organisms during and following open-water placement. Since the time the DM is suspended in the water column is relatively short, as is the time to disperse the dissolved contaminant plume, elutriate toxicity tests employ USEPA-standardized acute toxicity test methods in order to expose invertebrate and vertebrate sentinel species for a relatively short duration (48 to 96 hours). In addition, compliance with the LPC must generally be achieved within four hours after placement. Elutriates are prepared in the laboratory, as illustrated in Figure 1C. Three or more concentrations (e.g., 100%, 50%, 10% and sometimes 1%) of the elutriate water are typically used in toxicity tests to allow the determination of the lethal median effect concentration (LC50) value for the survival endpoint or median effective concentration (EC50) value for zooplankton development endpoints. Approaches for testing and interpretation are specific to CWA or MPRSA regulations. In a CWA water column evaluation, it is recommended that at least two different test species be used, although the number can be more or less. Flexibility exists for interpreting water column toxicity tests and in selecting toxicity

endpoints and thresholds under the CWA, as previously discussed by Clarke et al. (2002). However, the MPRSA regulations explicitly require that elutriate toxicity tests be conducted representing three different types of organisms: a vertebrate (fish), an invertebrate/crustacean, and plankton. Also under MPRSA, the LPC is determined by applying an AF to an acutely toxic concentration. The remainder of this publication focuses on evaluations conducted under the MPRSA regulations.

Determining the limiting permissible concentration ("safe" concentration)

The median effect values (LC50, EC50) generated from the acute elutriate toxicity tests referenced above by definition adversely impact 50% of the test organisms and thus do not provide a protective level. The no-observed-effect concentration (NOEC) generated from the toxicity tests represents a level not expected to impart an acutely toxic response in a 48-hour or 96-hour exposure (Clarke et al. 2002). However, LC50 (or EC50) values multiplied by an AF have been historically employed to generate a protective concentration (i.e., the LPC) of the suspended phase dredged material (NAS 1972, USEPA / USACE 1991, 1998) that is not expected to impart a chronic toxicology response. In practice, DM placement operations typically yield exposure dosages less than the 48-hour or 96-hour exposure durations of standard acute toxicity bioassays. In DM management, the LPC is derived by multiplying the acute LC50/EC50 value by the AF, expressed as a percentage of the elutriate prepared for testing (i.e., the 100% treatment). The Code of Federal Regulations (CFR, Title 40 § 227.27) defines the LPC as the:

"...concentration of a constituent which, after allowance for initial mixing as provided in § 227.29, [that] does not exceed applicable marine water quality criteria; or, when there are no applicable marine water quality criteria, that concentration of waste or dredged material in the receiving water which, after allowance for initial mixing, as specified in § 227.29, will not exceed a toxicity threshold defined as 0.01 of a concentration shown to be acutely toxic to appropriate sensitive marine organisms in a bioassay carried out in accordance with approved EPA procedures."

--- 40 CFR 227.27(1)(2)

The LPC is compared to the predicted concentration of DM in the water column at the designated DM placement site, determined through the use of a numerical mixing model such as STFATE (USEPA / USACE 1991, 1998). If the LPC is greater than the modeled dredged material concentration, the material passes the water column toxicity evaluation. However, if the LPC is exceeded by the modeled dredged material concentration, the material must be managed differently (Fava et al. 1984), which may include volume restrictions.

Historic and current use of application factors

When the elutriate water prepared in the laboratory is toxic to test organisms, the selection of the AF clearly influences the LPC since the LPC is a product of the AF and the LC50/EC50 (i.e., the lower the AF the lower the LPC). Consequently, the selection of the AF influences the outcome of the water column toxicity evaluation and the need for alternative management or operational controls (Fava et al. 1984). The National Academy of Science (NAS) recommended a generic AF of 0.01 to provide continuous protection for a steady discharge/exposure (NAS 1972); this AF was further referenced (Mount et al., 1977, Fava et al., 1984) and adopted in the MPRSA (in 40CFR 227.29) and dredged material testing guidance (USEPA / USACE, 1991, 1998). The AF of 0.01 is similar to safety factors (e.g., 1/100) used in mammalian toxicity to determine acceptable daily food intake levels for applying animal test data to humans (21 CFR 170.22; US FDA 2000). An AF is a value used to mitigate uncertainty due to a lack of data and available science to conservatively estimate a concentration that is protective to organisms. The AF approach is similar to use of uncertainty factors (UFs) for incomplete datasets; uncertainty factors are typically in multiples of 10 to reduce toxicity endpoint concentrations down to lower levels, adding an element of conservatism to account for unknowns and interspecies variability in chemical sensitivity (NAS 2013). According to NAS (2013), "the accuracies of the UFs used are largely unknown, so quantitative characterization of the uncertainties associated with any given RfD (reference dose) is generally not possible." It is important to reinforce that 40CFR 227.29(3) clearly states that use of AFs other than the default 0.01 for specific contaminants of concern is acceptable when scientifically defensible.

"When there is reasonable scientific evidence on a specific waste material to justify the use of an application factor other than 0.01 as specified in paragraph (a)(2) of this section, such alternative application factor shall be used in calculating the LPC."

---- 40 CFR 227.27(a)(3)

An issue needing clarification is whether the AF factor approach, when applied to acute bioassay data, is intended to be protective of short-term acute exposure and its associated toxicity or long-term (chronic) exposure. According to 40CFR 227.29 (3b), the LPC "...will not cause unreasonable acute or chronic toxicity or other sublethal adverse effects based on bioassay results..." An improved technical understanding with regard to the level of protection provided by multiplying 0.01 X the LC50 (or EC50) is needed. While it is generally recognized that the exposure to DM is short term, as implied by use of acute toxicity tests in the water column evaluation, the use of the generic 0.01 AF provides a conservative level that is protective of longer term (chronic) exposure durations.

Importance of ammonia in determining elutriate toxicity results

Differentiating persistent from nonpersistent contaminants in water column evaluations is also important for determining exposure duration and protective concentrations. Nonpersistent contaminants (defined as chemicals with a half life less than 8 weeks; NAS 1972) dissipate more rapidly and may require less dilution to reduce toxicity. Ammonia is a common nonpersistent contaminant that often causes toxicity in elutriate tests. Elevated ammonia concentrations occur naturally in sediment and ammonia toxicity, and the condition is well documented in toxicity testing of field-collected materials (Ferretti et al. 2000, Postma et al. 2002). Unionized ammonia (NH₃) is more toxic relative to the ammonium ion (NH_4^+) (USEPA 1989, 1999a, 2009). The fraction of total ammonia (i.e., sum concentration of NH₄⁺ and NH₃) that is unionized ammonia (UIA) is contingent on the pH, temperature, and salinity of the test water (Figure 2). Generally, UIA increases with higher pH and temperature but it decreases with higher salinity. Depending on the site of interest, ammonia may be considered a confounding factor or a contaminant of concern (CoC). Classifying ammonia as a CoC may be contingent on how quickly it dissipates from exposure media and the type of exposure media. It is generally agreed that ammonia is not a CoC in benthic sediments (USEPA / USACE 1994, 1998). For instance, standard USEPA marine sediment toxicity guidance (USEPA 1994) recommends reducing sediment porewater concentrations of ammonia that exceed 30 to 60 mg/L prior to exposing test organisms. Also, there is recognition in NAS (1972) that it is appropriate to use larger AFs for nonpersistent contaminants, such as ammonia.

For persistent (half-life in water > 8 weeks) and non-persistent (half-life in water < 8 weeks) chemicals, application factors of 0.01 and 0.05 are recommended, respectively.

--- NAS 1972

For ammonia and certain other pollutants, levels below 0.1 of the lethal concentration do not seem to contribute to the lethal action of a mixture.

--- NAS 1972

Concentration of materials that are nonpersistent or have noncumulative effects should not exceed 0.1 of the 96-hour LC50 at any time or place after mixing with the receiving waters. The 24-hour average of the concentration of these materials should not exceed 0.05 of the LC50 after mixing.

--- NAS 1972

Figure 2. Chemical speciation of ammonia (a) and idealized species contribution to toxicity (b) from USEPA (1999a).



Development toxicity tests and use of application factors

MPRSA regulations (40 CFR 227.27 (c)) for ocean disposal require testing of zooplankton. To address this requirement, the most commonly employed test organisms are bivalve and echinoderm larvae, used in standard (48- to 72-hour) larval development toxicity tests (e.g., ASTM E1563, ASTM E724-98). While mussel and echinoderm larval are planktonic, one should consider whether either adult or larval forms of these organisms are present and relevant at an open ocean placement site. Nonpersistent contaminants, such as ammonia and sulfides, are well-known confounding factors in sediment porewater testing using these organisms and development methods (Losso et al. 2007). Therefore, it should be recognized that it will often not be possible to specifically determine the toxicity of persistent contaminants using development toxicity tests when the ammonia concentration in the elutriate is elevated. Development tests may not be the best method to select for USACE districts / USEPA regions that do not classify ammonia as a CoC or wish to characterize the toxicity of more persistent contaminants (e.g., metals) when elevated levels of ammonia are present. Further, there is expressed concern regarding the lack of technical relevance of applying the 0.01 AF to the embryonic development EC50 endpoint.

"Note that the 0.01 factor is intended for acute mortality data (e.g., relating acute to chronic toxicity) and not for more subtle effects such as abnormalities, growth or reproduction, including EC50 data (NAS 1972). However, in the absence of other alternatives, the 0.01 application factor should be applied to EC50 data although it is recognized that these results will be conservative and that derivation of this historic application factor was largely a matter of "best professional judgment" by the NAS (1972). Thus, site-specific review may be required in some cases to determine compliance."

--- From USEPA / USACE (1998)

Thus, there is logic and precedent for applying AFs other than the 0.01 default in cases where (1) the contaminant causing toxicity is identified and justification for a contaminant-specific AF can be presented; (2) the nonpersistent contaminant ammonia is the driver of toxicity; and (3) the EC50 development toxicity test endpoint is generated, especially when high ammonia concentrations are present in the elutriate water. Alternative AFs are used on a case-by-case basis at the discretion of USEPA regions and USACE districts and the rationale for their selection is not applied consistently. Unfortunately, the use of the default 0.01 AF in DM evaluations has been more expedient in practice due to the absence of clearly provided alternatives. Therefore, this technical report proposes alternatives accomplished through (1) a literature review on the use of AFs; (2) a literature review of relative species and life stage sensitivity; (3) data

compilation of larval development elutriate toxicity tests where ammonia likely caused toxicity; and (4) laboratory testing of adult and larval mussels exposed to ammonia (to be presented in separate publication).

2 Methods

Calculation of Application Factors and Acute-to-Chronic Ratios

To understand the technical relevance and implications of the 0.01 AF, a retroactive assessment was performed through literature review. While AFs are discussed in the literature (e.g., NAS 1972, Mount et al. 1977, Verma 1981; Giesy and Graney 1989), a more common term in the ecotoxicology literature is the acute-to-chronic ratio (ACR). The AF is effectively the inverse of the ACR (Mount 1977; Kenaga 1982; Giesy and Graney 1989, USEPA 1999b). The calculation of the ACR is discussed in the literature (Kenaga 1982, Raimondo et al. 2007, Hoff et al. 2010). Generally, the ACR is defined as the ratio between the LC50 generated from an acute exposure and no observable effects concentration (NOEC) generated from a chronic exposure. Thus, the ACR approach is intended to derive a concentration protective of chronic exposure using data generated from an acute exposure. The NOEC is a statistically derived value, defined as the highest concentration at which the test endpoint is not statistically significantly ($\alpha = 0.05$) reduced relative to the control (or reference) treatment. It should be noted that the NOEC value is one of the test concentrations that was arbitrarily selected for the experiment; thus, it is not interpolated or otherwise mathematically or empirically based and it is not a recommended regulatory value (Crane and Newman 2000, Chapman et al. 1996). The ACR can alternatively be determined by the ratio between the LC50 and the maximum allowable toxicant concentration (MATC), which is the geometric mean of the NOEC and lowest observable effect concentration (LOEC) generated from a chronic exposure. While the NOEC has flaws (Crane et al. 2000, Chapman et al. 1996), its use as the divisor in this case provides a more conservative ACR since the NOEC is, by definition, a lower concentration than the MATC; further, in practice the MATC may relate to a partial effect (Crane et al. 2000).

$$AF = \frac{1}{ACR}; \quad ACR = \frac{LC50}{NOEC \text{ (or MATC)}}$$

Elutriate toxicity database

A database was built using elutriate toxicity data previously generated in DM evaluations for the USACE South Atlantic Division (SAD). The data

included in the database were limited to larval development toxicity test results in which ammonia was most likely the cause of toxicity and high enough to cause an effect, as supported in the reports (determined by comparison to ammonium chloride toxicity and/or stripping of ammonia concentrations). Twelve DM evaluations involving testing of 58 elutriate samples were included. Dredging evaluation reports were provided by USACE districts at Jacksonville (5 dredging evaluations; 18 elutriates), Mobile (2 dredging evaluations; 12 elutriates) Savannah (1 dredging evaluation; 5 elutriates) and Wilmington (3 dredging evaluations; 23 elutriates). AFs for the development tests were determined from the dataset using the ratio of the EC50 and NOEC; the NOEC from the acute development test was used in the equation presented above due to the high sensitivity of these tests (supported by the discussion below). To provide more conservative values, the database was expressed as the 10th percentile of the calculated AFs, as performed by previous authors for the inverse of the AF; the 90th percentile of the ACR (Lange et al. 1998; Raimondo et al. 2007).

3 Results and Discussion

Application factors and acute to chronic ratios

By definition, ACRs / AFs are designed to convert acute lethality concentrations — specifically LC50s — to concentrations that will not cause a longterm (chronic) impact; this includes no significant impairment to sublethal endpoints such as growth and reproduction that are generated from longerterm chronic lifecycle testing. The generic 0.01 AF approach is problematic in that it is nonspecific. That is, AFs are variable across taxonomic groups and classes of chemicals (Mount et al. 1977, Verma 1981, Kenaga 1982, Giesy and Graney 1989, Raimondo et al. 2007). Application factors that are admittedly arbitrary were suggested by various organizations in the United States and Europe, and fall within the range of 0.00001 to 0.9, thus highlighting the great degree of uncertainty in the generic AF/ACR approach (Verma 1981, Kenaga 1982; Giesy and Graney 1989).

Some authors made efforts to determine generic ACRs that are acceptably protective of many species exposed to many chemicals by percentile ranks of the data distribution (Lange et al. 1998, Heger et al. 1995, Giesy and Graney 1989, Raimondo et al. 2007). According to Kenga (1982), ACRs of 99 (AF < 0.01) were chronically protective of 87% test species for all chemicals they considered; they also reported that 93% of organic chemicals have ACRs less than 125 (i.e., AF > 0.008) and 67% have ACRs less than 25 (i.e., AF > 0.04). Similarly, Heger et al. (1995) determined that ACRs derived from full life-cycle chronic toxicity testing of 40 (AF = 0.025) and 100 (AF = 0.01) are protective of 72% and 85% of fish and crustacean species, respectively. Lange et al. (1998) employed a large database of analytically confirmed toxicity testing generating ACR data and described a 90^{th} percentile ACR of 73 (AF = 0.014) for all chemicals. Raimondo et al. (2007) derived generalist ACRs across species and chemical classes, presenting median, 10th percentile and 90th percentile ACRs of 8.3 (AF = (0.12), 2.5 (AF = 0.40) and 79.5 (AF = 0.01), respectively. Note the similar 90th percentile ACRs presented by these studies, corresponding to an AF of 0.01.

While it is desirable to protect all species, the 90th percentile in toxicological studies may be skewed by extreme outliers in the literature (which is biased by low reporting of negative or low toxicity data results).

Some metals (e.g., Cd, Zn, Co) may have very high ACRs, ranging from 83 to 382 (AFs from 0.0026 to 0.012), while other metals (e.g., Ni, Cu, Fe, Pb, Se) have much lower ACR ranges of 2 to 48 (AFs from 0.021 to 0.5). Lange et al. (1998) concluded that ACRs of 15 - 25 (AF = 0.040 - 0.067) should be sufficient to be protective of chronic effects due to 90% of chemicals in risk assessments; these authors stated that while organo-metals / metals and pesticides have much higher ACRs (lower AF), such chemical classes should be considered on a case-by-case basis (when present at levels of concern), rather than being applied as a generic value. Kenaga (1982) also stated use of ACRs of 25 or above (AF = 0.04 or less) is reasonable for protecting against chronic toxicity. Thus, a higher general AF (e.g., 0.05) should be considered for dredging disposal operations where DM will only be in suspension for short periods of time (less than the 48- to 96-hour bioassay duration). Kenaga (1982) and Lange et al. (1998) provide tables with ACRs that are specific to organic chemicals and metals; this is a better approach than a generic ACR/AF approach when the contaminant(s) of concern is known. For example, Lange et al. (1998) defined chemicalclass-specific 90th percentile ACRs for pesticides, other organics, metals/organo-metals and other inorganics of 84 (AF = 0.012), 16 - 25(AF = 0.04 - 0.062), 192 (AF = 0.005) and 20 (AF = 0.05), respectively.

In all of the above cases, ACRs were determined specifically using a ratio of acute lethality data and chronic sublethal concentrations for arthropods and fish. The chronic testing endpoints have exposure durations of 7 days, 28 days, and longer. Thus, while the above literature review provides some support for the 0.01 default AF when comparing to conservative 90th percentile ACRs, it is not clear that such a conservative AF is appropriate in water column evaluations (elutriate toxicity testing) of suspended dredged material for which the exposure duration is so short (< 4 hours); compliance is determined with plume concentrations no more than 4 hours after placement.

Sensitivity of the embryonic life stage

In general, adverse effects following long-term exposures are observed at lower levels than short-term or acute exposures. Sublethal effects such as growth, reproduction, and development are expected to occur at lower levels of exposure than will measures of lethality. However, the bivalve and echinoderm development toxicity bioassays are exceptions. While these are short duration tests that are defined as acute exposures (ASTM E1563, ASTM E724-98), they involve the most sensitive life stage of the animal that is exposed during the highly susceptible embryonic development period. Fertilization and development toxicity tests are sensitive to noncontaminant factors such as total suspended solids and it is generally recommended that suspended or settled sediment particles be centrifuged from the test treatments prior to test initiation (Carr and Chapman 1995). Ammonia toxicity is also a major consideration in urchin and mussel embryo and larval testing. While the gametes used in fertilization tests are not overly sensitive to ammonia, the embryos used in development toxicity testing are very sensitive to ammonia (Carr et al. 2006, Losso et al. 2007); thus, ammonia must always be considered when interpreting these test results (Carr et al. 2006).

Overall, embryonic development tests using bivalves and echinoderms are suggested to be the most sensitive saltwater bioassays available (Rosen et al. 2005). This greater sensitivity of embryos is attributed to higher metabolism, greater surface area to volume ratios (for interaction with chemicals), lesser capacity for detoxification, and the inability of embryos to reduce exposure via valve closure, as observed in adult bivalves (Williams and Hall 1999, Connor 1972, Ringwood 1992, Calabrese et al. 1973, Pavicic et al. 1994, Millward et al. 2007, Howell et al. 1984). Larvae were previously reported to be 14 to 1000 times more sensitive than adults of the same species (Connor et al. 1972). Depending on the toxicant, mussel larvae were 2 to 80 times more sensitive than juveniles and adults in our literature review (Table 1). *Mytilus* development was the most sensitive test endpoint to copper in the USEPA saltwater criteria database, followed by the *Crassostrea* and *Arbacia* development tests; all were more sensitive than fish or invertebrate survivorship tests (Arnold et al. 2005). There are also differences in sensitivity between the life stages of mussels during the development test (Figure 3), with the trochophore larval stage typically being more sensitive than the D-stage (Millward et al. 2007). Mussels are in the embryo and trochophore stages during elutriate toxicity tests.

Use of application factors for the larval development toxicity test

While the bivalve and echinoderm development tests are considered acute tests (ASTM E1563, ASTM E724-98), they differ from typical acute survival tests in that they generate an embryonic development endpoint at the most sensitive life stage. Thus, as stated in USEPA / USACE 1998), the technical relevance of applying the 0.01 AF to the embryonic development endpoint is questionable since the development endpoint (EC50) is

fundamentally different than acute lethality test endpoints (LC50). Further, the use alternative AFs is allowable when justified (40 CFR 227.27(a)(3)), as discussed in the background section.

Table 1. Summary of available data from the peer-reviewed literature supporting the greater sensitivity of the
development toxicity tests relative to juveniles and adults of the same species.

Organism	Chemical	Larval 48 h EC50 (in mg/L)	Reference	Adult test duration (h)	Adult LC50 (mg/L)	Increase in adult tolerance	Reference
<i>Mytilus</i> spp.	Bayluscide	0.012	Millward et al. (2007)	96	0.226 - 0.500	19 to 42X	Millward et al. (2007)
<i>Mytilus</i> spp.	Cadmium	1.200	Martin et al. (1981)	96	0.960 - 1.550	1X	Amiard- Triquet et al. (1986); Nelson et al. (1988)
<i>Mytilus</i> spp.	Chlorine	0.5	Millward et al. (2007)	96	>7.5	>15X	Millward et al. (2007)
<i>Mytilus</i> spp.	Copper	0.041	Millward et al. (2007)	96	3.15	77X	Millward et al. (2007)
<i>Mytilus</i> spp.	Copper	0.006 - 0.008	Martin et al. (1981); Rosen et al. (2005)	336	0.146	18 to 24X	Weber et al. (1992)
<i>Mytilus</i> spp.	Copper	0.006 - 0.008	Martin et al. (1981); Rosen et al. (2005)	96	0.122 - 0.480	15 to 80X	Amiard- Triquet et al. (1986); Nelson et al. (1988)
Urchin	Copper	0.011	Bielmyer et al. (2005)	96	0.025	2X	Bielmyer et al. (2005)
<i>Mytilus</i> spp.	Mercury	0.006	Martin et al. (1981)	96	0.161	27X	Nelson et al. (1988)
<i>Mytilus</i> spp.	Silver	0.002	Martin et al.(1981)	96	0.011	6X	Nelson et al. (1988)
<i>Mytilus</i> spp.	Trinitrotoluene	0.75	Rosen et al. (2007)	96	19.5	26X	Rosen et al. (2007)
<i>Mytilus</i> spp.	Zinc	0.175	Martin et al. (1981)	96	5.000	29X	Amiard- Triquet et al. (1986)



Figure 3. Illustration of the various life stages the mussel *Mytilus galloprovincialis*. This figure was reproduced from Millward et al. (2007).

In most cases, exposure to the suspended particulate phase during DM placement at a disposal site is expected to be for an acute duration. However, even when water column (elutriate) evaluations should be protective of chronic (sublethal) exposure durations and population-level effects, the 0.01 AF is only relevant to survival-based LC50 values (48- to 96-hour survival tests) generated for invertebrates and fish, not the zooplankton development EC50 value. An EC50 value is more subjective than a LC50 value (dead vs. alive) and the Ocean Testing Manual (USEPA/USACE 1991) does not discuss EC50s in context with the generic 0.01 AF. The use of the 0.01 AF as a multiplier for development test EC50 values was more recently developed in evaluations conducted under the CWA (USEPA / USACE 1998) and MPRSA evaluations in the SAD (USEPA / USACE 2008); use of a development toxicity endpoint was not originally considered in MPRSA regulations or the Ocean Testing Manual (USEPA / USACE 1991). The literature cited above on ACR and AFs does not consider mollusks, echinoderms, or developmental endpoints. Multiple authors (Mount et al. 1977; Kenga 1982, Ahlers et al. 2006, Raimondo et al. 2007) have stated that the AF / ACR approach is undermined when comparing across species (i.e., mollusks and urchins versus fish and shrimp) and different modes of toxic action or different endpoints (e.g., embryonic development vs. lethality). Since development bioassays are more sensitive, an alternative approach or AF should be considered to ensure the EC50s generated from development tests are protective, but not overprotective of the species.

Relative species sensitivity to ammonia

Ammonia naturally occurs in sediment; thus, it is a common contaminant that causes test organism mortality in elutriate tests. Toxicity reference values for ammonia for four of the most commonly tested species in MPRSA DM evaluations are summarized in Figure 4, and specific values are presented in supplemental Table 2, Table 3, and Table 4. Sensitivity to ammonia was compared between fish (Cyprinondon variegatus, Menidia species) and shrimp (*Americamysis bahia*) used in the 96-hour lethality tests relative to the mussel larvae and urchin larvae used in the 48-hour development toxicity tests. Mussel and urchin larval development was substantially more sensitive than any 96-hour lethality endpoint. The median development test endpoint (48-hour EC50 values) was more than 17 times more sensitive to ammonia than the lethality tests (as 96-hour LC50 values). The literature review on ACRs/AFs above provides support that the 0.01 AF is most relevant for estimating a protective chronic value for persistent contaminants, such as some metals, when applied to the acute survival endpoint (LC50). Therefore, there is a strong argument that alternative AFs should be applied since the generic 0.01 AF was not intended, and thus is not directly relevant, for a development endpoint (EC50) or nonpersistent contaminants, such as ammonia. These data, in combination with knowledge that later mussel life stages are more tolerant than their larval development stages, suggest that the 0.01 AF is overprotective (overly restrictive LPCs) for organisms unlikely to reside at the disposal site when applied to a development EC50 value driven by the nonpersistent contaminant ammonia.

Chemical specific application factors, including ammonia

The above derivations of generic ACRs (applied as AFs) may be useful in cases where the chemicals potentially driving water column (elutriate) toxicity are persistent or unknown. However, in cases in which there are strong lines of evidence suggesting that specific chemicals or analytes are the likely cause of toxicity, greater effort should be made to determine and/or apply a more specific, technically defensible AF. Such lines of evidence include the presence of elevated ammonia concentrations (above known toxicity reference values for the test species of interest), low concentrations of other contaminants (e.g., metals and organics below water quality criteria), and removal of the toxicity when ammonia concentrations are reduced in a toxicity identification (and reduction) evaluation.

	Exposure	Temperature		Solipity	NOEC	LOEC	LC50		
Organism	(hours)	(°C)	рН	(ppt)	mg/	L (*union	ized)	Reference	
Mysidopsis bahia	48	20	8.2	25	*0.69		*1.03	Boardman et al. (2004)	
Mysidopsis bahia	96	20	8.2	25	*0.22		*0.76	Boardman et al. (2004)	
Mysidopsis bahia	96	25	6.9	11			51.08, *0.23	Miller et al. (1990)	
Mysidopsis bahia	96	25	7.1	31			98.9, *0.5	Miller et al. (1990)	
Mysidopsis bahia	96	24	8	11			24.36, *1.04	Miller et al. (1990)	
Mysidopsis bahia	96	26	7.9	30			36.6, *1.7	Miller et al. (1990)	
Mysidopsis bahia	96	24	9.1	11			6.16, *2.02	Miller et al. (1990)	
Mysidopsis bahia	96	25	9	30			8.92, *2.87	Miller et al. (1990)	
Holmesimysis costata	96	[not reported]			*0.757	*1.179	*0.839	Phillips et al. (2005)	

Table 2. Toxicity reference values in the available literature for the mysid shrimp (*Americamysis bahia*, formerly *Mysidopsis bahia*, and other species). Total ammonia levels are provided when available, and unionized ammonia values are indicated with asterisks.

Table 3. Toxicity reference values in the available literature for the *Menidia* fish species and a close surrogate. Total ammonia levels are provided when available, and unionized ammonia values are indicated with asterisks.

	Exposure	Tomporatura			NOEC	LOEC	LC50	
Organism	duration (hours)	(°C)	рН	Salinity (ppt)	mg/	/L (*union	ized)	Reference
Menidia menidia	48	20	8.2	30	*0.92		*1.08	Boardman et al. (2004)
Menidia beryllina	96	26	7.9	19			42.9, *1.94	Miller et al. (1990)
Menidia beryllina	96	25.5	7	29.5			193.7, *0.97	Miller et al. (1990)
Menidia beryllina	96	25.5	7	30			192.9, *0.93	Miller et al. (1990)
Menidia beryllina	96	18	8	30.5			38, *0.98	Miller et al. (1990)
Menidia beryllina	96	25.5	8	30.5			45.6, *1.77	Miller et al. (1990)
Menidia beryllina	96	32.5	8	30			27.1, *1.7	Miller et al. (1990)

	Exposure	Tomporaturo		0.11.11	NOEC	LOEC	LC50	
Organism	duration (hours)	(°C)	рН	(ppt)	mg/	/L (*union	ized)	Reference
Menidia beryllina	96	26	8.9	30			2.01, *0.75	Miller et al. (1990)
Menidia beryllina	96	25	7	11			338.4, *1.64	Miller et al. (1990)
Menidia beryllina	96	24	7.9	11			20.32, *0.88	Miller et al. (1990)
Menidia beryllina	96	24	9	11			3.55, *1.16	Miller et al. (1990)
Menidia menidia	48	20	8.2	14	20, *1.08		27.8, *1.50	Li (1997)
Menidia menidia	48	20	8.1	22	20, *0.90		24.93, *1.17	Li (1997)
Atherinops affinis	96	[not reported]			*<0.424	*0.587	*0.560	Phillips et al. (2005)

Table 4. Toxicity reference values in the available literature for the *Mytilus* mussel and surrogate bivalve species.Total ammonia levels are provided, where available, and unionized ammonia values are indicated with asterisks.BLD = bivalve larval development.

	Exposure	Temperature		O a line iter	NOEC	LOEC	LC50	EC50	
Organism	duration (hours)	(°C)	pН	(ppt)		mg/L (*u	nionized)		Reference
Mercenaria mercenaria	48	20	8.2	27	*38		*218		Boardman et al. (2004)
Mercenaria mercenaria	96	20	8.2	27	*9.6		*37.9		Boardman et al. (2004)
Ruditapes decussatus	3 (scope for growth)	20	7.9	35	*0.11				Sobal and Fernandes (2004)
Mytilus galloprovincialis	48 (BLD)	[measured but not reported]						*0.044	McDonald (2005)
Mytilus galloprovincialis	48 (BLD)	[not reported]			*0.090	*0.152		*0.120	Phillips et al. (2005)
Mytilus galloprovincialis	48 (BLD)	[not reported]			*0.097	*0.182		*0.231	Tang et al. (2007), cited in Phillips et al. (2005)

Figure 4. Relative Species Sensitivity to ammonia. Box plots represent the 25th and 75th percentiles of the data, bars represent the 10th and 90th percentiles of the data distribution, and solid dots represent outliers. The mean and median of the data distribution are represented by dashed and solid lines, respectively. Data were summarized from USEPA (1989), Miller et al. (1990), Greenstein et al. (1995), Basuyaux and Mathieu (1999), Boardman et al. (2004), Carr et al. (1996), Carr et al. (2006) and the dredging evaluations cited in the methods.



As stated in the introduction, it is generally recognized in guidance and peer-reviewed publications that less restrictive AFs (0.05 to 0.1) can be applied to nonpersistent chemicals (NAS 1972; Fava et al. 1984; USEPA/USACE 1991, 1998). The 0.05 AF for nonpersistent chemicals presented in NAS (1972) is lower (i.e., more restrictive) than various AFs derived from ACRs, specifically for ammonia. There is literature support for a specific AF of 0.11 for ammonia, derived using the freshwater fish *Pimephales promelas* (Thurston et al. 1983, 1986). Diamond et al. (1993) presented a mean ACR of 7.23 (AF = 0.14) for nine freshwater species exposed to ammonia. Values for the marine organisms used in elutriate tests also suggest higher AFs for ammonia are warranted. Miller et al. (1990) reported ACRs for *A. bahia* and *M. beryllina* of 7.2 (AF = 0.14) and 21.3 (AF = 0.05), respectively. Therefore, since these AF are specific to test species exposed to ammonia, they should be considered for use in

determining the LPC in future dredging evaluations where the toxicity driver is ammonia. Some alternative AFs for consideration are provided in Table 5. Use of these higher AFs is expected to be protective of long-term chronic exposure, and thus overprotective of the acute exposure typical of DM placement operations.

Table 5. Nonexhaustive list of alternative application factors that provide chronic protection when applied to acute lethal median concentration (LC50) values. The 10th percentile values are expected to be overprotective in most cases (Lange et al. 1998). Asterisks indicate values that have previously been approved for use in specific dredging evaluation projects. The Engineer Research and Development Center (ERDC) is developing a more comprehensive, sortable database containing chemical and species-specific application factors available <u>e</u>}. а

Chemical	Type of AF	Organism (Genus)	AF (1 / ACR)	Source
Persistent contaminants	MPRSA default	-	0.01*	NAS 1972
Metals, organometals, pesticides	10 th percentile	-	0.01	Lange et al. (1998) Raimondo et al. (2007)
Aromatic hydrocarbons	10 th percentile		0.05	Lange et al. (1998)
Nonpersistent contaminants (ammonia)	24 h average at any time (after mixing)		0.05* 0.1	NAS 1972
Ammonia	Mean		0.14	Diamond et al. (1993)
Ammonia	Species-specific	Pimephales Americamysis Menidia	0.11 0.14 0.05	Thurston et al. (1983, 1986) Miller et al. (1990) Miller et al. (1990)

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Specific AF to larval development tests and ammonia

Data from development toxicity tests were extracted and summarized from USACE SAD evaluation reports to generate a database. Only elutriates with toxic concentrations of ammonia were included based on UIA concentrations exceeding the EC50 for ammonium chloride. It was further confirmed that ammonia was the primary driver of the toxicity for these elutriate tests by regressing normal development with unionized ammonia concentrations; the result was a strong fit (Figure 5), providing an additional line of evidence that UIA was responsible for the toxicity.



Figure 5. Relationship between normal development and ionized ammonia concentration in USACE elutriate tests.

Acute-to-chronic comparisons typically employ acute bioassays using juvenile test organisms and compare the resulting acute lethality value to sublethal endpoints derived from longer, chronic exposures. Since the larval development toxicity tests use the most sensitive developmental life stage of bivalves and echinoderms, the NOEC value derived from these tests is expected to be protective of longer term effects to older, subsequent life stages. Thus, acute NOEC/EC50 ratios were calculated for each individual elutriate toxicity test (Table 6) to determine Development Test Factors (DTFs) for ammonia. In addition, the average DTFs were calculated for each dredging evaluation, each USACE district, and overall for each test species across USACE districts. More elutriate toxicity data were available for mussels relative to urchins in the particular reports provided by USACE districts. The mean DTFs for the mussel test ranged from 0.39 to 0.76, while mean DTFs for the urchin test ranged from 0.23 to 0.45. In order to provide more conservative, easier-to-apply factors, 10th percentile DTFs were calculated individually for mussels and urchins, and overall for both species. This approach is comparable to the 90th percentile ACR approach previously applied by others (e.g., Lange et al. 1998). The 10th percentile DTF for mussels and urchins was 0.37 and 0.19, respectively. The overall 10th percentile DTF for both species was 0.22. Thus, a generic DTF of 0.2 may be applied to development toxicity tests when the predominant cause of toxicity is ammonia.

Table 6. Summary of acute-to-chronic ratios (ACRs) and development test factors (DTFs) for ammonia. Means
(1 standard deviation), minimum and maximum values are summarized. More conservative 10 th percentile
DTFs are provided, with the number of values (n) used to calculate those indicated in parentheses.

Description	Number of Elutriates	DTF Mean	DTF StDev	DTF (minimum)	DTF (maximum)	DTF (10 th percentile)
						0.37
Mussel (overall mean)	33	0.56	0.17	0.12	0.79	(n = 33)
SAJ (overall mean)	10	0.48	0.19	0.12	0.73	
Fort Pierce	2	0.68	0.04	0.65	0.71	
Mayport Event 1	4	0.40	0.22	0.12	0.64	
Mayport Event 2	3	0.39	0.01	0.37	0.39	
Port Everglades	1	0.73		0.73	0.73	
SAW (overall mean)	23	0.59	0.15	0.33	0.79	
Charleston Harbor	18	0.57	0.15	0.33	0.79	
Morehead City	1	0.76		0.76	0.76	
Wilmington Harbor	4	0.64	0.13	0.45	0.73	
						0.19
Urchin (overall mean)	25	0.33	0.11	0.07	0.48	(n = 26)
SAJ (overall mean)	8	0.33	0.14	0.07	0.45	
Port Manatee	4	0.43	0.03	0.40	0.45	
Canaveral Harbor	4	0.23	0.13	0.07	0.33	
SAM (overall mean)	12	0.27	0.06	0.18	0.34	
Gulfport Harbor	11	0.27	0.05	0.18	0.34	
Mobile Bay	1	0.33		0.33	0.33	
SAS (overall mean)	5	0.45	0.03	0.41	0.48	
Brunswick	5	0.45	0.03	0.41	0.48	
Grand mean	58	0.46	0.18	0.07	0.79	0.22 (n = 58)

Figure 6 compares the ammonia-specific DTFs from development test database to the default survival test DTFs for persistent and nonpersistent contaminants. Further, ERDC is conducting laboratory studies comparing the 48-hour development test sensitivity to longer-term juvenile mussel exposure to directly derive a DTF; preliminary results also indicate a DTF of 0.2 applied to the 48-hour EC50 is protective.

Figure 6. Comparison between the default application factors (AF) for persistent (0.01) and nonpersistent chemicals, proposed factors for development tests in which ammonia is the toxicity driver. Note that the proposed factor is the 10th percentile of the data set and thus lower (more protective). Data were extracted from USACE-South Atlantic Division dredging evaluation reports. Horizontal solid and dashed lines represent the median and mean, respectively. The upper and lower margins of the boxes represent 90th and 25th data percentiles, respectively. Upper and lower bars represent 90th and 10th data percentiles, respectively. Points represent values outside 90% of the data range.



4 **Conclusions and Recommendations**

Based on the cited literature, applying an AF of 0.01 to acute elutriate lethality data results in levels that are protective to overprotective of chronic impacts of persistent contaminants, such as metals. The practice of determining the LPC by applying 0.01 multiplied by an acute toxic concentration is particularly over protective when the DM is only suspended in the water column for an acute exposure duration (< 48 hours). Further, the retroactive assessment of the 0.01 AF indicated that it is overly conservative for some chemicals (e.g., many organics, some metals), and particularly ammonia. Since MPRSA regulations allow use of scientifically justified alternative AFs, the most technically relevant and defensible approach would be to generate such values for the relevant elutriate test organisms for each potential contaminant of concern (or find such values in the literature when available). However, in absence of such data and in cases where the specific chemical(s) driving the toxicity in elutriate bioassays cannot be identified, a generic AF approach may be expedient. However, it must be understood that applying the 0.01 AF to an acute LC50 or EC50 generated from elutriate toxicity bioassays is overprotective of acute impacts. If the desire is to protect against only acute impacts of the suspended phase DM due to the short-term exposure duration, an order of magnitude higher AF or use of an acute NOEC value would be more relevant; such a strategy was previously proposed and is generally allowable under CWA (Clark et al. 2002). The dredging method, placement method, and duration of time (i.e., representative of acute or chronic exposure duration) that the DM remains in the water column should be considered when assessing the implications of applying the 0.01 AF. At the very least, a larger AF of 0.05 to 0.1 should be used more consistently in dredging evaluations for nonpersistent contaminants such as ammonia (NAS 1972, Thurston 1986, USEPA/USACE 1991, 1998, 2008); the default value is more relevant to chronic, long-term protection from persistent contaminants such as metals.

The larval development toxicity tests that assess a normal development endpoint are much more sensitive tests as compared to acute fish and invertebrate lethality tests. Further, development test endpoints are extremely sensitive to ammonia (substantially more so than other test species; Figure 4) and can be confounded by the physical effects of sediment

particles (Carr and Chapman 1995). It must be recognized that available default- and literature-reported ACRs and AFs were intended to be applied to acute lethality data (LC50) to estimate a chronically protective concentration; these AFs were not intended for embryonic development endpoints, such as the normal larval development endpoint used in water column (elutriate) toxicity evaluations. Applying the same generic AF of 0.01 used for acute survival data is not relevant for the much more sensitive embryonic development tests, as stated in USEPA / USACE (1998); use of the default AFs to the larval development endpoint (EC50) is expected to result in an overly conservative LPC, especially for nonpersistent contaminants such as ammonia. Most simply stated, the NOEC from development tests is expected to be protective of acute and longer-term, delayed effects of the DM exposure. When there is evidence that ammonia is the cause of elutriate toxicity in larval development toxicity tests, a higher AF (such as 0.2 presented herein) should be considered. A toxicity identification evaluation (TIE) may be needed to determine whether ammonia is the driver of the toxicity rather than a different contaminant of concern in the sediment elutriate. ERDC has adopted currently available toxicity identification/reduction evaluation methods for water (USEPA 1991) and sediments (USEPA 2007) to be specifically applicable to dredging evaluations and is publishing a toxicity reduction evaluation (TRE) method specifically for elutriates.

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REPORT DOCUMENTATION PAGE

Form Approved OMB No. 0704-0188

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				5c.	PROGRAM ELEMENT NUMBER			
6. AUTHOR(S)				5d.	PROJECT NUMBER			
Alan J. Kennedy, Gu	fery A. Steevens		5e.	TASK NUMBER				
				5f. '	WORK UNIT NUMBER			
7. PERFORMING ORG	ANIZATION NAME(S) AN	D ADDRESS(ES)		8. F	PERFORMING ORGANIZATION REPORT			
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9. SPONSORING / MOI	NITORING AGENCY NAI	ME(S) AND ADDRESS(E	S)	10.	SPONSOR/MONITOR'S ACRONYM(S)			
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12. DISTRIBUTION / AV	AILABILITY STATEME	NT						
Approved for public	release; distribution ur	limited.						
13. SUPPLEMENTARY NOTES								
 14. ABSTRACT The purpose of this technical report is to review the use of application factors (AFs) in determining the limiting permissible concentration (LPC) for water column (elutriate) toxicity evaluations for dredged material placement operations. Application factors are used as multipliers; AFs are applied to median effect toxicity endpoints in an effort to determine "safe" contaminant levels in the open water environment. While the default AF in the Marine Protection Research and Sanctuaries Act is 0.01, Federal regulations allow use of justifiable alternatives. An underprotective, high AF would underestimate the toxic effects of contaminants and potentially impact the organisms at the open-water, dredged material placement site. However, an overly conservative, low (excessively restrictive) AF would needlessly impose volume restrictions and increase dredged material management needs and costs. Herein the authors identify cases where use of the default AF is too conservative or even inappropriate: examples include situations in which it is applied to short-exposure-duration dredging placement operations (e.g., < 24 hours) where the exposure concentrations are decreasing steadily over time; instances in which it is applied to non-persistent contaminants and to inappropriate toxicological endpoints. Particularly, use of the default AF is overly conservative when high levels of non-persistent chemicals such as ammonia are present in elutriate water, and when toxicity is assessed using the very sensitive embryo/larval development tests. Since the default AF was not originally intended to be applied to nonpersistent contaminants or larval development endpoints, the authors propose alternatives to the default AF that is specific to ammonia toxicity and the development toxicity test endpoint. 15 SUBJECT TERMS 								
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Dredging Safe contaminant levels								
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