DEPARTMENT OF THE AIR FORCE
59TH MEDICAL WING (AETC)
JOINT BASE SAN ANTONIO - LACKLAND TEXAS

14 FEB 2017

MEMORANDUM FOR SGVUL
ATTN: THOMAS GIBBONS

FROM: 59 MDW/SGVU

SUBJECT: Professional Presentation Approval

1. Your paper, entitled **Genome Sequencing at the 59th Medical Wing Clinical Research Division** presented at/published to **News Letter Style Article to be submitted to Office of the Air Force Surgeon General (SG5M, Research/Innovations)** in accordance with MDWI 41-108, has been approved and assigned local file #**17071.**

2. Pertinent biographic information (name of author(s), title, etc.) has been entered into our computer file. Please advise us (by phone or mail) that your presentation was given. At that time, we will need the date (month, day and year) along with the location of your presentation. It is important to update this information so that we can provide quality support for you, your department, and the Medical Center commander. This information is used to document the scholarly activities of our professional staff and students, which is an essential component of Wilford Hall Ambulatory Surgical Center (WHASC) internship and residency programs.

3. Please know that if you are a Graduate Health Sciences Education student and your department has told you they cannot fund your publication, the 59th Clinical Research Division may pay for your basic journal publishing charges (to include costs for tables and black and white photos). We cannot pay for reprints. If you are 59 MDW staff member, we can forward your request for funds to the designated wing POC.

4. Congratulations, and thank you for your efforts and time. Your contributions are vital to the medical mission. We look forward to assisting you in your future publication/presentation efforts.

Linda Steel-Goodwin

LINDA STEEL-GOODWIN, Col, USAF, BSC
Director, Clinical Investigations & Research Support

PROCESSING OF PROFESSIONAL MEDICAL RESEARCH/TECHNICAL PUBLICATIONS/PRESENTATIONS

INSTRUCTIONS

USE ONLY THE MOST CURRENT 59 MDW FORM 3039 LOCATED ON AF E-PUBLISHING

1. The author must complete page two of this form:
   a. In Section 2, add the funding source for your study [e.g., 59 MDW CRD Graduate Health Sciences Education (GHSE) (SG5 O&M); SG5 R&D; Tri-Service Nursing Research Program (TSNRP); Defense Medical Research & Development Program (DMRDP); NIH; Congressionally Directed Medical Research Program (CDMRP) ; Grants; etc.]

   b. In Section 2, there may be funding available for journal costs, if your department is not paying for figures, tables or photographs for your publication. Please state "YES" or "NO" in Section 2 of the form, if you need publication funding support.

2. Print your name, rank/grade, sign and date the form in the author's signature block or use an electronic signature.

3. Attach a copy of the 59 MDW IRB or IACUC approval letter for the research related study. If this is a technical publication/presentation, state the type (e.g. case report, QA/QI study, program evaluation study, informational report/briefing, etc.) in the "Protocol Title" box.

4. Attach a copy of your abstract, paper, poster and other supporting documentation.

5. Save and forward, via email, the processing form and all supporting documentation to your unit commander, program director or immediate supervisor for review/approval.

6. On page 2, have either your unit commander, program director or immediate supervisor:
   a. Print their name, rank/grade, title; sign and date the form in the approving authority's signature block or use an electronic signature.

7. Submit your completed form and all supporting documentation to the CRD for processing (59crdpubspres@us.af.mil). **This should be accomplished no later than 30 days before final clearance is required to publish/present your materials.** If you have any questions or concerns, please contact the 59 CRD/Publications and Presentations Section at 292-7141 for assistance.

8. The 59 CRD/Publications and Presentations Section will route the request form to clinical investigations, 502 ISG/JAC (Ethics Review) and Public Affairs (59 MDW/PA) for review and then forward you a final letter of approval or disapproval.

9. Once your manuscript, poster or presentation has been approved for a one-time public release, you may proceed with your publication or presentation submission activities, as stated on this form. **Note:** For each new release of medical research or technical information as a publication/presentation, a new 59 MDW Form 3039 must be submitted for review and approval.

10. If your manuscript is accepted for scientific publication, please contact the 59 CRD/Publications and Presentations Section at 292-7141. This information is reported to the 59 MDW/CC. All medical research or technical information publications/presentations must be reported to the Defense Technical Information Center (DITC). See 59 MDWI 41-108, *Presentation and Publication of Medical and Technical Papers,* for additional information.

11. The Joint Ethics Regulation (JER) DoD 5500.07-R, *Standards of Conduct*, provides standards of ethical conduct for all DoD personnel and their interactions with other non-DoD entities, organizations, societies, conferences, etc. Part of the Form 3039 review and approval process includes a legal ethics review to address any potential conflicts related to DoD personnel participating in non-DoD sponsored conferences, professional meetings, publication/presentation disclosures to domestic and foreign audiences, DoD personnel accepting non-DoD contributions, awards, honoraria, gifts, etc. The specific circumstances for your presentation will determine whether a legal review is necessary. **If you (as the author) or your supervisor check "NO" in block 17 of the Form 3039, your research or technical documents will not be forwarded to the 502 ISG/JAC legal office for an ethics review.** To assist you in making this decision about whether to request a legal review, the following examples are provided as a guideline:

    For presentations before professional societies and like organizations, the 59 MDW Public Affairs Office (PAO) will provide the needed review to ensure proper disclaimers are included and the subject matter of the presentation does not create any cause for DoD concern.

    If the sponsor of a conference or meeting is a DoD entity, an ethics review of your presentation is not required, since the DoD entity is responsible to obtain all approvals for the event.

    If the sponsor of a conference or meeting is a non-DoD commercial entity or an entity seeking to do business with the government, then your presentation should have an ethics review.

    If your travel is being paid for (in whole or in part) by a non-Federal entity (someone other than the government), a legal ethics review is needed. These requests for legal review should come through the 59 MDW Gifts and Grants Office to 502 ISG/JAC.

    If you are receiving an honorarium or payment for speaking, a legal ethics review is required.

    If you (as the author) or your supervisor check "YES" in block 17 of the Form 3039, your research or technical documents will be forwarded simultaneously to the 502 ISG/JAC legal office and PAO for review to help reduce turn-around time. If you have any questions regarding legal reviews, please contact the legal office at (210) 671-5795/3365, DSN 473.

NOTE: All abstracts, papers, posters, etc., should contain the following disclaimer statement:
"The views expressed are those of the [author(s)] [presenter(s)] and do not reflect the official views or policy of the Department of Defense or its Components"

NOTE: All abstracts, papers, posters, etc., should contain the following disclaimer statement for research involving humans:
"The voluntary, fully informed consent of the subjects used in this research was obtained as required by 32 CFR 219 and DODI 3216.02_AFI 40-402."

NOTE: All abstracts, papers, posters, etc., should contain the following disclaimer statement for research involving animals, as required by AFMAN 40-401_IP :
"The experiments reported herein were conducted according to the principles set forth in the National Institute of Health Publication No. 80-23, Guide for the Care and Use of Laboratory Animals and the Animal Welfare Act of 1966, as amended."

PROCESSING OF PROFESSIONAL MEDICAL RESEARCH/TECHNICAL PUBLICATIONS/PRESENTATIONS

1. TO: CLINICAL RESEARCH	2. FROM: (Author's Name, Rank, Grade, Office Symbol)	3. GME/GHSE STUDENT:	4. PROTOCOL NUMBER:
	Thomas Gibbons, GS-14, SGVUL	☐ YES ☒ NO	NA

5. PROTOCOL TITLE: (**NOTE:** For each new release of medical research or technical information as a publication/presentation, a new 59 MDW Form 3039 must be submitted for review and approval.)

NA

6. TITLE OF MATERIAL TO BE PUBLISHED OR PRESENTED:

Genome Sequencing at the 59th Medical Wing Clinical Research Division

7. FUNDING RECEIVED FOR THIS STUDY? ☐ YES ☒ NO FUNDING SOURCE:

8. DO YOU NEED FUNDING SUPPORT FOR PUBLICATION PURPOSES: ☐ YES ☒ NO

9. IS THIS MATERIAL CLASSIFIED? ☐ YES ☒ NO

10. IS THIS MATERIAL SUBJECT TO ANY LEGAL RESTRICTIONS FOR PUBLICATION OR PRESENTATION THROUGH A COLLABORATIVE RESEARCH AND DEVELOPMENT AGREEMENT (CRADA), MATERIAL TRANSFER AGREEMENT (MTA), INTELLECTUAL PROPERTY RIGHTS AGREEMENT ETC.?

☐ YES ☒ NO **NOTE:** If the answer is YES then attach a copy of the Agreement to the Publications/Presentations Request Form.

11. MATERIAL IS FOR: ☒ DOMESTIC RELEASE ☐ FOREIGN RELEASE

CHECK APPROPRIATE BOX OR BOXES FOR APPROVAL WITH THIS REQUEST. ATTACH COPY OF MATERIAL TO BE PUBLISHED/PRESENTED.

☐ 11a. PUBLICATION/JOURNAL (List intended publication/journal.)

☐ 11b. PUBLISHED ABSTRACT (List intended journal.)

☐ 11c. POSTER (To be demonstrated at meeting: name of meeting, city, state, and date of meeting.)

☐ 11d. PLATFORM PRESENTATION (At civilian institutions: name of meeting, state, and date of meting.)

☒ 11e. OTHER (Describe: name of meeting, city, state, and date of meeting.)

News Letter Style Article to be submitted to Office of the Air Force Surgeon General (SG5M, Research and Innovations)

12. HAVE YOUR ATTACHED RESEARCH/TECHNICAL MATERIALS BEEN PREVIOUSLY APPROVED TO BE PUBLISHED/PRESENTED?

☐ YES ☒ NO ASSIGNED FILE # _____ DATE _____

13. EXPECTED DATE WHEN YOU WILL NEED THE CRD TO SUBMIT YOUR CLEARED PRESENTATION/PUBLICATION TO DTIC

NOTE: All publications/presentations are required to be placed in the Defense Technical Information Center (DTIC).

DATE	

14. 59 MDW PRIMARY POINT OF CONTACT (Last Name, First Name, M.I., email)	15. DUTY PHONE/PAGER NUMBER
Gibbons, Thomas, F. thomas.f.gibbons6.civ@mail.mil	210-292-7363

16. AUTHORSHIP AND CO-AUTHOR(S) List in the order they will appear in the manuscript.

LAST NAME, FIRST NAME AND M.I.	GRADE/RANK	SQUADRON/GROUP/OFFICE SYMBOL	INSTITUTION (If not 59 MDW)
a. Primary/Corresponding Author			
b.			
c.			
d.			
e.			

17. IS A 502 ISG/JAC ETHICS REVIEW REQUIRED (JER DOD 5500.07-R)? ☐ YES ☒ NO

I CERTIFY ANY HUMAN OR ANIMAL RESEARCH RELATED STUDIES WERE APPROVED AND PERFORMED IN STRICT ACCORDANCE WITH 32 CFR 219, AFMAN 40-401_IP, AND 59 MDWI 41-108. I HAVE READ THE FINAL VERSION OF THE ATTACHED MATERIAL AND CERTIFY THAT IT IS AN ACCURATE MANUSCRIPT FOR PUBLICATION AND/OR PRESENTATION.

18. AUTHOR'S PRINTED NAME, RANK, GRADE	19. AUTHOR'S SIGNATURE	20. DATE
Thomas F. Gibbons, GS-14	GIBBONS.THOMAS.F.1128005281	January 27, 2017
21. APPROVING AUTHORITY'S PRINTED NAME, RANK, TITLE	**22. APPROVING AUTHORITY'S SIGNATURE**	**23. DATE**
Paul T Barnicott, GS-15, DAF, Dep Dir, 59th CRD	BARNICOTT.PAUL.T.1102318273	January 27, 2017

PROCESSING OF PROFESSIONAL MEDICAL RESEARCH/TECHNICAL PUBLICATIONS/PRESENTATIONS

1st ENDORSEMENT (59 MDW/SGVU Use Only)

TO: Clinical Research Division 59 MDW/CRD Contact 292-7141 for email instructions.	24. DATE RECEIVED January 27, 2017	25. ASSIGNED PROCESSING REQUEST FILE NUMBER 17071

26. DATE REVIEWED February 09, 2017	27. DATE FORWARDED TO 502 ISG/JAC

28. AUTHOR CONTACTED FOR RECOMMENDED OR NECESSARY CHANGES: ☒ NO ☐ YES If yes, give date. _____ ☐ N/A

29. COMMENTS ☒ APPROVED ☐ DISAPPROVED

Informational brochure/publication for CRD technologies. Appropriate disclaimers included

30. PRINTED NAME, RANK/GRADE, TITLE OF REVIEWER	31. REVIEWER SIGNATURE	32. DATE
Kevin Kupferer/GS13/Human Research Subject Protection Expert	KUPFERER.KEVIN.R.1086667270 Digitally signed by KUPFERER.KEVIN.R.1086667270 DN: c=US, o=U.S. Government, ou=DoD, ou=PKI, ou=USAF, cn=KUPFERER.KEVIN.R.1086667270 Date: 2017.02.09 12:34:20 -08'00'	February 09, 2017

2nd ENDORSEMENT (502 ISG/JAC Use Only)

33. DATE RECEIVED	34. DATE FORWARDED TO 59 MDW/PA

35. COMMENTS ☐ APPROVED (In compliance with security and policy review directives.) ☐ DISAPPROVED

36. PRINTED NAME, RANK/GRADE, TITLE OF REVIEWER	37. REVIEWER SIGNATURE	38. DATE

3rd ENDORSEMENT (59 MDW/PA Use Only)

39. DATE RECEIVED February 10, 2017	40. DATE FORWARDED TO 59 MDW/SGVU February 13, 2017

41. COMMENTS ☒ APPROVED (In compliance with security and policy review directives.) ☐ DISAPPROVED

42. PRINTED NAME, RANK/GRADE, TITLE OF REVIEWER	43. REVIEWER SIGNATURE	44. DATE
Kevin Iinuma, SSgt/E-5, 59 MDW Public Affairs	IINUMA.KEVIN.MITSUGU.1296227613 Digitally signed by IINUMA.KEVIN.MITSUGU.1296227613 DN: c=US, o=U.S. Government, ou=DoD, ou=PKI, ou=USAF, cn=IINUMA.KEVIN.MITSUGU.1296227613 Date: 2017.02.13 09:55:11 -08'00'	February 13, 2017

4th ENDORSEMENT (59 MDW/SGVU Use Only)

45. DATE RECEIVED	46. SENIOR AUTHOR NOTIFIED BY PHONE OF APPROVAL OR DISAPPROVAL ☐ YES ☐ NO ☐ COULD NOT BE REACHED ☐ LEFT MESSAGE

47. COMMENTS ☐ APPROVED ☐ DISAPPROVED

48. PRINTED NAME, RANK/GRADE, TITLE OF REVIEWER	49. REVIEWER SIGNATURE	50. DATE

Genome Sequencing at the 59th Medical Wing Clinical Research Division

The 59th MDW Clinical Research Division (CRD) has successfully sequenced the entire human genome. Human genome sequencing was first completed in 2003 at a cost of approximately 3 Billion dollars. It took the combined effort of 20 laboratories across 7 countries working for 13 years to complete. Since then, remarkable advancements in both molecular techniques and computer technology have enabled smaller laboratories to bring this tool into the hands of more and more researchers.

Picture: Preparation of deoxyribose nucleic acid (DNA) for analysis.

Sequencing of the human genome at the CRD was possible due to the recent purchase of an Illumina NextSeq® 500 sequencing system. This system greatly expanded our sequencing capability from 2 gigabases (Gb) to 120 Gb. For reference, one gigabase is one billion base pairs of DNA. Previously, only a few human genes or small genomes such as bacterial genomes could be sequenced in a single run. With this new sequencer, the entire human genome which contains 3 billion base pairs of DNA can be sequenced on a single run with an average of 30x coverage. 30x coverage means that, on average, each base pair of DNA is sequenced 30 times which helps to ensure the accuracy of results when utilizing this technology.

For this specific study, A-431 cells, from a human epidermoid carcinoma, were acquired from the American Type Culture Collection (ATCC CRL 1555). The CRD Cell Biology Department then grew the cells as a monolayer in Dulbecco's modified Eagle's medium with 10% fetal bovine serum. The cells were grown to confluence and the monolayer removed from the flask

via standard trypsinization. A cell count was performed and cells were frozen at -80^{O}C at 1x10^{6} cells/mL. DNA was extracted from the cells using Qiagen's AllPrep DNA/RNA isolation Kit and then fragmented to 550 bp in length.

Picture: Dr. Jody Noe analyzing the human genome.

Once the DNA was fragmented to the correct length, a sequence ready "library" was created using an Illumina NeoPrep™ automatic library preparation instrument. This device, automatically prepares up to 16 libraries of genomic DNA (gDNA) by adding adapter sequences and barcodes onto the ends of DNA fragments in preparation for subsequent cluster generation and sequencing. Once sequencing was complete, a pre-configured onsite bioinformatics platform was used to analyze the results of the genomic sequencing experiment without the need for internet access to cloud data storage.

The CRD laboratory can now provide sequencing flexibility across a broad range of applications and study sizes. Other potential sequencing applications include exome, transcriptome, gene expression comparisons, metagenomic and epigenetic studies. For many of these studies several specimens can be run simultaneously on a single run which maximizes resources. For example up to eight transcriptomes (all messenger RNA from the genome or parts of the genome that are active) or exomes (the exons of the genome which constitute the part of the genome that are typically expressed as proteins) can be run simultaneously.

The mission of the 59th CRD Laboratory Branch is to Provide Laboratory Support for the development, performance, and dissemination of human and animal research. This research support laboratory maintains the latest state of the art equipment in each of its 5 departments: Hematology, Chemistry, Microbiology, Cell Biology and Molecular Biology. Each department is run by a laboratory scientist dedicated to providing cutting edge laboratory support to Air Force Graduate Medical Education students conducting research. In many cases the research being conducted requires the support and cooperation of more than one department. For example, sequencing the human genome in this study required the tissue culture expertise of Ms. Dixon who runs the Cell Biology Department and the molecular expertise of Dr Jody Noe who runs the Molecular Department. This latest acquisition by the Molecular Department of the CRD opens up a host of research capabilities for the average Graduate Medical Education student at an affordable level, using local subject matter experts within the 59th CRD. If you are interested in utilizing the CRD Laboratory to support your GME research you should contact the Lab Branch Chief, Dr Thomas Gibbons at 210-292-7363.