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CONTRACTING ORGANIZATION: Stevens Institute of Technology

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14. ABSTRACT Currently, no studies have investigated excised wound beds as a supportive matrix for a well-defined population of stem cells cultured directly within the wound bed niche. To capitalize on the idea of a wound bed-stem cell synergy, the discovery science phase of work in this proposal will rest on the premise that the native wound microenvironment will be the optimal bioreactor for a stem cell-based dermal graft model. Over the past year (Year 1) of the project, the PI team has completed the initial electrohydrodynamic printing process development phase towards fabricating high resolution 3D biological templates for stem cell-wound bed culture. The systematic design of experiments carried out in the current reporting period yielded significant results, including the quantitative effect of deposition time and collector temperature on fiber accumulation geometry, the effect of collector temperature on the interaction between two adjacent fibers, and that, to print 3D template structures, a toolpath optimization step is required to ensure straight, ordered fiber prints. For the next reporting report, systematic studies for the 3D template printing as a function of collector temperature will be analyzed. Stem cell -3D template interactions will be assessed for potential as a scalable in situ dermal graft model.					
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1. INTRODUCTION: *Narrative that briefly (one paragraph) describes the subject, purpose and scope of the research.*

The subject of this research is developing an innovative therapeutic strategy for chronic cutaneous wound care that is complicated by an active military and Veteran patient population with a heterogeneous ulcer etiology. Furthermore, sub-optimal patient outcomes with existing wound care technologies and approaches reflect the inadequacy of single, rudimentary agents to mediate the complex interplay of wound healing. Therefore, the purpose of this research is to develop an in-situ scalable therapeutic strategy that reliably harnesses stem cell regenerative and immunomodulatory properties that effectively reverse the debilitating consequences of chronic ulcer wound healing. The scope of this discovery phase of research will be the design and implementation of a precision, super-resolution printing process that yields microscale fiber-based templates to program MSC expansion and modify the wound bed niche towards subverting the chronic wound phenotype that is currently refractory to existing treatment modalities.

2. KEYWORDS: *Provide a brief list of keywords (limit to 20 words).*

Additive manufacturing, 3D printing, dermal graft, stem cell, wound, ulcer, immunomodulation, in situ printing

3. ACCOMPLISHMENTS: *The PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction.*

What were the major goals of the project?

List the major goals of the project as stated in the approved SOW. If the application listed milestones/target dates for important activities or phases of the project, identify these dates and show actual completion dates or the percentage of completion.

The major goals of the project as stated in the approved SOW are partitioned into two Specific Aims: 1) Test the identified key process and material parameters with optimization for uniform mechanical stretching and alignment of printed fibers, and 2) Design, fabricate, and evaluate precision dermal graft model under varying microscale geometrical cues. For the associated two major tasks, the milestones and percentage of completion are as follows:

Milestone 1 Achieved (60% completion) at 8 months: Identification of process and material parameter regimes that enable fiber stretching and alignment in an in situ phantom wound bed.
Milestone 2 Achieved (20% completion) at 18 months: Production of a homogeneous self-renewing MSC population as a dermal graft model.

What was accomplished under these goals?

For this reporting period describe: 1) major activities; 2) specific objectives; 3) significant results or key outcomes, including major findings, developments, or conclusions (both positive and negative); and/or 4) other achievements. Include a discussion of stated goals not met. Description shall include pertinent data and graphs in sufficient detail to explain any significant results achieved. A succinct description of the methodology used shall be provided. As the project progresses to completion, the emphasis in reporting in this section should shift from reporting activities to reporting accomplishments.

Major Activities:

Over the past year (Year 1) of the project, the PI team has completed the initial electrohydrodynamic printing process development phase towards fabricating high resolution 3D biological templates for stem cell-wound bed culture. In the MEW process, a polymer melt material is drawn from a needle to a collector plate, and the processed polymer from the needle tip constitutes the charged fiber. The temperature differential between the needle tip and the plate is defined as temperature gradient; and it affects fiber deformation (that determines fiber geometry) and electrostatic repulsion between fibers (that, through relative alignment of multiple fibers, determines pore geometry in a 3D biological substrate).

A. Building high-resolution electrohydrodynamic process with heat-exchange collector plate

The 3D electrohydrodynamic printing system configuration is schematized in Figure 1. The barrel of the syringe is heated by the industrial heat gun set at 340 °F through a heating tunnel. The bottom of the heating tunnel is penetrated by the needle of the syringe and the exposed needle length is 2 mm. The plunger of the syringe is propelled by a programmable extrusion pump. The needle of the syringe and the aluminum collector plate (203 mm×203 mm×3.3 mm) are connected to the positive and negative terminals of the high voltage supply (Gamma, USA), respectively. The collector plate (shown in Figure 1) is fixed on a heat exchange element, which is mounted on XY moving platform consisting of two linear actuators. The inlet of the heat exchange element is connected to the faucet, which can adjust the temperature of the inflow water. The outflow water is discharged down the drain. Each time the situation of inflow water is altered by turning the knob of the faucet, the temperature of the collector plate is monitored until it stabilizes at the target level for 15 min. The ambient temperature is kept at $23.5\pm 1^{\circ}\text{C}$. The PI team was able to establish the operational values/ranges for the key process variables for the process as shown in Table 1.

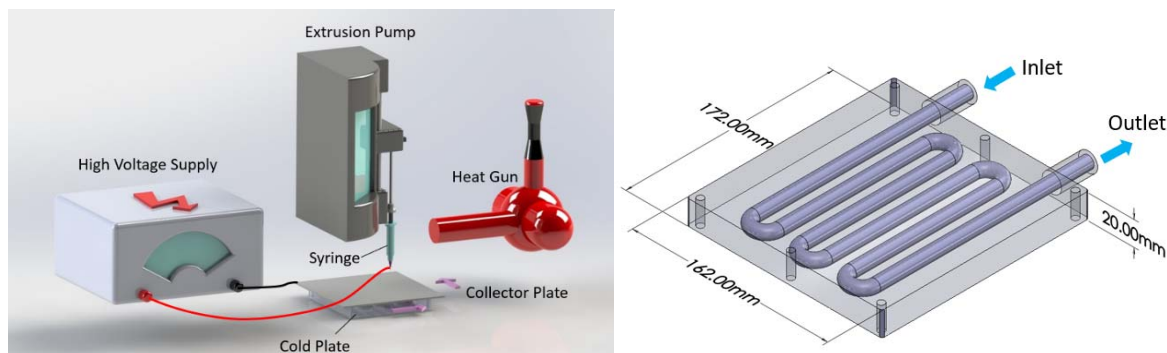


Figure 1. Schematic of MEW system and the heat-exchange plate

Table 1. The parameters and their values or range in our MEW process

Parameters	Values
Polymer temperature (T_p)	94.7±1 °C (measured)
Applied Voltage (V)	12 kV, unless otherwise stated
Stage translational speed (v_s)	15-20mm/s
Volumetric flow rate (Q_v)	25 μ L/h
Tip to collector distance (D_{tc})	20 mm
Inter-fiber distance (S_f)	60-500 μ m
Set temperature range (T_s)	320 -160 °F

B. Studying the effect of collector temperature on printed fiber disposition

During the melt-based 3D printing process, control of temperature is always critically important, including the material temperature, the ambient temperature and the collector temperature, or more generally, the temperature gradient during the flight phase of the jet, which will affect the material printability, viscosity, flowrate, jet speed, and degradation rate, and plays an important role in taming the jet effectively. However, the effect of temperature on the extent of interaction among the fibers during the printing process hasn't been systematically studied. To address these problems, a design of experiments for three modes of printing, namely 1) stationary printing, 2) single fiber printing, and 3) 3D template printing, are adopted with variable collector temperature settings. The effect of collector temperature on the bulk shape of the fiber accumulation, the fiber interaction and the wall morphology in the layered 3D template are studied in these three printing modes, respectively. In addition, the printing toolpath is preliminarily studied regarding the peripheral area beyond the target template area. In Year 1, the PI team was able to complete studies for the stationary and single fiber printing modes with preliminary 3D template printing results, but the systematic studies and comprehensive results for the 3D template printing as a function of collector temperature will be reported in Year 2 of this research.

Significant Results:

A. Effect of collector temperature in stationary printing mode experiments

The PI team completed a stationary printing mode study to quantitatively measure the effect of varying collector temperature on the evolution of the bulk geometry on a bare aluminum collector plate with uniform conductivity. In order to tune the collector temperature parameter, water channel lines within the heat exchange element are prescribed varying temperatures so as to reach steady-state collector temperatures of 16±0.5°C, 23.5±0.5°C and 30±0.5°C, with other parameters prescribed according to Table 1. The general observation here is that the shape of the bulk fiber accumulation geometry is typically a high central cone encircled by an outer ring. Between the central cone and outer ring, several lower cones can also be vaguely identified upon closer inspection. When the temperature is 16°C, the height difference between the central cone and outer ring is insignificant and the bulk geometry assumes a disk (Figure 2a), while as the temperature increases, the identification of a characteristic cone-and-ring feature becomes apparent (Figure 2c). A side view profile comparison is shown in Figure 2d. To quantify the bulk geometry of the fiber accumulation, three parameters are measured or calculated including

its height (H_{ac}), base diameter (D_{ac}) and their ratio (H_{ac}/D_{ac}). From Figure 3, both H_{ac} and D_{ac} increase as the deposition time increases, while H_{ac}/D_{ac} remains relatively constant. As the collector temperature increases, H_{ac} significantly increases and D_{ac} decreases, which results in the increase of H_{ac}/D_{ac} (Figure 3d).

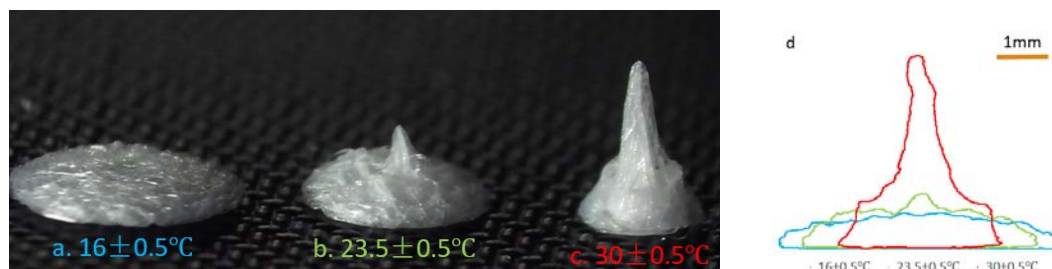


Figure 2. Comparison of fiber accumulation shape at different collector temperatures (Deposition time: 6 min. a, b, c are the photographs of samples at different collector temperatures. d shows the profiles of them from a side view by digitalizing the corresponding photographs)

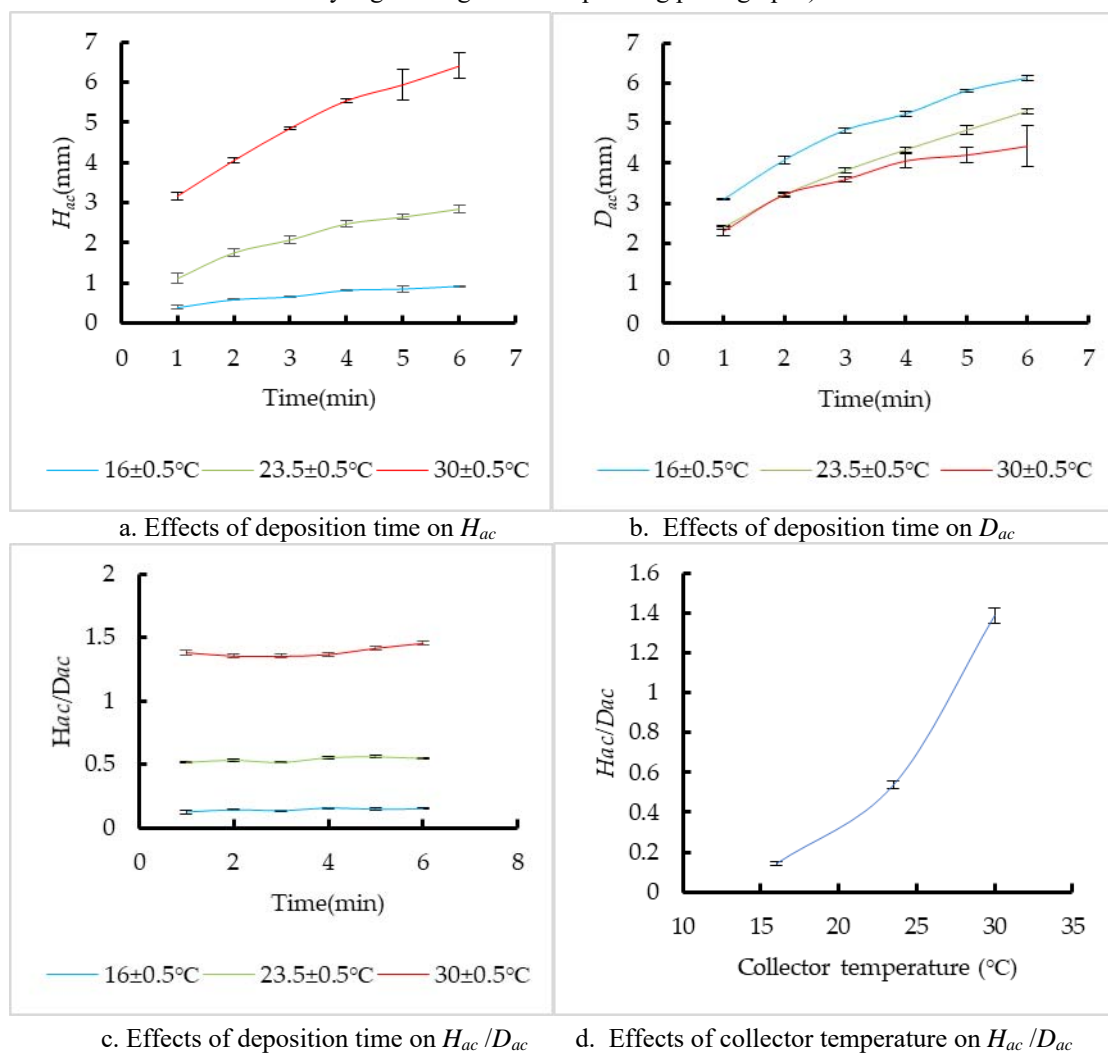


Figure 3. Effect of deposition time and collector temperature on the formation of fiber accumulation geometry

B. Effect of collector temperature in stationary printing mode experiments

Another significant result in Year 1 is understanding the effect of collector temperature on the interaction between two adjacent single fibers. The interaction between the adjacent fibers in each set is studied through setting sSf (i.e. the inter-fiber distance) at different levels (sSf , from $500\ \mu\text{m}$ to $60\ \mu\text{m}$), and measuring the actual Sf (mSf), as is shown in Figure 4. The result is the average of 3 sets of fibers at each level, and for each set of fibers, the mSf is measured 5 times with sampling points equally distributed on the fibers. It shows that regardless of the collector temperature (set at 16 , 23.5 , and 30°C), the mSf is consistently smaller than the sSf , and the relative error becomes increasingly significant when sSf is smaller than $140\ \mu\text{m}$, as is shown in the dashed box in Figure 4. When the sSf is smaller than $80\ \mu\text{m}$, the adjacent fibers in each sample all stick together with one onto another. It should be noted that when sSf is between 100 and $120\ \mu\text{m}$, the large negative deviation is attributed to the phenomenon where some sets of fibers at this level stick together, while others maintain separation, although the distance between these separate fibers is only slightly smaller than the corresponding sSf . The effect of the collector temperature on the mSf is insignificant with $p > 0.3$ between any two selected data groups.

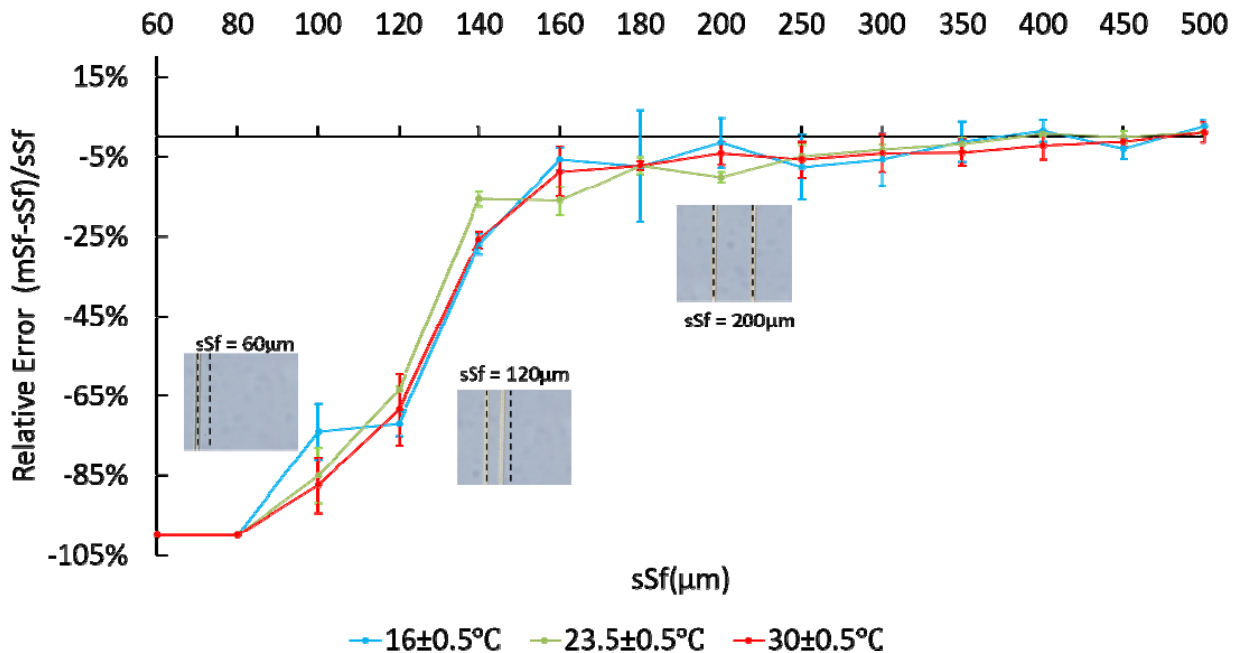


Figure 4. Dependence of Relative Error $(mSf - sSf)/sSf$ on sSf

(The typical image of the adjacent fibers when $sSf = 200, 120, 60\ \mu\text{m}$ are shown to exemplify the case of no significant attraction, significant attraction and complete vertical overlap of the fibers, respectively. The black dashed line denotes the corresponding sSf . The imaged fibers are printed at 23.5°C , but the result is mirrored at other collector temperatures.)

C. Optimization of toolpath for 3D template printing experiments

Another significant result in Year 1 is the preliminary finding that, to printing 3D template structures, a toolpath optimization step is required to ensure that straight, ordered fiber prints are obtainable. In order to produce straight fibers, it's necessary to make sure that the stage exerts a stretching, instead of a compressive buckling force on the jet. Therefore, the movement of the point of contact between the jetted material and the collector has to lag behind the movement of the printhead, namely the jet lag. The jet lag can be defined as the relative displacement between the point B and A in Figure 6g, and the lag length is defined as the magnitude of it. If the printing process is in steady state (i.e. no changes in printing speed and direction, and other process parameters; for example, the stage is moving uniformly in the middle of a long travel), the prerequisite of straight fiber printing or jet lag is that the translational stage speed exceeds a critical value, otherwise the fiber coiling will arise (Figure 5). At the start and end of a travel, the situation is different. The stage will experience an acceleration and deceleration process, respectively, therefore there will be short periods during which the stage speed is lower than jet speed, but mostly these periods are not long enough for the initiation of fiber coiling as long as the steady stage speed is higher than the jet speed, the jet lag, although smaller in magnitude than its steady state, still exists near the start and end of a travel. At turning points where toolpath direction changes, the jet lag will deviate the printed fibers from the prescribed toolpath, resulting in disorder (Figure 6c, f). This phenomenon is intractable in three ways. First, the jet instability may be attributed to many unpredictable factors, including material nonuniformities, along with any variations in the heating conditions and electric field parameters. For example, the electric field is affected by the relative motion of the printhead and collector. Second, whatever the motor and movement setup (a movable printing head with a stationary stage or a movable translational stage with a stationary printing head) is applied, an acceleration and deceleration process is unavoidable for the start and end of each travel, which makes the jet lag at turning points different from elsewhere (Figure 6g). If the travel distance is not small enough, the jet lag will increase, stabilize and decrease during the travel (Figure 6h blue curve). In contrast, if the travel distance is too short, the jet lag will decrease before it stabilizes (Figure 6h red curve). However, the design of short travel distance is inevitable in template printing, which makes the jet lag even more unpredictable. Lastly, with additional layering, the electric field will be increasingly weakened by the residual charge. Correspondingly, the jet speed will decrease, and jet lag increases, therefore Figure 6a-c is expected to arise sequentially as more layers are added. However, there is still a lack of model describing how the jet speed changes with layer number. In summary, the jet lag phenomenon is attributed to an unpredictable but transient fluctuation as well as a predictable but chronic increase.

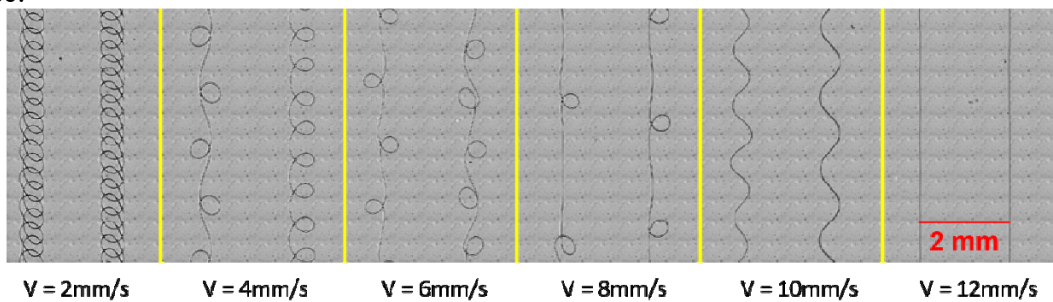


Figure 5. Fiber curling at a low translational stage speed relative to the fiber jet speed (Two fibers are printed for each speed level, which is 2, 4, 6, 8, 10, 12 mm/s from left to right.)

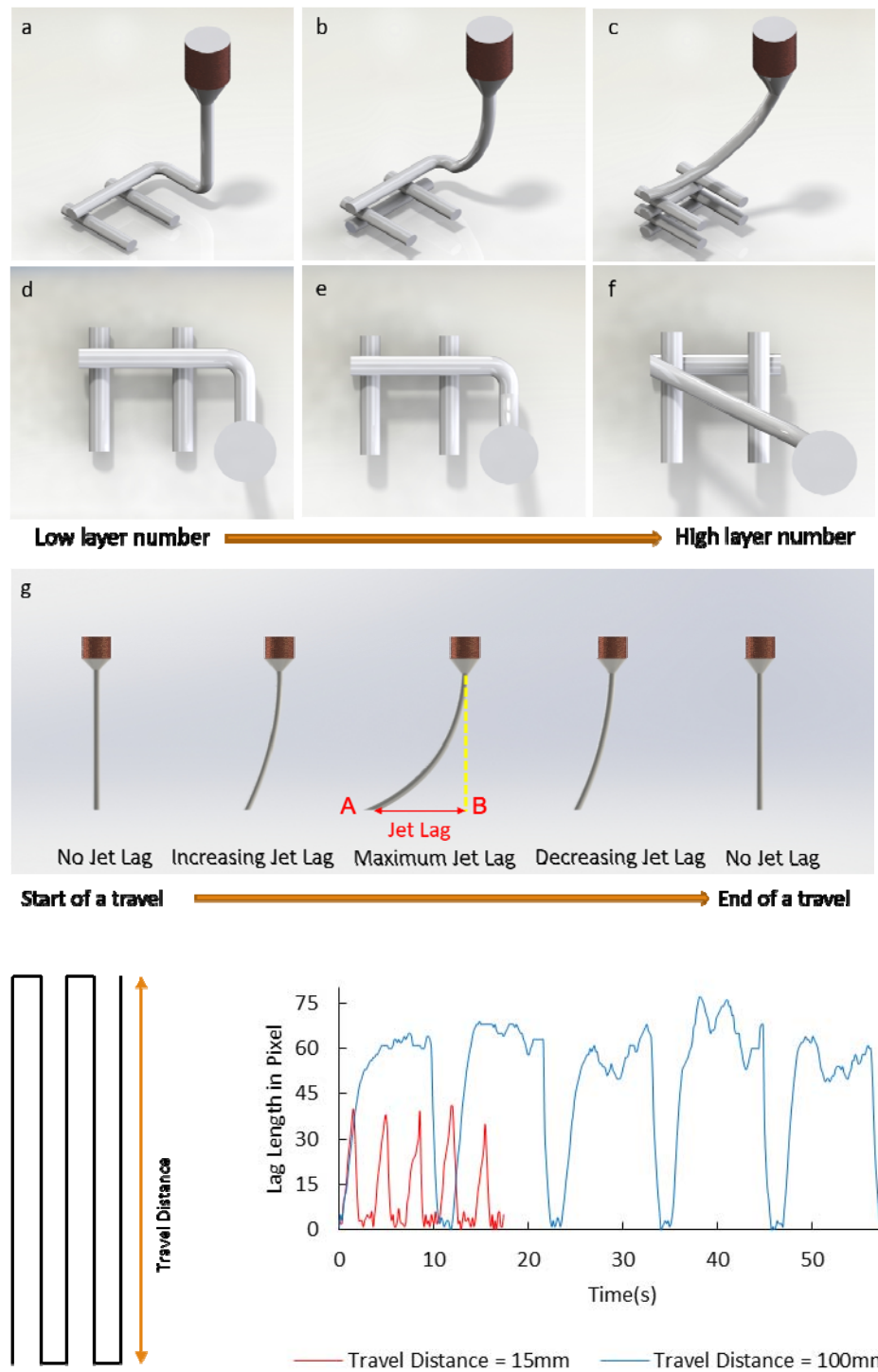


Figure 6. Evolution of lag length at turning points and during the travel (a-c represents the increase of jet lag at turning points when layer number increases. d, e and f are the top view of a, b and c, respectively. g represents the evolution of jet lag as a function of travel distance. h shows the back-and-forth path and the evolution of jet lag for long and short travel distance. At the end of each travel, the stage pauses for 1s to avoid the interplay between the adjacent travel. Jet speed is determined to be around 12 mm/s, and the translational stage speed is set at 20 mm/s. The travel distance is 15 and 100 mm for short and long travel distance, respectively. The lag length is recorded as the number of pixels between point A and point B in g.)

To address this challenge, three toolpaths are designed and tested in 3D template printing mode as shown in Figure 7a-c. Figure 7a is considered as the most intuitive toolpath for an orthogonally arranged template. In Figure 7b, considering the effect of jet lag, a larger orange area, namely the peripheral area, is expanded to avoid the occurrence of situation depicted in Figure 6c and f. In Figure 7c, the peripheral area is even larger, and the stage is designed to pause for 0.2 seconds (and 0.3 seconds after more than 100 layers are built, namely dwelling time) at the turning points (namely dwelling points). Additionally, the turns in the toolpath are made in an oblique way instead of an orthogonal way, so that the dwelling points are staggered spatially to avoid interplay. These three modifications are introduced based on the abovementioned two problems. A comparison of the printed 40-layer templates that implement these three distinct toolpaths in Figures 7a-c is shown in Figures 7d-h. To evaluate the orderliness of the template, a top view imaged is obtained and binarized within the template. The orderliness is measured by the percentage of the pixel number representing the template (or PPS). From these figures, the 40-layer template prints by Figure 7b and c (PPS = 35.2% and 36.2%, respectively) show more ordered layering than that by Figure 7a (PPS = 51.8%). Figures 7g-h are the resultant 200-layer prints corresponding to the template toolpaths depicted by Figures 7b and c, respectively. These two comparative template prints demonstrate that the toolpath in Figure 7c (PPS = 57.9%) results in a more ordered outcome than that prescribed by its counterparts in Figures 7b for higher template printing (PPS = 78.5%).

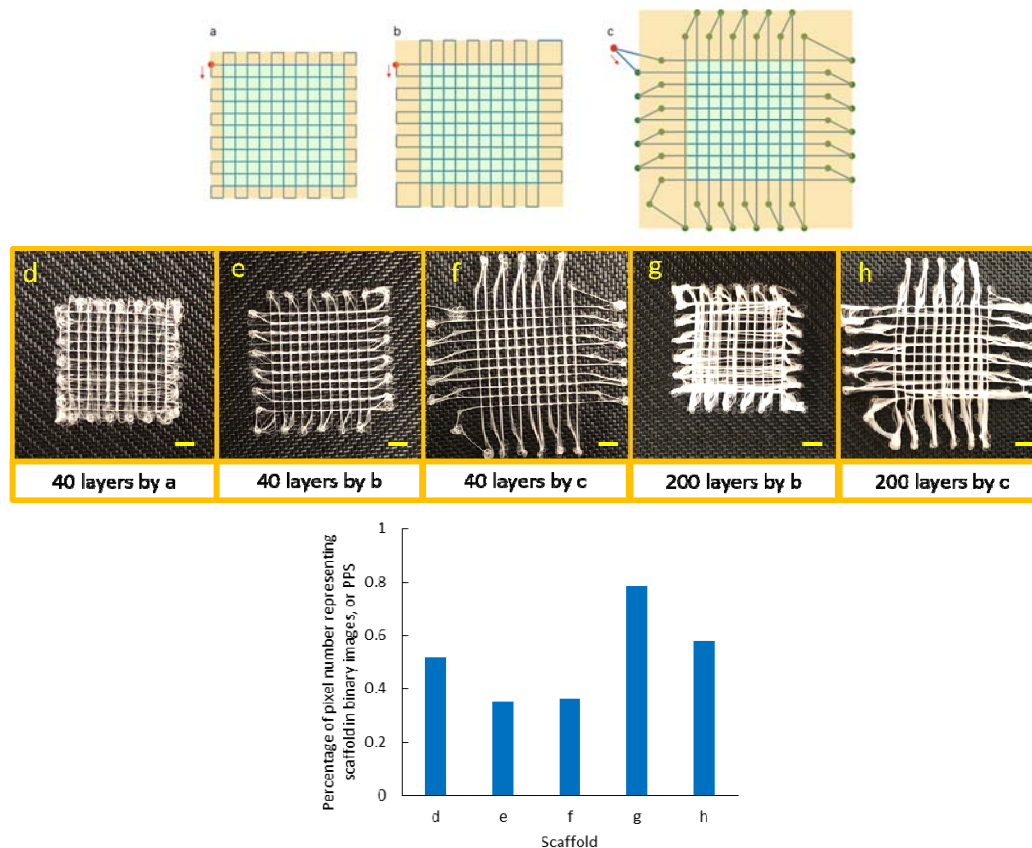


Figure 7. Toolpath of 3D template or scaffold printing and the corresponding templates with their PPS (Orange area designates the peripheral area and the green area designated the template print area of interest. Red dot denotes the starting point with the red arrow denotes printing direction. d, e and f are printed by a, b and c for 40 layers, respectively. g and h are printed by b and c for 200 layers, respectively. Scale bar in the pictures denotes 2 mm.)

What opportunities for training and professional development has the project provided?

If the project was not intended to provide training and professional development opportunities or there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe opportunities for training and professional development provided to anyone who worked on the project or anyone who was involved in the activities supported by the project. “Training” activities are those in which individuals with advanced professional skills and experience assist others in attaining greater proficiency. Training activities may include, for example, courses or one-on-one work with a mentor. “Professional development” activities result in increased knowledge or skill in one’s area of expertise and may include workshops, conferences, seminars, study groups, and individual study. Include participation in conferences, workshops, and seminars not listed under major activities.

In Year 1, this project has provided support of independent research training and development opportunities for one full-time PhD student. A senior PhD student (Mr. Houzhu Ding) and a junior PhD student (Mr. Kai Cao) have led the development of the melt-based electrohydrodynamic printing process for producing scalable 3D biological substrates at the microscale level; Houzhu has since graduated in November 2019 and Kai have continued working and being supported on the project.

The project has also provided research training exposure and training for one (1) female high school student who has worked with the PI during the summer and academic months.

How were the results disseminated to communities of interest?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe how the results were disseminated to communities of interest. Include any outreach activities that were undertaken to reach members of communities who are not usually aware of these project activities, for the purpose of enhancing public understanding and increasing interest in learning and careers in science, technology, and the humanities.

Our abstract was accepted for oral presentation (entitled “Effect of collector temperature in electrohydrodynamic 3D printing processes) at the 2020 ASME Manufacturing Science and Engineering Conference (MSEC) in Cincinnati, Ohio. However, the conference originally scheduled for early June was canceled due to COVID-19.

What do you plan to do during the next reporting period to accomplish the goals?

If this is the final report, state “Nothing to Report.”

Describe briefly what you plan to do during the next reporting period to accomplish the goals and objectives.

Based on the PI team’s findings in Year 1, a major task for the next reporting period will be to precisely control the layered fiber alignment and pore geometries within the produced 3D template. A related task will be to measure the effect of successive layering on the pore geometries. The resultant fiber alignment pore geometry outcomes will be measured with optical/scanning electron microscopy. Fabrication and characterization of a microscale 3D template will serve as the bridge between completion of Specific Aim 1 and enabling the biological studies proposed in Specific Aim 2. Due to the PI team’s research lab being shuttered since March 2020 with an unspecified re-opening time frame, the PI team will temporarily focus on advancing 3D non-planar toolpath algorithms and workflows required to simulate image-guided in situ bioprinting of a skin graft onto a phantom ulcer wound bed. Although this may expand the scope of the originally proposed specific aims, this will enable the PI team to augment the experimental robustness from both the manufacturing and biological standpoint when the experimental phase of work is permitted to resume.

- 4. IMPACT:** *Describe distinctive contributions, major accomplishments, innovations, successes, or any change in practice or behavior that has come about as a result of the project relative to:*

What was the impact on the development of the principal discipline(s) of the project?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe how findings, results, techniques that were developed or extended, or other products from the project made an impact or are likely to make an impact on the base of knowledge, theory, and research in the principal disciplinary field(s) of the project. Summarize using language that an intelligent lay audience can understand (Scientific American style).

Reliably engineering 3D biological templates poses a significant manufacturing challenge. To address this challenge, additive manufacturing (AM) has emerged as a promising approach to attribute unprecedented design complexity to 3D biological systems. Fundamentally, most 3D biological systems in engineered tissue applications are 3D structured biomaterial substrates as cell culture platforms. Current AM methods are able to fabricate biological substrates with complex geometrical features, but not at small scales. Specifically, the limited process resolutions of existing AM methods limits the range of downstream cell functions that can be engineered. Therefore, new manufacturing tools to probe cell-substrate interactions at small scales demands a new manufacturing process design. In this project, the PI advances a high-resolution melt-based electrohydrodynamic AM process towards the scalable (fast with small biologically relevant feature sizes) manufacturing of 3D templates for skin grafting. From the standpoint of biological or clinical impact, transplantation of self-renewing stem cells as therapeutic agents is poised to offer new treatment option for recalcitrant ulcer wounds. However, the biological complexity of cells has hampered the translation into the reliable, cost-effective manufacturing of stem cell-

based therapies. Although MSCs show great promise for engineered tissues and cell-based regenerative therapies, challenges to clinical adoption of MSC-based products are currently hampered by considerable heterogeneity in the stem cell populations, resulting in significant uncertainty associated with their therapeutic outcomes. The overall premise of this proposed project is accelerated microscale printing of a 3D ordered dermal graft will enable the scalable production of homogeneous MSC populations with a targeted self-renewing phenotype. An innovative strategy based on a novel hybrid electrospinning and 3D printing based additive manufacturing process is proposed herein to fabricate precision-structured three-dimensional geometries as decisive triggers of downstream MSC phenotypes. Furthermore, the novel compression of the time and space scales will be concurrently achieved by identifying the unusual processing conditions and material property ranges that mechanically stretch and align jetted fibers towards fast printing of microscale fibers. Moreover, the ability to quantitatively measure stem cell adhesion as a function of enforced geometrical parameters within a structured material matrix environment promises to open up new avenues of quantitative inquiry into the design of robust preclinical models. Finally, the successful outcome of the proposed research will have significant impact for public health by providing essential insight about the homogeneity of stem cell lineage commitment and their potential for improving therapeutic outcomes. In order to illustrate how fast, small-scale printing can be translated for chronic wound care in the clinical setting, an *in situ* printing-based methodological workflow is proposed to directly fabricate a custom dermal graft onto a phantom excised ulcer wound bed with a contour-matching print process toolpath.

What was the impact on other disciplines?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe how the findings, results, or techniques that were developed or improved, or other products from the project made an impact or are likely to make an impact on other disciplines.

“Nothing to Report”

What was the impact on technology transfer?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe ways in which the project made an impact, or is likely to make an impact, on commercial technology or public use, including:

- *transfer of results to entities in government or industry;*
- *instances where the research has led to the initiation of a start-up company; or*
- *adoption of new practices.*

“Nothing to Report”

What was the impact on society beyond science and technology?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe how results from the project made an impact, or are likely to make an impact, beyond the bounds of science, engineering, and the academic world on areas such as:

- *improving public knowledge, attitudes, skills, and abilities;*
- *changing behavior, practices, decision making, policies (including regulatory policies), or social actions; or*
- *improving social, economic, civic, or environmental conditions.*

“Nothing to Report”

- 5. CHANGES/PROBLEMS:** *The PD/PI is reminded that the recipient organization is required to obtain prior written approval from the awarding agency grants official whenever there are significant changes in the project or its direction. If not previously reported in writing, provide the following additional information or state, “Nothing to Report,” if applicable:*

Changes in approach and reasons for change

Describe any changes in approach during the reporting period and reasons for these changes. Remember that significant changes in objectives and scope require prior approval of the agency.

“Nothing to Report”

Actual or anticipated problems or delays and actions or plans to resolve them

Describe problems or delays encountered during the reporting period and actions or plans to resolve them.

Due to the PI team’s research lab being shuttered since March 2020 with an unspecified re-opening time frame, the PI team has been unable to make significant progress to the cell-template interactions (i.e. biological studies) being addressed in Specific Aim 2. As such, the PI team will likely submit a request for a 1-year no-cost extension in order to complete Milestone 2 upon resumption of the experimental work. In the meantime, the PI team will temporarily focus on advancing 3D non-planar toolpath algorithms and workflows required to simulate image-guided in situ bioprinting of a skin graft onto a phantom ulcer wound bed. Although this may expand the scope of the originally proposed specific aims, this will enable the PI team to augment the experimental robustness from both the manufacturing and biological standpoint when the experimental phase of work is permitted to resume.

Changes that had a significant impact on expenditures

Describe changes during the reporting period that may have had a significant impact on expenditures, for example, delays in hiring staff or favorable developments that enable meeting objectives at less cost than anticipated.

The post-doctoral funding allocation originally intended to support the PI’s senior PhD student (Mr. Houzhu Ding) upon graduation (as a post-doc) has been shifted to support another PhD student (Mr. Kai Cao) since the senior PhD student left upon securing employment upon graduation.

Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents

Describe significant deviations, unexpected outcomes, or changes in approved protocols for the use or care of human subjects, vertebrate animals, biohazards, and/or select agents during the reporting period. If required, were these changes approved by the applicable institution committee (or equivalent) and reported to the agency? Also specify the applicable Institutional Review Board/Institutional Animal Care and Use Committee approval dates.

“Nothing to Report”

Significant changes in use or care of human subjects

“Nothing to Report”

Significant changes in use or care of vertebrate animals

“Nothing to Report”

Significant changes in use of biohazards and/or select agents

“Nothing to Report”

6. PRODUCTS: *List any products resulting from the project during the reporting period. If there is nothing to report under a particular item, state “Nothing to Report.”*

- **Publications, conference papers, and presentations**

Report only the major publication(s) resulting from the work under this award.

Journal publications. *List peer-reviewed articles or papers appearing in scientific, technical, or professional journals. Identify for each publication: Author(s); title; journal; volume; year; page numbers; status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).*

The PI team is preparing a following journal manuscript (to be submitted by end of July 2020) supported by this grant:

Authors: Kai Cao, Fucheng Zhang, Robert C. Chang

Title: Effect of collector temperature on charge phenomena during the melt electrowriting process

Journal: Materials and Design

Acknowledgement of federal support: Yes

Books or other non-periodical, one-time publications. *Report any book, monograph, dissertation, abstract, or the like published as or in a separate publication, rather than a periodical or series. Include any significant publication in the proceedings of a one-time conference or in the report of a one-time study, commission, or the like. Identify for each one-time publication: author(s); title; editor; title of collection, if applicable; bibliographic information; year; type of publication (e.g., book, thesis or dissertation); status of publication (published; accepted, awaiting publication; submitted, under review; other); acknowledgement of federal support (yes/no).*

“Nothing to Report”

Other publications, conference papers and presentations. *Identify any other publications, conference papers and/or presentations not reported above. Specify the status of the publication as noted above. List presentations made during the last year (international, national, local societies, military meetings, etc.). Use an asterisk (*) if presentation produced a manuscript.*

“Nothing to Report”

- **Website(s) or other Internet site(s)**

List the URL for any Internet site(s) that disseminates the results of the research activities. A short description of each site should be provided. It is not necessary to include the publications already specified above in this section.

“Nothing to Report”

- **Technologies or techniques**

Identify technologies or techniques that resulted from the research activities. Describe the technologies or techniques were shared.

“Nothing to Report”

- **Inventions, patent applications, and/or licenses**

Identify inventions, patent applications with date, and/or licenses that have resulted from the research. Submission of this information as part of an interim research performance progress report is not a substitute for any other invention reporting required under the terms and conditions of an award.

“Nothing to Report”

- **Other Products**

Identify any other reportable outcomes that were developed under this project. Reportable outcomes are defined as a research result that is or relates to a product, scientific advance, or research tool that makes a meaningful contribution toward the understanding, prevention, diagnosis, prognosis, treatment and /or rehabilitation of a disease, injury or condition, or to improve the quality of life. Examples include:

- *data or databases;*
- *physical collections;*
- *audio or video products;*
- *software;*
- *models;*
- *educational aids or curricula;*
- *instruments or equipment;*
- *research material (e.g., Germplasm; cell lines, DNA probes, animal models);*
- *clinical interventions;*
- *new business creation; and*
- *other.*

“Nothing to Report”

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Provide the following information for: (1) PDs/PIs; and (2) each person who has worked at least one person month per year on the project during the reporting period, regardless of the source of compensation (a person month equals approximately 160 hours of effort). If information is unchanged from a previous submission, provide the name only and indicate “no change”.

Example:

Name: Mary Smith

Project Role: Graduate Student

Researcher Identifier (e.g. ORCID ID): 1234567

Nearest person month worked: 5

Contribution to Project: Ms. Smith has performed work in the area of combined error-control and constrained coding.

Funding Support: The Ford Foundation (Complete only if the funding support is provided from other than this award.)

1. Name: Houzhu Ding
Project Role: Graduate Student
Researcher Identifier (e.g. ORCID ID): N/A
Nearest person month worked: 3
Contribution to Project: melt-based electrohydrodynamic printing process development; programming of translational stage; early process parametric studies
Funding Support: DOD Award # W81XWH-19-1-0158
2. Name: Kai Cao
Project Role: Graduate Student
Researcher Identifier (e.g. ORCID ID): N/A
Nearest person month worked: 3
Contribution to Project: melt-based electrohydrodynamic printing process optimization; design and build of variable temperature collector module; ongoing parametric studies for microscale layered 3D templates
Funding Support: Internal funding & DOD Award # W81XWH-19-1-0158

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

If the active support has changed for the PD/PI(s) or senior/key personnel, then describe what the change has been. Changes may occur, for example, if a previously active grant has closed and/or if a previously pending grant is now active. Annotate this information so it is clear what has changed from the previous submission. Submission of other support information is not necessary for pending changes or for changes in the level of effort for active support reported previously. The awarding agency may require prior written approval if a change in active other support significantly impacts the effort on the project that is the subject of the project report.

“Nothing to Report”

What other organizations were involved as partners?

If there is nothing significant to report during this reporting period, state “Nothing to Report.”

Describe partner organizations – academic institutions, other nonprofits, industrial or commercial firms, state or local governments, schools or school systems, or other organizations (foreign or domestic) – that were involved with the project. Partner organizations may have provided financial or in-kind support, supplied facilities or equipment, collaborated in the research, exchanged personnel, or otherwise contributed.

Provide the following information for each partnership:

Organization Name:

Location of Organization: (if foreign location list country)

Partner’s contribution to the project (identify one or more)

- *Financial support;*
- *In-kind support (e.g., partner makes software, computers, equipment, etc., available to project staff);*
- *Facilities (e.g., project staff use the partner’s facilities for project activities);*
- *Collaboration (e.g., partner’s staff work with project staff on the project);*
- *Personnel exchanges (e.g., project staff and/or partner’s staff use each other’s facilities, work at each other’s site); and*
- *Other.*

“Nothing to Report”

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS: *For collaborative awards, independent reports are required from BOTH the Initiating Principal Investigator (PI) and the Collaborating/Partnering PI. A duplicative report is acceptable; however, tasks shall be clearly marked with the responsible PI and research site. A report shall be submitted to <https://ers.amedd.army.mil> for each unique award.*

QUAD CHARTS: *If applicable, the Quad Chart (available on <https://www.usamraa.army.mil>) should be updated and submitted with attachments.*

9. **APPENDICES:** *Attach all appendices that contain information that supplements, clarifies or supports the text. Examples include original copies of journal articles, reprints of manuscripts and abstracts, a curriculum vitae, patent applications, study questionnaires, and surveys, etc.*