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A portable, low-cost practice model for microsurgical skills training

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Abstract

Purpose We describe a portable practice model for acquisition of microsurgical skills using widely available inexpensive tools and materials as a model in learning ophthalmic corneal suturing skills.

Methods Interested participants without prior microsurgery experience affiliated with the Jacobs School of Medicine and Biomedical Sciences with no prior microsurgical experience qualified to participate. Each participant completed written informed consent. We

Trial Registration: ClinicalTrials.gov NCT04093063

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developed a 3-dimensional micro-stellated icosahedron model using microtubules, monofilament fishing line, jewelers' forceps, and a basic laboratory dissection microscope. We tested this model in improving microsurgical skills in a randomized, controlled intervention trial. Following a pre-assessment task of passing a microsurgical needle and performing a tie, participants were randomized to a control or an intervention (building the micro-stellated icosahedrons) group. The assessment task was repeated after two weeks. Videos of pre- and post-assessments were rated by two masked ophthalmologists. Technique scores and time to complete microsurgical tasks were analyzed to determine improvement in skills.

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Results A total of 27 microsurgically naïve participants were recruited and randomized (14 Intervention / 13 Control). Comparing pre- and post-assessments, the intervention group showed significant decrease in time required to pass the needle $(P = 0.018)$ and significant improvement in technical scores. $(P = 0.001)$. In the control group, there was no significant decrease in time or improvement in technical scores.

Conclusions The portable inexpensive micro-stellated icosahedron skills acquisition model is an effective practice model to acquire skills necessary to perform a microsurgical tie. The similarity in dimensions between the model and the eye suggests translatability to ophthalmic surgery.

Keywords Medical education - Microsurgery - Microsurgical practice - Ophthalmology - Resident education - Surgical education - Surgical skills - Suturing

Introduction

Microsurgery is a vital skill in many surgical fields including ophthalmology, neurosurgery, otolaryngology, urology, and transplantation surgery and providing its training is a requirement of ACGME [\[1](#page-9-0)]. Training in surgical fields has heavily depended on the apprenticeship model, in which senior surgeons work with trainees in clinical settings. This type of personal training requires time and resources that are increasingly limited by constraints in real-world clinical settings. Therefore, this classic model runs the risk of not providing the amount of repetition required to acquire skills [\[2](#page-9-0)].

Deliberate practice is the most important factor that determines a surgeon's performance, and not his or her innate ability $[3, 4]$ $[3, 4]$ $[3, 4]$ $[3, 4]$. Trainees who practice the most are typically the ones who show fastest improvement in skills [[5\]](#page-9-0). In addition, the stress level of the trainee, which directly affects surgical outcome, has been shown to decrease with practice of the different steps of the surgery [[6\]](#page-9-0). There is also evidence that discontinuing continuous practice for a prolonged time can significantly affect microsurgical skill [\[7](#page-9-0)]. These findings stress the importance of continuous deliberate practice.

Different practice methods based on nonliving models have been developed for acquisition of microsurgical skills. These include the use of surgical gauze, silicone tubes, glove boxes, etc. [\[1](#page-9-0), [2,](#page-9-0) [8–10](#page-9-0)]. Other methods are based on the use of animal models [\[1](#page-9-0)]. In ophthalmology, the use of wet laboratories is widely used to teach skills on animal tissues, such as porcine eyes which resemble the size of a human eye [\[4](#page-9-0)]. Some methods have creatively tried to simulate specific tissues; one example is the use of bacon as a substitute for extraocular muscles to practice strabismus surgery [\[11](#page-9-0)]. Virtual surgical simulators, such as the EyeSi simulator for ophthalmic surgery, are also available for practice. These devices use recent advances in computer power to create three-dimensional interactive platforms without the need of animal tissue [[12\]](#page-9-0).

However, there are barriers to implementation and consistent use of these models. An important limitation of wet laboratories and virtual simulators is the high costs incurred by these methods [\[13](#page-9-0)]. Initial costs of wet laboratories are estimated to be \$306,400 with annual costs of \$62,480. The cost of acquiring an EyeSi simulator is estimated to be \$169,000 with annual costs of \$26,590 (hardware/software maintenance, space rental costs, and personnel costs). These costs vary by hospital space required, equipment, and size of the training program [[13\]](#page-9-0). Physical access to these resources (e.g., timing of laboratory access, number of students that can practice at a time) can further interfere with the continuous and regimented practice required for proficient training.

Virtual simulators have been shown to have appropriate validity in acquiring specific microsurgical skills [[12\]](#page-9-0). Studies have shown that skills related to one specific microsurgical task do not necessarily translate to improved skills in other tasks [[14\]](#page-9-0). In other words, practicing skills in virtual simulators for cataract surgery are unlikely to improve corneal suturing skills. In addition, it is still not clear if practicing specific skills for parts of a complete surgery are sufficient to reduce surgical complications [\[15](#page-10-0)].

Because of the limitations in access and cost with current microsurgical skills practice methods, we developed a portable, inexpensive microsurgery training model with simple materials available in most parts of the world. We performed a randomized controlled trial to test the hypothesis that building this micro-stellated icosahedron model (Fig. 1) would result in improved surgical technique and efficiency for completing a microsurgical suture pass and tie.

Methods

This study is a single-center, prospective, randomized trial approved by the University at Buffalo Institutional Review Board (STUDY00001491). Medical students, residents, and other interested individuals without prior microsurgery experience affiliated with the Jacobs School of Medicine and Biomedical Sciences were invited to participate. All of the participants included in the study were asked at the beginning of the study whether or not they had had prior microsurgical experience; only those who reported no prior experience qualified to participate. Each participant completed written informed consent. This study adhered to the tenets of the Declaration of Helsinki.

Materials

The materials used for developing the microsurgical skills model and that were provided to each participant in the intervention group to complete the study task are detailed in Fig. 1.

The materials for the video-recorded pre- and postassessments were: silicon baking sheet (10 cm \times 7 $cm \times 2$ mm) adhered to a plastic container lid $(11.3 \text{ cm} \times 7.8 \text{ cm} \times 2.5 \text{ cm})$, and a stereoscopic dissecting microscope on a boom stand (Bausch & Lomb, Rochester, NY). The participants were provided with a 9–0 nylon suture (Epsilon USA, Chino, CA) and standard ophthalmic microsurgical instruments: curved non-locking needle holder, 0.12 mm toothed forceps, and curved and straight tiers. Video recordings were taken with a personal cell phone camera attached by clamp on a laboratory stand.

Study protocol

Pre- and post-assessments were performed in vacant classrooms at the Jacobs School of Medicine and Biomedical Sciences, University at Buffalo.

Participants first watched an instructional video made by the Principal Investigator on how to pass a 9–0 nylon suture needle, and how to perform a microsurgical tie for a linear incision in a silicon baking mat using the same materials as would be provided to them for the pre- and post-assessments (Video 1). After watching the video three consecutive times, participants were tasked to pass and tie a 9–0 nylon suture in the same manner as the video recording. Their attempt at performing the microsurgical task was video-recorded for a pre-assessment with a cellphone camera focusing on the microscope field ensuring a clear view of the surgical field and the participant's instrument manipulation. Each subject

Fig. 1 A, Micro-stellated icosahedron. B, Stereoscopic dissecting microscope (Bausch & Lomb, Rochester, NY). C, Scalpel blade (#15) (1), jeweler style forceps (Economy Tweezers #3, World Precision Instruments, Sarasota, FL) (2), curved-tip micro scissors (Curved Iris Scissors with Spring 4.75'', Sona Enterprises, Mumbai, India) (3), polyimide microtubule material (Code 175-I.5, Inner diameter 0.44 mm, Outer diameter

0.52 mm, Microlumen, Oldsmar, FL) (4), double-sided tape (5), monofilament nylon thread (NanoFil Uni-Filament Fishing Line 1 lb break-strength, Berkley, Spirit Lake, IA) (6), 15 cm metal ruler (7). D, Comparison of the dimensions of 9–0 nylon suture (Epsilon USA, Chino, CA) (1) to dimensions of materials provided: polyimide microtubule material (2), monofilament nylon thread (3)

wore gloves during the assessment to ensure anonymity.

After completion of the video-recorded pre-assessment, simple randomization was performed by allocating participants to either the intervention or the non-intervention control group by flip of a coin. All subjects in both groups were required to return in two weeks for a second in-person meeting. At the second meeting, they were presented with the same instructional video as in the first meeting on how to pass a needle and perform a microsurgical tie. After watching the video three consecutive times, their attempt at completing a microsurgical tie was video-recorded for a post-assessment, using similar instruments and setup as those used two weeks prior. Subjects in the intervention group were asked to return borrowed materials and their completed micro-stellated icosahedrons.

Intervention group

Subjects in the intervention group were tasked with building a micro-stellated icosahedron using a detailed instruction manual (supplementary material). They were each provided with the materials listed above in the Materials section to complete the task at home at their leisure.

Control group

Subjects in the non-intervention control group were not given any task or any materials. Each participant in the intervention and control group was given a followup call at one week. During this call, participants were reminded to return at the end of the two-week period for post-assessments. Participants in the intervention group were reminded to complete the icosahedron model at their leisure. Participants in the control group were reminded to avoid any microsurgical practice until the post-assessment.

Video randomization and de-identification

After all subjects were recruited, and their corresponding microsurgical tasks were video-recorded for pre- and post-assessments, each of the videos was edited on Wondershare Filmora (Wondershare Technology Group Co., Ltd., Shenzhen, China) to mute all audio in order to ensure anonymity. Each of the videos was de-identified by naming it with a random number and saved in a file folder in random order. This file folder was sent separately to two ophthalmologists, masked to the identity of the subjects (intervention vs control) and time of recording (pre- or postassessment).

Video-based skills assessment

The raters used the Video-based Modified Objective Structure Assessment of Technical Skill (OSATS) Scoring Criteria, which scores four criteria with scores from 1 to 5: Economy of Movement, Confidence of Movement, Respect for Materials, and Precision of Operative Technique [[4\]](#page-9-0). The raters assigned separate scores for the participant's attempt at passing the needle through the incision, and for their attempt at tying a microsurgical tie. The attempt at passing the needle (Pass: Total) had a maximum possible score of 20, and the attempt at tying a microsurgical tie (Tie: Total) had a maximum possible score of 20, making 40 the maximum possible total score for each video (Total: Pass $+$ Tie). Both raters initially viewed six random non-identified assessment videos together to ensure that the scoring criteria were interpreted consistently prior to independent review.

The time each subject took to pass the needle, and the time each subject took to attempt tying a microsurgical tie was measured in number of seconds. The time in seconds each subject took to pass the needle was measured from the time subject started manipulating the needle holder and toothed forceps to the time the subject began tying maneuvers. The time in seconds each subject took to attempt tying a microsurgical tie was measured from the time the subject started manipulations for tying to the time the subject completed the microsurgical tie.

Main outcomes and measures

Time to complete microsurgical assessment tasks, and scores for passing a needle and completing a microsurgical tie.

Sample size calculation

A significant difference in mean scores for either passing a needle or completing a microsurgical tie between the control and intervention groups was determined to be a difference of 5 points. Significance level alpha was chosen at 0.05, and beta was chosen at 0.20 for detection power of 80%. Using these values, a sample size of 10 participants in each of the control and intervention groups was determined as appropriate for the study. The formula: 2 x [(alpha multiplier + beta multiplier)²x (standard deviation of scores) 2] / (mean intervention group – mean control group^2 , was used for sample size calculation for a continuous outcome [\[16](#page-10-0)].

Data analysis

For every participant, the scores for each of the individual OSATS criteria given by both raters were added. The sums were combined for all participants by intervention/control group and pre/post assessment. The averages of the sums for each group were compared. Paired t test was used in IBM SPSS Statistics (IBM Corporation, Armonk, NY) to calculate difference in means in time and grading between pre- and post-assessments among participants in the intervention and control groups, respectively. Wilcoxon signed-rank test was conducted in R (University of Auckland, Auckland, New Zealand) to calculate if improvement in time and grading among subjects in the intervention group was greater than improvement among subjects in the control group.

Cronbach's alpha was used in SPSS to calculate inter-rater reliability between the total score (Pass:To $tal + Tie:Total$) given by each of the raters for each of the videos. Intra Class Correlation (ICC) [\[17](#page-10-0)] was used in R to calculate consistency in scores in each of the groups between the two raters.

Results

Subject recruitment and randomization

A total of 27 participants were recruited and consented for the study from May 2017 to July 2018 (Fig. [2](#page-5-0)). After pre-assessments were recorded, participants were randomized by flip of a coin. Fourteen participants were assigned to the intervention group. Of these, three were withdrawn from the study: one voluntarily withdrew, another was not able to start building the stellated icosahedron within two weeks and the last was not able to return for the second inperson meeting within the allotted two weeks. Thirteen participants were assigned to the control group. Of these, three were withdrawn from the study: one voluntarily withdrew and two were not able to return for second in-person meeting within the allotted two weeks.

After video recording all pre- and post-assessments of subjects enrolled in the study, a total of 42 videos were analyzed by masked graders. The 42 videos included: 11 pre-assessment and 11 post-assessment videos from the intervention group, and ten preassessment and ten post-assessment videos from the control group.

Inter-rater reliability data

The scores were initially analyzed to calculate difference in means and standard deviation between each rater's pre- and post-assessment scores. The total score assigned by each rater for three of the videos was found to be more than two standard deviations away from each other. These videos were de-identified and randomly sent to the raters for re-scoring. For the scores given by each of the raters to all of the 42 videos, Cronbach's alpha was 0.89. The ICC results for the following groups of videos are: Intervention group pre-test, 0.88 (95% CI 0.54 to 0.97, $P = 0.001$); Control group pre-test, 0.92 (95% CI 0.68 to 0.98, $P < 0.001$); Intervention group post-test, 0.86 (95%) CI 0.49 to 0.96, $P = 0.002$; Control group post-test, 0.81 (95% CI 0.25 to 0.95, $P = 0.01$).

Needle Pass

If a participant took multiple attempts to pass the needle during either the pre- or post-assessment videos, only the duration for the first attempt was measured, and only the first attempt was scored. The number of subjects in each group who made this error was: Intervention, $n = 2$; Control, $n = 2$ (Table [1](#page-5-0)). These participants were excluded from the calculations for time required to pass the needle.

Intervention group: There was a significant decrease in time required to pass the needle between pre- and post-assessments ($n = 9$; $P = 0.018$). There was no significant improvement in technical scores for any of the OSATS criteria for passing the needle between pre- and post-assessments $(n = 11,$ Table [2](#page-6-0)).

Figure 2 Flow diagram displaying subject recruitment and randomization

Table 1 Errors in Intervention and Control groups. Each row represents a single participant. These participants were excluded from calculation for time

	Intervention group errors			
Pass		Tie		
Pre	Post	Pre	Post	
		X		
		X		
		X		
		X		
X		X		
X		X		
	Control group errors			
		X		
X		X		
		X		
X	X	X	X	
		X	X	

Control group: There was no significant decrease in time required to pass the needle between pre- and postassessments $(n = 8)$. There was no significant improvement in scores for any of the OSATS criteria for passing the needle between pre- and post-assessments ($n = 10$, Table [3\)](#page-6-0).

The decrease in time required to pass the needle among subjects in the intervention group was not significantly greater than the decrease in time in the control group. The increase in score for passing the needle among subjects in the intervention group was

not significantly higher than the increase in score in the control group (Table [4](#page-7-0)).

Microsurgical tie

If a subject took multiple attempts at microsurgical tie during either the pre- or post-assessment videos, only the duration for the first attempt was measured, and only the first attempt was scored. If a subject did not attempt tying a microsurgical tie during either pre- or post-assessment videos because suture was pulled through while passing the needle, a technical score of 0 was assigned for ''Tie: Total.'' If subject was not able to complete a microsurgical tie during either preor post-assessments, but attempted tying, a technical score of 0 was assigned for ''Tie: Precision of Operative Technique.'' The number of subjects in each group who made these errors was: Intervention, $n = 6$; Control, $n = 5$. These participants were excluded from the calculations for time required to make a microsurgical tie (Table 1).

Intervention group: There was no significant difference in the time required to complete the microsurgical tie between pre- and post-assessments $(n = 5)$. There was significant improvement in technical scores for all the OSATS criteria and in the total score for attempting a microsurgical tie between preand post-assessments ($n = 11$, Table [2\)](#page-6-0).

Control group: There was no significant decrease in the time required to complete the microsurgical tie

Table 3 Summary of results for control group (${}^*p \le 0.05$)

between the Pre- and the Post-assessments $(n = 5)$. There was significant improvement in technical scores for only one OSATS criterion for attempting a microsurgical tie between Pre- and Post-assessments: Precision of Operative Technique (Table 3). There was no significant improvement in any of the other scores or in the total score.

Subjects who committed the errors mentioned above were excluded from the calculations in time to complete the tasks. These subjects were excluded to

	Mean decrease in time (SD)	P	Mean Increase in total OSATS score (SD)	P
		value		value
Control:	60.25 (93.76)	0.407	1.60(4.78)	0.275
Needle Pass				
Intervention:	72.67 (73.30)		1.98(4.45)	
Needle Pass				
Control: Microsurgical Tie	133.00 (222.20)	0.726	2.70(4.25)	$0.031*$
77.20 (333.69) Intervention: Microsurgical Tie			6.09(4.37)	

Table 4 Comparison of improvement in time and improvement in total objective structured assessment of technical skills (OSATS) scores for needle pass and microsurgical tie between intervention and control groups. (*p ≤ 0.05)

avoid skewing the averages toward faster times. For example, recording time required to complete a microsurgical tie as zero seconds would make a subject appear extremely fast at this task. However, there was no attempt at making a tie because they pulled the suture through completely across the incision. Subjects who pulled suture through while attempting to make a tie were also excluded because there was no way of controlling for how quickly they could have pulled the suture through. Subjects who had multiple attempts at a task were also excluded because it was not possible to control for the large variation in times between the different attempts.

The decrease in time required to make a microsurgical tie among subjects in the intervention group was not significantly greater than the decrease in time in the control group. However, the increase in technical scores for microsurgical tie was significantly greater among subjects in the intervention group than the increase in score in the control group (Table 4).

Discussion

Face validity exists when simulated tasks resemble maneuvers performed during a real-life surgical procedure [[1\]](#page-9-0). Our data shows that after two weeks of practicing on our model, there was significant improvement in all aspects of completing a microsurgical tie. Our model requires at least 55 ties to complete, demonstrating its face validity in attempting to resemble the maneuvers to form a microsurgical tie. It attempts to simulate passing a needle by passing fine fishing line through the microtubules, but the model does not contain a needle that is passed through any type of simulated tissue. This could explain why there was no significant improvement in needle pass after two weeks of practice. These results support the idea that skills training is highly specific, and that skills in one task do not cross over to other tasks. [[15\]](#page-10-0).

We further increased the face validity of our model for ophthalmic surgery by mimicking the dimensions of the human eye and using tools and materials as close to those of standard ophthalmic microsurgical supplies. The horizontal diameter of the cornea in normal eyes ranges from 11 to 12.6 mm [[18\]](#page-10-0). The approximate diameter of our model is approximately 16 mm in its final form. In addition, the anterior chamber depth is an average of 3.24 mm among normal eyes in the United States [\[19](#page-10-0)]. Each segment of microtubule material required to build our practice model has a length of 4 mm, which approximates the anterior chamber depth of normal eyes. Mimicking the dimensions of the eye allows trainees to develop their micromanipulation skills within the range of distances necessary to manipulate within the structures of the eye.

Our results are generalizable to any trainees without prior microsurgical experience. While we recruited broadly, our enrolled microsurgically naïve individuals were medical students at various stages in their training. It has been demonstrated that eye-hand coordination specifically relies on precise control of ocular and appendicular sensorimotor systems to accomplish a single goal, such as touching a visual target [\[20](#page-10-0)]. Several studies have reported that both brief and extensive exposure to oculo-manual activities such as video-game play can result in a broad range of enhancements to various cognitive faculties [\[21](#page-10-0)]. The driving principle for this study is that microsurgical skill is not determined by innate characteristics of the microsurgeon, but can be learned with practice $[3, 4]$ $[3, 4]$ $[3, 4]$.

Because subjects interested in microsurgery are more likely to volunteer, volunteer bias was an important factor to address. By using flip of a coin, allocation of subjects into the control and intervention groups was done randomly to avoid any bias from the subject or the study manager. Therefore, randomization allowed participants who were likely to be eager to learn a new skill to have an equal chance to be in either group, reducing the effect of volunteer bias in our study.

Our results show that control subjects showed improvement in certain aspects of the microsurgical tasks, namely improvement in time required to pass the needle, and improvement in Tie:Precision of Operative Technique. This result suggests that any intervention can be valuable for the microsurgically naïve specially when this intervention is a complex tridimensional manual task. Table [1](#page-5-0) shows that subjects in the intervention group were less likely than those in the control group to require multiple attempts for a task, or pull the suture through after practicing on our model for two weeks. Subjects in the intervention group made these ''errors'' only during the pre-assessment videos, while those in the control group made them during either pre- or post-assessment videos, or during both. One limitation of our study is that due to those errors, the data for those participants were excluded from the calculations for time. Therefore, the sample size used in those calculations was smaller.

One of the major drawbacks of other systems for microsurgical skills practice is cost and availability. Our model circumvents these issues by its low cost and high portability. The cost of materials may vary by supplier, but a complete set of instruments and materials for practice for one individual as described in the Methods section (excluding the microscope) was approximately \$20. This is less than the cost of most 10–0 nylon sutures that may be used in a practice laboratory. The dissecting microscopes, which could potentially be the costliest item required for our model were easy to obtain. Emails to faculty at the Jacobs School of Medicine and Biomedical Sciences turned up several similar microscopes there were not being used. We also noted that similar microscopes were available online in used condition for less than \$100.

The microscopes are not consumable items and could be cycled between trainees. Finally, the size and weight of materials are of low burden, adding to the model's portability, thus allowing trainees to practice at their own leisure in convenient locations. Combining these benefits promotes the constant and deliberate practice required to acquire microsurgical skills.

It is important to note that our practice model has the limitation of improving only the microsurgical skill it reinforces, making a microsurgical tie. Because skills training is highly specific, our model may not be applicable to acquire other micromanipulations such as needle pass, or cataract surgery micromanipulation techniques [[12\]](#page-9-0). However, we hope to design future studies evaluating whether building a micro-stellated icosahedron helps in accelerating the acquisition of future microsurgical skills.

Although many practice models have been developed in microsurgery, there is lack of evidence in their validity as effective tools of skill acquisition. Most of these models have not been validated by randomized controlled trials that compare skills before and after their use by volunteer subjects. One systematic review published in 2014 determined that only nine studies, out of 238 published reports on surgical simulation models, used subject groups to analyze validity and effectiveness on skill acquisition [\[2](#page-9-0)]. Our study was designed as a randomized control trial, adding to the validity of our model as a tool to acquire skills.

The most detailed recent studies on surgical skills models have reported on microsurgical skills acquisition using the Eyesi virtual surgical simulator. There are several similarities and differences between our model and the Eyesi. One big difference is cost. Our model was developed at a cost of approximately \$20 per set of materials, whereas the Eyesi cost is over \$100,000. Our model is also portable, allowing trainees access to practice at their convenience in any location while Eyesi instrumentation is limited to practice at the institution where it is located. With regards to skills acquisition, similar to our model, training with Eyesi results in improvement of the skills that are practiced. The multiple modules available with the Eyesi allow practice of multiple skills such as navigation, antitremor, forceps, bimanual training, capsulorhexis, and phacoemulsification [[22–27](#page-10-0)], whereas our current model is focused on skills of micromanipulation and microsurgical suture ties, and interestingly the latter skill has not been included in Eyesi training platform. We have not tested the impact of training with our model on real-life surgical outcomes. Non-randomized studies with the Eyesi have shown improvement in scores for Objective Structured Assessment of Cataract Surgical Skill and reduced complications in actual surgeries post- compared to pre-Eyesi training. [\[22–25](#page-10-0)].

The apprenticeship model in clinical settings can present ethical concerns when practicing on real patients. As trainees experience a learning curve, patients suffer consequences in the form of inevitable complications [4]. For these reasons, surgical training has shifted toward using safer methods that simulate surgical environments. This strategy promotes the development of skills in a graduated process, in which nonliving models are used to acquire skills before practicing in living models and in real scenarios [1, 4, 10]. Our model offers the benefit of regimented practice without the need to use living models or real patients. It allows trainees to increase their skills and become more confident under the microscope before attempting microsurgery on real patients.

Author contributions HDG-A and SPP designed the study. HDG-A, AMF, and SPP performed the study. HDG-A, CC and JZ analyzed the data. HDG-A wrote the first draft of the manuscript which was edited by all authors. SPP supervised all stages of the study and manuscript preparation.

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Data availability All data are available upon request.

Declarations

Conflict of interest The authors have no relevant financial or non-financial interests to disclose.

Ethical approval This study was approved by the Institutional Review Board at the University at Buffalo (STUDY00001491) and adhered to the principles of the Declaration of Helsinki.

Consent for participate Informed consent was obtained from all individual participants included in the study.

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