Prevention of Surgical Site Infection in Colorectal Surgery Through the Use of Silver Nanoparticle Absorbable Sutures

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ABSTRACT

Surgical site infections (SSIs) are a pressing clinical issue that costs the U.S. billions of dollars annually and increases mortality rates significantly. To prevent such infections, doctors often prescribe oral antibiotics before and after surgical procedures. Antibiotics, such as triclosan, can also be applied to suture coatings and woven into suture fibers to prevent infection. A major issue with antibiotic use is that some bacteria have mutations that lead to antibiotic resistance. Bacteria can then spread these resistance genes within microbial communities, decreasing the effectiveness of a drug against fighting infections. The use of silver nanoparticles (AgNPs) is a newly researched alternative to antibiotic treatment to prevent SSIs. While it is known that AgNPs have antimicrobial properties, their specific infection-fighting mechanisms are unknown. In this proposal, we seek to conduct an *in-vivo* experiment on mice to test the effectiveness of triclosan-coated Vicryl sutures versus AgNP-coated Vicryl sutures in preventing SSIs during colorectal surgery. Our methods consist of three main stages: suture preparation, *in-vivo* suture application in mice, and a bacterial enumeration assay. We hope to find that the AgNP-coated sutures are as effective as the triclosan-sutures in preventing SSIs in mice. We believe our study addresses the One Health goal by seeking alternatives to antibiotics in medicine that negatively impact the environment as well as animal and human health.

SECTION I

Surgical site infections (SSIs) are post-operative infections caused by the growth of bacteria on the operated body. They occur in about 1-3% of patients that have received a surgery[10]. While that percentage may seem low, there are serious implications for SSIs; they cost the U.S. an additional $10 billion annually, lead to increased mortality rates, and result in an extra 400,000 days of hospitalization[12]. Out of the numerous surgical procedures, colorectal surgery, a procedure used to treat disorders in the rectum, anus, and colon, has a relatively high SSI rate due to the

dispersal of microorganisms from contents within the bowel. The chances of developing an SSI post-colorectal surgery ranges from 3% to 30%, and it can reach as high as 40% if antibiotics are not used[6].

Traditionally, antibiotics given before and during surgery can limit the risks of SSIs by as much as 75%[6]. These antibiotics are typically administered orally or via antibiotic-coated sutures. Current treatments for colorectal surgery, along with many other surgical procedures, include triclosan, an antimicrobial agent that is commonly found in soap and toothpaste. However, both gram-negative and gram-positive bacteria are growing increasingly resistant to antibiotics like triclosan, and this has led to 2.8 million infections and 35,000 deaths in the U.S. annually[2]. Additionally, triclosan has a multitude of harmful environmental effects. Due to triclosan's widespread use in consumer products, it is one of the most frequent contaminants in our country's waterways. Thus, there is a push to find antibiotic alternatives for the treatment of SSIs as it affects the health of people, animals, and the environment. Silver nanoparticle (AgNP) treatment is a newly arising method in reducing SSIs. These are small particles of silver that range between 1 nm and 100 nm in size[17]. AgNPs have antimicrobial properties as seen in laboratory settings. Due to their nanoscale size, they have the ability to penetrate the cell walls of bacteria that have acquired antibiotic resistance. In addition, AgNPs are not yet widely used in clinical settings, so there is low risk of bacterial resistance.

We propose that AgNP-coated Vicryl sutures are as effective in combating SSIs in colorectal surgery as triclosan-coated Vicryl sutures. While nanotechnology is rapidly emerging in clinical applications, much is still unknown about its specific capabilities in targeting SSIs. What is known is that AgNPs can serve as an antimicrobial agent, and it has relatively few adverse effects. By conducting more research, we hope to discover that AgNPs will both decrease the rate of SSIs in colorectal surgery and have minimal harmful impacts on the environment.

SECTION II

The antiseptic triclosan is used in many quotidian products. It kills bacteria by inhibiting fatty acid synthesis, which consequently inhibits lipid synthesis and kills the cell[7]. However, bacterial resistance to triclosan has been observed. There have been numerous occurrences of triclosan resistance in the skin, intestine, and environmental

microorganisms since 2000[13]. Triclosan may also contribute to reduced susceptibility to antimicrobials in health settings through cross-resistance or co-resistance mechanisms[13]. It also poses many environmental threats. When triclosan is exposed to sunlight in an aqueous environment, such as our waterways, it is converted into a toxic compound called dioxin. Triclosan can also react with chlorine to form chloroform, which is a possible human carcinogen. Triclosan is highly toxic to algae, which are keystone species in many aquatic ecosystems, and it may serve as potential endocrine disruptors in other aquatic organisms, which would lead to reproductive defects[9]. With increased exposure to triclosan, bacteria will learn how to bear normally deadly amounts of this antibiotic[11]. This is a significant threat to the global population as it encourages the rise of superbugs and endangers the efficacy of infectious disease treatments. Additionally, triclosan accumulates in the body and can be traced in the urine of three-quarters of the American population. Thus, it is imperative to stray away from increased triclosan use.

With increasing antibiotic resistance developing in bacteria, scientists are looking towards alternatives that exhibit both antimicrobial properties and a low likelihood of bacteria developing resistance. One such alternative is silver nanoparticles (AgNPs) which have been known to have antimicrobial properties for centuries. But only recently were researchers able to produce them as an antimicrobial agent at a nanoscale[8]. Much of the exact mechanism of how AgNPs can kill bacteria remains unclear; however, it is known that AgNPs can conglomerate on bacterial cell walls, causing the cell membrane to denature. This denaturation can disrupt the function of organelles and even cause the cell to lyse. AgNPs can also impact signal transduction pathways in the bacteria by dephosphorylating tyrosine residues which can lead to cell apoptosis. Compared to gram-positive bacteria, gram-negative bacteria are more prone to penetration by AgNPs due to their thinner cell wall[14].

As the rise of SSIs and antimicrobial resistance become more of a problem in post-surgical procedures, novel research has explored alternatives, such as silver nanoparticle coated sutures, to go beyond traditional antibiotic suture use. A study done in 2011 examined the efficacy of AgNPs layered onto surgical sutures against *Staphylococcus aureus*, a gram-positive bacteria. The AgNPs were capped with sodium alginate in concentrations from 0.1 mM to 0.5 mM and deposited in the sutures in a layer-by-layer fashion. Ultimately, the 0.1 mM

concentration of sodium alginate was found to have the greatest antimicrobial activity[4]. More research must be done to examine the effectiveness of silver nanoparticle-coated sutures in decreasing SSIs.

The effectiveness of silver nanoparticles coated absorbable sutures compared to triclosan coated absorbable sutures in combating SSIs from colorectal surgery remains unknown, and thus the comparison of these treatments will be the focus of our study.

SECTION III

Material and Methods: Based on previous research involving *in-vitro* use of AgNPs against bacterial strains, we chose to design an *in-vivo* experiment in mice testing absorbable sutures compared to triclosan antibiotic coated absorbable sutures. As shown in Figure 1, our methods consist of three main stages: 1. Suture Solution Preparation, 2. In-Vivo Application of Sutures within the Colons of Mice, and 3. A Bacterial Enumeration Assay.

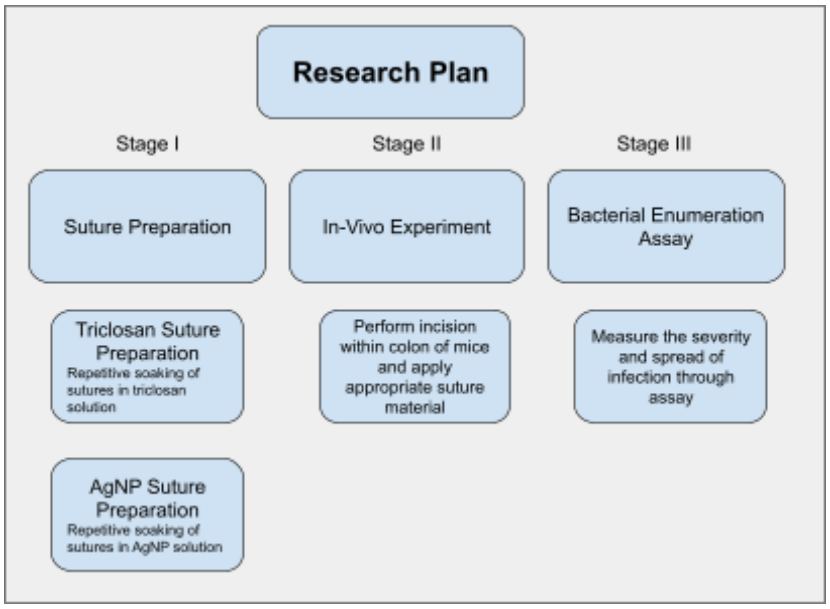

Figure 1. Research plan summary.

Stage 1: Suture Preparation

Three types of sutures will be prepared as shown in Figure 2. These groups are non-coated Vicryl absorbable sutures (control group), triclosan coated Vicryl absorbable sutures, and silver nanoparticle coated Vicryl absorbable sutures.

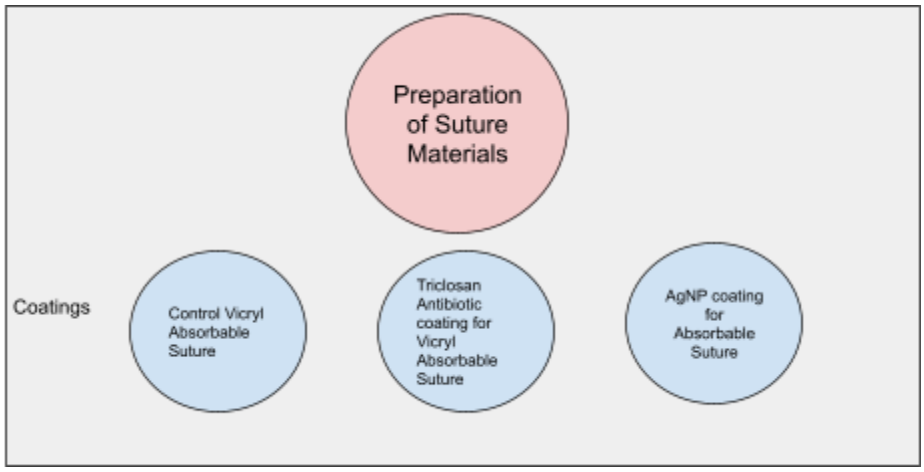

Figure 2. Suture preparation.

Triclosan Coated Vicryl Sutures Preparation

We will prepare a set concentration of triclosan solution. Sterile Vicryl absorbable sutures will then be dipped into the triclosan solution, dried, and dipped again. This process will be repeated for multiple sutures.

Silver Nanoparticle Solution Preparation

The environmental impact of the AgNP solution can be minimized by using non-chemical procedures. One such non-chemical procedure allows for the production of AgNPs at a constant size. During this procedure, silver graduates are first distilled in silver vapor with a ceramic conductive element. This distillation is then used to produce AgNP liquid droplets. To produce condensed AgNP spheres, the droplets are placed in flowing helium gas[8]. For our research purpose, the last step will be omitted to produce a final product of AgNP liquid droplets.

Silver Nanoparticle Coated Vicryl Sutures Preparation

The AgNP droplets will be diluted in a solution of distilled water. Sterile Vicryl sutures will be dipped in this solution, dried, then re-dipped to produce AgNP coated sutures. This procedure was similarly done by Dubas et al., 2011 with a varying concentration of AgNP and capping agents. For our research purposes, we will keep a constant concentration of AgNPs dipping solution.

Stage 2: Animal Experiment

30 mice of similar weight and age range were chosen and separated into 3 different groups. The first group will be a control group where plain, sterile Vicryl sutures are used. The second group of mice will be treated with

triclosan-coated Vicryl sutures. The third group will be treated with silver nanoparticle-coated Vicryl sutures. Approval from the animal ethics committee is needed before proceeding with *in-vivo* research on mice.

Stage 3: Bacterial Enumeration Assay

We will quantify the number of bacterial colonies through the use of a bacterial enumeration assay. In addition, we will qualitatively assess the colon tissue reaction from the sutures and monitor the spread of infection.

Results and Discussion: Due to the growing body of evidence of the antimicrobial activity of silver nanoparticles, we expect that the silver nanoparticle coated sutures will perform as well as triclosan coated sutures in preventing SSIs. We will be able to compare the antibacterial properties of each treatment through a bacterial enumeration assay and then use statistical analysis to determine the significance of the results. If our hypothesis is correct, we will observe statistically more or equal counts of bacterial colonies in mice treated with triclosan coated sutures than those treated with silver nanoparticle coated sutures. If our hypothesis is incorrect, we will observe statistically fewer counts of bacteria colonies in mice treated with triclosan-coated sutures than those treated with silver nanoparticle coated sutures.

If the expected results are not met and the silver nanoparticle coated sutures are ineffective or less effective, three possible changes can be made to the protocol. First, if the silver nanoparticle coated sutures show antimicrobial activity, we could combine silver nanoparticle coated suture use with an additional antimicrobial drug to reach the same level of microbial killing as triclosan and other common antibiotics. This synergistic approach could potentially improve the ability of these antibiotic alternatives and serve as a safer and equally effective approach to killing infectious agents.

A second way in which our method can be altered is by performing trials *in vitro* rather than *in vivo* with mice to understand the mechanism of silver nanoparticles. Moreover, this change in the protocol could potentially show what conditions or environments allow silver nanoparticle use to be the most beneficial, providing greater insight into the limitations of silver nanoparticles when used with sutures.

Finally, we could alter the protocol to test different sutures with various types of drugs and bacteria, such as gram-negative and gram-positive bacteria. We would isolate strains known to specifically cause SSIs in patients

receiving colorectal surgery and test current antibiotics and silver nanoparticle use on these strains while also observing their efficacy with different suture materials. This way, we could develop an understanding of which combinations of antimicrobial drugs interact well with certain suture materials to prevent infection against dangerous bacterial strains.

Conclusion and Future Studies: The next step of this study would be to research the use of silver nanoparticle-coated sutures in humans. If our experimentation in mice is successful, meaning that there is clear prevention of infection with the use of silver nanoparticle coated sutures and no observed toxicity effects, then our results would support additional research in human trials. Human trials would demonstrate the true potential of silver nanoparticle sutures for colorectal surgery and others. If the trials move forward successfully, then these sutures can be applied to multiple types of surgery, including veterinary procedures as well. Additionally, the use of silver nanoparticle sutures would decrease the risk of antibiotic resistance, thus stopping the cycle of antibiotic use which negatively impacts environmental, animal, and human health.

It is important to mention that although mice models are helpful to determine the biocompatibility and effectiveness of the proposed sutures, they do not appropriately represent the biological conditions of the human body, and thus, suture use requires further experimentation in human models as well. For example, there is a large difference in gut microbiota, which can cause a difference in susceptibility in contracting SSIs. There are also clear anatomical differences, hence the location of a colon in mice might not accurately represent the environment surrounding a human colon. Humans also have more uncontrollable factors, such as pre-existing health conditions. The mice used are limited in their differences from each other, but when compared to humans, additional considerations need to be taken into account.

Moreover, the toxicity of silver nanoparticles is conflicted amongst the scientific community. Some studies state that silver nanoparticles pose little to no harm to the human body, whereas others state that they do pose harm to the human body. Therefore, more studies on the toxicity of silver nanoparticles must be done to safely apply silver nanoparticles in the clinical setting.

Overall, our study will contribute to the field of suture development and support the One Health goal of providing solutions to positively impact multiple communities. Our research of an antibiotic alternative in sutures has the potential to reduce antibiotic resistance threats, thus benefiting the environment, animal, and human health.

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