



SILVER NANOPARTICLES AS TOPICAL MISSILE FOR WOUND HEALING

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ABSTRACT

Introduction: Recently the Nano-silver particles coated on conventional silk stitch as the new emerging technique in the nanotechnology industry have been the fastest growing products. Its antimicrobial feature, led to increasing number of its medical applications. Some of such products already available in the market include wound dressings, contraceptive devices, surgical instruments and bone prostheses

Aim: To evaluate physical, antimicrobial and healing effect of nano-silver suture on periodontal flap.

Objectives: To compare tensile strength, coating of silver, clinical and microbiological parameters of nano-silver and silk suture.

Methodology: In this split mouth randomized controlled study, 10 patients were selected indicated for periodontal flap surgery. One quadrant was given a silk suture (control group) and the other quadrant was given nano-silver suture (test group). Sutures were removed at 7th day post operatively, added to the normal saline for microbial colony count for both the groups. This suspension was then used for streaking the blood agar plates. Wound healing index was assessed at baseline and 1 month. Assessment of Ag NPs impregnated suture threads was performed through inverted electron microscopy and spectrophotometry.

Results: Nano-silver suture showed enhanced physical, clinical and microbiological parameters as compared to silk suture. Conclusion: Silver particles due to their beneficial effects as antibacterial and wound healing accelerator in the periodontal surgeries could reduce inflammation and promote healing process.

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INTRODUCTION

Recently the Nano-silver particles (NS) coated on conventional silk stitch as the new emerging technique in the nanotechnology industry have been the fastest growing products. Its antimicrobial feature, led to increasing number of its medical applications. With the help of the improving technologies, it is now possible to incorporate functional bioactive substances to enhance the potential capability of the sutures. Silver has recently been used as an effective antimicrobial agent that is thought to be a potential solution to the problem of multidrug-resistant microorganisms. When compared to current antibiotics, silver performs against microorganisms by attaching the cell membrane and penetrate inside. The nanoparticles preferably attack the respiratory chain of the microorganisms, cell division and cause to cell death.¹

The term “Nano-Silver” refers to the nanoparticles of silver with a size between 5-50 nm. The active surface of Nano-Silver is 4 m²/g compared to 1-2 m²/g of commercial silver powder². During the chemical reduction, the reducing agent donates electrons to the silver ions (Ag⁺), causing silver to revert to its metallic form (Ag⁰). By controlling the experimental conditions including the temperature, energy input, presence of capping agents, the reaction kinetics could be influenced including the clustering silver atoms form NS of Nano scale dimensions Oral cavity is a normal habitat for several bacterial species, many of which are capable of causing acute and chronic infection if any cut or wound appears on the epithelium.

Patients with Immuno deficiencies and cancerous patients, due to weak immune system experienced more delay in their wound healing. Improving wound healing and decreasing the microbial flora enhance defense system so antiseptic materials for periodontal surgeries and oral wounds have received important attention. Regenerative, respective surgeries, oral implants and reconstructive dentistry, without controlling the microbial activity will fail.³

Tian *et al.* investigated the wound-healing properties of silver nanoparticles in an animal model and found that rapid healing and improved cosmetic appearance occur in a dose dependent manner. Furthermore, through quantitative PCR, immunohistochemistry, and proteomic studies, they showed that silver nanoparticles exert positive effects through their antimicrobial properties, reduction in wound inflammation, and modulation of fibrogenic cytokines.

The mechanisms of action and binding of silver nanoparticles to microbes remain unclear, but it is known that silver binds to the bacterial cell wall and cell membrane and inhibits the respiration process by which the chemical energy of molecules is released and partially captured in the form of adenosine triphosphate. Silver nanoparticles interact with sulfur-containing proteins of the bacterial membrane, as well as with phosphorus-containing compounds such as DNA, to inhibit replication. The bactericidal effect of silver has also been attributed to inactivation of the enzyme phosphomannose isomerase, which catalyzes the conversion of mannose-6-phosphate to fructose-6-phosphate, an important intermediate of glycolysis, the most common pathway in bacteria for sugar catabolism.²

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Furthermore, sutures can be exposed to a bacterial insult because of the nature of the wound itself or by later inoculation events. Surgical wounds are frequently complicated by infection so different severity, so giving rise to a range of clinical problems and prolonging hospitalization with increased healthcare costs.⁴ Thus, the definition of preventive strategies results essential, also considering the potential sources of contamination and the onset of significant complications. Such risks can occur because of many factors, such as the type of surgery performed, the general conditions of the patient or the duration of surgery. Moreover, the use of many antibiotics or antimicrobial agents to prevent post-surgery infections contributes to the phenomenon of antibiotic resistance. Surgical wounds infected by antibiotic-resistant bacteria may cause further morbidity in the patient and result in additional treatment costs. The use of antiseptic sutures may constitute an interesting way to prevent early contamination of surgical wounds by environmental microorganisms.

The aim of this study to evaluate physical, antimicrobial and healing effect of nano-silver suture on periodontal flap.

MATERIALS AND METHOD

Non-absorbable multifilament silk sutures (0.3 mm diameter) were used. Silver nanoparticles and deionized water were adopted for the preparation of the silver solution. The silver solution was obtained by dissolving 0.5 wt/v % of silver nanoparticles in a mixture of 5 v/v % of deionized water.

Silver treatment on the silk surgical sutures

The samples were dipped in the silver solution for 24 hr and then exposed to UV lamp ($k = 365 \text{ nm}$, $t = 20 \text{ min}$, distance 20 cm) in order to induce the synthesis of silver clusters on the surface of the suture. After the treatment, the samples were washed in deionized water to remove the unreacted salt.⁵

Morphological analysis

Inverted electron microscopy was performed in order to analyse the braided structure of the multifilament silk sutures and to verify the presence of silver on the surface. At this purpose, also the cross section of the materials was observed. A quantitative spectrophotometry analysis was performed to quantify the percentage of silver deposited.⁵

Tensile test⁵

Tensile strength was measured by Universal Testing Machine. The "universal" part of the name reflects that it can perform many standard tensile and compression tests on materials, components, and structures. The specimen was placed in the machine between the grips and an extensometer. Once the machine was started it begins to apply an increasing load on specimen. Throughout the tests the control system and its associated software recorded the load and extension or compression of the specimen.

Antibacterial test⁵

The antibacterial activity of the silver-treated sutures was tested on *P.gingivalis* inoculating bacterial density and *A.a* through agar diffusion tests, according to Standard.. From each bacterial suspension, 100 ul were plated on blood agar plates for *P.gingivalis* and *A.a*. Then, the samples were placed over the bacteria and the dishes were incubated in oven at 37 °C for 24 h. After this time, the width of the inhibition are at bacteria

growth was evaluated and labelled according to the levels of antibacterial capability defined by the Standard.

In this split mouth randomized controlled study, 10 patients were selected indicated for periodontal flap surgery. One quadrant was given a silk suture (control group) and the other quadrant was given nano-silver suture (test group). Sutures were removed at 7th day post operatively, added to the normal saline for microbial colony count for both the groups. This suspension was then used for streaking the blood agar plates. Then plates are incubated at 37 C for 24 hrs. Wound healing index was assessed at baseline and 1 month.

RESULTS

Morphological analysis

The distribution of the silver particles on the silk suture fibres and on the cross-section of the suture was evaluated through inverted electron microscope. Silver particles was observed on silver coated suture and were visible among the filaments of the suture after washing with water.

A quantitative spectrophotometry analysis was performed to quantify the percentage of silver deposited on silver nano suture and conventional silk suture. It resulted 44 % on the surface of the suture and 17 % on the cross section, with respect to the components of the substrate.

Tensile tests

This analysis was aimed to compare the tensile strength of untreated and silver treated sutures, in order to determine if the strengths of the materials resulted affected by the nanosilver coating. Tensile strength of silver coating suture was higher than conventional silk suture.

Antibacterial test

When compared with the untreated samples, a reduced growth of bacteria was visible for when incubated in presence of the silver treated sample than untreated.

Clinical parameter

Wound healing index was recorded and compared between silk suture and silver coated suture. It was observed that healing was better in silver coated suture group score. There was significant reduction seen in colony forming units in silver coated suture group.



Figure 1 Silk Suture Dipped In SNP Suspension



Figure 2 Sample Exposed To Uv Light

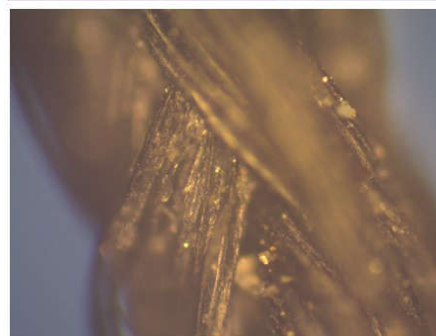
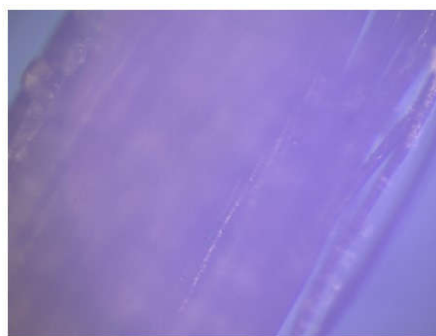


Figure 5 Inverted Electron Microscopic Image Of Silk Suture And Nano Silver Suture



Figure 3 Microhardness Machine And Inverted Electron Microscope

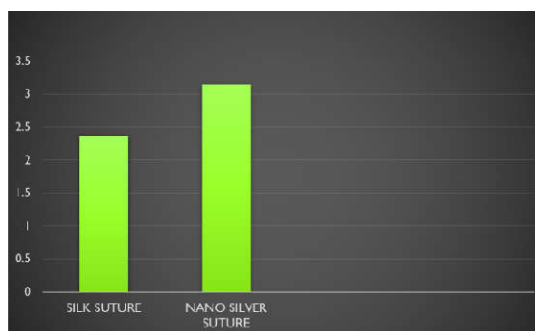
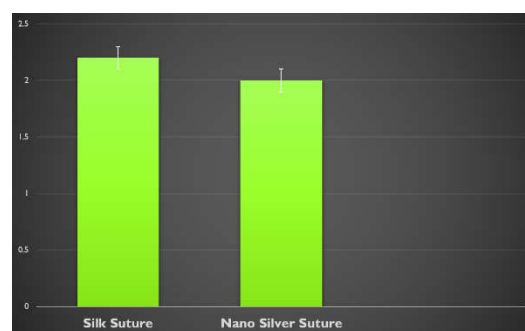


Figure 6 wound Healing Index And Tensile Strength Graph



Figure 4 Spectrophotometry

DISCUSSION

Surgical sutures used to close scared tissues are often source of microbial infection. Despite their sterility before using, the sutures can become a fertile ground for microbial colonization once implanted, thus resulting in delayed healing, susceptibility to infection, poor tissurepair with potential pathological scarring, and also affecting the quality of life of patients and the health care costs. Several authors have suggested that the risk of infection depends on the degree of bacterial adherence and on the nature of the suture.⁶ In particular, monofilament sutures produced smaller inflammatory reactions than multifilament materials with respect to the tissue response.⁷ Antibiotic-resistant bacteria also represent an increasing concern in the management of

surgical infections, as wound bacterial colonization is often associated to an aggressive management wound and to a limited choice of therapeutic antibiotics. Bacterial adhesion to surgical sutures might serve as an infection focus, thus causing a delay in wound healing.⁸ In this work novel silver coated silk sutures with antimicrobial properties have been developed for the prevention of surgical infections.

Nanosilver has been recognized as the nanomaterial with the highest degree of commercialization in medical and healthcare fields. Another advantage of this method is associated to the long-term antibacterial properties that can be obtained even with very low contents of silver. In this study, a silver solution containing 0.5 wt/v % of silver nanoparticles dissolved in a mixture 5 v/v % of water was adopted for the treatment and resulted effective in providing good antibacterial capability. Inverted electron microscope analysis demonstrated the effectiveness of the technology in providing a homogeneous distribution of silver particles on the silk filaments, as clearly visible. Spectrophotometry analysis also confirmed the presence of silver and allowed its quantification, while no peak was visible in the spectrum of untreated sample.

The mechanical properties of the treated silk suture were evaluated because the reduction in the strength of the suture material can result in untimely suture breakage, so inducing complications in healing tissues

The capability of the silver treated silk sutures in reducing the bacterial viability, proliferation and adhesion to the substrate was evaluated through qualitative and quantitative tests on *P.gingivalis* and *A.a* as examples of Gram negative bacteria. Also the bacterial adhesion to the silk suture resulted reduced by the presence of the silver coating. The percentages of reduction of bacteria adhered to the substrate were 40 and 35 % for *P.gingivalis* and *A.a*, respectively which improves healing process. Silver nanoparticles have proved to have an inhibitory effect on the growth of *P.gingivalis* and *A.actinomycetemcomitans*.⁵

All antibacterial studies clearly demonstrate that the use of silver coated sutures can offer clinical advantages in terms of the prevention of surgical infection against bacterial colonization, without inducing cytotoxic effects on fibroblasts. Moreover, low percentages of silver have been adopted for the treatment, thus indicating that the introduction of these devices in hospital practice can be proposed with no significant impact on the health care costs.

Its clinical application can reduce the inflammation, improve healing process and reduce scar in periodontal flap surgery.

The strong adhesion and the durability of the silver treatment suggested that the sterilization techniques commonly adopted in biomedical field should not adversely affect the properties of the silver coating. However, future studies will focus on the effect of the sterilization techniques on the silver coated sutures developed

CONCLUSION

Sutures coated with biosynthesized silver nanoparticles may provide antimicrobial and antibiofilm functionality that is related to the success of the clinical applications and wound healing process.

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