Bacterial adherence to silk and expanded polytatrafluorethilene sutures: an *in vivo* human study

A. Scarano[1], F. Inchingolo[2], L. Leo[3], C. Buggea[3], A. Crisante[3], A. Greco Lucchina[4] and G. Scogna[3].

[1]Director of graduate School in Oral Surgery, Department of Innovative Technologies in Medicine & Dentistry and CAST, University of Chieti-Pescara, Italy; [2]Department of Interdisciplinary Medicine, University of Bari "Aldo Moro", Bari, Italy; [3]Department of Innovative Technologies in Medicine & Dentistry, University of Chieti-Pescara, Italy; [4]Oral Surgery Unit, University of Chieti-Pescara, Italy.

After oral surgery, bacterial adhesion to suture can cause surgical site infections and delay wound healing. Microbial adherence to the suture is influenced by its physical configuration and chemical structure. The aim of this study was to compare in vivo the bacterial adhesion to two suture materials used in oral surgery: silk and monofilament expanded polytetrafluoethilene (e-PTFE). After sinus lift surgery, 15 flaps were sutured with silk (nonabsorbable, organic, braided, 4.0) and 15 were sutured with e-PTFE (nonabsorbable, synthetic, monofilament, 4.0). Seven days after surgery, bacterial adherence, in terms of percentage of the surface covered, was evaluated for each suture material by scanning electron microscope (SEM). Onto silk suture, plaque consisted of a few cocci and a higher proportion of rods and filamentous-shaped bacteria, with some mineralized plaque. Onto e-PTFE speciments, only small colonies of a few cocci or no bacteria were observed, with empty spaces between the colonies and no plaque mineralization. The surface covered by bacteria on e-PTFE specimens was significantly lower than that of silk sutures. (22.1% ±4.96% vs 54.3% ± 7.9%; P =0.0001). The results of the present study suggest that multifilament structure of silk favours a greater bacterial adherence, proliferation, and persistence, so monofilament and e- PTFE suture should be preferred in oral surgery.

There are numerous suture materials for use in the oral environment, with different compositions. Many of these sutures elicit an inflammatory reaction, that can also delay the surgical healing. The biological response to suture materials has been studied in animals, implanting suture materials subcutaneously, intramuscularly or in the abdominal wall, both in aseptic and septic conditions (1,2). The results of these investigations suggested that monofilamented sutures evoke a less-intense reaction than multifilamented sutures (3) and absorbable sutures produced more tissue reaction than nonabsorbable sutures (4).

But the oral environment is unique since it is humid and infected. These factors enhance the likelihood of bacterial migration along the suture into the tissues. Tissue reactions to suture materials in the oral cavity have been studied, but the factual information appears to be incomplete and conflicting (5). Castelli et al. observed that the inflammatory reaction to silk was more intense in mucosa than in gingiva at first 4 days, being similar at later periods. Monofilament materials are associated with less severe tissue response tha multifilament materials (6,7). Racey et al. compared silk, polyglactin 910

Key words: microbial adherence, silk suture, monofilament expanded polytetrafluoethilene, e-PTFE suture, bacterial adhesion, oral surgery

Corresponding Author:
Prof. Antonio Scarano
Department of Medical, Oral and Biotechnological Sciences,
University of Chieti-Pescara, Chieti, Italy.
E-mail: ascarano@unich.it

0393-974X (2020)

DISCLOSURE: ALL AUTHORS REPORT NO CONFLICTS OF INTEREST RELEVANT TO THIS ARTICLE.

and plain catgut in human oral tissues (8). After 7 days, silk and polyglactin 910 produced a moderate and similar inflammation, but plain catgut induced a more severe reaction.

Leknes et al. compared silk (nonabsorbable, organic, braided material), catgut (absorbable organic monofilament), expanded polytetrafluorethilene (e-PTFE, nonabsorbable, synthetic, monofilament) and polyglactin 910 (absorbable synthetic, undyed braided material), in an experimental canine model (9). After 14 days, the observations in light micorscopy showed that silk speciments had bacterial plaque perisuturally as well as betwwen the threads, with a dense inflammatory response. The gut sutures were lost or absorbed. The e-PTFE sutures showed limited inflammatory reaction and polyglactin 910 was intact, but some cellular invasion was noticed (10).

Braided sutures seem to conduct bacterial migration to a greater extent than monofilament sutures, and the presence of immobile bacteria inside the multifilament suture hinders the cellular and immunological defense against them (11).

The purpose of this study was to compare *in vivo* the bacterial adhesion of two suture materials used in oral surgery: silk and monofilamented expanded polytetrafluoethilene (e-PTFE).

MATERIALS AND METHODS

The suture material of thirty patients submitted to oral surgical procedures were evaluated. The surgical wounds of fifteen patients were sutured with nonabsorbable organic material: silk (black braided silk, 4.0, cutting needle FS-2, Ethicon), and fifteen were sutured with expanded polytetrafluoethilene (e-PTFE) (nonabsorbable synthetic monofilament, reverse cutting needle RT-18, 3i). All patients were treated in the Outpatient Department of Oral Implantology, Center for Advanced Studies, Dental Research Division, UNINGÁ—Cachoeiro de Itapemirim, Brazil. The patients underwent sinus lifting with a surgical protocol has been published previously (12). During operative surgeon all operators wears the surgical mask (13) and after each surgical procedure we have sanitized the environment with a recently published protocol (14). The sutures were removed after seven days. All suture has diameter to 4.0. The bacterial adherence was calculated for each suture material by scanning electron microscope (SEM) (Fig. 1).

Prefixation

Prefixation took place for 20 h at 4° C in a 5 ml of glutaraldehyde at 2% in 0.05M phosphate buffer pH 7.4, 366 mOsm. The washing was made in 0.1M phosphate buffer with 0.15M of sucrose to ensure the osmolarity remained at about 360 mOsm. Following prefixation the specimens were treated with OTOTO method of post fixation as MALIK-WILSON involving repeated exposure to osmium tetroxide and thiocarbohydrazide as follows:

- 1% OsO_4 in phosphate buffer for 2h,
- six washes in distilled water for 15 minutes,
- incubation in 1% of thiocarbohydrazide solution,
- six washes in distilled water for 15 minutes,
- 1% OsO_4 in distilled water for 2h,

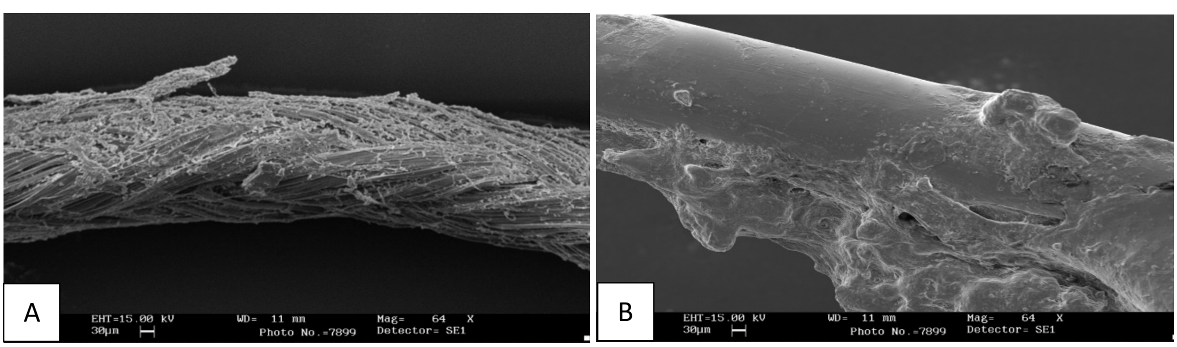

Fig. 1. ***A)****: Scanning electron microscopy images of suture removed after seven days. Numerous cocci or filamentous bacteria covers the silk suture surface;* ***B)****: A few numbers of bacteria cover the e-PTFE suture removed after seven days.*

- six washes in distilled water for 15 minutes,
- incubation in 1% of thiocarbohydrazide solution,
- six washes in distilled water for 15 minutes,
- 1% OsO4 in distilled water for 2h,
- six washes in distilled water for 15 minutes,
- dehydration using graded alcohol series,
- Critical Point Dried from liquid CO2 in an Emitech K 850 (Emitech Ltd. Ashford, Kent, UK) critical point drier,
- mounted onto aluminium specimens holders using carbon adhesive discs, and
- lightly coated with gold in an Emitech K 550 (Emitech Ltd. Ashford, Kent,UK) sputter coater.

All specimens were examined and photographed using SEM (LEO 435Vp Cambridge, UK) operating at 15-20 kV**.** Quantitation of the percentage of the surface covered by bacteria was done on the JPEG images using a personal computer associated with a dedicate software package with image-capturing capabilities. Ten areas of 200 μm along the suture were evaluated for each suture and an image in JPEG format was created. The identification of bacterial cells was based on the morphological characteristics as the protocol used in a previous research (15).

Statistical Evaluation

The data means were recorded and analyzed by the software package GraphPad 8 (Prism, San Diego CA-USA). The differences in the percentages of surface covered by bacteria in the two groups were evaluated with the unpaired t-Student test. The percentage of suture surface covered by bacteria was expressed as a mean ± SD. The descriptive statistic included interquartile percentage and 95% confidence intervals. Statistically significant differences were set at P <0.05.

RESULTS

Silk suture

The all-multifilament silk sutures appeared with typical braided structure and its intrinsically unstructured conformation in aqueous solution and in some field appeared self-assemble into nanofibrils. The largest presence of bacterial, extracellular polysaccharides and cellular detritus to be contained a biofilm on the silk suture were observed. The adhesion of the bacterial were low to the silk group and consisting of a few cocci and a higher proportion of rods and filamentous-shaped bacteria. A thin, regular layer of cocci was found in many areas of the surface. Salivary proteins, in contact with the suture surface, were found in a large portion of the surface of e-EPTFE. Some mineralized plaque was noted on most specimen. The area covered by bacteria was 54.3% ± 7.9% (Table I).

Expanded polytatrafluorethilene sutures

EPTFE sutures appeared as typical microporous monofilament structure. The scanning electron microscopic study showed the differences in bacterial colonization between silk and e-EPTE types of suture material. Inmany areas no bacteria or salivary proteins were observed. In other areas, only small

Table I. *Summary of the surface covered by bacteria (%) of silk sutures vs. EPTFE sutures. A significant difference was present between the study groups. (p<0.01. t-Student Test).*

Surface covered by bacteria (%)	Silk sutures	EPTFE sutures
Mean	54.30%	22.10%
Standard deviation	± 7.9%	±4.96%
95% Confidence Interval	(48.34- 60.26)	(18.36-25.84)
Interquartile Range	(46.4- 62.2)	(17.14-27.06)
Unpaired t-Student		
Difference between means ± SEM	(-32,20 ± 3,109)	
95% confidence interval	(-38,73 to -25,67)	
R^2 (eta squared)	0.8563	
p value	P<0.01 t=10.36, df=18	

colonies of a few cocci were found. In the major portion of the disk surface, empty spaces between the colonies of bacteria were present. Polymorphous aggregates of bacteria were present, consisting mainly in cocci and short rods. No calcification of the bacteria was present. The surface covered by bacteria was 22.1% ±4.96% (Table I).

Statistical Evaluation

The specimen surface covered by bacteria on e-PTEF specimens was significantly lower than that of silk sutures. (P =0.0001) (Fig. 2).

DISCUSSION

The outcomes of the present study have shown that statistically significant difference exist in bacterial adhesion between silk suture and e-PTFE suture surfaces. Primary soft tissue closure is a required condition for the success of oral surgery procedures for minimizes postoperative discomfort and reduction soft tissue dehiscence (16). Surgical site infections (SSIs) are common complications that occur after oral surgery and microbial adherence to the suture is a main cause (17). Bacterial adhesion onto suture is related the structure and chemical composition of the suture material. This bacterial adhesion cannot be eradicated by chemical or biologic agents, or other mechanisms of wound decontamination. Silk suture as extensively used in oral surgery for its easy manipulation and knot security and is preferred to monofilament suture. However, many studies reported that multifilament sutures favor bacterial adherence and can cause severe wound infections (18–20). In fact, in oral cavity there are a larger number of bacteria can cause wounds infection with delayed healing.

Several previous studies have examined the bacterial adherence properties of sutures and have showed that adherence of bacteria to suture was directly correlates to the ability to cause a wound infection (21, 22). The selection of surgical suture is very important in oral and implant surgery. Our results confirm that the physical configuration played a relatively important role in surg bacterial. However also the chemical structure of the suture was found to be the important factor for biofilm formation (21, 23) and vary coating was developed for reduction of bacterial adhesion (18, 24-26).

The results of the present study suggest that the structure of silk suture provide a hospitable niche for bacterial growth and proliferation. The microorganisms present in site of multifilament are resistant to immune response, antimicrobial therapy and can produce a biofilm, thus allowing microbial persistence. In conclusion, based on the results, the structure of suture influences the bacterial adhesion and strongly indicate that, whenever possible, the first choice of suture should be monofilament and in PTFE.

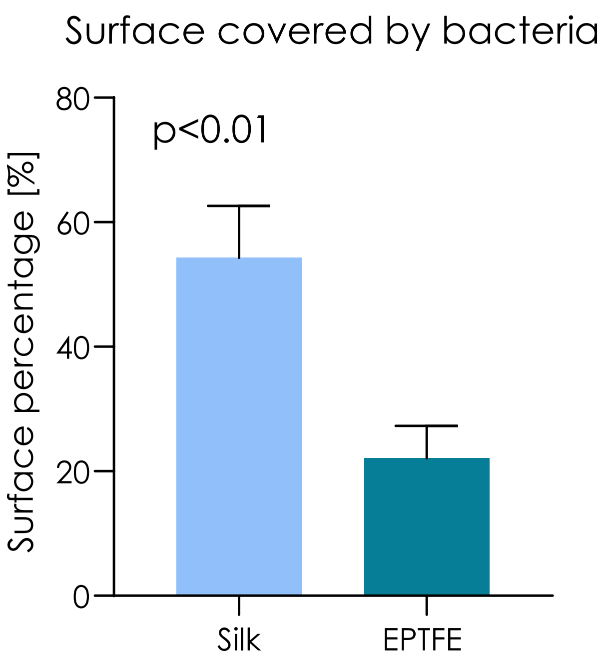

Fig. 2. *Comparative evaluation of the surface covered by bacteria (%) between silk sutures vs. EPTFE sutures (p<0.01 t-Student Test).*

REFERENCES

1. Edlich RF, Panek PH, Rodeheaver GT, Kurtz LD, Edgerton MT. Surgical sutures and infection: a biomaterial evaluation. J Biomed Mater Res 1974; 8(3):115–26.
2. Setzen G, Williams EF. Tissue response to suture materials implanted subcutaneously in a rabbit model. Plast Reconstr Surg 1997; 100(7):1788–95.
3. Everett WG. Sutures, incisions, and anastomoses.

Ann R Coll Surg Engl 1974; 55(1):31–8.

4. Hastings JC, Van Winkle W, Barker E, Hines D, Nichols W. The effect of suture materials on healing wounds of the bladder. Surg Gynecol Obstet 1975; 140(6):933–37.
5. Javed F, Al-Askar M, Almas K, Romanos GE, Al-Hezaimi K. Tissue reactions to various suture materials used in oral surgical interventions. ISRN Dent 2012; 2012:762095.
6. Lilly GE, Cutcher JL, Jones JC, Armstrong JH. Reaction of oral tissues to suture materials. IV. Oral Surg Oral Med Oral Pathol 1972; 33(1):152–7.
7. Lilly GE. Reaction of oral tissues to suture materials. Oral Surg Oral Med Oral Pathol 1968; 26(1):128–33.
8. Racey GL, Wallace WR, Cavalaris CJ, Marguard JV. Comparison of a polyglycolic-polylactic acid suture to black silk and plain catgut in human oral tissues. J Oral Surg 1978; 36(10):766–70.
9. Leknes KN, Røynstrand IT, Selvig KA. Human gingival tissue reactions to silk and expanded polytetrafluoroethylene sutures. J Periodontol 2005; 76(1):34–42.
10. Cartmill BT, Parham DM, Strike PW, Griffiths L, Parkin B. How Do Absorbable Sutures Absorb? A Prospective Double-Blind Randomized Clinical Study of Tissue Reaction to Polyglactin 910 Sutures in Human Skin. Orbit 2014; 33(6):437–43.
11. Dahl E. Wound infections on board ship--prevention, pathogens, and treatment. Int Marit Health 2011; 62(3):186–90.
12. Scarano A, Lorusso F, Arcangelo M, D'Arcangelo C, Celletti R, de Oliveira PS. Lateral Sinus Floor Elevation Performed with Trapezoidal and Modified Triangular Flap Designs: A Randomized Pilot Study of Post-Operative Pain Using Thermal Infrared Imaging. Int J Environ Res Public Health 2018; 15(6):1277.
13. Scarano A, Inchingolo F, Lorusso F. Facial Skin Temperature and Discomfort When Wearing Protective Face Masks: Thermal Infrared Imaging Evaluation and Hands Moving the Mask. Int J Environ Res Public Health 2020; 17(13):4624.
14. Scarano A, Inchingolo F, Lorusso F. Environmental Disinfection of a Dental Clinic during the Covid-19 Pandemic: A Narrative Insight. Biomed Res Int 2020; 2020:8896812.
15. Scarano A, Piattelli A, Polimeni A, Di Iorio D, Carinci F. Bacterial adhesion on commercially pure titanium and anatase-coated titanium healing screws: an in vivo human study. J Periodontol 2010; 81(10):1466–71.
16. Scarano A, Ceccarelli M, Marchetti M, Piattelli A, Mortellaro C. Soft Tissue Augmentation with Autologous Platelet Gel and β-TCP: A Histologic and Histometric Study in Mice. Biomed Res Int 2016; 2016:2078104.
17. Edmiston CE, Seabrook GR, Goheen MP, Krepel CJ, Johnson CP, Lewis BD, et al. Bacterial adherence to surgical sutures: can antibacterial-coated sutures reduce the risk of microbial contamination? J Am Coll Surg 2006; 203(4):481–9.
18. Baygar T. Characterization of silk sutures coated with propolis and biogenic silver nanoparticles (AgNPs); an eco-friendly solution with wound healing potential against surgical site infections (SSIs). Turk J Med Sci 2020; 50(1):258–66.
19. Dhas SP, Anbarasan S, Mukherjee A, Chandrasekaran N. Biobased silver nanocolloid coating on silk fibers for prevention of post-surgical wound infections. Int J Nanomedicine 2015; 10(Suppl 1):159–70.
20. Ananthakrishnan N, Rao RS, Shivam S. Bacterial adherence to cotton and silk sutures. Natl Med J India 1992; 5(5):217–8.
21. Banche G, Roana J, Mandras N, et al. Microbial adherence on various intraoral suture materials in patients undergoing dental surgery. J Oral Maxillofac Surg 2007; 65(8):1503–7.
22. Katz S, Izhar M, Mirelman D. Bacterial adherence to surgical sutures. A possible factor in suture induced infection. Ann Surg 1981; 194(1):35–41.
23. Sortino F, Lombardo C, Sciacca A. Silk and polyglycolic acid in oral surgery: a comparative study. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2008; 105(3):e15-18.
24. Mahesh L, Kumar VR, Jain A, et al. Bacterial Adherence Around Sutures of Different Material at Grafted Site: A Microbiological Analysis. Materials (Basel) 2019; 12(18):2848.
25. Asher R, Chacartchi T, Tandlich M, Shapira L, Polak D. Microbial accumulation on different suture materials following oral surgery: a randomized controlled study. Clin Oral Investig 2019; 23(2):559–65.

26. Henriksen NA, Deerenberg EB, Venclauskas L, et al. Triclosan-coated sutures and surgical site infection in abdominal surgery: the TRISTAN review, meta-analysis and trial sequential analysis. Hernia 2017; 21(6):833–41.