IN VITRO ANTIBACTERIAL EFFICACY OF VICRYL PLUS SUTURE (COATED POLYGLACTIN 910 WITH TRICLOSAN) USING ZONE OF INIBITION ASSAYS

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SUMMARY

In vitro antibacterial efficacy of Vicryl plus suture (coated Polyglactin 910 with triclosan) using zone of inibition assays

Objectives: This study evaluates the *in vitro* antimicrobial efficacy, of an absorbable suture coated with triclosan (Vicryl Plus®) against two bacteria, potential responsible for the development of oral diseases: *Pseudomonas aeruginosa* and *Streptococcus mutans*.

Methods: Vicryl Plus 3-0 and Vicryl 3-0 were tested for their efficiency against P. aeruginosa and S. Mutans. 27 segments 10 cm long each, of every suture, have been tested against P. Aeruginosa and S. Mutans respectively. Every sample has been dipped in a broth culture containing pure dried stocks of P. aeruginosa and S. mutans and placed in a Petri dish right after. Four hours later the sutures have been aseptically removed and placed in a selective culture. The incubation time was 18 hours for P. aeruginosa and 43 hours for S. mutans at 37°C. The antimicrobial efficacy of both sutures was performed by measuring the length of the bacteria-free suture segment. Results: A statistically significant difference between Vicryl Plus 3-0 and Vicryl 3-0 has been observed, with an higher bacterial growth on Vicryl 3-0 for both bacteria (P. aeruginosa and S. mutans).

Conclusions: Vicryl Plus presented an antibacterial effectiveness *in vitro* against both *P. aeruginosa* and *S. mutans*.

Key words: suture, vicryl plus, polyglactin 910, pseudomonas aeruginosa, streptococcus mutans.

RIASSUNTO

Valutazione in vitro dell'efficacia antibatterica della sutura Vicryl Plus (polyglactin 910 rivestito con triclosan) mediante analisi delle zone di inibizione

Obiettivi: Lo scopo di questo studio è stato quello di valutare *in vitro* l'efficacia antibatterica di una sutura assorbibile rivestita con Triclosan (Vicryl Plus®) su due batteri potenzialmente responsabili dello sviluppo di patologie nel cavo orale: *Pseudomonas aeruginosa* and *Streptococcus mutans*.

Metodi: Le suture analizzate nello studio sono state il Vicryl Plus 3-0 (sutura test) e Vicryl 3-0 (sutura controllo). Per ciascun tipo di sutura sono stati testati 54 frammenti da 10 cm, 27 per *P. aeruginosa* e 27 per *S. mutans*. I segmenti di sutura sono stati immersi in una brodocoltura contenente ceppi puri liofilizzati di *P. aeruginosa* e di *S. mutans* e poi posti, ciascuno, in una capsula di Petri. Dopo 4 ore sono stati rimossi asetticamente e posizionati su terreni selettivi per ciascuno dei due tipi di batteri. Il tempo di incubazione a 37° C è stato 18 ore per *P.aeruginosa* e 43 ore per *S. mutans*. Il grado di efficacia della sutura test rispetto al controllo è stato valutato in base alla lunghezza di filo priva di patina batterica ben evidenziabile.

Risultati: Abbiamo riscontrato una differenza statisticamente significativa, con minore estensione della patina batterica sul Vicryl plus rispetto a quella proliferante sui segmenti di filo di sutura in Vicryl, sia nella valutazione dell'efficacia nei confronti di *P. aeruginosa*, sia nei confronti di *S. mutans*.

Conclusioni: Il Vicryl Plus presenta *in vitro* efficacia antibatterica sia verso *P. aeruginosa* sia verso *S. mutans.*

Parole chiave: sutura, vicryl plus, polyglactin 910, pseudomonas aeruginosa, streptococcus mutans.







Introduction

Sutures performed in oral cavity are highly exposed to bacterial colonization. In fact sutures cause themselves an interruption of the mucosal barrier and offer an adhesion surface to bacteria with subsequent tissue colonization. In dental extractions and in periodontal surgery when suturing is necessary, the possible complications due to bacterial colonization are dehiscence of the suture, slower wound healing due to edema, inflammation and suppuration (1). In implant surgery, in particular with GBR, another possible complication that should be considered is bone loss associated with osteitis and potential no osteointegration (2-4).

The surface characteristics, the coating of the suture and its own abilities to inhibit bacterial proliferation due to the material the thread is made of, play a great role in plaque formation during the post-surgical period.

Bacterial adhesion is particularly facilitated by the presence of an irregular surface. Synthetic monofilament sutures, thanks to their smooth coating, do not facilitate bacterial adhesion and the consequent biofilm formation compared multifilament (5).

Vicryl Plus is an adsorbable synthetic multi-filament suture where the characteristics of the material are combined with a potential antiseptic activity due to surface coating with triclosan (6).

The Vicryl Plus, *in vitro*, has shown to be able to reduce the colonization of *S. aureus* and *S. epidermidis* bacterial stocks (7), and *in vivo*, in animal model, inhibits the *S. aureus* bacterial colonization of the suture (8, 9).

The aim of this work was to evaluate, *in vitro*, the antimicrobial activity of coated polyglactin 910 suture with triclosan, relatively to the degree of inhibition of the bacterial growth of two potential pathogenic agents of the oral cavity (10-12): *Pseudomonas aeruginosa* and *Streptococcus mutans*.



Methods

The sutures evaluated were:

- Vicryl Plus 3-0 (Ethicon, Inc., Somerville, NJ), [test group].
- Vicryl 3-0 (Ethicon, Inc., Somerville, NJ), [control group].

For each suture, 54 fragments (10 cm long) have been tested, 27 for P. aeruginosa and 27 for S. mutans. In aseptic conditions dehydrated stocks of *P. aeruginosa* (cod.73/36 former ATCC 9027) and S. mutans (cod. 93/2, produced by Sieroterapico Soc. Coop ARL Diagnostici, Milan, Italy) have been rehydrated with 300 µl of sterile distilled water and left for 4-5 hours at room temperature. Each sample was then placed in 10 ml of nutritive liquid and divided in smaller shares (0,3 milliliter) that were immediately frozen. Prior to their use, the shares were left at room temperature for 4 hours and each added in a liquid culture of 20 milliliter each. The shares were then incubated at 37°C for 24 hours. In aseptic condition, the positive broth cultures were gradually diluted with a progressive 1:10 dilution, identifying the 1:100 dilution as the optimal for the contamination of both the sutures studied. Both sterile sutures were aseptically divided in segments 10 cm long. Two segments of thread, one treated with triclosan (study group) and one non-treated (control group) were joined and then contaminated with broth culture 1:100 dilution. After a few minutes the suture was aseptically removed and put in a sterile Petri dish for 4 hours at room temperature, ensuring an ideal humidity by adding a drop of saline solution in the capsule. This can allow a sufficient contact time of the surface of the treated suture with the microorganisms in order to guarantee the proper result. Subsequently the thread was aseptically placed on a selective culture specific for the growth of the survived microorganisms, and incubated at 37°C for 18 hours in agar Cetrimide by (Italian Biolife Company, srl, Milan, Italy) (P. aeruginosa) and for 43 hours in agar Nutrient by (Italian the Biolife Company s.r.l., Milan, Italy)

(*S. mutans*). The degree of efficacy of the Vicryl Plus was estimated by measuring the length of suture segment that was bacteria free and tested by counting-colony product (Barloworld Scientific s.r.l., Milan, Italy).

For the statistical analysis of the efficacy of the suture treated with triclosan a non parametric test has been used since the relatively small group tested couldn't allow an exact Gaussian curve of the bacterial population. In this case, a Wilcoxon-Mann-Whitney test also known as test U (Mann-Whitney) or simply as test of Wilcoxon has been chosen.

For the calculation of the test and the verification of the hypothesis the SAS System program has been employed, specific for this type of test, choosing as level of significance $\alpha = 0.01$.

Results

The hypothesis to be verified is represented by the different extension of the bacterial plaque on the Vicryl Plus in comparisons to the bacteria plaque present on the segments of Vicryl of suture. Therefore the aim of this study is to test the efficacy of the thread with triclosan on its surface.

Pseudomonas aeruginosa

The data obtained against *P. aeruginosa* are shown in Table 1. The inibition zone were present on all the length of the cut suture pieces coated with tri-



Figure 1Contaminated Vicryl with *P. aeruginosa*; it is evident the bacterial proliferation all along its length.



Vicryl Plus: halo of inhibition showed through fluorescence in *P. aeruginosa* contamined samples.

closan and only on few portion of samples control (Fig. 1, 2).

All the variables considered are shown in Table 2.

Table 1 - P. aeruginosa bacterial contamination expressed in cm of suture.																											
									Len	ght	of s	utur	e in	cen	time	etre	S										
Vicryl	8	8	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
Vicryl plus	0	1	8	0	4	0	4	1	0	0	0	0	2	1	5	1	0	0	2	0	0	0	2	3	0	0	0
Difference	8	7	2	10	6	10	6	9	10	10	10	10	8	9	5	9	10	10	8	10	10	10	8	7	10	10	10



Table 2 - Variation studied for P. aeruginosa.											
P. aeruginosa	Vicryl	Vicryl plus	Difference								
Average	9.9	1.3	8.6								
Mid-point	10.0	0.0	10.0								
Standard Deviation	0.5	2.0	2.0								

Streptococcus mutans

The data obtained against *S. mutans* are shown in table 3. Also in this experiment, the inibition zone

were present on all the length of the cut suture pieces coated with triclosan and only on few portion of samples control (Fig.s 3, 4).

The variations studied are shown in table 4.

Table 3 -	S. n	nuta	ns	cont	ami	natio	n e	xpre	esse	d in	cm	of s	utu	re.													
									Len	ght	of s	utur	e in	cer	ntime	etre	S										
Vicryl	10	10	8	7.5	10	8.5	10	10	9	10	10	10	9	9	9.5	10	10	10	5	8	10	5	8	5	6	9	10
Vicryl plus	5	5	6	6	0	6.5	7	5	7	5	9	3	6	3	4	8	7	2.5	2	2	6	2	5	2	3	1	2
Difference	5	5	2	1.5	10	2	3	5	2	5	1	7	3	6	5.5	2	3	7.5	3	6	4	3	3	3	3	8	8



Figure 3Contaminated Vicryl with *S. mutans:* it is evident the bacterial proliferation all along its length.

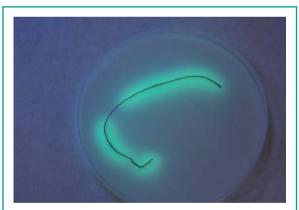


Figure 4Vicryl Plus: it is evident the bacterial proliferation all along its length.

Table 4 - Variations studied for S. mutans.										
S. mutans	Vicryl	Vicryl plus	Difference							
Average	8.9	4.4	4.2							
Mid-point	9.5	5.0	3.0							
Standard Deviation	1.7	2.3	2.3							

research article

Discussion

Many authors have investigated the relationship of suture construction and chemical composition as it relates to microbiological colonization (6-9, 11). Any suture product on natural or synthetic composition, and of mono- or multi-filament construction, is susceptible to bacterial attachment and colonization. It is clear that colonization in potential associated with sugical site infection (7). Any device that can reduce the potential for colonization of a surgical suture in a safe ana efficacious manner is a potentially useful addition in surgery, also in dentistry, in particolar in implantology and periodontology.

The data obtained in this paper shown a good *in vitro* antibacterial efficacy of coated polyglactin 910 suture with triclosan (vicryl plus) against the two bacteria examined, with a response better on *P. aeruginosa* than *S. mutans*.

The employment of a suture with a limited pathological activity and during the hole post-surgical period can avoid many complications and improve the result of the therapeutic treatment. Although, in other studies, the suture coated with triclosan has demonstrated antimicrobial effects on *S. aureus* e *S. epidermidis* (7,11) and in our tests its antimicrobial activity appears greater against *P. aeruginosa*, (gram negative), than on *S. mutans*, (gram positive).

The results obtained, *in vitro*, regarding Vicryl Plus antimicrobial activity against *P. aeruginosa* and *S. mutans* could be particularly encouraging in order to effectuate an, *in vivo*, test even though in this case a proper valuation might be strongly conditioned by the local bacterial population and the oral hygiene of the single subject.

These data cleary support the conclusion that coated polyglactin 910 suture with triclosan (vicryl plus) provides an antibacterial effect sufficient to prevent *in vitro* colonization of the suture by *P. aeruginosa* e *S. mutans*.

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