ORIGINAL ARTICLE

Annals of Agricultural and Environmental Medicine 2015, Vol 22, No 3, 551–555
www.aaem.pl

Comparison of antibacterial-coated and non-coated suture material in intraoral surgery by isolation of adherent bacteria

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Pelz K, Tödtmann N, Otten JE. Comparison of antibacterial-coated and non-coated suture material in intraoral surgery by isolation of adherent bacteria. Ann Agric Environ Med. 2015; 22(3): 551–555. doi: 10.5604/12321966.1167733

Abstract

Objectives. In general surgery the incidence of postoperative wound infections is reported to be lower using triclosan-coated sutures. In intraoral surgery, sutures are faced with different bacterial species and the question arises whether the antibacterial-coated suture material has the same positive effects.

Materials and Methods. Triclosan-coated and uncoated suture materials were applied in 17 patients undergoing wisdom tooth extraction. Postoperatively, sutures were removed and adherent bacteria were isolated, colony-forming units (cfu) were counted, and species identified.

Results. Oral bacteria were found in high numbers (cfu>10^7) on both Vicryl and the triclosan-coated Vicryl Plus. The total number of bacteria isolated from Vicryl Plus was 37% higher than for Vicryl, mainly due to increased numbers of anaerobes. The number of bacterial strains identified was higher for Vicryl (*n*=203) than for Vicryl Plus (*n*=198), but the number of pathogens was higher on Vicryl Plus (*n*=100) than on Vicryl (*n*=97). Fewer Gram-positive strains were found on Vicryl Plus (*n*=95) than on Vicryl (*n*=107) and, conversely, more Gram-negative strains on Vicryl Plus (103vs.96).

Conclusions. In terms of the total number of oral bacteria, and especially oral pathogens, that adhered to suture material, no reduction was demonstrated for Vicryl Plus. The use of triclosan-coated suture material offers no advantage in intraoral surgery.

Key words

Oral and maxillofacial surgery, oral bacteria, suture, triclosan

INTRODUCTION

Suture materials used for treating wounds were originally natural materials, such as animal tendons and cotton fibres. Usage of these materials often resulted in severe infections. Sterilisation reduced these complications significantly. However, sutures are still foreign materials, which tend to attract bacteria. Postoperative wound infections are still the second most common perioperative complication. In view of this risk of infection, much recent academic and industrial research in this area has focused on avoiding bacterial colonisation of medical materials from the beginning, especially by the use of antibacterial coating. Although antibacterial coating prevents the build-up of bacteria on medical materials to some degree, it is almost impossible to clear or kill bacteria that adhere to suture material once a biofilm has formed [1]. Hence, suture materials applied during surgery carry an intrinsic risk of postoperative wound infections and associated complications, such as bone infection, bacteraemia, organ abscess, endocarditis, or even sepsis [2, 3, 4]. Several studies have shown that wound infections cause longer stays in hospital, require additional treatment, antibiotics, and wound treatment at home, and add to disability. All of these result in substantial additional expense [5].

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Received: 01 May 2014; accepted: 10 October 2014

Triclosan is an antibacterial phenol derivative that has *in vitro* activity against Gram-positive and, to a lesser extent, Gram-negative bacteria. *In vitro* adherence of *Staphylococcus aureus*, *Staphylococcus epidermidis* and *Escherichia coli* is significantly reduced on triclosan-impregnated as compared to untreated suture material [6]. When tested by the agar-diffusion test, a triclosan-coated suture produced inhibition zones against *S. aureus* and *S. epidermidis* even when the suture had been immersed in aqueous fluid for 7 days [7]. In guinea pigs, an inoculum of 21,000 colony-forming units (cfu) of *S. aureus* was reduced to 559 cfu on a triclosan-coated suture that was implanted subcutaneously, whereas an uncoated suture contained 16,831 cfu [8]. A clinical evaluation of cerebrospinal fluid shunts showed a reduction in the percentage of shunt infections from 21 % with uncoated suture material to 4.3% with coated material [9].

To the best of our knowledge, there have been no studies on triclosan suture material in intraoral surgery or on the effect of triclosan on the anaerobic bacteria predominantly found in this region. A report of an *in vitro* study [10] on triclosan-coated suture that was incubated with human saliva concluded that 'sutures coated with triclosan do not provide a sufficient antimicrobial effect to prevent *in vitro* colonisation by oral bacteria'. Walker has investigated dentifrices containing triclosan [11] and described a significant reduction in the total cultivable flora, but found no significant reduction of anaerobic and strict anaerobic counts.

The aim of the presented study was comparative analysis of bacterial colonisation on conventional suture material Vicryl and the antibacterial suture material Vicryl Plus in

routine wound treatment during dentoalveolar surgery. Investigations were concentrated on the total number of viable bacteria and the spectrum of aerobes (facultative anaerobes) and strict anaerobes on the two suture materials.

MATERIALS AND METHOD

The study examined two different suture materials (Vicryl and Vicryl Plus; Ethicon, Norderstedt, Germany) of the same strength and needle type as used in dentoalveolar surgery: needle type V-5, length 70 cm, strength 4–0, undyed. According to the manufacturer, Vicryl is a synthetic sterile resorbable polyfil surgical suture material that consists of a copolymer of 90% glycolid and 10% L-lactide (polyglactin 910), and is coated with a copolymer of glycolid and lactide (polyglactin 370) and calcium stearate. Vicryl Plus has the same characteristics as Vicryl but is coated with the antibacterial agent triclosan (Irgacare MP) at a density not exceeding 150 µg /100 cm.

Over a period of 2 years, both suture materials were used for routine wound treatment in 17 patients (18–43 years old) who were undergoing wisdom tooth extraction. All 17 patients underwent extraction of two wisdom teeth on the same side at the same time. One operation site was closed by a Vicryl suture and the other by a Vicryl Plus suture; in addition, these were alternated between the maxillary and mandibular sites, respectively. No additional methods of disinfection (such as insertion of iodine-containing strips) were used. The sutures were removed after 7 days and immediately transferred into sterile tubes containing reduced transport fluid.

Isolation and differentiation of microorganisms. Microorganisms on the sutures were isolated by culture and identified. For that purpose, the sutures were agitated in peptone–yeast extract bouillon by using a vortex mixer. Dilutions (10^{-1} to 10^{-6}) were prepared, and 100 µl of each dilution were plated on yeast–cysteine blood agar (HCB) or Columbia blood agar (CoBl). The length of each suture was measured after any knots had been removed (average length of sutures: 2.9 cm). The HCB plates were used to cultivate anaerobic bacteria under strictly anaerobic conditions at 36 °C for 12 d (Anaerocult; Merck, Darmstadt, Germany). The CoBl plates were incubated at 36 °C in a 5–10% CO_2 atmosphere for 4 d to cultivate aerobic (facultative anaerobic) bacteria. All colonies with differing morphologies, colours, sizes or haemolytic reactions were selected to obtain as many of the predominant bacterial types as possible. The identified colonies were isolated by subculture on HCB or CoBl.

Morphological analysis, Gram staining was performed and cellular morphologies were determined by light microscopy.

Biochemical analysis. Fermentation of sugars and measurement of enzymatic activities were used to identify the isolated aerobic bacteria. Biochemical tests were performed with routine methods (e.g. detection of catalase or oxidase) and commercially available kits for microbial identification (e.g. API Strep; bioMérieux, Marcy-l'Etoile, France). Biochemical differentiation of isolated anaerobic bacterial strains was performed by routine tests (detection of indole, alkaline phosphatase, glucosidase and galactosidase) and commercially available tests (RapID ANAII; Oxoid, Wesel, Germany). The tests were performed according to the manufacturer's instructions. In addition, gas chromatographic analysis of fatty acid methyl esters (4) were used in some cases to identify anaerobic species.

***In vitro* susceptibility tests.** To determine the antibacterial effect of the Vicryl Plus suture in comparison with the conventional Vicryl suture, the diameters of the inhibition zones for *Aggregatibacter actinomycetemcomitans* (aerobic), *Actinomyces naeslundii* (aerobic), *Prevotella intermedia* (anaerobic), *Parvimonas micra* (*Peptostreptococcus micros*, anaerobic) and *Fusobacterium nucleatum* (anaerobic) were measured using the agar diffusion method [10]. For the inhibition of bacterial growth by Vicryl Plus sutures, the reported activity against *S. aureus* was used as a reference. The three aerobic species were cultured on CoBl plates, as well as on blood-free diagnostic sensitivity test (DST) plates (CM261; Oxoid, Wesel, Germany), and the three anaerobic species on HCB plates and on Wilkins-Chalgreen Agar. For each plate, a sterile, ~2-cm long fragment of Vicryl (USP 4–0) or Vicryl Plus (USP 4–0) was applied.

Statistical analysis. Pair-wise comparison of the data was carried out using the Mann-Whitney-U-test. Non-normal distribution was checked with the Kolmogoroff-Smirnoff-test. The maximum difference between the cumulative distribution is D: 0.2353 with a corresponding P of 0.673 for the data in Table 1 and Table 2 aerob. For Table 2 anaerob D: 0.1765 and P of 0930.

RESULTS

The results obtained show differences in the number and in the type of bacteria that adhered to the two types of suture material.

Table 1 shows the total number of cfu found on Vicryl and Vicryl Plus sutures that had been removed from 17 patients. The number varied from 1.05×10^5 – 1.4×10^8 with Vicryl and 1.5×10^4 – 1.8×10^8 with Vicryl Plus. Intra-individual

Table 1. Comparison of the total number of bacterial colonies on Vicryl and Vicryl Plus sutures in 17 patients

Patient No.	Vicryl *No. of colonies ×10³*	Vicryl Plus *No. of colonies ×10³*
1	8 020	2 176
2	1 204	1 908
3	18 158	7 194
4	9 454	16 886
5	23 300	180 200
6	30 640	40 860
7	2 641	92 620
8	21 286	9 769
9	105	15
10	6 060	3 618
11	75 720	94 420
12	16 266	4 000
13	140 150	67 470
14	40 334	5 935
15	1 950	4 000
16	97 510	58 320
17	40 010	141 910
Sum	532 808	731 301
Mean	31 342	43 018
Sd	38 846	55 221
Median	18 158	9 769

variations regarding the number of colonies were found. The total number of bacteria was 37% higher on the 17 Vicryl Plus sutures (7.3×10^8 colonies) than on those made of Vicryl (5.3×10^8 colonies). Table 2 compares the number of aerobic and anaerobic bacterial colonies that were grown from the Vicryl and Vicryl Plus sutures. Looking at the results for aerobic and anaerobic bacteria separately, in both cases, the mean total counts were higher for Vicryl Plus. Although the mean colony count for aerobic bacteria was only 2.6% higher, this value was 75% higher for anaerobic bacteria on Vicryl Plus. For evaluation of the two sutures, this finding is particularly important because the dominant role of anaerobic bacteria in infections of the mouth and in bacteremia is well-documented [12].

Table 2. Comparison of the number of aerobic and anaerobic bacterial colonies on Vicryl and Vicryl Plus sutures of 17 patients

Patient No.	Vicryl *No. of colonies ×10³*		Vicryl Plus *No. of colonies ×10³*	
	aerobic	anaerobic	aerobic	anaerobic
1	5 100	2 920	1 880	296
2	922	282	1 700	208
3	14 158	4 000	4 734	2 460
4	5 034	4 420	11 486	5 400
5	18 060	5 240	70 200	10 000
6	14 200	16 440	27620	13240
7	2 568	73	17 420	75 200
8	3 686	17 600	927	8 842
9	32	73	5	10
10	2 900	3 160	258	3 364
11	22 520	53 200	92 20	85 200
12	2 660	13 606	1 156	2 844
13	57 150	83 000	36 070	31 400
14	37 942	2 392	4 505	1 430
15	900	1 050	250	3 750
16	64 010	33 500	53 110	5 210
17	26 000	14 010	44 510	97 400
Sum	277,842	254 966	285 051	446 254
Mean	16 344	14 998	16 768	26 250
Sd	19 783	22 455	21 774	38 830
Median	5 100	4 420	4 734	5 210

To understand the implications of these differences in colony counts, it is necessary to characterise the bacterial species that were isolated in the various situations. Table 3 lists the aerobic species isolated and Table 4 the anaerobic species. Both Tables contain species regarded as pathogenic and those that belong to the normal flora. To evaluate the benefits of the different suture materials, it is helpful to analyse the pathogenic bacteria separately and assess their numbers.

The number of aerobic species found on Vicryl Plus (n=95) was lower than that on Vicryl (n=100) but the number of pathogenic aerobic species on Vicryl Plus (n=19) was higher than that on Vicryl (n=18), albeit only slightly (Tab. 3). The number of bacterial species of the normal flora on Vicryl (n=82) exceeded that on Vicryl Plus (n=76) more obviously. Therefore, not only is the number of pathogenic species higher, but the protective normal flora is reduced more markedly on Vicryl Plus. The differences were not significant statistically.

The same total number of anaerobic species (n=103) was found on both types of suture; however, the number of pathogenic anaerobic bacteria was again higher on Vicryl Plus (n=81) than on Vicryl (n=79) (Tab. 4). The number of Gram-positive and Gram-negative bacterial strains is listed separately in Table 5. The reduction in Gram-positive bacteria, particularly staphylococci, on Vicryl Plus has been noted in previous studies by other authors [8,9]. As expected

Table 3. Aerobic bacterial strains isolated from Vicryl and Vicryl Plus sutures in 17 patients

Aerobic bacteria	No. of strains isolated from the sutures	
	Vicryl	Vicryl Plus
Gram-positive cocci		
Streptococcus		
sanguis/oralis/mitis	**17**	**17**
salivarius	**13**	**10**
equisimilis[a]	0	1
mutans[a]	0	1
anginosus[a]	1	0
Stomatococcus	**0**	**1**
Gram-negative cocci		
***Neisseria* spp.**	**16**	**16**
Gram-positive rods		
Actinomyces		
odontolyticus	**11**	**10**
***Actinomyces* spp.**	**15**	**14**
Bacterionema/Rothia	**10**	**8**
Gram-negative rods		
Eikenella corrodens[a]	3	4
Capnocytophaga spp.[a]	11	12
Kingella spp.[a]	2	1
Enterobacter spp.[a]	1	0
No. of isolates in total	100	95
No. of pathogens	18	19
No. of normal flora	82	76

[a] Pathogens, **bold = normal flora**

Table 4. Anaerobic bacterial strains isolated from Vicryl and Vicryl Plus sutures in 17 patients

Anaerobic bacteria	No. of strains isolated from the sutures	
	Vicryl	Vicryl Plus
Gram-positive cocci		
Parv. micra[a]	17	13
Gram-negative cocci		
Veillonella parvula	**15**	**16**
Gram-positive rods		
Eubacterium spp.[a]	14	14
***Actinomyces* spp.**	**8**	**5**
***Bifidobacterium* spp.**	**1**	**1**
Gram-negative rods		
Prevotella		
intermedia[a]	9	13
corporis[a]	3	4
tannerae[a]	1	0
Prevotella spp.[a]	1	1
F. nucleatum[a]	17	17
Campylobacter rectus[a]	15	16
T. forsythia[a]	1	0
Selenomonas spp.[a]	1	3
No. of isolates in total	103	103
No. of pathogens	79	81
No. of normal flora	**24**	**22**

[a] Pathogens, **bold = normal flora**

Table 5. Comparison of the number of Gram-positive and Gram-negative bacteria on Vicryl and Vicryl Plus sutures in 17 patients

Patient No.	Vicryl: No. of bacterial strains		Vicryl Plus: No. of bacterial strains	
	Gram-positive	*Gram-negative*	*Gram-positive*	*Gram-negative*
1	7	5	5	5
2	7	8	6	7
3	5	5	5	6
4	7	6	8	6
5	8	4	5	5
6	8	6	5	7
7	5	8	5	6
8	7	5	7	6
9	5	5	4	6
10	5	6	2	7
11	8	5	7	9
12	7	7	6	6
13	7	6	6	6
14	7	6	7	6
15	2	4	4	4
16	6	6	5	6
17	6	4	8	5
Sum	107	96	95	103
Mean	6.3	5.6	5.6	6.1

Table 6. Zones of inhibition around bacteria from Vicryl and Vicryl Plus sutures

Aerobic species	Medium	Zone of inhibition (mm)	
		Vicryl	Vicryl Plus
S. aureus	CoBl	0	3
	DST	0	12
Agg. actinomycetemcomitans	CoBl	0	1
	DST	0	0
Act. naeslundii	CoBl	0	0
	DST	0	0
Anaerobic species			
Prev. intermedia	HCB	0	0
	WC	0	0
Parv. micra	HCB	0	0
	WC		0
F. nucleatum	HCB		0
	WC		0

WC – Wilkins-Chalgreen Agar

from these previous studies, in the current study fewer Gram-positive bacterial strains were found on Vicryl Plus (n=96) than on Vicryl (n=107). The opposite was found for Gram-negative bacteria: on Vicryl Plus, more Gram-negative strains (n=103) were detected than on Vicryl (n=95). The differences were also statistically not significant.

To test the effectiveness of the coating of the suture material, exploratory *in vitro* experiments were conducted with representative bacterial species (Tab. 6). The growth of the reference bacterium *S. aureus* was inhibited considerably on the Vicryl Plus suture (growth inhibition: 3 mm on CoB1, 12 mm on DST). However, inhibition of the growth of the oral bacteria tested was seen only with *Agg. actinomycetemcomitans* on blood agar (a small inhibition zone of 1 mm). With *Act. naeslundii* and the anaerobic bacteria *Prev. intermedia*, *Parv. micra* and *F. nucleatum*, no inhibition of growth was observed around the antibacterial-coated Vicryl Plus.

DISCUSSION

The triclosan-coated suture material Vicryl Plus has been in use in the USA since 2003. The antibacterial agent triclosan has been used as an additive to hygiene products, such as toothpaste and mouth-rinses, for about 20 years [13]. *In vitro* studies and animal experiments have documented the biocompatibility of the active ingredient triclosan [14], as well as the antibacterial efficacy of the suture against the bacteria that typically cause surgical wound infections [9]. Most studies have been limited to skin bacteria, such as *S. aureus, S. epidermis*, methicillin-resistant *S. aureus*, methicillin-resistant *S. epidermidis,* and *E. coli* (fewer studies) [6, 13]. However, to-date, only a few studies have examined the efficacy of the suture material in humans. This is particularly relevant to the special field of dentoalveolar surgery with its peculiar bacterial spectrum.

Under healthy conditions, the normal flora of the oral cavity encompasses both aerobic and anaerobic bacteria. At the onset of inflammation, and especially in infection, anaerobic bacteria predominate. A major difference between dentoalveolar infections and skin infections is the fact that the former are typically mixed infections with a multitude of aerobic and anaerobic microorganisms [4, 12]. The question arose: whether triclosan-coated sutures can be used successfully under these entirely different conditions? To investigate the issue, patients were selected for the present study who were undergoing simultaneous extraction of two wisdom teeth. One wound was closed with triclosan-coated Vicryl Plus suture and the other with uncoated Vicryl.

There were no differences in clinical course, other than redness and swelling around a few Vicryl Plus sutures. The significant reduction in the total number of bacteria by triclosan coating that has been described in the literature [6, 15] was not confirmed in the current study. There was no apparent advantage of one type of suture material over the other for this type of medical use (Tab. 2). The mean number of colonies of anaerobic bacteria was around 75% higher on Vicryl Plus than Vicryl, whereas for the aerobic bacteria, the difference was only 2.6%. No antibacterial effect of the coated suture was seen with the anaerobes which, on the contrary, adhered in greater numbers. No comparable results have been reported previously with triclosan-coated sutures. In a study with dentifrices that contained triclosan [11], the total cultivable flora was reduced significantly, but the results for strict anaerobic bacteria were not significantly different. On the other hand, the numbers of *Porphyromonas gingivalis, Prevotella intermedia* and *Tannerella forsythia* were found to be 1.5-, 5.8- and 2.1-fold greater, respectively, during the brushing period of 6 months with the triclosan-containing dentifrice, compared with the standard dentifrice.

The total number of adherent bacteria is not the only relevant parameter when considering the potential of an antibacterial agent to reduce wound infection. The agent's effect on each of (a) bacterial species that are considered pathogenic and (b) the normal flora in the oral cavity, needs to be determined. Ideal antibacterial treatment should have no effect on the normal flora, which is thought to have a protective effect. Analysis of the distribution of pathogenic bacteria versus normal flora yielded an unfavourable result for Vicryl Plus. On Vicryl sutures, 82 out of 100 aerobic strains belonged to normal flora, whereas on Vicryl Plus, only 76 out of 95 strains were from the normal flora. Furthermore,

pathogenic strains of bacteria were isolated more frequently from Vicryl Plus (*n*=19) than from Vicryl (*n*=18).

Comparison of the two suture materials for anaerobic species gave a similar but less pronounced result. Among 103 strains for both sutures, more belonged to the normal flora on Vicryl (*n*=24) than on Vicryl Plus (*n*=22). Fewer pathogenic strains were found on Vicryl (*n*=79) than Vicryl Plus (*n*=81). Thus, on sutures in the oral cavity, increased numbers of pathogenic species were detected on Vicryl Plus compared with Vicryl, which is in contrast to the results obtained with Vicryl Plus sutures in the skin [8,9]. In addition, the number of species from the normal oral flora was reduced on Vicryl Plus.

According to reports in the literature, Gram-positive bacteria are affected by triclosan up to 10 times more strongly than Gram-negative bacteria [8]. Therefore, the presented study examined the isolated strains with regard to Gram staining behaviour (Tab. 5). Gram-positive bacterial strains were found in smaller numbers on Vicryl Plus (*n*=95) than on Vicryl (*n*=107). However, as discussed above, this difference was due to a reduction in the normal flora and therefore was probably not an advantage of the coated material. In contrast, Gram-negative bacteria, predominantly pathogenic species, were recovered in higher numbers on Vicryl Plus (*n*=103) than Vicryl (*n*=96).

CONCLUSIONS

The *in vivo* results for Vicryl Plus triclosan-coated sutures did not meet expectations for their use in the oral cavity. Vicryl Plus sutures had the disadvantage of not reducing the number of Gram-negative pathogenic bacteria while reducing the number of protective bacteria of the normal flora. Because of the costs, the possibility of allergy and developing resistance, the use of triclosan-coated sutures is not recommended. Triclosan resistance is associated with multidrug-resistant bacteria in animals and humans [16]. If it is harmful for the environment, its unnecessary usage should be avoided.

Acknowledgements
The study was supported by in-house funds of the University Medical Centre in Freiburg, Germany. Ethicon provided free suture material to be used in the study which was performed in accordance with the Ethics Commission of the Medical Faculty of Freiburg University, and in accordance with the Helsinki Declaration. The submitted research is original and not presently under consideration for publication elsewhere.

Conflict of Interest
The authors have no conflicts of interest to declare.

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