Anti-bacterial Action of Multi-component Bioactive Glass Coating for Surgical Suture

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Abstract

Surgical sutures are textile biomaterial used for wound closure and ligate blood vessels. One of disadvantages of this material is establishment of infection at the surgical site. Coating sutures with bioactive glasses can be used to overcome such side effect. In this study, multi-component bioactive glass was prepared by melt-derived route and used to coat Mersilk suture by in-house slurry dipping technique. The antimicrobial action of this coating was investigated *in vitro* against *Staphylococcus aurous*, *Streptococcus mutans* and *Lactobacillus* and compared with tertiary-component (45S5) glass. The results indicated that multi-component bioactive glass coating exerted antibacterial action and this action increased with increasing glass concentration.

Key words: Bioactive glasses, Anti-bacterial, suture, coating.

الخلاصة

الخيوط الجراحيه هي مواد قماشيه تستخدم لخياطة الجروح وعقد الاوعيه الدمويه . ولكن واحدة من سلبياتها هو انتشار الالتهاب البكتيري في المنطقه الجراحيه .عمل غطاء للخيوط الجراحيه من مادة الزجاج الحيوي (البايولوجي) من الممكن ان يساعد في التغلب على هذة السلبيه .في هذه الدراسه الزجاج الحيوي المتعدد العناصر قد تم تحضيره بطريقة الذوبان الحراري لجعله غطاء ا الى الخيط الجراحي بطريقة التغطيس في المحلول المحليه .الفعل المضاد للبكتريا لهذا الغطاء قد تم دراسته ضد انواع من البكتريا *Streptococcus mutans and Lactobacillus Staphylococcus*

وقد تم مقارنته بالزجاج الحيوي الرباعي العناصر ٥ ٤٥ ٥ .النتائج اثبتت ان الزجاج الحيوي المتعدد العناصر كان فعالا ضد البكتريا المستخدمة في هذة الدراسه وان فعله يزداد بزيادة تركيز الزجاج الحيوي .

الكلمات المفتاحيه :الزجاج الحيوي المتعدد العناصرٍ,المضاد البكتيري ,الخيوط المغطاة.

1. Introduction

Sutures are textile biomaterials widely used for wound closure, to approximate tissue together and ligate blood vessels (Chue , 2002). Sutures can also be used in the field of tissue engineering for construction of 3D structures and meshes of controlled and variable pore size; using textile technology in the production of resorb able scaffold (Stamboulis *et al*, 2002). The materials of surgical sutures can be classified into synthetic or natural; absorbable or non-absorbable and monofilament or multifilament (twisted or braided).

One of disadvantages of surgical suture is establishment of infection at the surgical site. The bacterial biofilm adheres to the suture surface exhibits a more virulent phenotypic properties and promotes surgical site infection (Donlan, and Costerton, 2002). In addition, the multifilament braided suture harbors bacteria in the interstices between its fibers and keeps them a distance from phagocytosis; thus, inducing infection and tissue reaction (Enab *et al*, 2014). It is stated that implantation of sutures increases the susceptibility of surgical field to infection, as pathogenic bacteria proliferate in the surgical wound result in impairment of wound healing and separation of wound edges (Mingmalairak, 2011).

For this reason, there is a growing need to develop a material or coating for sutures that are able to reduce bacterial contamination and prevent surgical site infection. Coating sutures with anti-bacterial agents such as anti-biotics have been developed to reduce the infection at the surgical site (Enab *et al*,2014). However,

prolonged use of anti-biotics will eventually cause bacterial resistance and increase their virulence (Ricco and Assadian, 2011). An alternative approach is to coat surgical sutures with bioactive glasses. Bioactive glasses are special glass system generally composed of SiO_2, CaO, Na_2O and P_2O_5. The bioactive behavior of these glasses are related to their ability to form chemical bond with hard and soft tissue through a series at the glass-tissue interface (Hench *et al,* 1972). Interestingly, studies showed that these glasses can exert anti-bacterial action through ionic dissolution and rapid change in the pH of the surrounding medium (Stoor, 1999; Allan, 2001). Though, this action depends on many factors such as, glass composition, glass concentration, particle size and bacterial strains (Ahmed *et al*, 2006; Xie *et al*, 2009).

The present study investigated *in vitro* the antibacterial effect of multi-component bioactive glass-coated sutures and compared this effect with sutures coated with tertiary-component bioactive glass (45S5 glass) as well as with uncoated sutures. The multi-component bioactive glass is modified from the original 45S5 glass by adding Mg, Zn and K into glass composition. The efficacy of the coatings was investigated against *Staphylococcus aurous, Streptococcus mutans,* and *Lactobacillus*. These bacterial strains were chosen due to their relevancy to surgical site infection in the oral cavity.

2.Materials and methods

2.1 Glass synthesis

The 45S5 glass was synthesized using reagent grade chemicals (SiO_2, $CaCO_3$,Na_2CO_3 and P_2O_5) , the multi-component bioactive glass was prepared by adding MgO, K_2CO_3 and ZnO to the composition of 45S5 glass in the appropriate proportions. The composition of these glasses are listed in table 1. The glass batch was melted in a 300 mL platinum–rhodium alloy crucible using an electric furnace (Hope Valley, Lenton Thermal Designs, UK) at temperatures between 1450 and 1460 °C for one and a half hours. The melts were then rapidly quenched in deionized water to prevent crystallization and phase separation. The glass frit produced was collected in a sieve and dried overnight at 120 °C. The dried frit was then ground in a Gyro Mill (Glen Creston, UK) for 14 min and sieved for 60min in a sieve shaker (Retsch,VS1000, Germany) and separated into more and less than 45 μm particle size groups.

Table-1: **Chemical composition (mole%) of multicomponent bioactive glass and 45S5 glass.**

Glass	SiO_2	CaO	MgO	Na_2O	K_2O	P_2O_5	ZNO
multicomponent	41.7	36.3	7.8	5.2	1.0	4.7	3.0
45S5	48	36	…….	12	…….	4	…….

2.2 Coating surgical suture

The coated surgical sutures were prepared using in-house slurry dipping technique, as described by (Blacker et al,2004). The slurry was prepared by dispersing (0.25, 0.5 and 1 g) of multi-component and tertiary 45S5 bioactive glass particles (<45 μm) in 5 ml distilled water by stirring thoroughly . A length (~5mm) of commercially available Mersilk® (3/0) suture was dipped in glass powder suspension for 1 hour and left for 72 hours at room temperature to dry slowly. In homogeneities of the coating surface was expected during drying process.

2.3Culture and identification

Three types of bacterial strains were isolated from oral cavity (*Staphylococcus aurous, Streptococcus mutans ,Lactobacillus)* by swab samples. All specimens were inoculated onto Columbia blood agar with 5% defibrinated horse blood and mannitol

salt agar After incubation, a selection , colonies were Gram stained and tested for catalase production .Catalase positive ,Gram positive cocci were tested for coagulase production by slide agglutination. *S. aureus* was identified by macroscopic characteristics, lecithinase production and the coagulase test. Typical colonies for *Lactobacillus* species appear as white, smooth, with a diameter > 1 mm), Gram-positive, catalase negative rods . Colonies grown on mitis salivaris -agar medium was spread on the blood agar plates and incubated anaerobically for 2-5 days, to identification of *S.mutans*.(Bergeys, 1994 ; Macfaddin, 2000).

2.4 Antimicrobial activity

Antibacterial action of coated surgical suture with multi-component and 45S5 bioactive glasses was investigated *in vitro* using agar diffusion test. Bacterial strains (*Staphylococcus aurous*, *Streptococcus mutans* and *Lactobacillus*) were used to investigate the antibacterial action of these glass coatings. The inoculums were prepared by adding five isolated colonies to five ml of brain heart agar broth and incubated at 37^{o}C for 18- 24 hours to compare with (0.5) McFarland standard tube. Then, the inoculums were streaked on a Mueller Hinton agar plate and left to dry. The coated surgical suture were placed on the surface of the medium of Mueller Hinton agar plate that streaked with these bacterial strains and incubated for 24 hours. After the incubation period, the inhibitory zone was measured in millimeter (mm) using a transparent ruler. Uncoated surgical suture was used as a control.

3. Results and discussion

The antibacterial action of surgical suture coated with multi-component and tertiary-component (45S5) bioactive glasses was investigated using *Staphylococcus aurous*, *Streptococcus mutans* and *Lactobacillus* using agar diffusion method. The results showed that multi-component bioactive glass had significant antimicrobial action on the bacterial strains used in this study compared to tertiary (45S5) glass. In addition to that, the antimicrobial action of this glass increased progressively with increasing glass concentration, as shown in figure 1 and tables 2, 3 and 4.

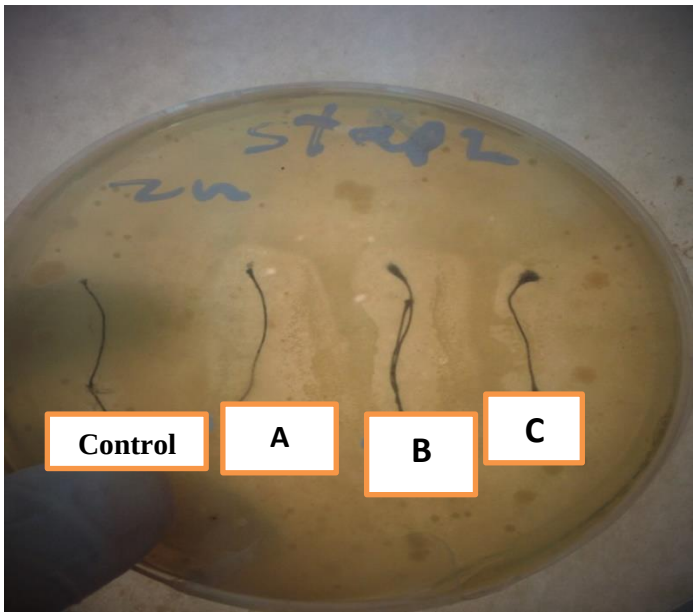

Figure (1) :- The inhibition zone produced by different concentration of multicomponent bioactive glass coating of Mersilk suture against *Staphylococcus aureus*A: 1gm/5mlD.W; B: 0.5 gm/5mlD.W ;C: 0.25 gm/5mlD.W.

The antimicrobial effect of multi-component bioactive glass coating of surgical suture could be attributed to the composition of this glass compared to tertiary-component 45S5 glass, as the former contains additional elements such as Mg, Zn and K in addition to the elements present in the composition of 45S5 glass. This increase in the alkali metals and alkaline earth ions in the composition of multi-component bioactive glass increases the alkalinity of the surrounding medium and hence exhibiting antibacterial action. Bellantone et al, 200 stated that the antimicrobial action of bioactive glasses is ascribed to the ionic dissolution of these glasses and subsequent increase in the pH and ionic concentration of the culture medium. However, this effect depends on glass composition, glass concentration and glass particles (Ahmed *et al*, 2006; Xie *et al*, 2009). Notably, in this study, particle size (< 45 µm) of multi-component and tertiary (45S5) glasses were used to assess their antimicrobial action against the bacterial strains.

The presence of zinc in the composition of multi-component bioactive glass could be another factor behind the antimicrobial action of this glass. Many studies postulated that zinc can exhibit antibacterial action at certain concentration (Scherer *et al*, 1989; Anaraku *et al*, 1975; Chrapil,1980; Simkin,1976). This effect of zinc could be associated with the release of hydrogen peroxide from zinc oxide surfaces (Sawai *et al*, 1998).

Table (2) Susceptibility of *Lactobacillus* according to type of bioactive glass coating.

Type of bioactive glass coating	Means at 0.25gm/5ml D.W	Means at 0.5gm/5ml D.W	Means at 1gm/5ml D.W
Multicomponent	1mm	4mm	5mm
45S5	0	0	0
control	0	0	0

Table (3) Susceptibility of *staphylococcus aureus* according to type of bioactive glass coating.

Type of bioactive glass	Means at 0.25gm/5ml D.W	Means at 0.5gm/5ml D.W	Means at 1gm/5ml D.W
Multicomponent	3mm	4mm	7mm
45S5	0	0	0
control	0	0	0

Table (4) Susceptibility of *Streptococcus mutans* according to type of bioactive glass coating.

Type of bioactive glass	Means at 0.25gm/5ml D.W	Means at 0.5gm/5ml D.W	Means at 1gm/5ml D.W
Multicomponent	2mm	2mm	4mm
45S5	0	0	0
control	0	0	0

The results of this study regarding 45S5 glass were consistent with other studies (Geyer *et al*, 1999; Stoor *et al*, 1996; Bellantone *et al*, 2002), as these studies indicated that this glass has no antibacterial action. However, (Brown *et al*, 2009) showed that 45S5 glass has antibacterial action against certain bacterial strains. This disagreement among different studies might be ascribed to the method used for glass synthesis, particle size and the tested bacterial strains.

4. Conclusion

Multi-component bioactive glass coating of surgical suture exerted antibacterial action against *Staphylococcus aurous*, *Streptococcus mutans* and *Lactobacillus*; whereas tertiary-component 45S5 glass coating did not. The antimicrobial action of multi-component bioactive glass coating increased with increasing glass concentration. Since multi-component bioactive glass coating exhibited antimicrobial action, it can be used for various clinical applications.

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