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Original Article

Anti-adhesion activity and physicochemical features of the surgical silk sutures coated with Liquidambar orientalis styrax

Liquidambar orientalis styrax ile kaplanan cerrahi ipek sütürlerin anti-adezyon aktivitesi ve fizikokimyasal özellikleri

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Abstract

Aim: The present study was aimed to characterize the surgical silk sutures coated with Styrax liquidus, Turkish sweetgum balsam obtained from Liquidambar orientalis, and to investigate their anti-adhesion capacity against oral pathogenic microorganisms.

Material and Methods: Fourier Transform Infrared Spectroscopy (FTIR) was applied to determine the chemical composition of the Liquidambar orientalis styrax (LOS)-coated sutures. Thermogravimetric Analysis (TGA) was performed to compare the thermal stability of the LOS-coated sutures. Scanning Electron Microscopy (SEM) was used to evaluate the morphological structure of the sutures. Anti-adhesion activity of the LOS-coated sutures was investigated against common oral pathogenic microorganisms.

Results: FTIR spectrum and SEM images revealed out that LOS was succesfully coated onto the silk sutures. TGA analysis showed that LOS coating moderately affected the thermal stability of the silk sutures. According to the anti-adhesion activity analysis, the highest activity was observed against S. aureus, a gram positive bacteria.

Conclusion: Coating the surgical silk sutures with LOS might be useful to prevent the surgical site infections in oral surgery.

Keywords: anti-adhesion; characterization; Liquidambar orientalis; styrax; suture

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Öz

Amaç: Bu çalışmada, Liquidambar orientalis'ten elde edilen Türk sığla balsamı olan Styrax liquidus ile kaplanmış cerrahi ipek sütürlerin karakterize edilmesi ve oral patojenik mikroorganizmalara karşı anti-adezyon kapasitelerinin araştırılması amaçlandı.

Gereç ve Yöntemler: Liquidambar orientalis styrax (LOS) kaplı sütürlerin kimyasal bileşimini belirlemek için Fourier Transform Kızılötesi Spektroskopisi (FTIR) uygulandı. LOS kaplı sütürlerin termal stabilitesini karşılaştırmak için Termogravimetrik Analiz (TGA) yapıldı. Sütürlerin morfolojik yapısını değerlendirmek için Taramalı Elektron Mikroskopisi (SEM) kullanıldı. LOS kaplı sütürlerin anti-adezyon aktivitesi, yaygın oral patojenik mikroorganizmalara karşı araştırıldı.

Bulgular: FTIR spektrumu ve SEM görüntüleri LOS'un ipek sütürlere başarılı bir şekilde kaplandığını ortaya koydu. TGA analizi, LOS kaplamanın ipek sütürlerin termal stabilitesini orta derecede etkilediğini gösterdi. Anti-adezyon aktivite analizine göre, en yüksek aktivitenin gram pozitif bakteri olan S. aureus'a karşı olduğu gözlendi.

Sonuç: Cerrahi ipek sütürlerin LOS ile kaplanması, oral cerrahide cerrahi bölge enfeksiyonlarını önlemek için yararlı olabilmektedir.

Anahtar kelimeler: anti-adezyon; karakterizasyon; Liquidambar orientalis; styrax; sütür

Introduction

Surgical site infections (SSIs) represent a major complication which occurs after surgical procedures [1, 2]. The reported rates of SSIs vary from 2% to 5%, accounting for 20% of all-health care-associated infections [1]. SSIs lead to longer hospital stays and greater health-care costs as well as substantial morbidity and mortality [3]. In case of infection, further surgical interventions may be required followed by a decrease in patient quality of life and work productivity [3, 4].

SSIs have a multifactorial nature. Bacterial colonization of the suture is one of the most important factors in the development of SSIs [1]. As a foreign body, suture materials may act as a potential surface for bacteria bioadherence and lead to microbial colonization on the incision site. The biofilm is established by the colonization and proliferation of microorganisms [2]. Once the biofilm is formed, the antibiotic treatment is often ineffective [3].

Silk has been widely used as a suture material due to its excellent internal performance. It is easy to use and provides a safe knot [5]. However silk sutures tend to cause a more intense and prolonged inflammatory response [6]. The braided nature of the silk suture facilitates bacterial accumulation, thereby increasing the risk of infection [5].

Several studies have demonstrated that developing silk sutures with good antibacterial properties is essential in order to avoid the suture being a risk factor for SSIs [5, 7]. Baygar et al. used silver nanoparticles (AgNPs) to coat the silk sutures [8]. Bide et al. reported the antimicrobial properties of silk sutures immobilized with ciprofloxacin [9]. Viju and Thilagavathi have studied the chitosan-based antimicrobial sutures [10].

The use of medicinal plants for the treatment of various infectious diseases has been known since ancient times. Medicinal plants are essential herbal products which represent an important source of biologically active compounds. The antimicrobial compounds of these plants prevent bacterial proliferation by mechanisms different from the commonly used antimicrobial agents. Therefore medicinal plants have a significant therapeutic value [11].

Liquidambar orientalis is a herbaceous plant which is known to have medicinal and cosmetic properties [12]. Liquidambar orientalis Mill tree is commonly known as “Sığla ağacı” or “Günlük ağacı” in Turkey. This species has locally distributed in the South-western coastal district of Turkey [13]. In the mediterranian region, it is commonly used in phytotherapy for treatment of various diseases including ulcer, stomach ache, mouth diseases, burn, wounds, cuts, whooping cough and skin diseases [14]. This herbaceous plant has good antiseptic properties [15]. The antimicrobial properties of the ethanolic extract of the leaves of Liquidambar orientalis have been previously studied [16, 17]. Styrax liquidus, locally named as “sığala or sığla yağı” is a resinous exudate obtained from the wounded trunk of Liquidambar orientalis Miller from Altingiaceae family (Hamamelidaceae) [18]. The balsam is not a natural part of the tree but is produced as a result of the stimulus from wounds in the bark. The outer bark is bruised, and then the inner bark becomes saturated with this pathological exudation. The outer bark is removed and the inner is boiled in water, the storax is skimmed off the surface as it rises, then afterward the boiled bark is pressed [15, 19]. Resin produced by injuring tree is a good antiseptic and has also been used as a topical parasiticide for the treatment of some skin diseases [20].

In the present study surgical silk sutures were coated with Liquidambar orientalis styrax (Styrax liquidus) using dip slurry technique. Coated sutures were characterized using Fourier Transform Infrared Spectroscopy (FTIR) and thermogravimetric analysis (TGA). Coated sutures were also investigated morphologically by Scanning Electron Microscopy (SEM). Antiadhesion activity was evaluated against oral pathogenic bacteria Candida albicans, Enterococcus faecalis, Staphylococcus aureus and Streptococcus mutans.

Material and Methods

LOS coating of the sutures

Liquidambar orientalis styrax used within the present study was obtained from a local company from Koycegiz Province, Mugla, Turkey and extracted with ethanol (1:10) then evaporated after filtration. Nonabsorbable 3.0 silk sutures (Dogsan, Turkey) silk sutures were dipped in LOS for 2 min, and dryed for 24h [8, 21].

Characterization of the LOS-coated sutures

Morphological and microanalytical characterization

FTIR Spectroscopy

FTIR spectrum of the LOS-coated sutures was obtained by FTIR (Thermo Scientific Nicolet iS10-ATR, USA) and compared. The spectra were recorded in the wavelength interval of 4000 and 400 cm^{-1}.

Thermogravimetric analysis (TGA)

Thermogravimetric analysis of non-coated and LOS-coated sutures were performed on a TGA instrument (Perkin Elmer TGA 4000, Perkin Elmer, Waltham, MA). Samples were heated from 30°C to 900°C at a rate of 10°C min^{-1} under a nitrogen flow rate of 20 mL min^{-1}.

Scanning Electron Microscopy (SEM)

Surface morphology of the LOS-coated sutures were evaluated using a JSM 7600F Field Emmission Scanning Electron Microscope (JEOL, Japan) and compared with the non-coated group. Non-coated and LOS-coated sutures were coated with gold before examining with SEM and monitorized under 15 kV accelaration voltage.

Anti-adhesion activity

Anti-adhesion activity of the LOS-coated silk sutures was determined against oral pathogenic bacteria obtained from American Type Culture Collection (ATCC); Staphylococcus aureus ATCC 25923, Enterococcus faecalis ATCC 29212, Candida albicans ATCC 10231 and Streptococcus mutans ATCC 25575. Suture fragments (1 cm) were incubated in inoculated broth media (Sabouroud Dextrose Broth for C. albicans, Mueller Hinton Broth for S. aureus and Brain Heart Infusion Broth for E. faecalis and S. mutans) under appropriate temperature for 24-48 h (37ºC for S. aureus and E. faecalis, 30ºC for C. albicans and 37ºC, 5% CO2 for S.mutans). After incubation periods, suture fragments were discarded and ultrasonicated in fresh broth mediums for 5 minutes. Ultrasonicated broths were incubated at appropriate periods again. Afterwards, the absorbances of the broth mediums were recorded at 540 nm using a UV-Vis spectrophotometer (Multiskan GO UV/Vis Microplate Spectrophotometer, Thermo-Fisher Scientific, USA) and the inhibition percentages were calculated.

Results

FTIR spectrum of the LOS-coated silk sutures is given at Figure 1. The peaks were obtained at 3277 cm^{-1}, 2939 cm-^{1}, 2160 cm^{-1}, 2027 cm-1, 1704 cm-1, 1633 cm-1, 1512 cm-1, 1448 cm-1, 1379 cm^{-1}, 1309 cm^{-1}, 1262 cm^{-1}, 1163 cm^{-1}, 1068 cm^{-1}, 971 cm^{-1}, 862 cm^{-1}, 765 cm^{-1} and 687 cm^{-1}.

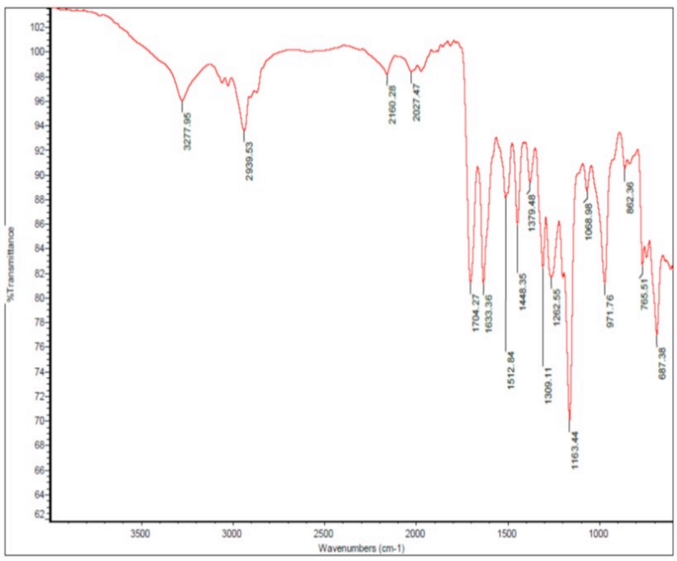

Figure 1. FTIR spectrum of the LOS-coated silk sutures.

TGA result of the LOS-coated suture is given at Figure 2. According to the TGA result, the initial decomposition stage which was marked at 0-100 °C was due to evaporation of water.

The second stage started at 225-260 °C. At 900°C, a total mass change of 85.79% was observed with a residual mass of 14.20%.

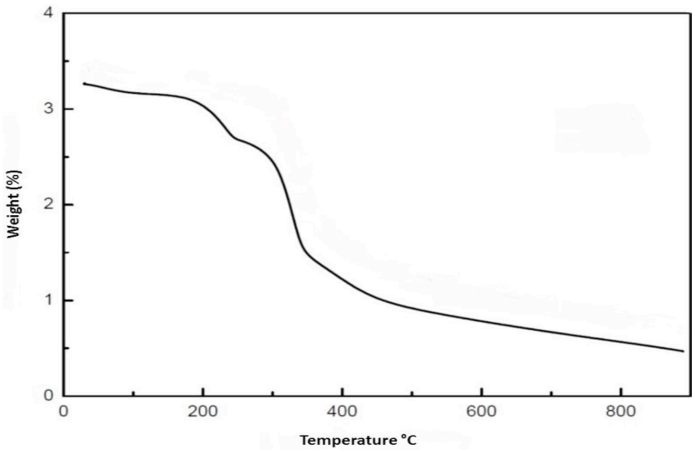

Figure 2. TGA spectrum of LOS-coated silk sutures.

The SEM micrographs of the non-coated and LOS-coated sutures are given at Figure 3. SEM images displayed that the diameter of the control group (non-coated) suture fragment was measured as 296 µm while the diamater of the LOS-coated suture fragment was 304 µm. The SEM images indicated the succesful coating process of the styrax onto the suture surface.

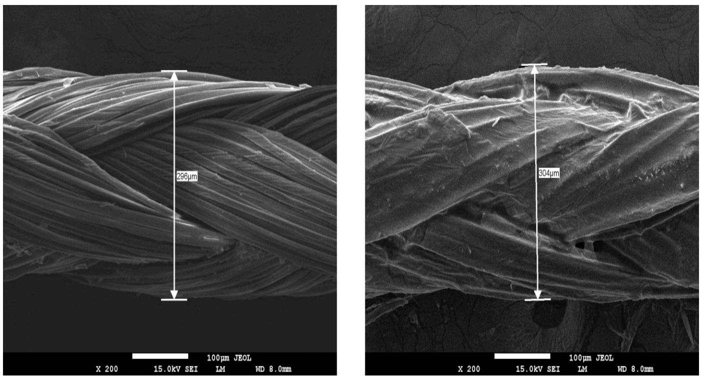

Figure 3. SEM images of the non-coated (left) and LOS-coated (right) silk sutures.

Anti-adhesion analysis indicated that, there was a 20.87% decrease in the biofilm amount onto the LOS-coated sutures (Table 1). There was not a significant inhibiton value for other microorganisms.

Table 1. Anti-adhesion activity of LOS-coated sutures

Microorganism	Inhibition %
S. mutans	4.66
E. faecalis	2.17
S. aureus	20.87
C. albicans	-

Discussion

Surgical silk sutures were coated with Liquidambar orientalis styrax and their anti-adhesion properties were characterized in the present study. Silk is commonly used as a suture material in oral surgery and accepted as a comparison standard for the assessment of suture properties [22]. Therefore, silk suture has been selected for this study. Similar to the SEM images, the FTIR spectrum of the LOS-coated sutures also indicated that the sutures were coated with LOS completely. The peak at 3277 cm^{-1} could be related to –OH group vibrations. The bands at 2939 cm^{-1}, 1448 cm^{-1}, 1379 cm^{-1} and 1309 cm^{-1} could be due to the C-H stretching vibration. The band at 1704 cm^{-1} could be probably related to the ketones. The bands between 687 cm^{-1} and 1262 cm^{-1} could be due to the steroidal or triterpenic structure [23]. According to the thermal decomposition stages, LOS-coating of the sutures moderately affected the thermal stability of the silk sutures. Elakkiya et al. (2014) reported that the weight loss was due to thermal decomposition of the antiparallel β-sheet structure of fibroin which forms the structural core of silk [24].

Surgical site infection remains one of the most common complications among surgical patients, which causes a significant amount of morbidity and increases medical costs [25]. In the presence of contamination, the sutures may act as a source of bacterial colonization and contributes to the development of surgical site infection [26]. The formation and growing of the bacterial biofilm on the surface of surgical sutures has been widely reported as an important causative factor for the SSIs [10, 25]. The oral cavity poses a high infectious potential due to the moist and vascularized environment [27, 28]. Multifilamentous and braided silk sutures cause a significant inflammatory reaction due to the bacterial adhesion in oral mucosa [28]. The suture related infections in the oral mucosa appear to be linked with the amount of contamination during the placement of sutures [27]. Therefore several studies have focused on the development of antimicrobial coated silk sutures. Janiga et al. (2012) employed the synergistic drug combination of levofloxacin-tinidazole to coat the silk suture [29]. They reported a good antibacterial activity and persistence against both Gram-positive and negative organisms. Baygar et al.(2019) used silver nanoparticles (AgNPs) obtained via a green synthesis approach [8]. The authors demonstrated a strong antimicrobial and antibiofilm capacity for AgNP-coated silk sutures. Pethile et al.(2014) concluded that coating silk sutures with a combination of poly(ε-caprolactone) (PCL) and sulfamethoxazole trimethroprim (SMZ) has a suitable antibacterial efficacy [5].

Liquidambar orientalis styrax, which is used traditionally to treat peptic ulcer disease by the inhabitants in the south-western Turkey, is a balsam obtained from the barks of Liquidambar orientalis tree [30]. Due to their potent antimicrobial activities, plant-derived secondary metabolites are known to be critical in the treatment of various diseases [31, 32]. Sağdıç et al. (2005) reported that the ethanolic extract of L. orientalis storax (styrax) had strong antibacterial activity against B. subtilis, E. coli, P. aeruginosa, S. aureus [15]. Within the present study, LOS-coated sutures had potent anti-adhesion activity against S. aureus, S. mutans and E. coli while there were no anti-adhesion activity against C. albicans strain. The major constituents of the L. orientalis styrax are reported as terpinen-4-ol, α-terpinol, sabinene and -terpinene along with cinnamyl cinnamete, phenylpropyl cinnamete, cinnamaldehyde, cinnamyl alcohol, ethyl cinnamate, methyl cinnamate and cinnamyl acetate [33-36].

The use of antimicrobially coated sutures presents a beneficial approach to deal with suture-associated infections [37]. By the year 2002, the Food and Drug Administration (FDA) approved the first antimicrobial surgical suture coated with triclosan which is a biocide that exhibits broad-spectrum activity against both gram-positive and gram-negative bacteria [38]. Since the introduction of triclosan-coated sutures, several studies have shown its efficacy for decreasing the rate of SSIs in various surgical operations [39-42]. However contradictory results have also been demonstrated and reported no change in terms of infection rates with the use of triclosan-coated sutures [43-45] In a current study, Tabrizi et al.(2019) compared the rate of SSI with the use of polyglactin 910 and polyglactin 910 coated with triclosan sutures in dental implant surgery. The authors found no significant difference between two groups and concluded that triclosan coated sutures had no influence for decreasing the incidence of SSIs in dental implant surgery[43].

Recently absorbable sutures coated with chlorhexidine have been developed and introduced as a commercial product into the markets. Studies on clorhexidine coated sutures have shown good antibacterial results and chlorhexidine was proposed as a promising agent for the prevention of SSIs [25, 46]. The antibacterial drug octenidine has also been investigated as a coating agent. The authors reported high antimicrobial efficacy and biocompatibility [37].

Based on the previous findings, the present study was designed to obtain an efficient surgical suture with potent antiadhesion activity. The results indicated that LOS-coated silk sutures may be beneficial for preventing SSIs following oral surgery operations.

Conclusion

The present study was conducted to characterize the surgical silk sutures coated with Liquidambar orientalis styrax and to display their antiadhesive potentials against oral pathogenic microorganisms. Sutures coated with LOS were found to have moderate antiadhesion activity. The results of the study figured out that, the strong biological activites of L. orientalis styrax may enhance the surface features of the sutures in respect to their antimicrobial and anti-adhesion capacities. The further studies are required to investigate the biomedical use of the LOS-coated sutures for dental applications and their clinical potential.

Declaration of conflict of interest

The authors received no financial support for the research and/or authorship of this article. There is no conflict of interest

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