Research Article

BIOMEDICAL
Journal of Scientific & Technical Research

ISSN: 2574 -1241

DOI: 10.26717/BJSTR.2023.53.008335

In Vivo Time-Lapse Observation of PLA/CL and PVDF Surgical Sutures by Optical Coherence Tomography

Fengcheng Wei[1]*, Hinata Nakakubo[1], and Masato Ohmi[1]

Department of Medical Physics and Engineering, Division of Health Sciences, Osaka University, Japan

***Corresponding author:** Fengcheng Wei, Department of Medical Physics and Engineering, Division of Health Sciences, Osaka University, Japan

ARTICLE INFO

Received: July 31, 2023
Published: September 26, 2023

Citation: Fengcheng Wei, Hinata Nakakubo, and Masato Ohmi. *In Vivo* Time-Lapse Observation of PLA/CL and PVDF Surgical Sutures by Optical Coherence Tomography. Biomed J Sci & Tech Res 53(1)-2023. BJSTR. MS.ID.008335.

ABSTRACT

We applied Optical Coherence Tomography (OCT), which has the advantages of high speed and high resolution on imaging, to perform *in vivo* animal experiments on surgical sutures. A Spectral-Domain OCT system was used to scan the absorbable PLA/CL surgical sutures and Non-absorbable PVDF surgical sutures, both of which had been sutured on a hairless rat. Two kinds of surgical sutures, polylactic acid/polycaprolactone (PLA/CL) compound surgical sutures, which is absorbable, and polyvinylidene fluoride (PVDF), which is non-absorbable, have been sutured on the back of a hairless rat. And a Spectral-Domain OCT was used to take the pictures of both surgical sutures and surrounding tissues every week till PLA/CL sutures was absorbed and broken in order to record and analyzed the changings. As a result, the deposits on the surface of the suture that were observed over time were decomposed substances of keratinocytes and fibers. Furthermore, with the decomposition, the crystal structure of PLA/CL fiber became amorphous, and reflected signal was detected. Some void also appeared which shows that the proliferated part detached from the sutures and the epidermal layer. PLA/CL sutures finally got broken at the 16th week, with the diameter decreased from 220 μm to 180 μm. *In vivo* time-lapse observation of surgical sutures can be accurately performed by OCT. It is also able to scan not only the surface but also the internals structure like crystal changes of surgical sutures. What's more, with 40 μm decrease on diameter, PLA/CL sutures broke after 15 weeks.

Keywords: *In Vivo* Observation; Optical Coherence Tomography; Surgical Suture; Biomaterials

Abbreviations: OCT: Optical Coherence Tomography; PLA: Polylactic Acid; PVDF: Polyvinylidene Fluoride; CL: Crystal Structure

Introduction

Since the latter half of the 20th century, various surgical sutures have been developed with advances in polymer materials science. Currently, clinically applied surgical sutures are mainly classified into two types: non-absorbable sutures and absorbable sutures. A non-absorbable suture needs to be removed after the wound is healed, and the suture itself is considered not to be decomposed or absorbed and mostly at a lower price range and has stable physical properties [1,2]. However, Poor healing and scarring of suture wounds have been reported when non-absorbable sutures are used [3]. Typical materials include nylon, polypropylene, and polyester. Absorbable sutures are produced using biodegradable materials and degrade with the passage of the time. The decomposition products fall off the skin surface or are excreted through the circulatory system. While there is no need to remove sutures, they are usually more expensive than non-absorbable sutures, their physical properties deteriorate with the passage of time, and there is a risk that they will break before the wound heal [4,5]. Typical materials include polylactic acid, silk, and catgut. There have been many studies on the physical and material properties of sutures. However, *in vivo* research on their biocompatibility is seldomly done [6-8]. However, as a biomaterial applied to the human body, animal experiments is still an irreplaceable method [9,10]. Therefore, in many cases, the suture is sutured to the living body, and after a certain period, the suture is taken out and measured [11]. If it is removed from the organism, it can be measured using specialized measuring equipment, but it has the disadvantage of using a destructive method which is cutting the surgical sutures and that may cause extra pain to the experimental subjects [12].

Until now, almost no studies have been conducted to avoid the above disadvantages and to observe and measure experimental subjects *in vivo*. Furthermore, a high resolution is required for observing such a fine structure, in which the diameter of the suture thread is basically only a few hundred micrometers. In the case of in vivo, there is movement due to heart pulsation and respiration of experimental animals, so high speed measurement is also required. On the other hand, since the 1990s, Fujimoto et al. have been able to observe eye and skin tissue using noninvasive optical coherence tomography (OCT) technology that detects backscattered and back reflected light [13]. Compared to conventional optical ranging measurements using femtosecond laser pulses, the energy of the light is lower, which makes it possible to apply it to *in vivo* experiments [14]. Furthermore, by using OCT, it is possible to observe more structures from both soft tissues such as retina layers and muscles and hard tissues such as bones and teeth [15]. In recent years, our laboratory has used OCT to analyze the behavior of the sweat glands on human fingertips during mental sweating [16]. Then, we conducted an *in vivo* animal experiment using guinea pigs and investigated the allergic dermatitis of skin tissues [17]. Based on the above previous studies, we strongly suggested that *in vivo* observation on surgical sutures by using high resolution rapid scanning OCT. Therefore, we sutured hairless rats with non-absorbable and absorbable sutures and performed time-lapse observation after surgery by OCT. From the image data, while confirming the change in the diameter of the yarn, it was compared with the reflected signal inside the yarn. Deposits on the outer circumference of the yarn were also examined.

Materials and Methods

Preparations of Experiment

A ten-week-old male hairless rat (SPF, HWY/slc, weight: 280 g) is used and kept in a constant temperature and humidity environment. During rearing, solid feed and tap water were freely taken. Animal experiments were conducted under the permission of the Osaka University Graduate School of Medicine Animal Experiment Ethics Committee. No. 6-0 (diameter approximately 70~100 μm) monofilament polyvinylidene fluoride (PVDF) non-absorbable suture (AR526, manufactured by Kono Seisakusho, blue) and polylactic acid/ polycaprolactone (PLA/CL) composite absorbable suture (LC516, manufactured by Kono Seisakusho, purple) was used in our suture surgery. Anesthesia by intraperitoneal administration of a three-kind mixed anesthetic was performed before the suturing surgery. Table 1 shows the amount of anesthetic used. Medetomidine (Nippon Zenyaku Kogyo CO., LTD.), Midazolam (Sandoz AG.), Butorphanol Tartrate (Meiji Animal Health Co., Ltd.) and Saline Solution (Otsuka Pharmaceutical Co., Ltd.) has been used in our research. After the animal›s nociceptive reflex disappeared, the suturing operation was started. Two parallel incisions of about 2 cm were made on the animal›s back, and four simple sutures were made at intervals of about 4 mm. (Figure 1) The whole suture operation took about 30 minutes.

Table 1: Injection Volume of drugs used in anesthesia

Drug	Injection Volume/mg/ml /kg
Medetomidine	0.15 mg/0.15 ml /kg
Midazolam	2 mg/0.4 ml /kg
Butorphanol Tartrate	2.5 mg/0.5 ml /kg
Saline Solution	1.45 ml/ kg

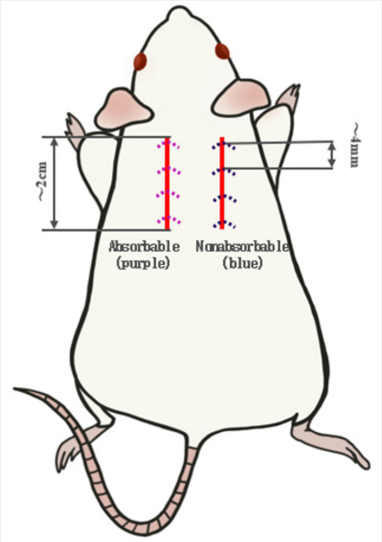

Figure 1: Schematic of the surgical surgeon. Two parallel incisions of about 2 cm were made on the animal's back, and four simple sutures were made by both absorbable(purple) and non-absorbable(blue) sutures at intervals of about 4 mm.

OCT Observation

Experiments were performed by using a Spectral Domain OCT (Telesto 320, manufactured by Throlab CO., LTD. Center wavelength 1300 nm, maximum imaging depth: air 3.6 μm /water 2.6 μm, axial resolution: air 5.5 μm /water 4.2 μm). 6 2D OCT images of each of the 8 sutured areas were recorded at the pace of once a week, and the absorbable suture was cut after 18 weeks. Finally, OCT images of 18 weeks' time lapse observation were taken. And the field of views of each OCT image is about 2 x 1.5 (X x Z axial) mm. 10 absorbable sutures and 10 non-absorbable sutures were selected from the acquired images of every week, and the diameter of the sutures in the vertical direction was measured by using Image J.

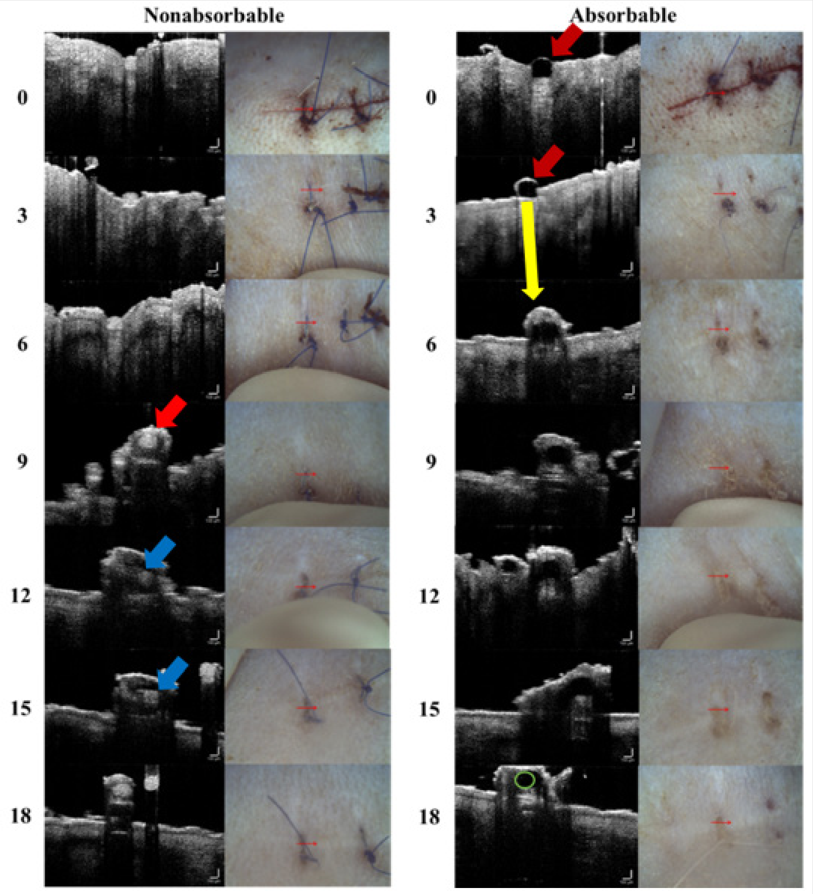

Figure 2:
a) Time lapse OCT images of nonabsorbable sutures. There are no obvious changes till 6th week. However, from 9th week upon the surface layer of the nonabsorbable sutures white signal area were appeared and indicated in red arrows. Voids (blue arrow) between non-absorbable sutures and white signal areas have appeared since 9^{th} week.
b) Time lapse OCT images of absorbable sutures. Upon the surface layer of the absorbable sutures white signal area were appeared and indicated in red arrows, and it increased with time (yellow arrow). White signal area also appeared inside the sutures at week 18 (green circle). For CCD camera pictures, the purple color faded with the decomposition of absorbable sutures.

Result

Time Lapse OCT Images

Figures 2(a) & 2(b) shows time-lapse images (0-18 weeks) of sutures sutured on the back of hairless rats. (a) is a non-absorbable suture and (b) is an absorbable suture. For the PVDF non-absorbable suture, a strong signal close to living tissue was detected and it appeared white. The PLA/CL absorbable composite suture appears black because it is close to air and the detected signal is rather weak [18]. Due to differences in their own properties and production processes, fiber materials vary greatly in internal microstructure [19-21]. It was suggested that the light scattering coefficients are also different for that reason. Furthermore, even at week 0, white areas could be confirmed upon the surface layer of the absorbable sutures (red arrow). It is regarded as the reflected light of incident OCT light from the surface. And the white parts increased with the lapse of time (yellow arrow). In addition to the reflected light from the light source, we presumed that with the absorbable suture thread decomposes, the fragments of PLA/CL, crusts and scales from tissues are also included. Furthermore, strong reflected signal was detected inside the absorbable suture over time (green circle), which indicates that the molecular chains inside the fiber were unraveled, and the homogeneous crystal became amorphous, and the decomposition progressed. On the other hand, from week 9, a strong signal was also clearly detected on the surface of the non-absorbable suture (orange arrow), so we verified that the white areas on the surface of the sutures were the exudates from the surrounding skin tissues, which were mainly thought to be crusts and scales.

And from week 12 onwards, there were too many deposits around the sutures, so it gradually dropped and peeled off from the surface of the sutures, and voids (blue arrow) between sutures and white signal area appeared. It has been demonstrated that PVDF non-absorbable sutures have better antibacterial and antifouling properties than the PLA/CL absorbable sutures we used in this research. In addition, from the CCD camera photograph, it was confirmed that the color of the absorbable suture gradually matched from the third week, while the non-absorbable suture remained almost unchanged. Polymer materials such as PLA and PCL are originally transparent or translucent materials. The purple suture used in this experiment was found to be dyed. It was also proved that the absorbable suture was decomposed by dropping the dye [22].

Time Lapse Diameter Changing of Absorbable Sutures

As absorbable sutures degrade over time, in theory, the diameter of the fiber is thought to decrease 22). Therefore, we decided to calculate the suture diameter (Figure 3). Absorbable sutures were shown in purple and non- absorbable sutures shown in blue. The non-absorbable sutures were about 160 µm at week 18, which was almost the same as what it was at week 0 (p>0.05). On the other hand, the diameter of absorbable sutures was reduced from 220 µm to 180 µm (p>0.05), which indicated that the diameter changes due to the decomposition and absorption of the PLA/CL composite surgical sutures was confirmed.

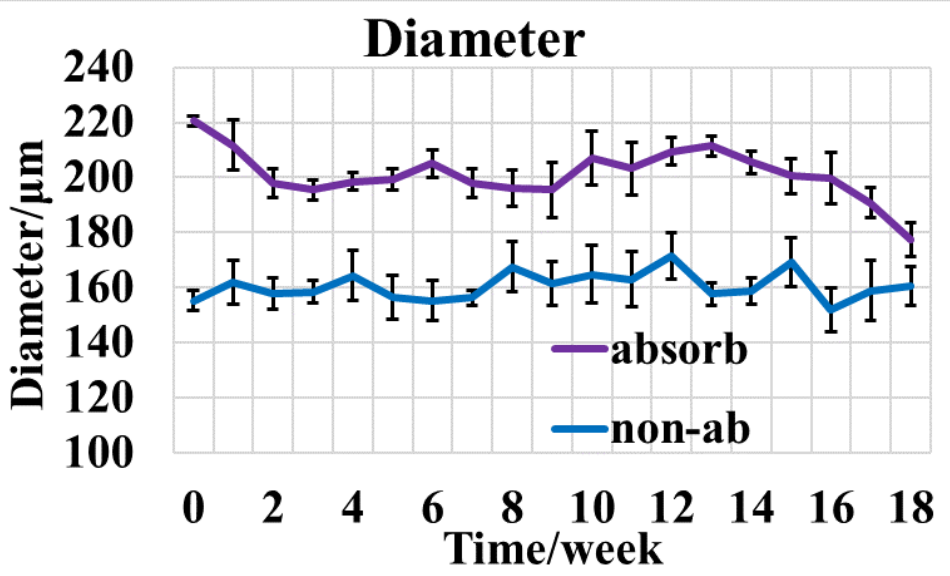

Figure 3: Diameter of absorbable sutures(purple) and non-absorbable sutures(blue). There is almost no change on non-absorbable sutures, but absorbable sutures got about 20 µm decrease.

Discussion and Conclusion

In this study, by using OCT, we were able to observe and measure non-absorbable and absorbable sutures sutured to a hairless rat *in vivo*. We confirmed that even hydrophobic materials such as PVDF have deposits from the surrounding skin tissues on the surface, but they would partly drop off over time. Due to the degradation of absorbable sutures, amorphization of surgical sutures' crystal happened and the signal of backscattered light and back reflected light inside the fiber increased, and reduction in diameter were also able to be evaluated. Through this research, we proved that even with ordinary OCT equipment, it is possible to measure minute level biomaterials and medical supplies, such as No. 6-0 sutures, which are several tens of µm in size. The measurement can be made not only for the body of the biological material, but also for the surrounding tissues. Furthermore, since OCT uses infrared wavelengths of light, it is possible to observe the reflection and scattering of light from the surface and internal structures of materials corresponding to that wavelength. In other words, it is strongly suggested that the use of OCT technology can also be able to analyze the internal structure of materials in *in vivo* research. In addition, now that the OCT equipment that can measure the refractive index is widely applied, it is considered possible to measure the refractive index change in the internal structure of the biomaterials like surgical sutures. From now on, by taking the advantage of high speed, high accuracy, and high resolution, we expect that we will be able to use OCT to make more accurate evaluations of *in vivo* animal experiment and medical materials under preclinical development. In the future, we aim to use an OCT device to measure the birefringence of surgical sutures, and to research not only surgical sutures but also artificial skin, hair regeneration, and percutaneous drug absorption.

Disclosures

The authors have no financial conflicts of interest to declare related to this study.

References

1. Travis L Callahan, William Lear, Jamie J Kruzic, Cory B Maughan (2017) Mechanical properties of commercially available nylon sutures in the United States. Journal of biomedical materials research 105(4): 815-819.
2. Moqaddaseh Afzali Naniz, Mahdi Bodaghi, Majid Safar Johari, Ali Zolfagharian (2020) Influence of hybridization on tensile behaviors of non-absorbable braided polymeric sutures. Polymers 12(3): 682.
3. Lin Xu, Yanan Liu, Wenhui Zhou, Dengguang Yu (2022) Electrospun Medical Sutures for Wound Healing: A Review. Polymers 14(9): 1637.
4. Christopher Dennis, Swaminathan Sethu, Sunita Nayak, Loganathan Mohan, Yosry Yos Morsi, et al. (2016) Suture materials — Current and emerging trends. Journal of biomedical materials research 104(6): 1544-1559.
5. Dong-Hoon Shin, Won Yeol Ryu, Jae Ho Jung (2020) The effect of absorbable and non-absorbable scleral suture on strabismus surgery in the rabbits. Curr Eye Res 45(10): 1252-1256.
6. John V Linberg, Linn M Mangano, J Vernon Odom (1991) Comparison of Non-absorbable and absorbable sutures for use in oculoplastic surgery. Ophthalmic plastic and reconstructive surgery 7(1): 1-7.
7. Mohammed A El-Bakary, Kermal A El-Farahaty, Nayera M El-Sayed (2019) Investigating the mechanical behavior of PGA/PCL copolymer surgical suture material using multiple-beam interference microscopy. Fibers and Polymers 20(6): 1116-1124.
8. Kai Shao, Baoqin Han, Jinning Gao, Zhiwen Jiang, Weizhi Liu, et al. (2016) Fabrication and feasibility study of an absorbable diacetyl chitin surgical suture for wound healing. Journal of biomedical materials research B: applied biomaterials 104(1): 116-125.
9. G Molea, F Schonauer, G Bifulco, D D'Angelo (2000) Comparative study on biocompatibility and absorption times of three absorbable monofilament suture materials (Polydioxanone, Poliglecaprone 25, Glycomer 631). British Journal of Plastic Surgery 53(2): 137-141.
10. Ying Yang, Heng-Bing Yang, Yu-Gang Wang, Shu-Hong Zhang, Zhi-Feng Yu, et al. (2017) Bacterial inhibition potential of quaternised chitosan-coated VICRYL absorbable suture: An *in vitro* and *in vivo* study. Journal of orthopaedic translation 8: 49-61.
11. KD Schlüter (2019) Tierversuche: Übertragbarkeit auf den Menschen und ihre Durchführbarkeit. Zeitschrift für Herz, Thoraxund Gefäßchirurgie 33: 67-71.
12. Jagdish Rai, Kuldeep Kaushik (2018) Reduction of Animal Sacrifice in Biomedical Science & Research through Alternative Design of Animal Experiments. Saudi pharmaceutical journal 26(6): 896-902.
13. David Huang, Eric A Swanson, Charles P Lin, Joel S Schuman, William G Stinson, et al. (1991) Optical coherence tomography. Science reports 254: 1178-1181.
14. S Bourquin, AD Aguirre, I Hartl, P Hsiung, TH Ko, et al. (2003) Ultrahigh resolution real time OCT imaging using a compact femtosecond Nd: Glass laser and nonlinear fiber. Optics express 11(24): 3290-3297.
15. Masato Ohmi, Yasuhito Ohnishi, Koji Yoden, Masamitsu Haruna (2000) *In vitro* simultaneous measurement of refractive index and thickness of biological tissue by the low coherence interferometry. IEEE transaction on biomedical engineering 47(9): 1266-1270.
16. Masato Ohmi, Motomu Tanigawa, Akihiro Yamada, Yoshihiro Ueda, Masamitsu Haruna (2009) Dynamic analysis of internal and external mental sweating by optical coherence tomography. Journal of biomedical optics 14(1): 14-26.
17. Masato Ohmi, Yuki Yoshida, Yoken Son, Kohji Abe (2022) *In vivo* observation of allergic dermatitis of the genuine pig by optical coherence tomography. Modern research in inflammation 11(1): 1-8.
18. Kelly M Brinsko (2010) Optical characterization of some modern "eco-friendly" fibers. J forensic Sci 55(4): 915-923.
19. Masatsugu Mochizuki (2010) Crystallization Behaviors of highly LLA-rich PLA effects of D-isomer ratio of PLA on the rate of crystallization, crystallinity, and melting point. Sen'I Gakkaishi 66(2): 70-77.
20. Marcus Himmler, Dirk W Schubert, Thomas A Fuchsluger (2021) Examining the transmission of visible light through electrospun nanofibrous PCL scaffolds for corneal tissue engineering. Nanomaterials 11: 13-19.
21. HM El-Dessouky, C A Lawrence, AM Voice, ELV Lewis, IM Ward (2007) An interferometric prediction of the intrinsic optical properties for cold-drawn iPP, PTFE and PVDF fibres. Journal of Optics A: Pure and Applied Optics 9(11): 1041-1047.
22. Olivier Claude, Thomas Gregory, Sebastiano Montemagno, Patrick Bruneval, Emmanuel H Masmejean (2007) Vascular microanastomosis in rat femoral arteries: experimental study comparing non-absorbable and absorbable sutures. Journal of reconstructive microsurgery 23(2): 87-91.

ISSN: 2574-1241

DOI: 10.26717/BJSTR.2023.53.008335

Fengcheng Wei. Biomed J Sci & Tech Res