Influence of suturing material on wound healing – An experimental study on dogs

Uticaj materijala za šivenje na zarastanje rana – eksperimentalna studija na psima

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Abstract

Background/Aim. The most common materials implanted in the human organism are suture materials that are classified on the basis of several criteria, usually the origin, structure, and properties. The properties of suture materials are related to its absorbability and non-absorbability. When using resorbable materials it is of great importance to determine whether its absorbability and tensile strength help wound healing in function of time. Sutures themselves can become a source of inflammation, that may reduce or compromise the potential of repairation and regeneration. The aim of this experimental study on dogs was to ascertain whether the absorption rate and the degree of local tissue reactions differ from information provided by the manufacturers, whether there are differences between the applied suture materials and which of the used suture materials have better effect on wound healing. Methods. Experimental testing of the selected suture materials basic characteristics was performed on 6 German Shepherd dogs, which, after induction of general anesthesia, were made 3 identical incisions each in all 4 quadrants (left and right side of the upper and lower jaws), so that 12 horizontal incisions were formed, 10 mm long, 20–25 mm rants (left and right side of the upper and lower jaws), so that there are differences between the applied suture materials and which of the used suture materials have better effect on wound healing. Results. The obtained results suggest that catgut has the fastest absorption rate, while Dexon® the lowest. Vicryl-Rapid® causes the lowest level of local reactions, while Dexon® the highest. Conclusion. There is no ideal suture material because various patient factors also influence the wound healing process.

Key words: oral surgical procedures; suture, techniques; catgut; wound healing; dogs; histological techniques.

Klučne reči: hirurgija, oralna, procedure; šavovi, tehnike; šavovi, materijali; rana, zarastanje; psi; histološke tehnike.

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Introduction

Surgical suture materials, in the strict sense suture, denote material used for reconstruction of tissue, hemostasis (ligation of blood vessels), fixing of tissue, as well as various transplants.

The most common materials implanted in the human organism are suture materials. It is estimated that 3 million individual stitches are placed worldwide on daily basis. Stitching up wounds is an ancient skill mentioned in Egyptian writings from 3,500 BC. Ancient Egyptian mummies are found with body cavities stitched up with suture materials made of animal tissue (tendons), braided horsehair, leather strips and plant fibers.

Centuries later, in the writings De Medicina, Celsus wrote about stitching up soft tissue with human hair. Galen, the physician of Roman gladiators, recommended using silk and hemp thread for hemostasis in the year 400 BC.

In Renaissance, Andreas Vesalius recommended stitching up fresh wounds, tendons and nerves. Physic, professor of surgery at the University of Pennsylvania, made absorbable suture materials of goat and deer skin in 1806. Catgut sutures were coated with chromium in 1876, which resulted in stronger stitch. Wound healing was enhanced when Joseph Lister introduced carbolic acid as suture disinfectant.

The first absorbable synthetic suture material made of polyvinyl alcohol was manufactured in 1931. In the same year, BASF laboratories produced the first polyamide suture (supramid), which was introduced in clinical practice after WWII. In the second half of the sixties it was discovered that polyglycolic acid could provide a material of particularly favorable features, so the first derivative of this substance called Dexon appeared in 1970.

Regardless of the fact that technological development of suture materials starts in the 30s of the 20th century, owing to development of new technologies and research, there is a great variety of these materials of different characteristics nowadays. However, the ideal suture material has not been found yet.

Suture materials are classified on the basis of several criteria, usually the origin, structure, and properties.

By origin, surgical sutures can be natural and synthetic; by structure they can be monofilament, multifilament (braided, twisted and spun) and pseudo-monofilament; by properties in tissue they can be absorbable and non-absorbable.

Healing of superficial wound tissue usually takes five to ten days, but some surgical procedures require sutures to persist 14 to 28 days.

Absorbable sutures are degraded and thus gradually lose strength in supporting tissues. As defined by the US Pharmacopoeia, most tensile strength of absorbable sutures is lost during the period of 60 days, as opposed to non-absorbable that retain it for longer than 60 days.

Synthetic absorbable sutures are dissolved by hydrolysis, which takes place in rather precise timeframe. Given that absorbable suture materials are dissolved under the influence of proteolytic enzymes or hydrolysis, it would be preferable not to remain in tissue longer than necessary.

Sutures themselves can become a source of inflammation, which may reduce or compromise the potential of repairation and regeneration. There are not many references comparing or discussing properties and quality of absorbable suture materials.

Oral surgery interventions are customarily finished by stitching surgical wound up, and choice of most appropriate suture material depends on the site and depth of tissue to stitch. Given that this type of intervention includes stitching up overlying tissue, routinely used are non-absorbable materials (usually silk sutures), which are removed after 5–7 days.

In some situations, however, when it is not possible to remove the placed sutures, absorbable materials are used for oral surgery interventions in case of persons with disability who are not able to cooperate, when removing sutures would imply induction of general anesthesia, for children and for soldiers from remote units, without adequate conditions for revisit and removing sutures (this can be quite common in states of emergency and during wartime).

From the viewpoint of their usage in oral surgery, absorbable suture materials are required to preserve their tensile strength long enough, not to have absorption time shorter than necessary for wound to heal, to keep adherence of soft sediments on sutures as slight as possible and, despite enzyme absorption process, to keep inflammatory reaction least intensive possible.

Information concerning these characteristics of specific absorbable materials is included in the manufacturers instruction manuals and stipulated prior to their mass production. However, neither measuring methods nor types of tissues used for placing these materials during tests, are known.

The aim of this study was to ascertain whether the absorption rate and degree of local tissue reactions differ from information provided by the manufacturers, whether there are differences between the applied suture materials and which of the used suture materials have better effect on wound healing.

Methods

Experimental testing of basic characteristics of the selected suture materials was performed on 6 German Shepherd dogs, which, after induction of general anesthesia, were made 3 identical incisions each in all 4 quadrants (left and right side of the upper and lower jaw), so that 12 horizontal incisions were formed, 10 mm long, 20–25 mm distant from one another, on each animal.

The experimental part of the study was fully implemented in accordance with the Act “The ethical principles of scientific experimental animals research”, Military Medical Academy, Act No. 282-10, issued on November 20, 2002.

All experimental animals were 6 years old. Dogs were marked randomly with numbers 1 to 6. Dog number 1 male, body weight 25 kg; dog number 2, female, body weight 30 kg; dog number 3, male, body weight 24 kg; dog number 4,
male, body weight 27 kg; dog number 5, male, body weight 21 kg and dog number 6, female, body weight 19 kg.

The animals were administered atropine in the dose 0.02–0.04 mg per kg of body weight, subcutaneously (SC), 15 min prior to administering anesthetics. After 15 min, they were administered propionylpromazine (Combelen®), 0.03 mL per kg of body weight intravenously (IV) and Ralatek® (ketamin) 5%, 0.3 mL per kg of body weight intramuscularly (IM) 20. Upon induction of anesthesia, 3 horizontal incisions, 1 cm long each were made in the vestibula of the animals, on the left and right side of upper and lower jaws, with equal spacing (Figure 1A).

Randomly, incisions were stitched up in the following order, starting from back to front: 1. catgut, 2. Dexon®, 3. Vicryl-Rapid®. Each incision was stitched up with individual stitch in the given order. Incisions were stitched up with catgut 3.0, Dexon® 3.0, Vicryl-Rapid® 3.0, with identical 3/8 circle needles (Figure 1B). Knots were tied using identical technique – the first knot was tied in clockwise direction, the second one counter-clockwise, and the third one in the same direction as the first one. So each animal had 4 incisions stitched up with each of the 3 types of material, or 12 incisions total. A total of 72 incisions were made and 72 individual stitches were tied with triple knots, or 24 per type of suture material on day zero when the experiment commenced. The appearance of the stitched up incisions on the postoperative day 7 is shown in Figure 1C.

Tissue samples for histological assessment of the speed of absorption and the degree of local reaction to implanted suture material were taken at predetermined time intervals. Each animal was taken biopsy on postoperative days 3, 7, 14 and 21 in the following way: biopsy from the upper right quadrant was taken on postoperative day 3, biopsy from the upper left quadrant was taken on postoperative day 7, biopsy from the lower left quadrant was taken on postoperative day 14 and biopsy from the lower right quadrant was taken on postoperative day 21 (Figure 1D) for all 3 types of suture material. Each section-taking on the preset days included 18 biopsies.

After reinduction of general anesthesia in the same way as on day zero, biopsy was taken from the upper jaw right on day 3, from the upper jaw left on day 7, from the lower jaw left on day 14 and from the lower jaw right on day 21, for all 3 types of suture materials by taking circular sections of gingiva and submucosal tissue, 2 cm diameter, including sutures.

Section wounds were stitched up (Figure 1E) with catgut 4.0, which was not removed, and sections were fixed in 10% neutral buffered formalin solution, packed in sterile vials and marked with letters A (catgut), B (Dexon®) and C (Vicryl-Rapid®) and the numbers 3, 7, 14 or 21. Prepared specimens were sent to the Institute of Pathology, Military Medical Academy, where they were embedded in paraffin molds (Figure 1F), cut with microtum 5–7 microns thick, and stained using hematoxylin and eosin, and the specific histochemical techniques: Masson trichrome, Paff Halmi periodic acid-schift (PAS) and PAS diastasis. Histopathological examination of specimens was then performed.

Fig. 1 – A) Incisions on experimental animals; B) Stitching up incisions on experimental animals; C) The appearance of the stitched up incisions on the postoperative day 7; D) The appearance after taken biopsies on the postoperative day 21; E) Stitched up biopsy sites; F) Vials with the taken sections soaked in formalin and in order for pathohistological analysis.

Results

Experimental part of the study was finalized with histopathological analysis of the specimens.

Histopathological findings on the postoperative day 3

On the postoperative day 3, on the sections taken from the postoperative wound sites stitched up with catgut, from all 6 dogs, a part of stratified squamous epithelium, and the connective tissue with blood vessels underneath were visible. Parts of suture were visible in one spot and gingival defects were also present in that area. In the connective tissue surrounding sutures, rare cells of chronic inflammation, predominantly lymphocytes were visible. On the postoperative day 3 intensive inflammation surrounding the implanted sutures was detectable, with inflammation cells including lymphocytes, leukocytes and plasma cells. There was no formation of new connective tissue nor the presence of new blood vessels (Figure 2a).

On the postoperative day 3, on the sections taken from all 6 dogs from the sites of postoperative wounds stitched up with Dexon®, preserved stratified squamous gingival epithel-
lium was visible, and connective tissue underneath with fresh bleeding spots. Gingival epithelium was missing in one spot and deep cavity protruding into the connective tissue was detectable, with remains of suture in it (Figure 2b).

In the wide area around the described cavity, rich non-specific chronic inflammation cells infiltration, prevalently lymphocytes were visible. There was sporadically a formation of foreign body type cells. On the postoperative day 3, intensive inflammation surrounding the implanted sutures was detectable, with inflammation cells that included lymphocytes, leukocytes and plasma cells. There was no formation of new connective tissue nor the presence of new blood vessels.

On the postoperative day 3, on the sections taken from all 6 dogs from the sites of postoperative wounds stitched up with Vicryl-Rapid®, partly preserved stratified squamous epithelium was visible. Remains of surgical sutures were visible in one spot. Around this area, as well as in somewhat broader range, in the connective tissue, rich non-specific chronic inflammation cells infiltration was present, mainly lymphocytes (Figure 2c).

On the postoperative day 3, intensive inflammation was detectable with inflammation cells that include lymphocytes, leukocytes and plasma cells. There was no formation of new connective tissue nor the presence of new blood vessels.

Besides taking sections for histopathological examination on the postoperative days 3, 7, 14 and 21 there were records about the presence of each single stitch implanted on the day-zero of the experiment (for all 3 types of suture materials).

Histopathological findings on the postoperative day 7

On the sections taken from all 6 dogs on sites of postoperative wounds stitched up with catgut, a part of stratified squamous gingival epithelium was visible. Rare lymphocytes were visible. The site of implanting surgical sutures in the connective tissue, while many fibroblasts and fibrocytes with initiated formation of granulation tissue were present (Figure 3c).

A large number of new blood vessels could be seen. Inflammatory changes were still present, as well as remnants of surgical sutures. One histological specimen (tissue section taken from dog number 2 showed a cavity in the connective gingival tissue with minor fragments of surgical sutures. This cavity was mainly surrounded by rich clusters of lymphocytes and leukocytes, with eosinophilic albuminous mass between them. The remaining connective and muscle tissue was permeated by inflammatory cells, prevalingly lymphocytes and leukocytes.

On the sections taken from all 6 dogs at the sites of postoperative wounds stitched up with Vicryl-Rapid®, the connective gingival tissue still included a significant number of lymphocytes, but also increased number of fibroblasts and fibrocytes, with formation of new granulation tissue. There was a spot with residues of surgical sutures in its center, which was surrounded by strongly increased number of fibroblasts, fibrocytes and new blood vessels with formation of young granulation tissue (Figure 3d).

Rare lymphocytes and leukocytes were visible. The site of Vicryl-Rapid® placement showed still present cell inflammation, but significantly milder. Rare fragments of surgical sutures and increased number of new blood vessels were visible.

Histopathological findings on the postoperative day 14

On the sections taken from all 6 dogs at the sites of postoperative wounds stitched up with catgut, mainly preserved gingival epithelium was visible. In the gingival connective tissue surrounding the site of implanted surgical sutures, proliferation of new connective tissue with large number of new blood vessels, fibroblasts and fibrocytes were

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found whose lumen was filled with blood. The granulation tissue was surrounded by clusters of chronic inflammation cells, mainly lymphocytes (Figure 4a). Rare giant cells were also present. Traces of chronic inflammation were still visible in the connective tissue, as well as remnants of surgical sutures. The number of new blood vessels was somewhat increased.

On the sections taken from all 6 dogs at the sites of postoperative wounds stitched up with Dexon®, cavity with fragments of surgical sutures was evident. This cavity was surrounded by multiplied fibroblasts and fibrocytes, as well as new blood vessels. Rare lymphocytes and giant cells were present. Traces of chronic inflammation in the connective tissue and fragments of surgical sutures were still visible. A somewhat increased number of new blood vessels were present (Figure 4b).

Histopathological findings on the postoperative day 21

On the sections taken from all 6 dogs at the sites of postoperative wounds stitched up with catgut, in the gingival connective tissue surrounding the site of implanted surgical sutures numerous macrophages, lymphocytes and leukocytes and slightly increased numbers of fibroblasts and fibrocytes were visible. Traces of chronic inflammation were still present. There were increasing numbers of new blood vessels and fibroblasts and the beginning of formation of new elastic tissue was evident (Figure 5a).

On the sections taken from all six dogs at the sites of postoperative wounds stitched up with Dexon®, the gingival connective tissue contained residues of surgical sutures, surrounded by proliferation of fibroblasts and fibrocytes, new blood vessels, phagocytes and still rich chronic inflammation cells infiltration (Figure 5b). Mature newly formed elastic
tissue with increased number of newly formed blood vessels in it was present. The connective tissue surrounding the sites of the newly formed elastic tissue was preserved, made of collagen fibers. Multiplication of new blood vessels and formation of elastic fibers with rich chronic inflammation cells infiltration was evident.

On the sections taken from all 6 dogs at the sites of postoperative wounds stitched up with Vicryl-Rapid®, the connective gingival tissue at the site of sutures contained new connective tissue, with spots towards mature connective tissue covered by a layer of epithelium and with rare lymphocytes (Figure 5c). Some connective tissue sections included spots of new granulation tissue with numerous fibroblasts, fibrocytes and new blood vessels. The number of chronic inflammation cells was still significant (Figure 5c). The presence of suture in the wound as a function of time is shown in Figure 6.

Discussion

Information concerning characteristics of specific absorbable suture materials is included in the instruction manuals of manufacturers and stipulated prior to their mass production. However, neither measuring methods nor types of tissues used for placing these materials during tests, are known.

There are not many references comparing or discussing properties and quality of absorbable suture materials 9–12.

There is no ideal suture material, and it is therefore necessary to conduct a large number of studies that will help clinicians consider all properties of suture materials, with the emphasis on biological properties, since manufacturers indicate them very concisely and without specifying the differences between implanting these materials in various human tissues 21–29.

Nary Filho et al. 21 conducted a comparative study on Wistar albino rats, comparing Poliglecaprone 25, Polynilactin 910 and Polytetrafluoroethylene suture materials. They observed inflammation, the degree of fibroblastic and angioblastic proliferation and the presence of fibrous tissue around implanted sutures. Nary Filho et al. 21 say that Polynilactin 910 is one of the preferred in dentistry. It shows no intensive local reaction. Its implantation includes mediocre acute inflammatory response and early fibroblastic and angioblastic proliferation. It also allows subsequent organization of connective tissue around suture filaments. Our experiences with Vicryl® are positive and we partly agree with these authors. A shortcoming of Vicryl-Rapid® is the possibility of loosening of knots (because it is coated). This shortcoming may be overcome by tying 3–5 knots. As for local reaction, Nary Filho et al. 21, 25 find biological reaction to Vicryl® good, including mediocre acute inflammatory response in the early postoperative period and early fibroblastic and angioblastic proliferation. It also allowed organization of connective tissue fibers around it subsequently.

Duprez et al. 27 conducted an experimental study, which included stitching up human cadaver skin grafts on the dorsal side of experimental animals (mice) with Vicryl-Rapid®. The absorption mechanism included hydrolysis. Inflammatory reaction was present, with abundant macrophages containing suture material fragments. It was also possible that inflammation cells released lytic enzymes, which increased spontaneous lysis and lead to fragmentation of suture material. Suture material absorption time depends on the degree of local
reaction, the amount of present electrolytes and basal metabolism. Duprez et al. 32 find Vicryl-Rapid® perfect, tolerant and breaking after 12 to 16 days, with moderately present reaction of macrophages.

All data from the literature suggest that inflammatory tissue reaction is strongest with catgut. Our research shows the strongest inflammatory response with Dexon®, then with catgut, and the mildest with Vicryl-Rapid®.

Sutures used in oral surgery behave differently than when implanted in other body parts, due to tissue quality, the presence of saliva and specific microorganisms, strong vascularization, as well as the present functions of speaking, chewing and swallowing. 22. Mirković and Đurđević-Mirković 30 have studied the way suture material affects accumulation of soft deposits, soft tissue decubitus and wound dehiscence, comparing silk, Vicryl® and nylon. They did not confirm superiority of Vicryl® over the other two, suggesting that silk, followed by nylon, causes the least decubital lesions, while silk accumulates mostly soft deposits, followed by Vicryl® and nylon. Suture dehiscence is equally present with nylon and silk, while it is somewhat more extensive with Vicryl® 30. Another study of Mirković et al. 31 examining influence of suture material on mechanical damage to the mouth, also suggest superiority of synthetic monofilaments (nylon).

Wallace et al. 32 compared polyglycolic acid, silk, chromic and flat catgut used on 52 respondents. Polyglycolic acid suture (Dexon®) caused mildest tissue reactions, less intensive than those caused by silk, flat and chromic catgut. Besides mentioning easier stitching up with Dexon® than with silk, Wallace et al. 32 suggest that Dexon® stays present in tissue 16–20 days after implanting. Catgut is absorbed after 3–5 days, and chromic after 7–10 days. According to these authors, Dexon® has properties closer to the ideal suture material than any other tested.

Besides being conditioned by the degree of local tissue reaction, suture material absorption period is also conditioned by present electrolyte concentrations and by basal metabolism. The only logical explanation of the difference in absorption period of Dexon® can be that Wallace et al. 32 perhaps equalize disappearing of knot from the mouth with complete absorption, although disappearing of knot does not necessarily imply complete absorption of suture material. Full absorption of suture material can be proven only by microscopic inspection of tissue sections taken from sites stitched up with suture material, which was not mentioned in this study as a research method.

In our experimental study, using microscopic inspection of tissue sections taken from sites stitched up with suture material, we find that catgut is absorbed most quickly, while Dexon® is absorbed most slowly.

The results of pathohistological examination of specimens taken from experimental animals suggest that Dexon® causes most intensive local tissue reaction, while reaction is a lot milder with catgut. Vicryl-Rapid® caused the least intensive tissue reaction, but it was not tested by Wallace et al. 32.

In our study, histopathological examination on postoperative day 21 of specimens taken from wounds stitched up with Dexon® suggests the presence of suture fragments visible in mucosa and submucosal tissue. The hypothesis that catgut is absorbed most quickly, while Dexon® is absorbed most slowly was confirmed. It was also confirmed that Vicryl-Rapid® causes the slightest local reaction, while reaction is strongest not with catgut, but with Dexon®, which differs from information suggested in most studies and manufacturers’ instructions. It is also confirmed that wounds heal more quickly with Vicryl-Rapid®, with lower dehiscence incidence and milder local reaction, than with catgut or Dexon®. Besides these properties, it is also important that local reaction is mildest possible (low antigenic potential), with the lowest possible wound dehiscence incidence. The displayed properties of Vicryl-Rapid® suggest its application whenever indication for usage of absorbable suture material is present.

Based on this study results, Vicryl-Rapid® ranks among the better suture materials.

Based on everything suggested here, we may conclude that the aim of this study was achieved, except in case of local reaction, which is not most intensive with catgut, but with Dexon®. This differs from the information suggested in most studies and manufacturers’ instructions. According to histopathological analysis, Vicryl-Rapid® has properties most suitable for oral surgery of all absorbable suture materials tested. For this reason, we suggest and recommend its usage whenever there is a need for application of absorbable suture materials, until some better material is launched.

Conclusion

There is no ideal suture material for the simple fact that various patient factors also influence the wound healing process.

Based on the results obtained in the experimental part of the study, it can be concluded that catgut has the highest absorption rate, while Dexon® has the lowest one. Vicryl-Rapid® causes the lowest level of local reactions, while the highest level of local reactions is not caused by catgut, but by Dexon®. Application of Vicryl-Rapid® makes wounds heal more quickly, with a lower dehiscence rate and milder local reactions than with catgut or Dexon®.

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