

Histological evaluation of capsules formed by silicon implants coated with polyurethane foam and with a textured surface in rats¹

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ABSTRACT

PURPOSE: To assess the capsules formed by silicone implants coated with polyurethane foam and with a textured surface.

METHODS: Sixty-four Wistar albinus rats were divided into two groups of 32 each using polyurethane foam and textured surface. The capsules around the implants were analyzed for 30, 50, 70 and 90 days. Were analyzed the following parameters: foreign body reaction, granulation tissue, presence of myofibroblasts, neoangiogenesis, presence of synovial metaplasia, capsular thickness, total area and collagen percentage of type I and III, in capsules formed around silicone implants in both groups.

RESULTS: The foreign body reaction was only present in the four polyurethane subgroups. The formation of granulation tissue and the presence of myofibroblasts were higher in the four polyurethane subgroups. Regarding to neoangiogenesis and synovial metaplasia, there was no statistical difference between the groups. Polyurethane group presented (all subgroups) a greater capsule thickness, a smaller total area and collagen percentage of type I and a higher percentage area of type III, with statistical difference.

CONCLUSION: The use of polyurethane-coated implants should be stimulated by the long-term results in a more stable capsule and a lower incidence of capsular contracture, despite developing a more intense and delayed inflammatory reaction in relation to implants with textured surface.

Key words: Implant Capsular Contracture. Breast Implants. Mammoplasty. Rats.

Introduction

Currently, the most performed plastic surgery worldwide is breast augmentation¹. Scales *et al.*² defined the properties of an ideal implant: no physical change in tissue; chemically inert; non carcinogenic; non allergic or foreign body reactive; mechanical forces resilience; and made into the desired shape. Unfortunately, there is no such implant, but a lot of progress has been made, since its introduction in 1962 with Cronin and Gerow, to the current generation's implants².

The most commonly used implants are those with textured surface and polyurethane-coated. All material introduced into the human body undergoes a foreign body reaction, which occurs in the formation of a fibrous capsule around itself³.

Unfortunately, there are some patients who undergo to breast augmentation with industrial liquid silicone, which is totally discouraged due to the high index of complications resulting in complex wounds⁴, often requiring therapy with subatmospheric pressure beyond the usual treatments^{4,5}.

In breast augmentation techniques using implants, the more frequent late complication is the capsular contracture. This situation is characterized by breast pain, discomfort, stress, deformations, stiffness and the exchange of implants is often necessary to resolve the problem^{6,7}.

The incidence of contracture varies depending on the coating of the silicone implant, being the polyurethane-coated at about 1%; the textured surface around 10%; and with smooth surface at rates higher than 50%. So, the use of smooth implants is being phased out^{2,8-10}.

The present study aims to assess the capsules formed by silicone implants coated with polyurethane foam and with a textured surface.

Methods

This study was performed in the *vivarium*, Universidade Estadual de Ponta Grossa, after was approved by the Ethics Committee on the Use of Animals of this university.

Sixty-four female Wistar rats were used (*Rattus norvegicus albinus*, Roentia mammalia), weighing 190-250 g, 30 to 90 days of life, distributed in acrylic boxes with 500 cm³ containing four rats in each with free access to water and specific diet for the species, room temperature and alternating 12 h cycles of light and dark. They were distributed into two groups of 32 and subdivided into four subgroups according to the time of death (30, 50, 70 and 90 days).

Experimental procedure

The rats were anesthetized by intramuscular injection consisting of a combination of 1% ketamine (40 mg/kg) and 2% xylazine (8 mg/kg)¹⁰. The effectiveness of the anesthesia was assessed by the lack of movement, corneal reflex and motor reaction after holding a clamp on the adipose cushion of one of the hind legs, and a good ventilatory pattern³.

The rats were placed in the prone position with shaving performed in the dorsal region followed by antiseptics.

For delimiting the incision, the crossing point of a horizontal line following the posterior inferior costal margin to the sagittal midline was defined. With a number 3 scalpel, coupled with a number 15 blade, a horizontal incision was made with a horizontal extension of 1 cm at the intersection of these virtual lines.

The pocket, which housed the implants, was made on the retromuscular plane with scissors in a cranial direction, starting 5 mm from the incision and square shape. After being made, the implant was inserted (Figure 1) and placed 5 mm from the incision (Figure 2), and skin closure was performed with three simple knots using mononylon 5-0.

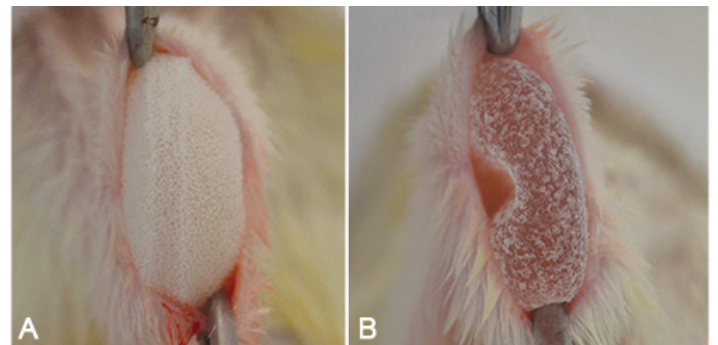


FIGURE 1 – Insertion of the implants coated with polyurethane foam and textured surface.

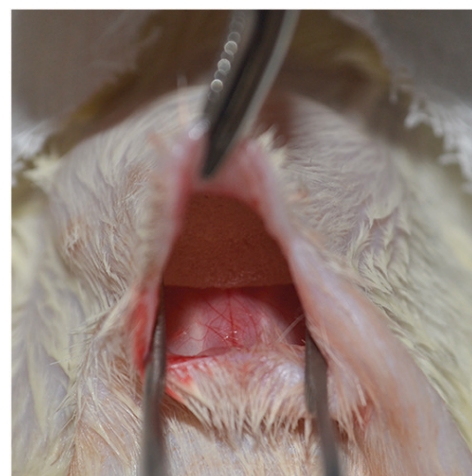


FIGURE 2 – Placement of the implant 5 mm far away from the incision.

The wound was kept exposed and postoperative analgesia was applied with a single, intramuscular injection of dipyrone (20 mg/kg) on the outside of the hind limb¹¹. There have been no postoperative dressings or stitches withdrawal.

The rats were killed according to their subgroups with an application of ketamine and xylazine, corresponding to four times the therapeutic dose and subsequent cervical dislocation. Because of implant extrusion in seven animals (four from the polyurethane group and three from the textured group), they were excluded from the study.

Histological evaluation

Hematoxylin-eosin staining

With this staining the following histological variables were evaluated: foreign body reaction, granulation tissue, myofibroblasts, neoangiogenesis and synovial metaplasia¹². The analyses of these variables were measured in four levels: absent, and discrete, moderate and intense presence.

Masson's trichrome acid staining

Through this staining, the measurement of the thickness of the capsule showing the muscle, collagen fibers, fibrin and erythrocytes were evaluated¹².

Picosirius staining

Through this staining, collagens type I and III were quantified. The study of the microscope slides was a closed-type, double-blind and read by polarized light (Figure 3) for further analysis by the software Image Pro-Plus (Figure 4)^{13,14}.

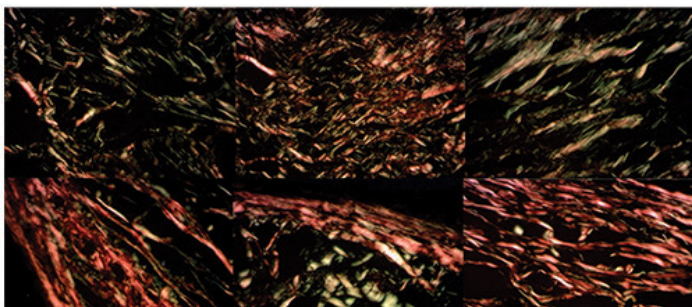


FIGURE 3 – Photo with six different photomicrographs (increase of x400) showing collagen fibers stained by Picosirius and visualized in the microscope with polarized light.

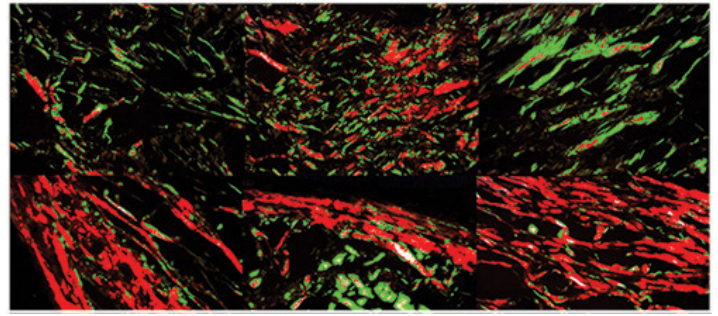


FIGURE 4 – The same six previous photomicrographs displayed on the computer after application of the mask by software Image Pro-Plus.

Statistical analysis

For the analysis of quantitative variables the nonparametric Mann-Whitney and Kruskal-Wallis were used. Qualitative variables were analyzed using Fisher's exact test. The significance level was 0.05, adjusted by Bonferroni for multiple comparisons of the groups ($p < 0.008$). Data was analyzed with the computer program IBM SPSS v.20.0.

RESULTS

Foreign body reaction

Most animals in polyurethane group had moderate or severe presence of a foreign body reaction (Figure 5), whereas in the textured group was not observed this variable.

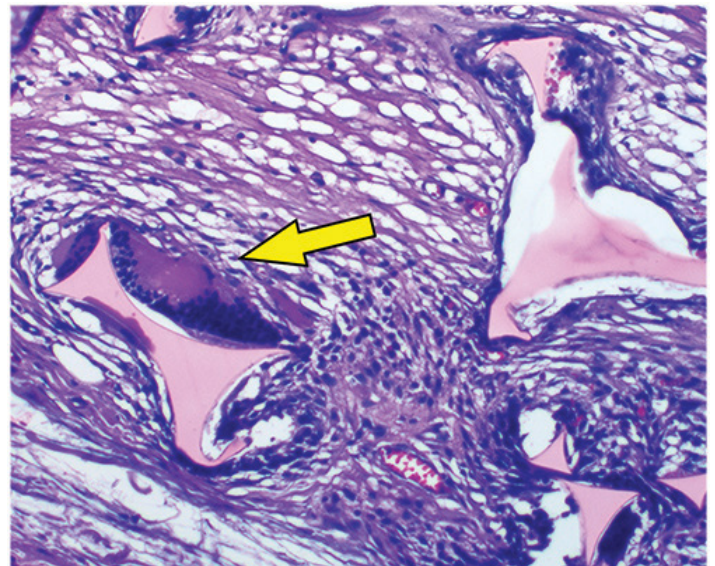


FIGURE 5 – Photomicrography of polyurethane foam showing the foreign body reaction, the arrow showing the foreign body reaction.

The foreign body reaction only was present in the polyurethane group within the four evaluated subgroups (30 days $p < 0.0001$ / 50 days $p < 0.002$ / 70 days $p < 0.001$ / 90 days $p < 0.0002$, Figure 6).

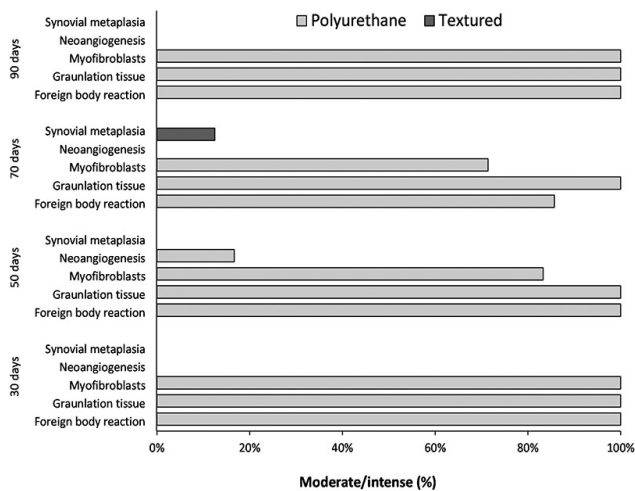


FIGURE 6 – Percentage of each variable in each subgroup in both groups.

Granulation tissue

While every rat of the polyurethane group had moderate or intense presence of the formation of granulation tissue, in the textured group absent or presence of this discrete variable was observed (Figure 7).

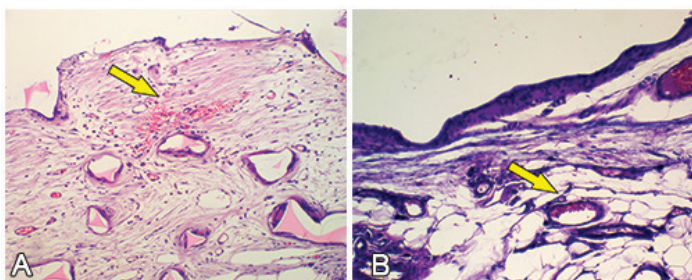


FIGURE 7 – Photomicrography of polyurethane foam (A) and textured surface (B) showing the granulation tissue with arrow showing fibroplasia and reactionary neovangiogenesis.

Granulation tissue formation was statistically significantly greater in the polyurethane group within the four evaluated subgroups (30 days $p < 0.0001$ / 50 days $p < 0.002$ / 70 days $p < 0.0002$ / 90 days $p < 0.0002$, Figure 6).

Myofibroblasts

In the polyurethane group most rats had moderate or severe presence of myofibroblasts, whereas in the textured group all rats had absent or discrete presence of the variable (Figure 8).

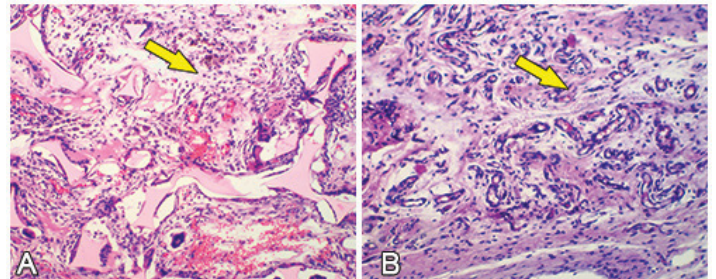


FIGURE 8 – Photomicrography of polyurethane foam (A) and textured surface (B) showing, in arrows, the myofibroblasts.

The presence of myofibroblasts was statistically significantly greater in the group polyurethane within the four evaluated in subgroups (30 days $p < 0.0001$ / 50 days $p < 0.015$ / 70 days $p < 0.007$ / 90 days $p < 0.0002$, Figure 6).

Neovangiogenesis

Only 16.7% of the animals in the 50 days subgroup of the polyurethane group had moderate or severe presence of neovangiogenesis, in the rest of the rats this variable was absent or had discrete presence (Figure 9).

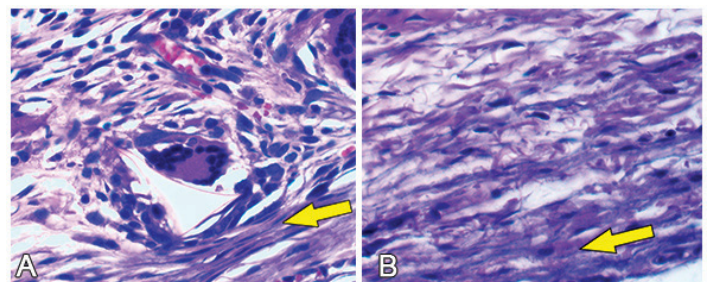


FIGURE 9 – Photomicrography of polyurethane foam (A) and textured surface (B) showing the neovangiogenesis and, in arrows, blood vessels.

No statistically significant differences were found between the two groups in the several subgroups (Figure 6).

Synovial metaplasia

Only 12.5% of the rats in the 70 days subgroup of the textured group had moderate or severe presence of synovial

metaplasia, the other rats had absent or discrete presence of this variable (Figure 10).

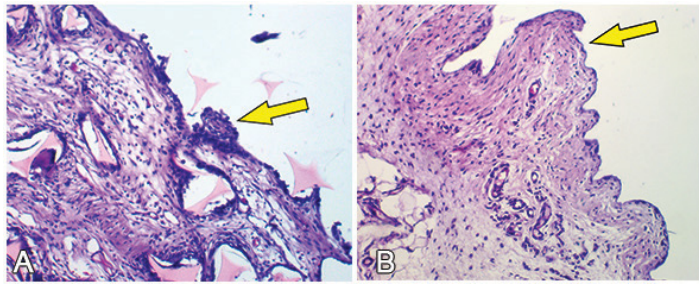


FIGURE 10 – Photomicrography of polyurethane foam (A) and textured surface (B) showing, in arrows, the synovial metaplasia.

There were no statistically significant differences being the two groups in all subgroups (Figure 6).

Measurement of the thickness of the capsule

The capsule in the polyurethane group was thicker compared to textured group (Figure 11), with statistical significance in all evaluated subgroups (30 days $p=0.001$ / 50 days $p=0.006$ / 70 days $p=0.001$ / 90 days $p=0.001$, Table 1).

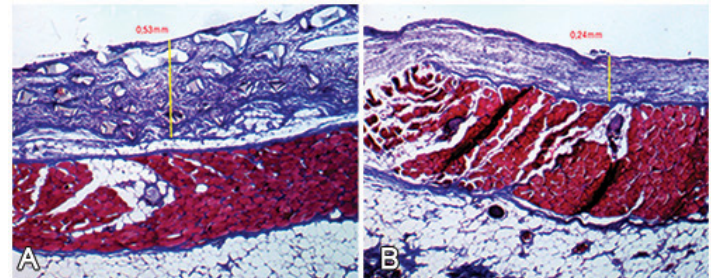


FIGURE 11 – Photomicrography of polyurethane foam (A) and textured surface (B) showing the capsule with 0.53 mm in A and 0.24 mm in B.

TABLE 1 – Comparison of the thickness of the capsule (mm) in each subgroup in both groups.

Measurement of the capsule (mm)	Group	n	Mean	Median	Minimum	Maximum	Standard deviation	p*
30 days	Polyurethane	8	0.76	0.73	0.45	1.00	0.20	0.001
	Textured	8	0.27	0.25	0.15	0.40	0.08	
50 days	Polyurethane	6	1.12	1.05	0.75	1.55	0.32	0.006
	Textured	5	0.28	0.30	0.20	0.35	0.06	
70 days	Polyurethane	7	1.06	1.10	0.90	1.35	0.17	0.001
	Textured	8	0.36	0.35	0.25	0.50	0.08	
90 days	Polyurethane	7	1.16	1.15	1.00	1.35	0.11	0.001
	Textured	8	0.36	0.35	0.25	0.50	0.08	

(*)The nonparametric Mann-Whitney test; $p<0.05$

The comparison of the measurement of the capsule in each subgroup (30, 50, 70 and 90 days) of the polyurethane group was statistically significant at $p=0.008$, while in the textured group there was no statistical significance (Table 2).

TABLE 2 – Comparison of the measurement of the thickness of the capsule (μm) in each subgroup within each group.

Group	Subgroup	n	Mean	Median	Minimum	Maximum	Standard deviation	p*
Polyurethane	30 days	8	0.76	0.73	0.45	1.00	0.20	0.008
	50 days	6	1.12	1.05	0.75	1.55	0.32	
	70 days	7	1.06	1.10	0.90	1.35	0.17	
	90 days	7	1.16	1.15	1.00	1.35	0.11	
Textured	30 days	8	0.27	0.25	0.15	0.40	0.08	0.165
	50 days	5	0.28	0.30	0.20	0.35	0.06	
	70 days	8	0.36	0.35	0.25	0.50	0.08	
	90 days	8	0.36	0.35	0.25	0.50	0.08	

(*)The nonparametric Kruskal-Wallis test. $p<0.05$

Due to the rejection of the hypothesis of equality to all subgroups for the polyurethane group, the subgroups were compared in pairs.

In the polyurethane group by comparing the different subgroups to each other. Statistical significance between 30 and 50 days in the polyurethane group was obtained ($p=0.009$), 30 and 70 days ($p=0.009$), and 30 and 90 days ($p<0.001$).

Measurement of the total and percentage areas of collagen type I and III

The total area of type I collagen was larger in the textured group (30 days $p=0.046$; 50 days, $p=0.045$; 70 days, $p=0.021$; 90 days, $p=0.037$). The area percentage of type I collagen was bigger in the textured group (30 days, $p=0.036$; 50 days, $p=0.011$; 70 days, $p=0.021$; 90 days, $p=0.028$). The total area of type III collagen was higher in the polyurethane group (50 days, $p=0.028$). The percentage area of type III collagen was larger in the polyurethane group (30 days, $p=0.036$; 50 days, $p=0.011$; 70 days, $p=0.021$; 90 days, $p=0.028$, Figures 12 and 13).

Discussion

Histological aspects

Since its discovery to the present day silicone implants have been defined as a biocompatible material. However, for Ratner¹⁵, the capsule formed around the implants is an attempt by the body to isolate it, which is a matter of incompatibility. According to this same author, the best concept that could be applied to implants in general, including silicone, is biotolerability, which is the ability of a material to reside in the body for long periods of time with only low-grade inflammation.

Evaluation of foreign body reaction

According to Ratner¹⁵, a foreign body reaction is more evident when the pores of the implant are either larger or smaller than 30-40 micrometers (μm). Disagreeing with the author, in the textured group in which the pore size exceeded this range at various times (200-500 μm), the foreign body reaction was absent in all subgroups analyzed in this research.

In accordance to Wagenführ², most of the rats in the polyurethane group had moderate or intense presence of a foreign body reaction in all subgroups evaluated. Although the same author obtained statistical significance only at 60 days subgroup, in this study, the results were statistically significant in the subgroups 30, 50, 70 and 90 days. This fact can be explained because Wagenführ² compared implants coated with polyurethane foam and with silicone foam. In this experiment the comparison was made with implants with textured surfaces.

Haddad Filho *et al.*¹⁶ after comparing silicone implants with a textured surface and coated PTFE-E, it was concluded that both groups induced a weak foreign body reaction, which was higher in textured group. Disagreeing with this author, in this study the foreign body reaction was absent in the textured group at all subgroups.

Corroborating with Balderrama *et al.*¹⁰ findings, it was confirmed with significant difference that the foreign body reaction was lower in textured group at the time of 60 days when compared to the foam silicone group. In this research lower foreign body reaction in textured group in all subgroups was also obtained, although the comparison was made with implants coated with polyurethane foam.

In according to Bassetto *et al.*¹⁷, the foreign body reaction was bigger in the polyurethane group. In the textured group, this

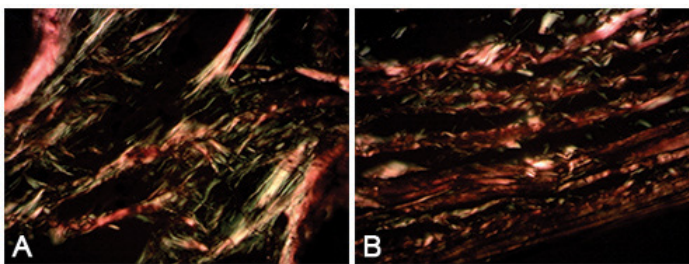


FIGURE 12 – Photomicrography of polyurethane foam (A) and textured surface (B) showing collagen fibers: in red type I and in green type III.

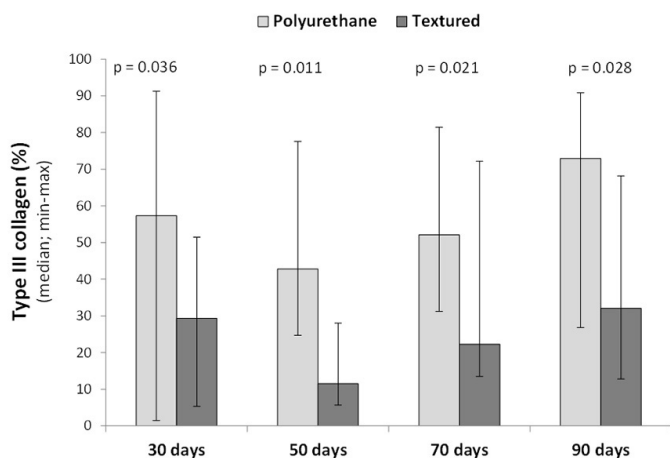


FIGURE 13 – Comparison of the percentage area of collagen type III

reaction was present in all members in a milder way, differing from the present study in which the foreign body reaction was absent in the textured group, although this study has been carried out in rats and the other in humans.

Evaluation of granulation tissue

Corroborating Vieira¹⁸, who found that the polyurethane group had increased formation of granulation tissue in relation to the textured group in subgroups of 30 and 90 days with statistical significance, in this study in all subgroups, this variable was more intense in the polyurethane group, whereas in the textured group it was absent or showed discrete presence, also with significant difference.

Evaluation of myofibroblasts

Disagreeing with Hwang *et al.*¹⁹ when saying that the preferential localization of myofibroblasts is in the outer layer of the capsule, in this study, like in Minami *et al.*²⁰ and Prantl *et al.*⁶, it was found in the inner layer.

According to Bui *et al.*²¹, in the current study all rats of the textured group had absent or mild presence of myofibroblasts, while in the polyurethane group most animals had moderate or severe presence of myofibroblasts, thereby strengthening the interdependence of the type of implant coating and the amount of myofibroblasts. An important observation was that these authors have conducted the study in humans.

Evaluation of neoangiogenesis

Disagreeing with Haddad Filho *et al.*¹⁶ that found greater number of capillaries in the textured and PTFE-E groups in relation to the control group with statistical significance in various subgroups, in this study all rats that took part in the textured group had absence or discrete presence of this variable with no significant difference.

Disagreeing with Wagenführ² and Vieira¹⁸ that found in the polyurethane group larger amount of vessels statistically significant, in this study the majority of the rats showed absence of neoangiogenesis or discrete presence, but without significant difference.

Evaluation of synovial metaplasia

In accordance with Prantl *et al.*^{6,22}, that found an inner layer with synovial metaplasia in 62.5% of cases, in this study

mild synovial metaplasia was found in 100% of the rats, however, there was no significant difference between groups.

Partially agreeing with Bassetto *et al.*¹⁷, who obtained the presence of synovial metaplasia in the polyurethane and textured groups, which was bigger in the polyurethane group, in this study was found this variable, however, there was no difference between the groups. The same authors concluded that synovial metaplasia is the most important protective factor against capsular contracture. This becomes clear when Bui *et al.*²¹ stated synovial metaplasia to be the most common in capsules Baker I/II, compared to Baker III/IV capsules.

Evaluation of the thickness of the capsule

Corroborating Minami *et al.*²⁰ and Vieira¹⁸ that observed variation in the thickness of the capsule due to increase in the non-collagenous layers (intermediate and internal), in this study this increase also occurred in these layers.

In accordance to Balderrama *et al.*¹⁰ that found thinner capsule in the textured group in the subgroups of 7 and 60 days compared to silicone foam group with statistical significance, all subgroups evaluated in this study (30, 50, 70 and 90 days) the capsule was thinner in the textured group with a significant difference, although the comparison has been made with polyurethane-coated implants.

Agreeing with Vieira¹⁸, who obtained ,statistically significant thinner capsule in the textured group in the subgroups of 30 and 90 days compared to polyurethane group, in all evaluated subgroups in this study the capsule was thinner in the textured group with statistical significance. Disagreeing with this author who found thinner layer of collagen in the polyurethane group compared to the textured group, in the current study were evaluated all subgroups and was found an opposite result.

Evaluation of the total and percentage areas of collagen type I and III

Disagreeing with Ratner¹⁵ that studied the coating of implants and concluded that a greater angiogenesis occurs when the pores on the surface of the implants are between 30-40 µm with less collagen deposition, in this study the pores measured 50-250 µm in the textured group and 250-350 µm in polyurethane group, and was obtained in both groups and in all subgroups, discrete angiogenesis with a significant deposition of collagen which was larger in the polyurethane group.

Regarding the characteristics of the collagen fibers according to Minami *et al.*²⁰ and Balderrama *et al.*¹⁰ that found a decline in the amount of type III collagen fibers with increased type I collagen fibers with significant differences, the same result was found in this study from 50 days on.

Disagreeing with Wagenführ² that obtained a greater amount of collagen in the subgroups of 28 and 60 days of the polyurethane group than in the silicone foam group with statistical significance, in this study all the textured subgroups evaluated had a higher amount of collagen, statistically significant, than the polyurethane group.

When analyzed the same fraction area, Minami *et al.*²⁰ found a greater increase of type I collagen fibers in the smooth implants for textured implants with a significant difference. In this study the textured group had a greater increase of type I collagen fibers in relation to the polyurethane group with statistical significance.

Conclusion

The use of polyurethane-coated implants should be stimulated by the long-term results in a more stable capsule and a lower incidence of capsular contracture, despite developing a more intense and delayed inflammatory reaction in relation to implants with textured surface.

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