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Original Article

INFLUENCE OF SCHINUS TEREBINTHIFOLIUS RADDI (AROEIRA) AND CARAPA GUIANENSIS AUBLET (ANDIROBA) IN THE HEALING PROCESS OF GASTRORRAPHIES

Efeito de Schinus terebinthifolius raddi (aroeira) e Carapa guianensis aublet (andiroba) na cicatrização de gastrorrafias

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ABSTRACT - ***Background*** - The gastrorraphy isolated or associated with the use of biological adhesives formed throughout the history of surgery the usual way to promote healing in gastric lesions; however, the use of herbal medicine has been increasingly employed to help the wound healing. ***Aim*** - To evaluate the wound healing caused in the stomach of rats using extract of *Schinus terebinthifolius* Raddi and *Carapa guianensis* Aublet oil. ***Methods*** - Ninety rats, adult males were divided into three groups: aroeira, andiroba and control group, which were subdivided into three subgroups of five animals according to the time of the deaths (seven, 14 and 21 days). All underwent the same surgical procedure (injury and suture the stomach) differing only to the animals in groups aroeira and andiroba that received a daily dose of 100 mg / kg of hydroalcoholic extract and oil, by gavage, while the control group received normal saline. The parameters evaluated were the macroscopic and microscopic test of resistance to air insufflation and test the traction force. ***Results*** - All animals showed good healing of gastric and abdominal wall without infection and dehiscence. Both groups presented neighboring organs adhesions on the gastric surface. The endurance test for air insufflation showed higher average pressure within seven days and the test revealed greater traction force of rupture between seven and 14 days in groups aroeira and andiroba. The intensity of chronic inflammation revealed statistically significant differences in angiogenesis and fibroblast proliferation. ***Conclusion*** - The use of extract of *Schinus terebinthifolius* Raddi and *Carapa guianensis* Aublet oil favored the gastric wound healing in rats.

HEADINGS - Rats. Healing. Insufflation. Traction.

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RESUMO - ***Racional*** - A gastrorrafia isolada ou associada ao uso de adesivos biológicos constituiu ao longo da história da cirurgia a forma usual de promover a cicatrização nas lesões gástricas; entretanto, o uso de fitoterápico tem sido cada vez mais empregado para auxiliar o processo cicatrical. ***Objetivo*** - Avaliar a cicatrização de ferida provocada no estômago de ratos com uso do extrato hidroalcoólico de *Schinus terebinthifolius* Raddi e óleo da *Carapa guianensis* Aublet. ***Métodos*** - Foram utilizados 90 ratos, adultos, machos, distribuídos em três grupos: grupo aroeira, grupo controle e grupo andiroba, os quais foram subdivididos em três subgrupos de cinco animais conforme o momento da morte induzida (sete, 14 e 21 dias). Todos foram submetidos ao mesmo procedimento cirúrgico (lesão e rafia do estômago) diferindo apenas que os animais dos grupos aroeira e andiroba receberam dose diária de 100 mg/kg do extrato hidroalcoólico e do óleo, via gavagem, enquanto o grupo controle recebeu solução salina isotônica. Os parâmetros avaliados foram: alterações macroscópicas e microscópicas, teste de resistência à insuflação de ar atmosférico e teste pela força de tração. ***Resultados*** - Todos os animais demonstraram boa cicatrização da parede abdominal e das gastrorrafias, sem infecção e deiscência. Ambos os grupos apresentaram aderências à superfície das gastrorrafias com órgãos vizinhos. O teste de resistência por insuflação de ar atmosférico mostrou maiores médias de pressão no período de sete dias e o teste de força de tração revelou maiores forças de ruptura no período de sete e 14 dias nos grupos aroeira e andiroba. A intensidade da inflamação crônica revelou diferença estatisticamente significante nas variáveis angiogênese e proliferação fibroblástica. ***Conclusão*** - O uso do extrato hidroalcoólico de *Schinus terebinthifolius* Raddi e do óleo da *Carapa guianensis* Aublet favoreceu a cicatrização do estômago de ratos.

DESCRITORES - Ratos. Cicatrização. Insuflação. Tração.

INTRODUCTION

The repair process of the injured tissue occurs when those cells are replaced by living cells. These can be derived from parenchyma or from stroma of the injured connective tissue. This event is a dynamic process that aims to ensure the restoration of tissue. At a certain period of time the phases coincide and occur simultaneously, thus allowing successful healing[7]. Celsius in the first century of the Christian era found that in the initial phase of tissue repair had inflammatory process responsible for redness, swelling, heat and pain. Today we know that the healing process is a chemical, physical and biological phenomenon. Consist of three interconnected phases: the inflammatory (including humoral and cellular phases), the formation of collagen and tissue remodeling[32].

The application of the herbal in wound healing of the stomach and other organs and tissues have been extensively assessed in experimental studies. Medical science at the end of the last millennium and the beginning of the XXI century observed increasing use of drugs in the treatment of several diseases. The investigations have not been limited to the development of synthetic drugs, but rather in ever more frequent attempts to isolate the active ingredients of plants and animals. Nevertheless, there is great scientific advance involving chemical and pharmacological studies on medicinal plants designed to obtain new compounds with therapeutic properties. With the development of new spectroscopic techniques, organic chemicals have managed to quickly discover complex structures of natural constituents, even once difficult to be elucidated[5].

Importantly, between 250-500 thousand existing species, only about 5% have been studied phytochemically and lower percentage was evaluated under biological aspects. Thus, the success of investigations of the active principles of plants and animals depends on the degree of multidisciplinary interaction, especially botany, chemistry and pharmacology[5].

The species *Schinus terebinthifolius* Raddi is an Anacardiaceous, common in northeastern Brazil, extending to the south and neighboring countries like Argentina, Paraguay and Peru. It is widely used in folk medicine and its first record in literature in 1926 by the Brazilian Pharmacopeia[6]. The shells of Aroeira of the beach revealed that they are rich in tannins, phenols and steroids. Many of its properties or its curative effects can be attributed to the different polyphenols that are distributed unevenly in different parts of the plant, such as bark, leaves, flowers, fruits and seeds. These polyphenols are substances of great importance to plant physiology, contributing to the mechanisms of defense[26]. Its use is mentioned against inflammatory diseases, infectious diseases, antimicrobial properties, anti-ulcerogenic and in gastritis. These presents astringent activities, tonic, haemostatic, anti-neuralgic, mutagenic effects, antioxidants and healing[4,6,10,26,27,28,29,30,34].

The *Carapa guianensis* Aublet, Meliaceae family, was first described by French botanist Jean-Baptiste Christopher Fusee Aublet in 1775 in French Guiana. Commonly known as andiroba (name derived from the Tupi-Guarani: "landi" - oil and "rob" - bitter), jandiroba, penaiba and carape. It is a 20-30 feet tall tree that has white flowers, round fruits and large and angular seeds. In Brazil, is distributed throughout the Amazon and from Pará to Bahia. Still occurs in southern Central America, Colombia, Venezuela, Suriname, French Guyana, Peru, Paraguay and the Caribbean islands[13].

The seeds contain 70% medicinal oil and against insects. The traditional process of extracting oil from the seeds of andiroba begins with the collection and selection of them, cooking for 1-3 h in water, kneading the seeds and placing the dough in sloping surface and leaving the oil drip[18]. In this oil we highlight the anti-inflammatory activity, antiparasitic, repellents, healing activity, anti-allergic, anti-toxic at the immature stage of mosquitoes *Aedes aegypti*[8,22,26].

The aim of this study is to analyze the healing effect of hydro alcoholic extract of aroeira and andiroba oil via orogastric in gastrorraphies in rats, through histological and tensiometric analysis.

METHODS

This study was performed at the Laboratory of Experimental Surgery at Federal University of Maranhao (UFMA). Were followed the rules for animal experimentation of the Sergio Arouca Law (No.-11794-2008) and was approved by the Ethics Committee of the State University of Maranhao, under the protocol (027-2007).

Botanical material and preparation of extract

The specimens of aroeira (*Schinus terebinthifolius* Raddi) and andiroba (*Carapa guianensis* Aublet) were cataloged in the Herbarium Atticus Seabra from the Department of Pharmacy, Center of Biological and Health Sciences, Federal University of Maranhão, under the registers numbers 488 and 01253 respectively. The aroeira in the form of bark was submitted to grinding, and the powder (2400 g) was diluted with absolute alcohol and distilled water to form 3,450 ml of crude extract, which its concentration was calculated in g / mL. The crude extract was concentrated on a rotary evaporator under reduced pressure and at a temperature of 55-60° C for complete removal of

solvent. The material obtained after concentration presented in paste form (596.85 g) which was removed 2.8 g of crude hydro alcoholic extract (ethanolic). It was diluted in 28 ml of saline solution resulting in concentration of 100 mg / ml. Then it was stored in a refrigerator at 10º C. This material (50 ml) underwent analysis at the Laboratory for Quality Control of Food and Water from the Department of Chemical Technology of Federal University of Maranhão, with concentrations of sugars (20.8%), protein (0.966%), lipid (22.93%), ash (0.5%), moisture (51.87%) and pH (3.9).

The sample of andiroba oil used was from the municipality of Axixá, MA, Brazil, and 50 ml was subjected to analysis by the same laboratory, with the following fat composition: myristic acid (18.1%), oleic acid (58, 9%) linoleic acid (9.2%) and palmitic acid (9.3%). Among the non fatty compounds was point out the triterpenes and tannins and two alkaloids: the andirobina and carapina.

Manipulation and distribution of animals in groups

Were used 90 rats (*Rattus norvegicus albinus*, Rodentia Mammalia) Wistar males, aged from 50 to 60 days, weighing on average 141.2 g. They were adapted for seven days in environmental conditions of temperature and humidity in day and night cycle of 12 hours.

The animals were randomized during the operation in six groups (G) of 15 rats: Aroeira insufflation group (GAI) 1-15, Aroeira traction group (GAT) 16-30, Insufflation control group (GCI) 31-45, Traction control group (GCT) 46-60, Andiroba insufflation group (GANI) 61-75 and Andiroba traction group (GANT) 76-90. Each of the subgroups of five animals was killed on interval of seven days (7, 14, 21) respectively.

After seven days of adaptation, was performed the surgery after the withdrawal of the diet six hours before keeping free access to water. The rats were weighed and anesthetized with intramuscular 20 mg / kg of ketamine 5% (Vetanarcol ®) and 10 mg / kg of xylazine 2% (Kensol®). The anesthesia was performed on the posterior surface of the thigh.

A longitudinal midline laparotomy was performed and the stomach was exteriorized. It was made a 1 cm longitudinal incision which was sutured with four sutures of 6-0 yarn blue polypropylene (Prolene®, Ethicon). Thereafter synthesis of the abdominal wall was performed with 5-0 monofilament nylon (Mononylon ®, Ethicon). The animals received analgesia with Dipyrone (25 mg / kg / dose) daily. They were kept in their cages, receiving standard rat feed and water. They were evaluated daily.

In animals from GA and GAN it was applied by gastric gavage 100 mg / kg / dose of hydroalcoholic extract of aroeira and 5 ml / kg / dose of andiroba oil daily, respectively, and the same volume of normal saline 0.9% in the animals of CG. The animals were clinically evaluated in accordance with the evaluation form.

The animals were killed on days previously established with a lethal dose of ketamine and xylazine. Then we withdrew the specimen containing from the stomach to pylorus with 4 cm of the distal esophagus, preserving the structures and organs adhered to gastrorraphy in order not to compromise the insufflation and tensile tests. Adhesions were classified by the score of Nair [20].

Was then perform the test of resistance to air insufflation. This procedure consisted of the introduction of a nº 6 silicone tube in esophageal segment which was fixed with cotton thread 2-0, connecting the probe to the manometer (Polzin ®, Watson Marlow, Berlin, Germany) submersion the piece in water, insufflation with air at a rate of 0.1 ml / s until the occurrence of release of air bubbles, the pressure being recorded in mmHg at the time of rupture.

To study the resistance by force of traction, the surgical specimens were sent to the Mechanical Testing Laboratory, of the Federal Institute of Education, Science and Technology of Maranhã (IFMA - MA). Was used the Universal Testing Machine, Model Tiratest 2420, TIRA Maschinenbau Gmbh. Immediately prior to mechanical testing, the stomach was opened through the posterior wall and transformed into rectangles with length and width (6 x 3 cm), with a central scar. Each piece received a preload of 2N, with accommodation time of 60 seconds. The set speed for all tests was 5 mm / min and measurements were taken every 0.5 mm. The tensile tests resulted graphs of maximum force (N) of rupture versus time elapsed, obtained from each test.

The stomachs underwent histological processing, the slides were stained with hematoxylin and eosin, Masson's trichrome and Picrosirius red and analyzed in three fields for double blind evaluation.

The histological evaluation included the following parameters: acute inflammatory infiltrate (polymorphonuclear), edema, vascular congestion and chronic inflammatory infiltrate (mononuclear cells), angiogenesis, fibroblast proliferation and collagen (fibrosis). Scores were adopted to quantify the variables in accordance with the intensity (absent-0, mild-1, moderate-2 and marked-3) and qualified collagen in immature and mature[36].

Statistical analysis

Data were evaluated by using the program BioEstat 5.0 (2007). They were expressed as mean and standard deviation in numerical variables, by analysis of variance (ANOVA). When there was significant difference the Dunn test was applied. In classificatory variables (histological) was applied the Mann -

Whitney, independent for each observation period (seven, 14 and 21 days). The level of significance for rejecting the hypothesis of nullity was 5% ($p < 0.05$).

RESULTS

All animals demonstrated good healing of the abdominal wall and stomach without clinical signs of infection or dehiscence. Adhesions on the surface of gastric sutures occurred nearby organs in the three study groups, mainly the liver, small intestine and abdominal wall.

The test of resistance to atmospheric air insufflation was performed on all rats, the rupture of the stomach has occurred in the gastrorraphy. The mean rupture pressures were higher in groups aroeira and andiroba than control group within seven days, but without showing statistically significant difference in this period. At 21 days there was a statistically significant difference (Figure 1).

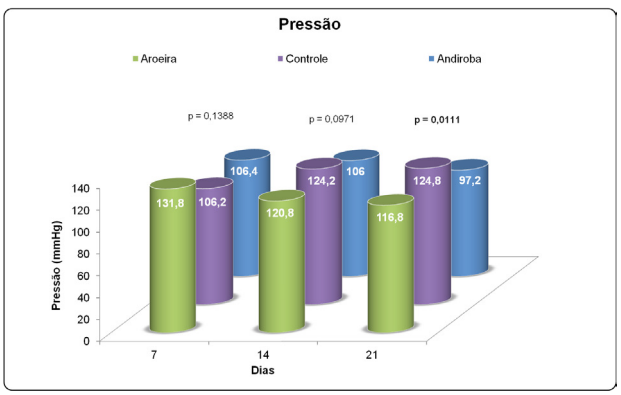

FIGURE 1 - Results of the mean rupture pressures of the test of resistance to atmospheric air insufflation

Regarding the resistance by force of traction, the means were higher in groups aroeira and andiroba on the 7th, 14th and 21st days, revealing a statistically significant difference in the groups of seven and 14 days, but not at day 21st (Figure 2), favoring therefore aroeira and andiroba groups.

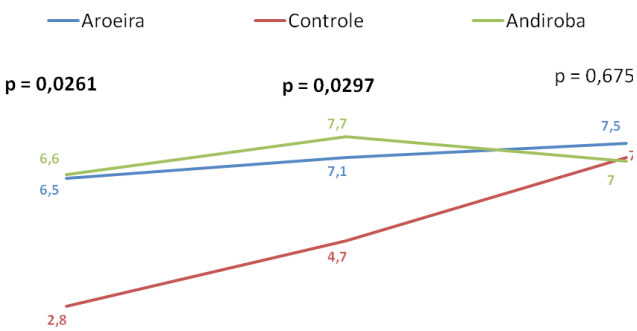

FIGURE 2 - Mean values of the disruptive forces of resistance to traction test

The intensity of acute inflammation and angiogenesis of the gastrorraphy in rats showed to be mild in the majority of animals in groups aroeira, control and andiroba, In the variable angiogenesis there was predominance of rats with moderate and severe intensity in the groups aroeira and andiroba over the control group. It was revealed thus statistically significant difference in variable acute inflammation within 14 days and angiogenesis on day 7, thus favoring groups aroeira and andiroba. The intensity of fibroblast proliferation and collagen deposition were moderate in most rats of the three groups. Considering fibroblast proliferation, there was a statistically significant difference on day 14 due to the fact that the control group in this period presented four rats classified as severe. Relative to collagenization it was observed a statistically significant difference in the 7th and 14th days. In the seven-day period were favored group aroeira and control in relation to the andiroba; within 14 days the favored groups were control and andiroba (Table 1). The intensity of acute and chronic inflammation are shown in Figure 3 (A, B, C, D, E and F).

TABLE 1 - Histological evaluation (Mann-Whitney) parameters of healing in gastrorraphy of the animals treated with hydroalcoholic extract of aroeira, the 0.9% saline (control) and oil andiroba

Variáveis Histológicas / P	7 dias			14 dias			21 dias		
	Aroeira	Controle	Andiroba	Aroeira	Controle	Andiroba	Aroeira	Controle	Andiroba
Inflamação aguda / p	+ (3)	+ (6)	+ (5)	+ (7)	+ (10)	- (1)	+ (9)	+ (6)	- (1)
	++ (7)	++ (4)	++ (5)	++ (3)		+ (9)	++ (1)	++ (4)	+ (8)
									++ (1)
		0,404			0,0374*			0,1313	
Angiogêese / p	+ (5)	- (1)	+ (3)	+ (7)	+ (10)	- (1)	+ (8)	+ (8)	+ (8)
	++ (4)	+ (8)	++ (7)	++ (3)		+ (7)	++ (2)	++ (2)	++ (2)
	+++ (1)	++ (1)				++ (2)			
		0,0213*			0,1532			1	
Proliferação fibroblástica / p	+ (2)	++ (10)	++ (10)	+ (2)	++ (6)	++ (9)	++ (10)	+ (1)	+ (1)
	++ (8)			++ (8)	+++ (4)	+++ (1)		++ (8)	++ (7)
								+++ (1)	+++ (2)
		0,1260			0,0206*			0,8123	
Colagenização / p	+ (5)	+ (3)	+ (9)	+ (2)	++ (5)	++ (6)	++ (7)	+ (1)	++ (9)
	++ (5)	++ (7)	++ (1)	++ (8)	+++ (5)	+++ (4)	+++ (3)	++ (5)	+++ (1)
								+++ (4)	
		0,0254*			0,0135*			0,5258	

Relative intensity of acute inflammation, angiogenesis, fibroblast proliferation and collagen formation (hematoxylin and eosin and Masson's trichrome): absent (-), mild (+), moderate (+ +) or severe; () = number of animals in each classification; * implies statistical significance between groups ($p < 0.05$)

The density of immature collagen showed up mainly very high (> 75%) on day 7, moderate in 14 and 21 days (25% to 50%) in all groups. Statistically significant difference was observed in the seven-day period showing larger formation of immature collagen in aroeira and control groups. Regarding mature collagen, the density on day 7 showed slightly (<25%) in the majority of rats

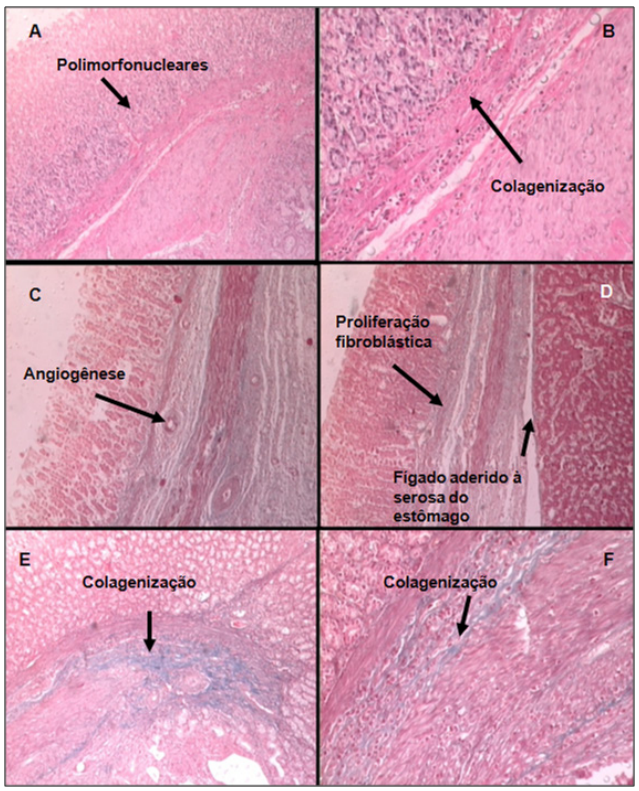

FIGURE 3 - Intensity of acute and chronic inflammation (hematoxylin and eosin and Masson's trichrome): A) polymorphonuclear aroeira group, 7 d, 100X, B) collagenization, the control group, 14 d, 200 X, C) angiogenesis, group andiroba, 14 d , 200 X, d) fibroblast proliferation, andiroba group, 21 d, 100 X, E) collagenization, aroeira group, 21 d, 100X, F) collagenization, andiroba group, 21 d, 200X

of the three groups. On day 14, this density was markedly (50% to 75%) in groups aroeira and andiroba and moderate in the control group. At day 21, was predominantly pronounced in aroeira and control groups and very pronounced in the group andiroba, demonstrating larger formation of mature collagen in the groups aroeira and andiroba. Thus, it was observed a statistically significant difference at 14 and 21 days (Table 2). The density of immature and mature collagen is shown in Figure 4 (A, B and C).

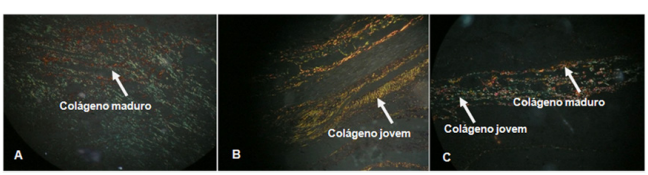

FIGURE 4 - Density of immature and mature collagen in picrosirius red: A) mature collagen in red, aroeira group; B) immature collagen in yellow or green, control group, C) immature and mature collagen in group andiroba

TABLE 2 - Histological evaluation (Mann-Whitney) parameters of immature and mature collagen in healing gastrorraphy of animals treated with extract of aroeira, control (saline with 0.9%) and andiroba oil

Grupos	7º dia		14º dia		21º dia	
	Jovem	Maduro	Jovem	Maduro	Jovem	Maduro
Aroeira	+++ (1)	**+ (5)**	+ (2)	++ (2)	+ (1)	**+++ (7)**
	++++ (9)	++ (3)	++ (4)	**+++ (7)**	++ (7)	++++ (3)
		+++ (1)	+++ (4)	++++ (1)	+++ (2)	
		++++ (1)				
Solução salina 0,9%	+++ (1)	- (1)	+ (2)	- (1)	+ (3)	+ (1)
	++++ (9)	**+ (9)**	**++ (4)**	+ (3)	**++ (4)**	++ (4)
			+++ (3)	**++ (6)**	+++ (2)	**+++ (5)**
			++++ (1)		++++ (1)	
Andiroba	+(1)	- (2)	+ (2)	+ (3)	+ (4)	++ (1)
	+++ (4)	**+ (4)**	++ (4)	**+++ (7)**	**++ (5)**	+++ (2)
	++++ (5)	+++ (2)	+++ (4)		+++ (1)	++++ (7)
		++++ (2)				
P	**0,0497***	0,1065	0,9826	**0,0002***	0,3947	**0,0021***

Relative density of immature and mature collagen by picrosirius red staining: absent (-), slight (+) <25%, moderate (+ +) (25-50%), strong (+ + +) (50-75%) or very strong (+ + + +) (> 75%) () = number of animals in each classification; * means statistical difference between groups (p <0.05)

DISCUSSION

In this study were chosen two plants commonly used by the population and the rat, as experimental model, for ease of handling and accommodation, resistance to manipulation, surgical aggressions and infection. Also because this animal has already been standardized by many researchers in the study of wound healing[19]. Males rats were chosen due to hormonal variations of the estrous cycle of females, which may interfere in mechanism of tissue repair. Stomach was used as experimental organ by having the same morphological characteristics of the human and the gastrorraphy held in the region of the body due to technical facilities. However, a study analyzing the pattern of healing in incisional wounds in the walls of the stomach and duodenum of rats with polypropylene thread, the authors found that in intact tissue, the fundus is more extensible than the portion of the body, antrum and duodenum, requiring more energy to rupture[3,28,29,31,33].

In this study the dose of hydroalcoholic extract of aroeira chosen was 100 mg / kg / dose orally (gavage) by calculating the DL-50. Another study used the backlands aroeira (M. urundeuva Allemão) of the same family of Schinus in the dosage of 200 mg / kg / dose[35]. The same dosage of andiroba was applied[8].

Were analyzed three moments in the evaluation: day 7, day 14 and day 21 postoperatively. The 7th day was the initial phase of the healing process while the 14th and 21th days were intermediate and advanced phase.

The gastrorraphy was performed in the body of the stomach involving all levels using Prolene® thread, do to being relatively inert, causing minimal

acute inflammatory reaction and not be subjected to degradation or weakening by enzymatic action tissue for a period of approximately two years[11]. The involvement of all levels of gastric wall in the confection of the suture is different from other studies that used only the serous and muscular layers. Was chosen to use the same gauge wire to standardize the method and avoid any possibility of variation in the response to tissue[3].

On the immediate postoperative, diet with water was introduced and the feed avoided in the first six hours after the surgery to avoid possible wound dehiscence[3], other authors indicate fasting for 48 hours[16]. The good acceptance of nutrition and maintenance of general health in daily clinical evaluation allowed characterization the good evolution of healing of gastric sutures, fact confirmed by macroscopic evaluation at necropsy.

Analysis of the healing parameters

This study aimed to analyze the healing process macroscopically, mechanical resistance of the scar and the morphological and morphometric study[1]. However, there are divergent opinions on the methods of measurement. There is no parallelism between the gain of resistance of an anastomosis to insufflation with the quantify rates of hydroxyproline[21], others showed good progress on the healing of abdominal wall and stomach using Mononylon® and Prolene® respectively for their advantageous features in the healing process. Studies with other threads (Maxon® and PDS®) in the synthesis of the abdominal wall, gastric and intestinal anastomosis stated that the main parameters in choosing the strands are: biological characteristics of the tissue, recovery rate of the tension resistance and speed with which the synthesis material loses its resistance[14]. Some authors have discussed extensively the establishment of standards for analysis of histocompatibility of the threads to the tissues. However, others do not define criteria, but compare the mucous folds at anastomotic site[12]; others authors provide ways to evaluate the presence of spur, necrosis and reaction the presence of threads in the suture line[3,21]. It was noted in this study the occurrence of adhesions at gastrorraphy level, and the structure that was most often involved was the liver, in contrast to other studies that was the greater omentum[3].

Test of resistance to atmospheric air insufflation and traction force

Here it was demonstrated that the group aroeira and andiroba presented higher mean insufflation pressures on the 7th day, but in this test, the control group was better than the experiments, when evaluated at the 21st day. The mean traction forces were greater in the periods of seven and 14 days, which showed better healing for aroeira and andiroba. The rupture has occurred at the level of the suture line in all animals, as well as the pressures and forces increased to in accordance with evaluated periods.

The mechanical resistance of the scar through the test of resistance to atmospheric air insufflation is one of main parameters for evaluating the integrity of anastomosis within the first postoperative days [21]. The determination of the mechanical strength of a scar may be achieved by two techniques: resistance to air insufflation or water and linear traction. When the subject of analysis is related to hollow viscera, such as the gastrointestinal tract, the test of resistance to air insufflation is physiological by reproducing the vectors of pressure that normally are transmitted over the wall of the organ, very similar to the real clinical situation, because the rupture occurs according to the distension[17]. In this study, both the measurement of resistance to air insufflation as the tensile force were carried out in preconized periods corroborating with authors that performed the research in similar period of time[24]. However, it is important to point out that the resistance of an anastomosis in the late period, after the 14th day, is best evaluated by linear tensile with precision dynamometers of an isolated band of tissue.

The measurement of the air insufflation pressure could be interpreted by the influence of testing *in situ* or *in vitro* by registering the exact site of gas escaped in relation to the anastomosis[17]. Most authors recommend removing the piece carefully without undo the adhesions on it and perform the test *in vitro* because it allows better identification of the exact location of the gas escaped - important information for analysis and conclusion of the mechanical resistance[9]. It is important to note that the rupture by insufflation test has occurred in a specific point of the suture line (the weakest point), and may conclude that at this location prevails immature collagen, whereas the tensile force showed linear rupture, ie, dependent on the break of immature and mature collagen.

Microscopic evaluation

The healing process which begins with the inflammatory response is a subject of continued interest in surgery due to the use of sutures, ligatures or biological adhesives as a form of approaching the tissues. One of the main criteria to be noted is the intensity of tissue reaction induced by thread, since the excessive inflammatory response may compromise the healing process. In the present study, less acute inflammatory reaction was observed in the groups aroeira and andiroba, compared with control group.

The inflammatory response excessive or missing compromises the tissue healing process. Some studies report that as the observation time extends occurs the regression of the acute inflammatory response and the chronic response prevails[23].

In this study, for staining of slides, was used

the hematoxylin and eosin, Masson's trichrome and picrosirius red in which different indicators were quantified and classified in accordance with its presence and intensity. It was adopted the classification of the inflammatory process in: acute, corresponding to stage I or inflammatory in healing process (neutrophilic infiltration, interstitial edema and vascular congestion), and chronic, corresponding to stage II or proliferative of the healing process (mononuclear infiltrate, fibroblast proliferation, fibrosis, collagenization and granulation tissue). The picrosirius red was used not only to check the area and density of total collagen, but also to assess the area and density of the fibers-forming collagen, differentiation of immature and mature fibers, different from other studies which use only hematoxylin and eosin and Masson's trichrome, which assess only the total collagen. In the present study, it was noticed that despite the variables, fibroblast proliferation and collagenization were favorable in the control group. It was noticed by picrosirius red staining, that collagen formed in this group was predominantly immature, whereas in groups aroeira and andiroba prevailed the mature collagen. This fact was responsible for the better healing process promoted in these last two groups.

CONCLUSION

The use of the hydroalcoholic extract of aroeira (*Schinus terebinthifolius* Raddi) and andiroba oil (*Carapa guianensis* Aublet) was able to accelerate the healing process of the stomach.

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