Original Article

Comparative evaluation of healing response between colo-colic invagination anastomosis and single-layer running suture. Experimental study in dogs

Miguel Augusto Arcoveder Nogueira\textsuperscript{a}, Francisco Sérgio Pinheiro Regadas\textsuperscript{b,\textdagger}, Carlos Renato Sales Bezerra\textsuperscript{a,c,d}, Wellington Ribeiro Figueiredo\textsuperscript{d,e}, Erbert Portela Martins Filho\textsuperscript{e}

\textsuperscript{a} Universidade Estadual do Piauí (UESPI), Escola de Medicina, Teresina, PI, Brazil
\textsuperscript{b} Universidade Federal do Ceará (UFC), Escola de Medicina, Fortaleza, CE, Brazil
\textsuperscript{c} Programa de Cirurgia Abdominal, Universidade Estadual do Piauí (UESPI), Teresina, PI, Brazil
\textsuperscript{d} Universidade Federal do Ceará (UFC), Fortaleza, CE, Brazil
\textsuperscript{e} Faculdade Integral Diferencial (FacidDevry), Teresina, PI, Brazil

\begin{abstract}
Objective: Evaluate healing response of colo-colic anastomosis by invagination vs. single-layer suture.

Methods: Sixty dogs were randomly distributed in two groups and anastomosed with single-layer suture (G-I, control) or by invagination and cardinal sutures (G-II, study). In the end, the animals were euthanized (10 from each group on POD7 and 20 on POD21) and the anastomosed segment was retrieved for histology and immunohistochemistry. Parameters included body weight, adhesions, edema, vasoproliferation, type I and III collagen, myeloperoxidase and nitric oxide. Findings were analyzed with Student's t test and the Mann–Whitney test.

Results: No animal died prior to euthanasia. The groups were similar with regard to all parameters: median weight 10.86 kg (G-I) and 9.98 kg (G-II) on POD7 \((p = 0.41)\) and 18.86 kg (G-I) and 11.55 kg (G-II) on POD21 \(p = 0.71\); abdominal adhesions \(p = 0.7383\) POD7; \(p = 0.5685\) POD21), level of edema \(p = 0.3006\) POD7; \(p = 0.7990\) POD21), vasoproliferation \(p = 0.1191\) POD7; \(p = 0.0758\) POD21), type I collagen \(p = 0.4591\) POD7; \(p = 0.3357\) POD21), type III collagen \(p = 0.2166\) POD7; \(p = 0.2712\) POD21), nitric oxide \(p = 0.3980\) POD7; \(p = 0.4796\) POD21) and myeloperoxidase \(p = 0.580\) POD7; \(p = 0.755\) POD21).

Conclusion: No significant difference in healing response was observed between the two anastomosis techniques (single-layer suture and invagination).

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\textsuperscript{\textdagger} Corresponding author.
E-mail: sregadas@hospitalsaocarlos.com.br (F.S.P. Regadas).

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Avaliação comparativa da resposta cicatricial entre anastomose colocólica por invaginação e sutura contínua em plano único. Estudo experimental em cães

R E S U M O

Objetivo: Avaliar a resposta cicatricial da anastomose colocólica por invaginação versus sutura em plano único.

Métodos: Sessenta cães foram randomicamente distribuídos em dois grupos e anastomosados com sutura em plano único (G-I, controle) ou por invaginação e suturas cardinais (G-II, estudo). Ao final, os animais foram submetidos à eutanásia (10 de cada grupo no 7º dia do pós-operatório [DPO7] e 20 em DPO21) e o segmento anastomosado foi recuperado para estudos histológicos e imunoistoquímicos. Os parâmetros foram: peso corpóreo, aderências, edema, vasoproliferação, colágeno dos tipos I e III, mieloperoxidase e óxido nítrico. Os achados foram analisados com as testes t de Student e de Mann-Whitney.

Resultados: Não ocorreram óbitos antes da eutanásia. Os grupos eram semelhantes com relação a todos os parâmetros considerados: peso mediano 10,86 Kg (G-I) e 9,98 Kg (G-II) em DPO7 (p = 0,41) e 11,86 Kg (G-I) e 11,55 Kg (G-II) em DPO21 (p = 0,71); aderências abdominais (p = 0,7383 DPO7; p = 0,5685 DPO21), nível de edema (p = 0,3006 DPO7; p = 0,7990 DPO21), vasoproliferação (p = 0,1191 DPO7; p = 0,0758 DPO21), colágeno tipo I (p = 0,4591 DPO7; p = 0,3357 DPO21), colágeno tipo III (p = 0,2166 DPO7; p = 0,2712 DPO21), óxido nítrico (p = 0,3980 DPO7; p = 0,4796 DPO21) e mieloperoxidase (p = 0,580 DPO7; p = 0,755 DPO21).

Conclusão: Não foi observada diferença significativa na resposta cicatricial entre as duas técnicas de anastomose (sutura em plano único e invaginação).

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6 and 12 h respectively during the first 24 h after the procedure. Meloxicam (0.2 mg/kg s.c.) was done every 12 h for 3 days and Tramadol (2 mg/kg i.m.) was administered preoperatively and at each 4 h during the first 24 h after the procedure. Following anesthesia with propofol (5 mg/kg i.v.) and orotracheal intubation, the animals were placed on mechanical ventilation with isoflurane in a semi-closed circuit. The anesthetized animals were submitted to digital rectal examination to evaluate the bowel preparation according to the classification of O’Dwyer et al.⁹ (excellent = no feces; good = minimal fecal residue; acceptable = liquid feces; soiled = solid feces). The grading was confirmed during the surgical procedure. Subsequently, the animals were placed in dorsal decubitus and the laparotomy was performed through a transumbilical, median incision measuring approximately 12 cm. The abdominal cavity was inspected to rule out hematogenic or gynecological disorders. Initially, the descending colon was identified at 20 cm from the anal margin and secured with a straight intestinal clamp (Fig. 1).

A linear transverse colotomy was performed using scissors. The animals from Group I were then submitted to colo-colic anastomosis, either with a single-layer running suture using polypropylene thread size 000 (Fig. 2) or by invagination in Group II, approximately 3 cm from the dissected mesocolon, inserting the proximal segment into the distal lumen and secured by four cardinal sutures using polypropylene thread size 000, involving all intestinal layers (Fig. 3). After the first postoperative evacuation, the animals were allowed access to standard feed and water ad libitum.

At the end of the study period, the animals were submitted to a second laparotomy to excise the anastomosed colon segment for histological and immunohistochemical analysis. The study parameters included body weight, presence of abdominal adhesions, edema, vasoproliferation, type I and III collagen, nitric oxide and myeloperoxidase. Subsequently, the animals were euthanized with 20% potassium chloride (i.v.) under anesthesia with ketamine. Ten from each group were euthanized on the 7th post-operative day (POD7) and twenty on the 21st post-operative day (POD21).

The findings were statistically analyzed using Student’s t test (non-paired samples, parametric data) and the Mann–Whitney test (non-parametric data). The level of statistical significance was set at 5% (p < 0.05).

Results

Body weight was statistically similar in both groups. The median weight was 10.86 kg and 9.98 kg in Groups I, II respectively on POD7 (p = 0.41) and 11.86 kg, 11.55 kg on POD21 (p = 0.71).

The abdominal adhesions incidence was statistically similar in both groups on POD7 (p = 0.7383) and POD21 (p = 0.5685), as well as for the levels of edema on POD7 (p = 0.3006); on POD21 (p = 0.7990), vasoproliferation on POD7 (p = 0.1191); POD21 (p = 0.0758), type I collagen on POD7 (p = 0.4591); on POD21 (p = 0.3357), type III collagen on POD7 (p = 0.2166); on POD21 (p = 0.2712) and nitric oxide on POD7 (p = 0.3980) and POD21 (p = 0.4796). There wasn’t also significant statistical difference of the myeloperoxidase levels between both groups on POD7 (p = 0.580) and POD21 (p = 0.755).

Fig. 3 – The animals from Group II secured by four cardinal sutures using polypropylene thread size 000, involving all intestinal layers.
Discussion

There are many studies discussing about the surgical techniques currently used to perform colorectal anastomosis. All types of suture (single-layered, double-layered, running, interrupted) and the different types of thread used trying to reduce the adherence rates.10–12 In this experimental study, it was used the invagination technique developed by Luchesi13 which is performed by invaginating the transected proximal wall into the lumen of the distal colon and comparing with single-layer running suture anastomosis regarding to the healing response. None experimental study was found in literature using the inflammatory and healing markers to evaluate colocolic invagination anastomosis.

There wasn’t statistical significant difference between the animals from both groups regarding the frequency of edema at the anastomosis site on POD 7 and POD21. Most animals from both groups presented very mild edema on POD7 and none on POD21 as previously demonstrated in another study16,15 also demonstrated that edema tends to subside after POD14. So, based on the findings in this study, the surgical technique did not change the frequency and intensity of edema at the anastomosis site.

According with previous studies8,16 a good vascularization plays an important role on the viability of intestinal anastomosis. And in this study, there wasn’t a significant statistical difference between both groups with regard to vasoproliferation on POD7 and POD21, suggesting that the surgical technique had no influence on the vascularization intensity, according with the findings previously demonstrated in another study.9

Moore et al.6 demonstrated that anastomosis invaginating the proximal segment into the lumen of the distal colon does not increase the risk of contamination of the perianastomotic tissues and peritoneum. In this study, the groups did not differ significantly with regard the presence of abdominal adhesions according with another study.12 Likewise, the amount of type I and III collagen at the anastomosis site on POD7 and POD21 was not significantly affected by the surgical technique used, as previously supported by Bezerra et al.8

Large amounts of iNOS are usually found inside macrophages up to one month after surgery.17 Moreover, increased levels of nitric oxide may be observed in endothelial cells in the anastomoses area, even in the absence of macrophages.18 Using immunohistochemistry exam, it was determined the levels of nitric oxide and myeloperoxidase in both groups of animals on POD7 and PODO21 but it was not found any significant difference between them, suggesting that the surgical technique has no measurable effect on these parameters as well.8

Conclusion

It was not demonstrated in this experimental study any significant difference on healing response between colocolic invagination anastomosis with single-layer running suture.

Conflicts of interest

The authors declare no conflicts of interest.

References

