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UNIVERSIDADE ESTADUAL PAULISTA

FACULDADE DE MEDICINA VETERINÁRIA E ZOOTECNIA

SUPLEMENTAÇÃO ORAL DE GLICOSAMINOGLICANOS E EXPRESSÃO DE PEQUENOS PROTEOGLICANOS RICOS EM LEUCINA APÓS CISTOTOMIA EM BEXIGAS SAUDÁVEIS VERSUS PARCIALMENTE OBSTRUÍDAS.

NATÁLIA CAROLINE NALESSO DE CASTRO

BOTUCATU-SP
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Dissertação para o título de Mestre no Programa de Biotecnologia Animal apresentada junto à Faculdade de Medicina Veterinária e Zootecnia, Universidade Estadual Paulista “Júlio de Mesquita Filho”, Campus de Botucatu.

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Palavras-chave: Bexiga; Colágeno; Glicosaminoglicanos; Imunofluorescência; Pequenos Proteoglicanos Ricos em Leucina.

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SUMÁRIO

1 CAPITULO 1..... Página 8
2 REVISÃO DE LITERATURA ..... Página 10
2.1 Micção e estrutura vesical ..... Página 10
2.2 Alterações estruturais e reflexo na dinâmica miccional ..... Página 11
2.3 Pequenos proteoglicanos ricos em leucina e seu papel na fibrose..... Página 13

3 OBJETIVO..... Página15
3.1 Objetivos específicos ..... Página 15
4 Referências..... Página 16
5- Capítulo 2..... Página 21
Artigo Científico..... Página 21

Castro, N.C.N., Suplementação oral de Glicosaminoglicanos e expressão de pequenos proteoglicanos ricos em leucina após cistotomia em bexigas saudáveis versus parcialmente obstruídas. Botucatu, 2018. 47 p. Dissertação de Mestrado - Faculdade de Medicina Veterinária e Zootecnia, Campus de Botucatu, Universidade Estadual Paulista "Júlio de Mesquita Filho".

RESUMO

A diminuição da complacência da bexiga está relacionada com modificações no equilíbrio dos componentes da matriz extracelular e na concentração das fibras de colágeno, acarretando em espessamento da parede vesical e fibrose. O objetivo do estudo é analisar a relação entre suplementação oral dos glicosaminogicanos (GAGs), distribuição de pequenos proteoglicanos ricos em leucina (SRLPs) e síntese de colágeno em bexigas saudáveis e fibrosadas por obstrução parcial submetidas a cistotomia. Foram utilizados 56 ratos da linhagem Wistar, fêmeas, divididos em cinco grupos experimentais- G1 (8) – controle, sem procedimento, G2 (12) – animais não suplementados, submetidos ao procedimento de obstrução vesical e posterior cistotomia, G3 (12) – animais suplementados e submetidos ao procedimento de obstrução vesical e posterior cistotomia, G4 (12) – animais não suplementados e submetidos ao procedimento de cistotomia e G5 (12) animais suplementados e submetidos ao procedimento de cistotomia. A suplementação de glicosaminoglicanos foi realizada a cada 24 horas por via oral durante 28 dias pós cistotomia, quando então os animais foram eutanasiados. O colágeno foi avaliado pela Microscopia Cars e biglican, decorina, lumican e fibromodulina pelo método de imunofluorescência. Para análise estatística foi utilizado o teste ANOVA e T-STUDENT, com nível de significância $p < 0,05$. Os animais dos grupos G2 e 3 foram os que apresentaram diferença estatística na seguinte constante: peso bexiga final /peso rato final, com o todos os grupos. A quantificação da imunomarcação da imunofluorescência foi avaliada com auxílio do software ImageJ, houve maior expressão no grupo G3 de todos os SRLPs, não havendo diferença estatística nos demais grupos. Na microscopia Cars (Coherent anti-Stokes Raman Scattering), houve diminuição do sinal de colágeno nos grupos G2 e 3. Concluímos que o aumento global dos proteoglicanos se deve ao tratamento com os GAGs durante 28 dias por via oral associado também a presença de fibrose tecidual. Ainda, não foi observado diferença aparente quanto ao colágeno após tratamento com os GAGs, no entanto, a obstrução uretral parcial leva a alteração do colágeno no tecido vesical.

Palavras-chave: pequenos proteoglicanos ricos em leucina, glicosaminoglicanos, colágeno, bexiga, obstrução vesical parcial, microscopia cars, imunofluorescência.

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ABSTRACT

Decreased bladder compliance is related with changes in the extracellular matrix components and collagen fibers concentration resulting in bladder wall thickening and fibrosis. The purpose of the study is the evaluation of the relationship between oral glycosaminoglycans (GAGs) supplementation, small rich-leucine proteoglycans distribution and collagen synthesis in healthy and fibrous bladder by partial obstruction after cystotomy procedure. Fifty-six female Wistar rats were divided into five experimental groups: G1 (8) – control, without procedure, G2 (12) – not supplemented animals, subjected to bladder obstruction procedure and after cystotomy, G3 (12) – supplemented animals subjected to bladder obstruction procedure and after cystotomy, G4 (12) – not supplemented animals subjected to cystotomy procedure and G5 (12) supplemented animals subjected to cystotomy procedure. Oral GAGs supplementation was done each 24 hours during 28 days after cystotomy, when we performed euthanasia. Collagen was evaluated by CARS (Coherent anti-Stokes Raman Scattering) microscopy and byglican, decorin, lumican and fibromodulin by immunofluorescent method. Statistical analysis results were done by ANOVA and T-STUDENT test with a significance level of $p<0,05$. G2 and 3 groups showed statistical difference on follow constant: bladder final weight/ rat final weight x 100. Immunofluorescent staining was measured by Image J software and there was statistical difference in all SRLPs on G3 group. Cars microscopy showed diminish of collagen signal on G2 and 3 groups. We concluded global increased of SRLPS are associated with GAGs supplementation for 28 days and tissue fibrosis. There wasn’t clearly modification on bladder collagen with supplementation, however partial bladder outlet causes bladder tissue changes.

Key words: small leucine rich proteoglycans, glycosaminoglycans, collagen, bladder, partial bladder outlet obstruction, Cars microscopy, immunofluorescence.

- Avaliar o efeito da suplementação oral de GAGs na distribuição de proteoglicanos (SLRPs) em bexigas de ratos saudáveis ou submetidos à obstrução parcial, tendo como comparativo os mesmos parâmetros obtidos de animais não suplementados;

- Avaliar a relação da distribuição de SLRPs e deposição de colágeno após cistotomia em bexigas de ratos saudáveis ou submetidos à obstrução parcial, tendo como comparativo os mesmos parâmetros obtidos de animais não suplementados;

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Conflito de Interesse

Os autores declaram que não há conflito de interesses.

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