

137 - PRESSURE PAIN EVALUATION IN KNEES OF RATS SUBMITTED TO RESISTANCE EXERCISE

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Universidade Estadual do Oeste do Paraná – campus Cascavel – PR – Brasilgladsonricardo@gmail.com**INTRODUCTION**

Joint pain occurs in response to tissue aggression (KIDD; LANGFORD; WODEHOUSE, 2007), an inflammatory reaction can occur in the synovial membrane with exudate associated with large numbers of inflammatory cells that express cyclooxygenase, interleukins, tumor necrosis factor- α , among other mediators (WOOLF, 2004; REZENDE; GOBBI, 2009).

Management such as medication, physiotherapy and rest are part of the indication of treatment for articular pain (MEDEIROS et al., 2006). The use of NSAIDs and analgesics has been associated with efficacy in the treatment of joint pain, with an inhibitory effect on inflammatory enzymes, such as PGE2 and prostacyclin (KIDD; LANGFORD; WODEHOUSE, 2007); opioids and corticosteroids are also suitable for inflammatory and analgesic treatment (KIDD; LANGFORD; WODEHOUSE, 2007; MATHER, 2001). However, the side effects of such therapies, limit its use (SCOTT et al., 2004). Therapeutic modalities such as TENS (KIDD; LANGFORD; WODEHOUSE, 2007) and low level laser therapy (CAMPANA et al., 2004; SORIANO et al., 2006; TASCIOGLU et al., 2004), are used to reduce the inflammation, especially with regard to pain.

Some studies in animals and humans have shown that submaximal exercise contribute to the alteration of pain perception (BILBERG; AHLMÉN; MANNERKORPI, 2005; LANA; PAULINO; GONÇALVES, 2006; KOLTYN, 2000; HOPKINS et al., 1998; BLUSTEIN; MCLAUGHLIN; HOFFMAN, 2006; MOGIL et al., 1996), one of the hypotheses is the neurophysiological analgesia by activation of a mechanism for endogenous opioid system induced by stress (KOLTYN, 2000; BENDER et al., 2007). However, it is possible that exercise might exacerbate a painful condition (KOLTYN, 2000; KOLTYN; UMEDA, 2006)

Aiming to get a better understanding of the effects of resistance exercise on joint pain, this study objective to evaluate the use of jumps in water, working in analgesia in the knees of rats submitted to joint pain induced by formalin 5%.

MATERIALS AND METHODS**Sample and Experimental Groups**

We used 14 male Wistar rats, 12 ± 2 weeks old, kept in plastic cages with free access to water and food ad libitum, with 12 hours light / dark cycle and room temperature controlled ($24 \pm 1^\circ \text{C}$). The study was conducted according to international standards of ethics in animal experiments (ANDERSEN et al., 2004), was approved by the Ethics Committee on Animal Experiments and Practical Lessons from the State University of West Paraná (Unioeste) under number 4810.

The animals were divided randomly into two groups:

- Placebo Group (PG, $n = 7$) – composed of animals submitted to the induction of nociception in the right knee, and not treated;
- Jump Group (JG, $n = 7$) – composed of animals submitted to nociception and performed as anaerobic exercise, jumping into water.

Nociception Experimental Model

The animals were restrained manually, in the supine position and was injected (27 gauge-needles) in the tibio-femoral joint space medial right knee, 50 μL of 5% formalin solution to induce nociception (MARTINS; BASTOS; TONUSSI, 2006).

Pain Evaluation

Von Frey filaments are used to assess nociceptive sensitivity, mechanical stimuli in animals (NEUGEBAUER et al., 2007). The equipment used to perform the test of pain sensitivity was the Von Frey type Digital Analgesymeter Insight®, the equipment consists of an arm transducer, with a polypropylene disposable tip with a capacity of 0.1-1000g, connected to a box amplifier, measuring the pressure made on the surface of the animal.

The test was performed in two ways. First the animal was contained in a wooden cage, with base metal grid, where the evaluator applied the filament on the plantar surface of the hind leg, immediately after the animals were restrained manually and the von Frey filament was applied on the medial joint tibio-femoral hind paw (BEYREUTHER; CALLIZOT; STÖHR, 2007). The tip of polypropylene filament was applied perpendicularly to the area, with gradual increase of pressure, and when the animal withdrew its paw, the test was interrupted to record the withdrawal threshold. There was a time of adjustment and training of animals for two days prior to the study. The evaluations were performed before induction of nociception (EV1), 15 (EV2) and 45 (EV3) minutes and 2 hours (EV4) after induction.

Treatment Protocol

The treatment protocol occurred after the evaluation of the moment EV2. The JG was subjected to jumps in water, using a PVC tube with 20 cm in diameter, loaded with 50% of the weight of the animal, performed by lead weights attached to a velcro strip positioned on the chest of the animal, aiming not impair movement. The animal performed 4 sets of 5 jumps each, with an interval of three minutes between each series. The count of the repetitions occurred each time the animal is projected toward the water surface to breathe. The site used was a water tank of 200 liters, oval, made of plastic, with a depth of 60 cm and water temperature maintained between $30\text{-}32^\circ \text{C}$. The placebo group did not undergo any therapeutic intervention, was not placed in water (less than 1 minute) to receive similar stress.

Statistical Analysis

Was checked data normality through the Kolmogorov-Smirnov test and analyzed within groups by ANOVA with repeated measures with post hoc of Tukey, and unpaired t test for comparison between groups. In all cases the level of significance was 5%.

RESULTS

Plantar Surface Pressure Evaluation

The results showed significant differences when comparing with the AV1 AV2 time for both groups ($p < 0.05$). Moreover, for the placebo group was also significant reduction in AV3 and AV4 (Fig. 1A). The treated group showed significant increase when compared with AV2 AV3 and AV4 (Fig. 1B)

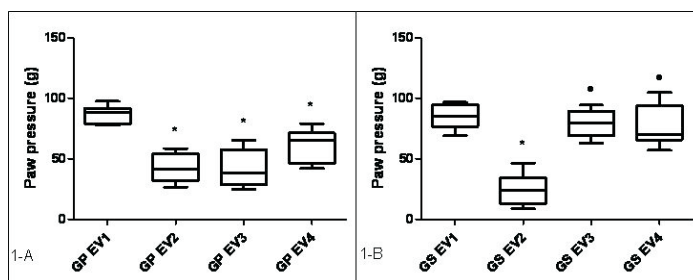


Figure 1 – graphical representation of the values of paw withdrawal threshold when held pressure on the plantar surface (quantity in grams) for the placebo group (1A) and jump group (1B), at different times of evaluation (EV1 – before induction of nociception, EV2 – after 15 minutes, EV3 – after 45 minutes EV4 – after two hours). * Significant difference when comparing with EV1. - significant difference when compared to EV2.

Local Pressure Evaluation

The results for local pressure were similar to those found for pressure on the plantar surface, both for the placebo group, as for the jump group (Fig. 2).

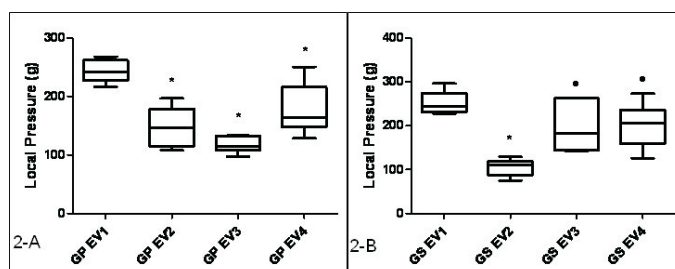


Figure 2 – graphical representation of the values of paw withdrawal threshold, when performed on the medial surface pressure of the knee joint (value in grams) for the placebo group (2A) and group jump (2B), at different times of evaluation (EV1 – before induction of nociception, EV2 – after 15 minutes, EV3 – after 45 minutes EV4 – after two hours). * Significant difference when comparing with EV1. - significant difference when compared to EV2.

DISCUSSION

The pain experimental model produced by formalin injection, is used in models with intraarticular injections, as performed in this study to evaluate the pain and procedures to reduce it. Since the nociception induced by formalin is characterized by two distinct phases, with a period of quiescence between them, around the 5th to 10th minute after induction (MARTINS; BASTOS; TONUSSI, 2006), was chosen in this study to compare pre-injection values with those found 15, 45 minutes and two hours after the pain induction. Thus, the evaluation after formalin injection was related to the second phase of nociception, and subsequent evaluation still related to this phase.

In this study the stress produced by resistance exercise (jump) produced the desired analgesic effect, whereas in two forms of pressure evaluation in this group, the results of EV3 and EV4 returned to baseline and significantly increased in the values observed in these ratings compared to those seen in Ev2.

According Mogil et al. (1996), the exercise is able to act upon the release of endogenous opioids when the stressor is of low intensity and a non-opioid agent when the stressor is of high intensity. As in the present study, we chose to use 30-32 ° C temperature and for a short period of time (four sets of 5 jumps), the analgesic effect observed could be explained by a non-opioid, although the lack of evaluation of the mechanism of analgesia, can be considered a limitation of this study.

However, for Lana, Paulino and Gonçalves (2006), the high-intensity exercise could, unlike the low-intensity stressor, to be a stimulus more intense and thus would be able to trigger neuroendocrine responses more evident in the body, with increased serum levels of corticotropin-releasing hormone (CRH), adrenocorticotrophic hormone (ACTH) and glucocorticoid hormones, and the rise of CRH could be the release of β -endorphin. Still, according to Cunha, Ribeiro and Oliveira (2008), overtraining produces inhibition of the endorphins release, that is, in this study believed that despite intense enough to produce stress and endorphins release, the exercise was not sufficient to produce such inhibition, or if it happened, non-opioid pathways were active.

The form of pain assessment with von Frey filaments is commonly found in literature, with nylon filaments used in the pain region, being used with increasing resistance (SLUKA, 1997; SLUKA et al., 2007; CHEN et al., 2010). In this study we used the Digital Analgesymeter, which behaves similarly to von Frey filaments, but with greater precision, since the force transducer records the peak pressure achieved during the removal of the member. Still, it is noted that were not correlated characteristics of th inflammation or endorphins level, with data from de Analgesymeter, being such limitations suggestions for future studies.

CONCLUSION

We concluded in this study that the jumps in water, used as a form of resistance exercise, produced analgesia in the rats' knees, induced the pain by formalin 5%.

ACKNOWLEDGMENTS

To National Council of Technological and Scientific Development (CNPq) by financial aid for the acquisition of Digital Analgesymeter equipment.

REFERENCES

- ANDERSEN, M. L.; D'ALMEIDA, V.; KO, G. M.; KAWAKAMI, R.; MARTINS, P. J.; MAGALHÃES, L. E.; TUFIK, D. **Princípios éticos e práticos do uso de animais de experimentação**. São Paulo: UNIFESP, 2004.
- BENDER, T.; NAGY, G.; BARNA, I.; TEFNER, I.; KÁDAS, É.; GÉHER, P. The effect of physical therapy on beta-endorphin levels. **European Journal of Applied Physiology**, v. 100, p. 371-382, 2007.
- BEYREUTHER, B.; CALLIZOT, N.; STÖHR, T. Antinociceptive efficacy of lacosamide in the monosodium iodoacetate rat model for osteoarthritis pain. **Arthritis Research & Therapy**, v. 9, n. 1, R14, 8 p., 2007. Disponível em: <http://arthritis-research.com/content/9/1/R14>.
- BILBERG, A.; AHLMÉN, M.; MANNERKORPI, K. Moderately intensive exercise in a temperate pool for patients with rheumatoid arthritis: a randomized controlled study. **Rheumatology**, v. 44, n. 4, p. 502-508, 2005.
- BLUSTEIN, J. E.; MCLAUGHLIN, M.; HOFFMAN, J. R. Exercise effects stress-induced analgesia and spatial learning in rats. **Physiology & Behavior**, v. 89, p. 582-586, 2006.
- CAMPANA, V. R.; MOYA, M.; GAVOTTO, A.; SPITALE, L.; SORIANO, F.; PALMA, J. A. Laser therapy on arthritis induced by urate crystals. **Photomedicine and Laser Surgery**, v. 22, n. 6, p. 499-503, 2004.
- CHEN, J.-J.; LUE, J.-H.; LIN, L.-H.; HUANG, C.-T.; CHIANG, R.P.-Y.; CHEN, C.-L.; TSAI, Y.-J. Effects of pre-emptive drug treatment on astrocyte activation in the cuneate nucleus following rat median nerve injury. **Pain**, v. 148, p. 158-166, 2010.
- CUNHA, G. S.; RIBEIRO, J. L.; OLIVEIRA, A. R. Níveis de beta-endorfina em resposta ao exercício e no sobre-treinamento. **Arquivos Brasileiros de Endocrinologia e Metabolismo**, v. 52, n. 4, p. 589-598, 2008.
- HOPKINS, E.; SPINELLA, M.; PAVLOVIC, Z. W.; BODNAR, R. J. Alterations in swim stress-induced analgesia and hypothermia following serotonergic or NMDA antagonists in the rostral ventromedial medulla of rats. **Physiology & Behavior**, v. 64, n. 3, p. 219-225, 1998.
- KIDD, B. L.; LANGFORD, R. M.; WODEHOUSE, T. Arthritis and pain: Current approaches in the treatment of arthritic pain. **Arthritis Research & Therapy**, v. 9, 214, 7 p., 2007. Disponível em: <http://arthritis-research.com/content/9/3/214>.
- KOLTYN, K. F.; UMEDA, M. Exercise, hypoalgesia and blood pressure. **Sports Medicine**, v. 36, n. 3, p. 207-214, 2006.
- KOLTYN, K. F. Analgesia following exercise. **Sports Medicine**, v. 29, n. 2, p. 85-98, 2000.
- LANA, A. C.; PAULINO, C. A.; GONÇALVES, I. D. Influência dos exercícios físicos de baixa e alta intensidade sobre o limiar de hipernocicepção e outros parâmetros em ratos. **Revista Brasileira de Medicina do Esporte**, v. 12, n. 4, p. 248-254, 2006.
- MARTINS, M. A.; BASTOS, L. C.; TONUSSI, C. R. Formalin injection into knee joints of rats: pharmacologic characterization of a deep somatic nociceptive model. **The Journal of Pain**, v. 7, n. 2, p. 100-107, 2006.
- MATHER, L. E. Trends in the pharmacology of opioids: implications for the pharmacotherapy of pain. **European Journal of Pain**, v. 5, Sup. A, p. 49-57, 2001.
- MEDEIROS, M. M. C.; FERRAZ, M. B.; VILAR, M. J. P.; SANTIAGO, M. B.; XAVIER, R. M.; LEVY, R. A.; CICONELLI, R. M.; KOWALSKI, S. C. Condutas Usuais entre os reumatologistas brasileiros: levantamento nacional. **Revista Brasileira de Reumatologia**, v. 46, n. 2, p. 82-92, 2006.
- MOGIL, J. S.; STERNBERG, W. F.; BALIAN, H.; LIEBESKIND, J. C.; SADOWSKI, B. Opioid and nonopioid swim stress-induced analgesia: A parametric analysis in mice. **Physiology & Behavior**, v. 59, n. 1, p. 123-132, 1996.
- NEUGEBAUER, V.; HAN, J. S.; ADWANIKAR, H.; FU, Y.; JI, G. Techniques for assessing knee joint pain in arthritis. **Molecular Pain**, v. 3, n. 8, 13 p., 2007. Disponível em <http://www.molecularpain.com/content/3/1/8>.
- REZENDE, U. M.; GOBBI, R. G. Tratamento medicamentoso da osteoartrose do joelho. **Revista Brasileira de Ortopedia**, v. 44, n. 1, p. 14-19, 2009.
- SCOTT, A.; KHAN, K. M.; ROBERTS, C. R.; COOK, J. L.; DURONIO, V. What do we mean by the term "inflammation"? A contemporary basic science update for sports medicine. **British Journal of Sports Medicine**, v. 38, p. 372-380, 2004.
- SLUKA, K. A. Blockade of calcium channels can prevent the onset of secondary hyperalgesia and allodynia induced by intradermal injection of capsaicin in rats. **Pain**, v. 71, p. 157-164, 1997.
- SLUKA, K. A.; RADHAKRISHNAN, R.; BENSON, C. J.; ESHCOL, J. O.; PRICE, M. P.; BABINSKI, K.; AUDETTE, K. M.; YEOMANS, D. C.; WILSON, S. P. ASIC3 in muscle mediates mechanical, but not heat, hyperalgesia associated with muscle inflammation. **Pain**, v. 129, p. 102-112, 2007.
- SORIANO, F.; CAMPANA, V.; MOYA, M.; GAVOTTO, A.; SIMES, J.; SORIANO, M.; SPITALE, L.; PALMA, J. Photobiomodulation of pain and inflammation in microcrystalline arthropathies: experimental and clinical results. **Photomedicine and Laser Surgery**, v. 24, n. 6, p. 140-150, 2006.
- TASCIOGLU, F.; ARMAGAN, O.; TABAK, Y.; CORAPCI, I.; ONER, C. Low power laser treatment in patients with knee osteoarthritis. **Swiss Medical Weekly**, v. 134, n. 17-18, p. 254-258, 2004.
- WOOLF, C. J. Pain: moving from symptom control toward mechanism-specific pharmacologic management. **Annals of Internal Medicine**, v. 140, n. 6, p. 441-451, 2004.

PRESSURE PAIN EVALUATION IN KNEES OF RATS SUBMITTED TO RESISTANCE EXERCISE

ABSTRACT:

Joint pain occurs in response to tissue aggression. The use of NSAIDS, steroids and analgesics has been associated with efficacy in the joint pain treatment, however, the side effects of such therapies, limit its use. Thus physical modalities earn their place in the treatment of such injuries, such as TENS, low-power laser and exercise, which presents some disagreement in the literature with respect to its real benefit. So the goal of this study was to evaluate the use of resistive exercise on joint pain in the knees of rats subjected to injection of formalin 5%. We used 14 male Wistar rats, they were divided into two groups: Placebo Group and Jump Group. To induce nociception was injected into the right knee medial tibio-femoral space, 50 µL of 5% formalin solution. Pain was assessed with use of Digital Analgesymeter, measuring the pressure required to produce limb withdrawal, either by stimulation of the plantar surface of the animal right paw, as in the lateral right knee. The evaluations were performed before induction of pain, after 15 and 45 minutes and finally after 2 hours of induction. For JG the exercise consisted of four sets of 5 jumps in water, loaded with 50% of body weight. In both forms of assessment used, for PG decreased the withdrawal threshold, no return to baseline, or increase with respect to the second evaluation. JG also decreased the pain threshold in the 2nd assessment, but after the exercise, the thresholds returned to pre-injury, with a significant increase when compared with the pre-exercise. We conclude that the pain produced by 5% formalin was attenuated with resistance exercise.

KEYWORDS: exercise, pain measurement, hydrotherapy.

VASOPRESSIVES ÉVALUATION DE LA DOULEUR DANS LES GENOUX DES RATS SOUMIS A L'EXERCICE DE RESISTANCE

RÉSUMÉ:

La douleur articulaire survient en réponse à l'agression tissulaire. L'utilisation des AINS, les corticoïdes et des analgésiques a été associée à l'efficacité dans le traitement des douleurs articulaires, cependant, les effets secondaires de ces traitements, en fin de compte de limiter son utilisation. Ainsi, les modalités physiques de gagner leur place dans le traitement de ces blessures, tels que les RTE, laser de faible puissance et de l'exercice, qui présente un certain désaccord dans la littérature par rapport à son bénéfice réel. Donc, le but de cette étude était d'évaluer l'utilisation d'exercice contre résistance sur les douleurs articulaires dans les genoux de rats soumis à une injection de formol à 5%. Nous avons utilisé 14 rats mâles Wistar ont été divisés en deux groupes: le groupe et groupe placebo Salto. Pour induire la nociception a été injecté dans l'espace interne des droits des animaux tibio-fémorale, 50 ml de solution à 5% de formol. La douleur a été évaluée à l'utilisation de analgesymeter numérique, mesure de la pression nécessaire pour produire le retrait du membre, soit par la stimulation de la surface plantaire de la patte droite de l'animal, comme dans le genou droit interne. Les évaluations ont été réalisées avant l'induction de la douleur, après 15 et 45 minutes et, enfin, après 2 heures d'induction. GS pour l'exercice se composait de quatre séries de 5 sauts dans l'eau, chargée avec 50% du poids corporel. Dans les deux formes d'évaluation utilisées pour le GP a diminué le seuil de retrait, pas de retour aux valeurs de base, ou une augmentation par rapport à la deuxième évaluation. GS a aussi diminué le seuil de la douleur dans l'évaluation des 2e, mais après l'exercice, les seuils de revenu avant l'accident, avec une augmentation significative par rapport à la pré-exercice. Nous concluons que la douleur produite par le formol à 5% a été atténuée par des exercices de résistance.

MOTS-CLÉS: exercice, la mesure de la douleur, balnéothérapie.

PRENSORAS EVALUACIÓN DEL DOLOR EN LAS RODILLAS DE RATONES SOMETIDOS A EJERCICIOS DE RESISTENCIA

RESUMEN:

El dolor articular se produce en respuesta a la agresión tisular. El uso de AINES, esteroides y analgésicos se ha asociado con la eficacia en el tratamiento del dolor en las articulaciones, pero los efectos secundarios de estas terapias, en última instancia, limitando su uso. Así, medidas físicas, ganarse su lugar en el tratamiento de estas lesiones, como TENS, láser de baja potencia y el ejercicio, que presenta un cierto desacuerdo en la literatura con respecto a su beneficio real. Así que el objetivo de este estudio fue evaluar el uso de ejercicios de resistencia en el dolor articular en las rodillas de ratas sometidas a la inyección de formalina al 5%. Se utilizaron 14 ratas macho Wistar se dividieron en dos grupos: Grupo placebo y Grupo de Salto. Para inducir la nocicepción fue inyectado en el espacio medial derechos de los animales femorotibial, 5 mL de solución de formol al 50%. El dolor fue evaluado con el uso de analgesymeter Digital, la medición de la presión requerida para producir retirada del miembro, ya sea mediante la estimulación de la superficie plantar de la pata derecha del animal, como en la rodilla derecha medial. Las evaluaciones se realizaron antes de la inducción del dolor, después de 15 y 45 minutos y finalmente después de 2 horas de la inducción. GS para el ejercicio consistió en cuatro series de 5 saltos en el agua, cargada con 50% del peso corporal. En ambas formas de evaluación utilizados para el GP disminuyó el umbral de retirada, sin retorno a los valores basales, o el aumento con respecto a la segunda evaluación. GS también disminuyó el umbral del dolor en la evaluación de segundo, pero después del ejercicio, los umbrales regresó a antes de la lesión, con un aumento significativo en comparación con el pre-ejercicio. Se concluye que el dolor producido por formalina al 5% se atenuó con el ejercicio de resistencia.

PALABRAS CLAVES: ejercicio, dimensión del dolor, hidroterapia.

AVALIAÇÃO DA DOR PRESSÓRICA EM JOELHOS DE RATOS SUBMETIDOS A EXERCÍCIO RESISTIDO

RESUMO:

A dor articular ocorre em resposta às agressões teciduais. O emprego de antiinflamatórios não esteróides, corticóides e analgésicos tem sido associado à eficácia no tratamento de dor articular, contudo, os efeitos colaterais de tais terapias, acabam limitando seu uso. Assim modalidades físicas ganham seu espaço no tratamento de tais lesões, como o TENS, laser de baixa potência e exercício físico, o qual apresenta certa discordância na literatura, com respeito a seu real benefício. Assim o objetivo deste estudo foi avaliar o uso de exercício físico resistido sobre a dor articular em joelhos de ratos Wistar submetidos à injeção de formalina 5%. Foram utilizados 14 ratos Wistar, machos, divididos em dois grupos: Grupo Placebo e Grupo Salto. Para induzir a nocicepção foi injetado no espaço tibio-femoral medial direito dos animais, 50 µL de solução de formalina 5%. A dor foi avaliada com uso do Analgesímetro Digital, mensurando-se a pressão necessária para produzir retirada do membro, tanto por estímulo na superfície plantar da pata direita do animal, quanto na região medial do joelho direito. As avaliações ocorreram antes da indução da dor, após 15 e 45 minutos e finalmente após 2 horas da indução. Para GS o exercício consistiu de 4 séries de 5 saltos em meio aquático, com sobrecarga de 50% do peso corporal. Nas duas formas de avaliação utilizada, para GP houve diminuição do limiar de retirada, sem retorno aos valores basais, ou aumento com relação à segunda avaliação. Para GS também houve redução do limiar de dor na 2ª avaliação, mas, após o exercício, os limiares retornaram aos valores pré-lesão, com aumento significativo ao comparar com o momento pré-exercício. Conclui-se que a dor produzida por formalina 5% foi atenuada com o exercício resistido.

PALAVRAS-CHAVE: exercício, medição da dor, hidroterapia.