



Thermographic and histological analysis of rabbit different tenorrhaphies techniques (4 and 6 strands) after early active mobilization

Análise termográfica e histológica de diferentes técnicas de tenorráfias (4 e 6 passadas) em coelhos após a mobilização ativa precoce

Rodrigo Arenhart^[a], Antônio Lourenço Severo^[b], Philipe Eduardo Carvalho Maia^[c], Daniela Augustin Silveira^[d], Rodrigo Roca Lopez^[e], Darian Bocaccio^[f]

^[a] MSc, professor, Departamento de Saúde, Universidade Regional Integrada do Alto Uruguai e das Missões (URI), Erechim, RS - Brazil, e-mail: rodrigoarenhart@gmail.com

^[b] PhD, Medical Residence in Orthopedics and Traumatology instructor, Medical Residence in Hand Surgery and Microsurgery instructor, Instituto de Ortopedia e Traumatologia, Passo Fundo, RS - Brazil, e-mail: antoniolsevero@gmail.com

^[c] Graduated, Medical Residence in Orthopedics and Traumatology instructor, Medical Residence in Hand Surgery and Microsurgery instructor, Hospital Mater Dei, Belo Horizonte, MG - Brazil, e-mail: philipemaia@gmail.com

^[d] MSc, pathologist, Hospital São Vicente de Paulo, professor, Universidade Federal da Fronteira Sul (UFFS) and Universidade de Passo Fundo (UPF), Passo Fundo, RS - Brazil, e-mail: dasilveira@upf.br

^[e] Graduated, Traumatology and Orthopedic Surgeon, Caja de Salud de La Banca Privada, Santa Cruz de la Sierra - Bolivia, e-mail: rodrigotrauma@gmail.com

^[f] Graduated, Traumatology and Orthopedic Surgeon, Department of Knee Surgery, Hospital São José, Jaraguá do Sul, SC - Brazil, e-mail: darianbs@gmail.com

Abstract

Introduction: This research is based on the results of the surgeries of tenorrhaphy, which have been improved due to the association between strong and not voluminous sutures and physiotherapeutic protocols, which preconize the early active motion to the postoperative period. **Objective:** To evaluate the healing process in vivo in different types of tenorrhaphies. **Methods:** Thirty-six rabbits that underwent early active

motion after tenorrhaphy. The sample was constituted of 3 groups of 12, in accordance with the 3 different types of suture (Brasil, Indiana and Tsai). **Results:** On the 15th and 30th days after the surgery, thermographic and histological analyses revealed similar results that all groups showed similar behaviors in the same time of surgical repair, just differentiating between the periods. On the 30th day analysis were observed that collagen fibers being more exuberant thickening, thus being able to offer higher tensile strength to the tendon. **Conclusion:** That suggests early active motion may be increased gradually to around the 30th day taking this as clinical relevance.

Keywords: Infrared thermography. Tenorrhaphy. Early active mobilization. Histology.

Resumo

Introdução: Esta pesquisa é baseada nos resultados das cirurgias de tenorrafia, as quais têm evoluído em virtude da associação de suturas fortes e não volumosas e protocolos fisioterápicos, os quais preconizam mobilização ativa precoce no período pós-operatório. **Objetivo:** Avaliar o processo de cicatrização *in vivo* em diferentes tipos de tenorrafias. **Método:** Trinta e seis coelhos foram submetidos a mobilidade ativa precoce após tenorrafia. A amostra foi constituída por 3 grupos de 12, de acordo com os 3 tipos de sutura utilizados (Brasil, Indiana e Tsai). **Resultados:** No 15^o e 30^o dia após a cirurgia, as análises termográficas e histológicas revelaram resultados similares evidenciando que todos os grupos apresentaram comportamentos semelhantes no mesmo tempo do reparo cirúrgico, diferenciando-se apenas entre os períodos. No trigésimo dia os estudos evidenciaram que as fibras de colágeno mostravam um exuberante afilamento, sendo assim, capazes de oferecer maior resistência tensil ao tendão. **Conclusão:** Isto sugere que o movimento ativo precoce pode ser gradativamente aumentado em torno do trigésimo dia, o que é de grande relevância clínica.

Palavras-chave: Termografia por infra-vermelho. Tenorrafia. Mobilização ativa precoce. Histologia.

Introduction

Lesions in the flexor tendons of the hands can result in great physical and emotional suffering and are a socio-economical setback to both the patient and the society because for the long period required for functional recovery (1).

The benefits of early active mobilization (EAM) include greater excursion of the osteofibrous tunnel and greater range of motion (ROM) and functionality of the hand (2, 3, 4, 5, 6). Understanding the behaviour of wound healing *in vivo*, particularly under tissue stress caused by exercises and during the rehabilitation period, is important and has great clinical relevance.

The formation of scar tissue makes this phase crucial in the healing process. However, the currently available assessment tools may not be able to help efficiently analyse human tendons *in vivo* during the first few weeks of the postoperative period (7).

This study aimed to evaluate the evolution of the healing process in rabbits that were subjected to different types of tenorrhaphies and submitted to early

active mobilization. Histological analysis showed an increase in the concentration and maturity of the collagen fibres, thereby indicating an improvement in the elastic properties of the tendon and good healing progress (7, 8). Because the inflammatory process is exothermic, infrared thermography can be used to assess the severity of inflammation (8, 9). Therefore, we believe that a combination of infrared thermography and histological analysis may be helpful in studying the evolution of the healing process in rabbits that were subjected to tenorrhaphy of the flexor tendons and submitted to early active mobilization.

Materials

The study sample comprised 36 rabbits (species *Oryctolagus cuniculus*; New Zealand). This project was approved by the Ethics Committee in Animal Experimentation (CETEA - protocol 1.01.09) of the State University of Santa Catarina (UDESC) Lages, SC. Adult rabbits (female and male rabbits; age, 8 ½ months; weight, 2–3 kg) were chosen because of the

similarities between the characteristics and properties of their tendinous tissue with the flexor tendon of the human hand. These rabbits were randomly divided into 3 groups of 12 individuals, and each group underwent a different technique for tenorrhaphy: 1) Brazil, 2) Indiana, and 3) Tsai (Table 1). The selection of techniques tenorrhaphy used the following criteria: techniques should allow early active mobilization, hence the choice of 4 and 6 strands, the search for possible interference of the position of the central knot or number of strands in the healing process: Suture Brazil – 4 strands and knot outside the central tendon; Suture Indiana – 4 strands and knot inside the central tendon; suture Tsai – 6 strands Knot outside the central tendon (Figure 1). Has been used mononylon 4.0 for core suture and mononyln 5.0 for peritendon suture.

Method

After tenorrhaphy was performed in the flexor tendon of the right rear paw, the area was immobilised

with a plaster cast extending up to the ankle joint of the operated limb at a 90° flexion to prevent overload in the area. Deambulation inside the confinement cage was considered as early active mobilization, since the subject was still active and physiologically mobile (10).

The following instruments were used: (a) Eletrophysics PV320A® computerised infrared thermographic camera, with the software Velocity version 2.0; (b) Leica Optical Microscope® - DMLS; (c) JB® precision digital scale, model 33; (d) MT® - 241 thermohygrometer (2 channels); and (e) a data acquisition form.

After each tenorrhaphy, a histological and thermographic analysis was performed to quantify the proportion of thick collagen fibres. It was compared the findings of the 3 groups on the 15th and 30th postoperative days. Half of the animals were sacrificed on the 15th day.

We used thermograms to record the maximum temperatures at the tenorrhaphy site (MTT), in the neighbouring area (MTN), and in the contralateral limb (MTC).

Table 1 - Distribution of groups (Source: Laboratory of Experimental Surgery of the Institute of Orthopedics and Traumatology, Passo Fundo, RS – Brazil)

Groups	Type of Tenorrhaphy	Number of rabbits/15* days postoperative	Number of rabbits/30* days postoperative	Total number of rabbits per group
1	Brazil – 4 strands	6	6	12
2	Indiana – 4 strands	6	6	12
3	Tsai – 6 strands	6	6	12

Source: Laboratory of Experimental Surgery of the Institute of Orthopedics and Traumatology, Passo Fundo, RS - Brazil.

*Note: Rabbits were euthanised for thermographic and histological analyses.

The statistical analysis of data was performed using a *t* test for paired variables and Pearson's chi-square test for the frequencies of the histological features. The association between temperature measurements and tenorrhaphy technique and that between temperature measurements and elapsed time after tenorrhaphy were evaluated using univariate analysis of variance (ANOVA). Test values showing $p \leq 0.05$ were considered significant.

Results

On the 15th and 30th postoperative days, the maximum temperature measured at the site of tenorrhaphy was significantly higher than that measured in the surrounding area, respectively (29.8 °C [SD 0.6] vs. 28.5 °C [SD 0.6] and 28.6 °C [SD 0.5] vs. 27.5 °C [SD 0.3]; $p < 0.001$); a similar trend was observed in the contralateral or control limb, respectively (29.8 °C

[SD 0.6] vs. 27.9 °C [SD 0.8] and 28.6 °C [SD 0.5] vs. 27.0 °C [SD 0.5]; $p < 0.001$) (Figure 1). It was also observed that the temperature of the area neighbouring the tenorrhaphy site was significantly higher than that in the control limb on the 15th day [28.5 °C [SD 0.6] vs. 27.9 °C [SD 0.8]; $p < 0.001$) and on the 30th day (27.5 °C [SD 0.3] vs. 27.0 °C [SD 0.5]; $p = 0.001$) after tenorrhaphy.

Still, MTT, MTN, and the MTC were significantly higher on the 15th day compared to those on the 30th day, respectively (29.8 °C [SD 0.6], 28.6 °C [SD 0.5], 28.5 °C [SD 0.6] vs. 27.5 °C [SD 0.3], 27.9 °C [SD 0.7], 27.0 °C [SD 0.5]; $p < 0.001$) (Figure 2).

The analysis of covariance showed that only the elapsed time from the 15th to 30th day after tenorrhaphy had an impact on the maximum temperature ($p < 0.001$), with no significant association with either suture technique ($p = 0.108$) or interaction between technique and time after surgery ($p = 0.820$) (Figure 3).

Among subjects who received the Brazil suture, the predominance of thick fibres on the 15th day was observed in 2 rabbits (33.3%) and on the 30th day in 4 (66.7%; $p = 0.248$). Among the rabbits that received the Indiana suture, the predominance of thick fibres on the 15th day was observed in 1 rabbit (16.7%) and on the 30th day in 4 (66.7%; $p = 0.079$). Among the subjects who received the Tsai suture, on the other hand, the predominance of thick fibres on the 15th day was observed in 4 rabbits (66.7%) and on the 30th day in 4 (66.7%; $p = 1.000$) (Figure 4).

Discussion

The postoperative inflammatory reaction associated with the healing of skeletal muscle tissue is a well-recognised phenomenon in the literature, and the number of cells returns to the normal count, with inhibition of reactional apoptosis around the 30th postoperative day. This reaction causes a physiological increase in the body temperature, which is rarely considered of clinical relevance (11, 12, 13). The thermographic analysis at day 30 is critical because in this period there is a decrease in temperature focal showing the evolution of the inflammatory process reparer and early remodeling phase which is proven by the flattening of the fibers in the histological analysis.

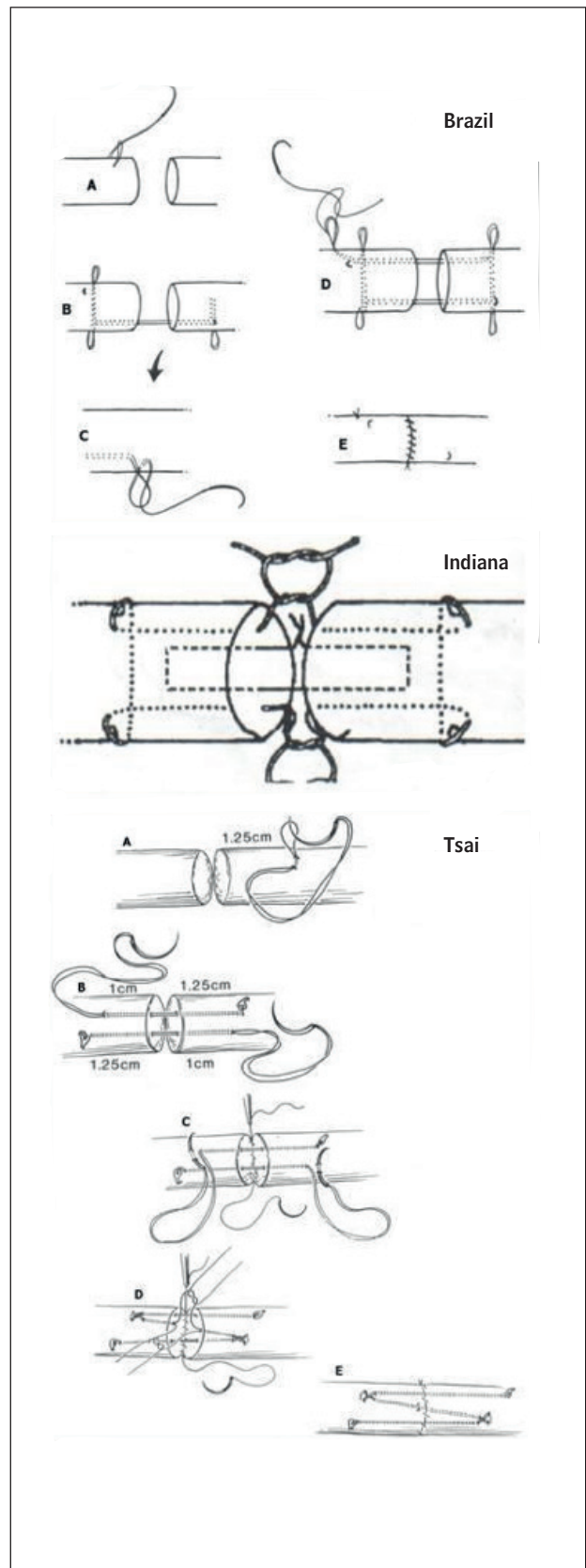


Figure 1 - Diagram of suture techniques: Brasil, Indiana and Tsai

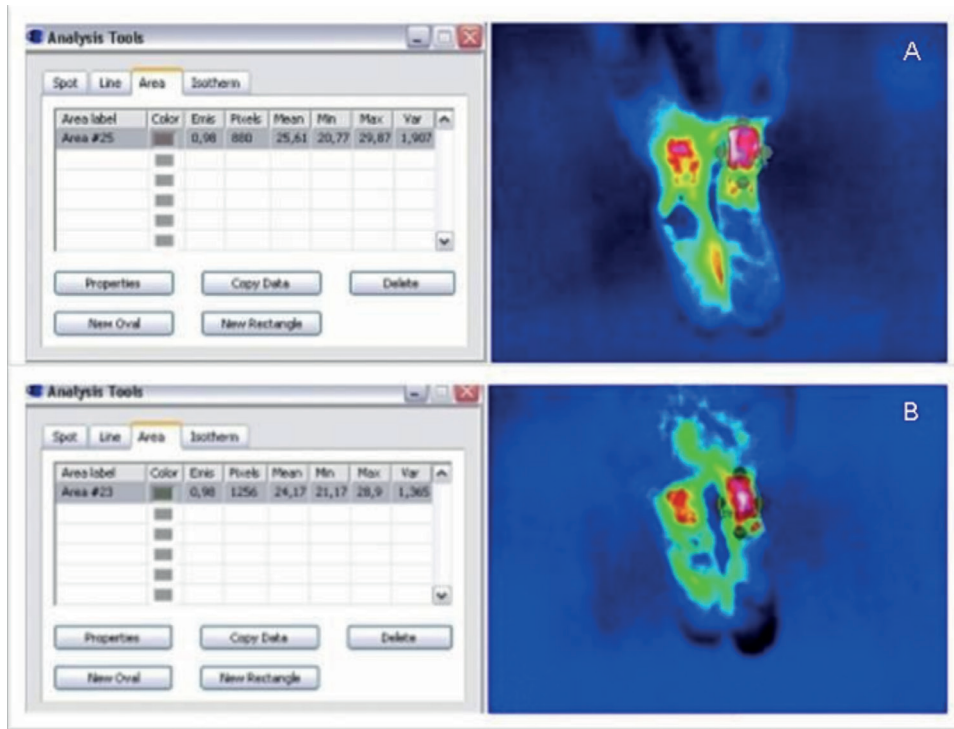


Figure 2 - A) Thermographic analysis MTT: Brazil suture – 15th day. B) Thermographic analysis MTT: Brazil suture – 30th day
Source: Laboratory of Experimental Surgery, Institute of Orthopedics and Traumatology, Passo Fundo, RS – Brazil.

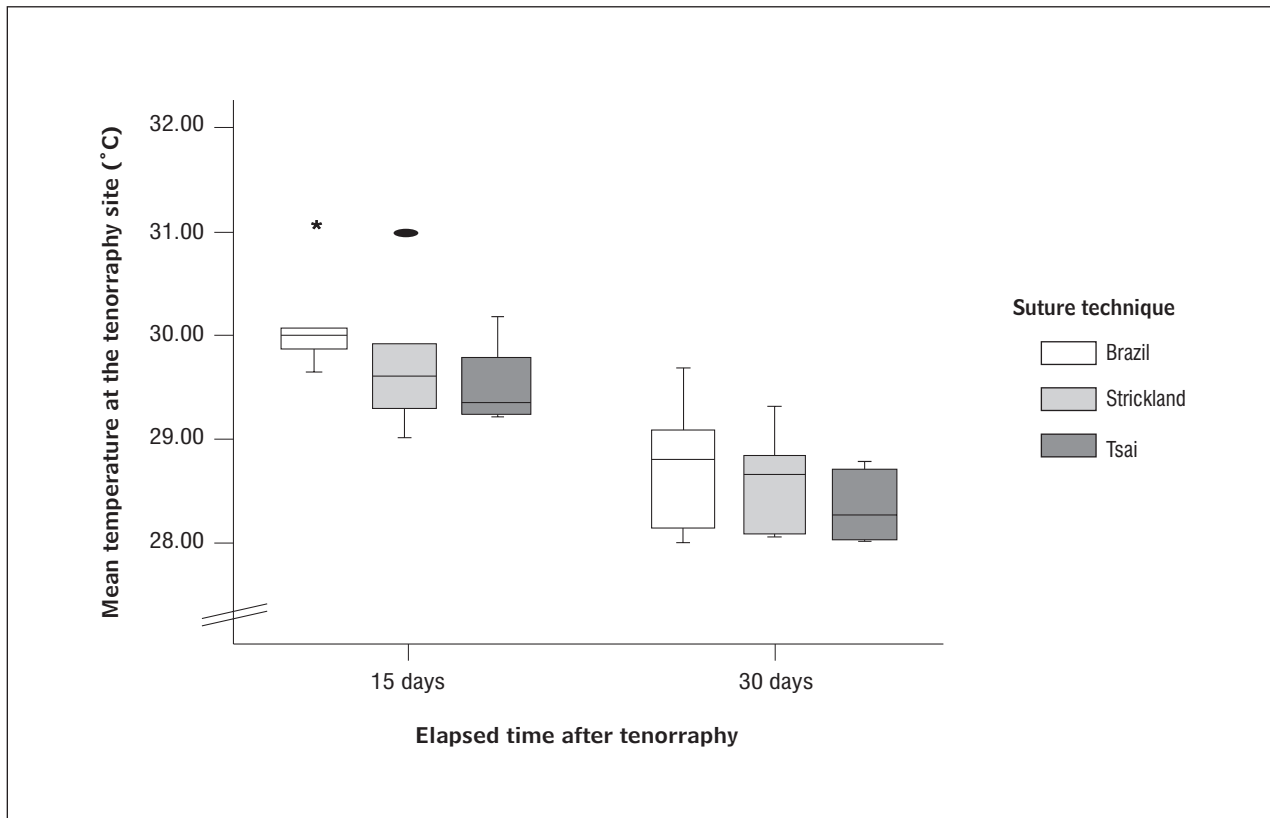


Figure 3 - Maximum temperature (°C) measured at the site of the tenorrhaphy: on the 15th and 30th days after the completion of the different techniques

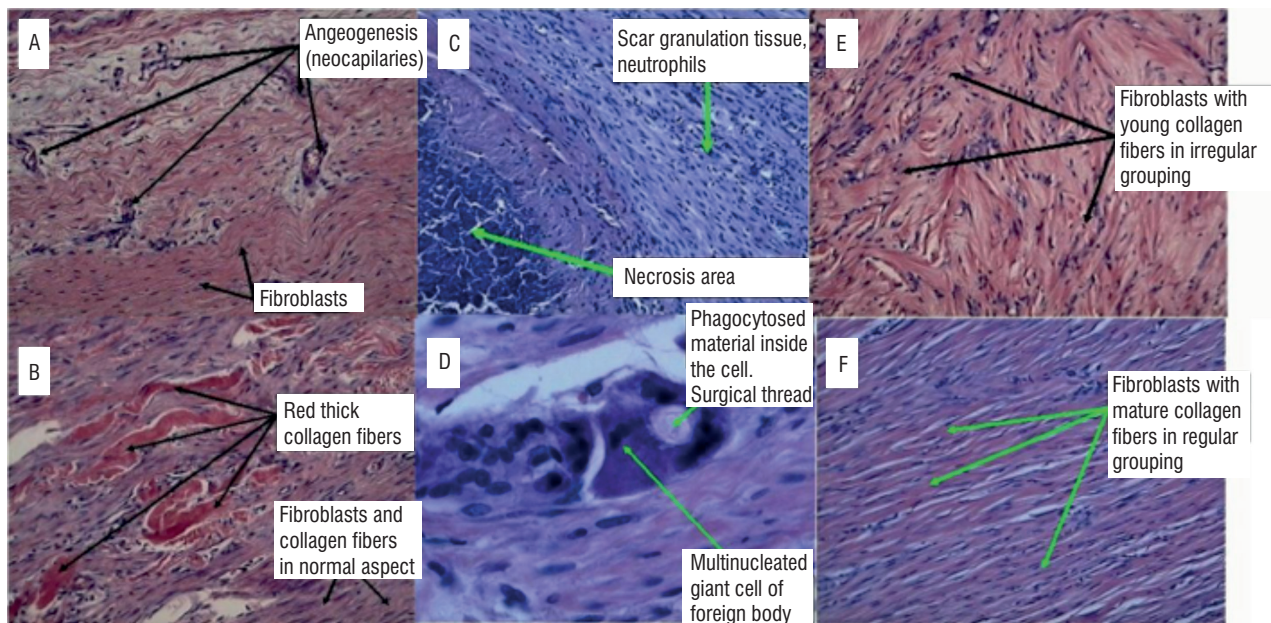


Figure 4 - A) The results of the Tsai technique (6-strands), displaying granulated tissue and angiogenesis on the 15th day (100x). B) Histological section of suture with Tsai technique (6-strands) – 15th postoperative day. Granulation tissue with deposit of thick collagen fibers (100x increase). C) The results of the Brazil technique (4-strands) on the 30th day, showing an area of necrosis in the periphery of the tendon without damage to the healing (100x). D) The results of the Indiana technique (4-strands) on the 15th day, showing a multinucleated giant cell from a foreign body (400x). E) The results of the Tsai technique (6-strands) on the 15th day, depicting the beginning of healing, immature collagen fibres (100x). F) The results of the Tsai technique (6-strands) on the 30th day, revealing the position of mature collagen fibres in the same direction of the axis of the tendon. Optimal healing (100x)

In this study, there was no measurement of internal body temperature. Ignoring this existing variation, the temperatures measured in the different areas in the 3 tenorrhaphy groups on the 15th and 30th day, significantly decreased over time, as compared to the healthy areas (control limb). The action of the natural homeostatic controls was confirmed as the evolution of the healing process.

When using the lower limbs during walking, there is a greater energy demand in the lower limb muscles to strengthen and stabilise the joints, while supporting bodyweight. Due to the initiation of early active mobilization, the left rear limb or control limb also showed an increase in temperature at an early stage (15th day). This rise in temperature occurs due to a compensatory activity when the limb is immobilised. However, it was still apparent that the MTT was considerably high during healing.

The studies of Head and Elliot (14) which used thermography to monitor the evolution of tumour metabolism by checking the angiogenesis, evidenced

hyperthermia in 86% of non-palpable breast cancers. In the present study, angiogenesis was observed in the histological sections collected from the tenorrhaphy sites. This intense neovascularization, which acts on the metabolism of cicatricial tissue, causes an increase in temperature (MTT) that was measured using thermography on the 15th day. On the other hand, during the evolution of tendon healing, MTT decreased on the 30th day.

By using thermography, Koop and Haraldson (15) observed a decrease in the temperature of the masseter muscle (cold spots) in patients with severe temporomandibular dysfunction. According to their findings, a temperature rise occurs during inflammation, but it decreases in instances of dental clenching caused by bruxism. Ring (9) reported the capacity and use of thermography in monitoring inflammation in rheumatic diseases, showing a decrease in the local temperature of the affected joints after the use of anti-inflammatory medication. These findings are similar to those of this study, in that during the initial phase of the healing process, a hyperthermic

sign was observed on the repaired region, and then, during the ensuing healing evolution, there was a drop in the local temperature due to a progressive decrease in metabolic activity. Thus, thermography is an effective and capable diagnostic tool useful for monitoring this condition.

Several authors report that the hyperthermic signal (hot spot) at the active trigger point can be attributed to a cutaneous, reflexive spinal response. This response involves vasodilation in the dermis, along with inhibition of α -adrenergic receptors due to the local nociceptive impulses that are caused by contractures, ischemic tissues, and nociceptive substances (16, 17, 18). Such a physiological response can occur from a painful or nociceptive sensation of the tendinous repaired area, or it could also be caused by EAM, although control measurements would ordinarily be taken, such as the use of analgesics for pain control. Therefore, it was believed that the hyperthermic signal at the site of tenorrhaphy was mainly due to the increased metabolic activity during healing.

The thermal asymmetry with a variation close to SD 0.3 °C is considered physiological in a thermo-neutral environment. The presence of a variation of ≥ 0.5 °C is already considered a significant thermal asymmetry that establishes a diagnosis of dysfunction or pathology (19). Significant differences such as these were found in this study, in which the temperature on the 15th day was significantly higher than the temperature on 30th day. This phenomenon was observed in all 3 types of sutures.

In a systematic review and meta-analysis of 29 studies by Dy et al. (20), who sought the prevention of complications such as adhesions and reruptures, it was observed that the rates of reoperation, rupture, and adherence were 6%, 4%, and 4%, respectively. Moreover, the use of epitendinous suture material decreased the rate of reoperation by 84%. As for the surgical technique, it was found that the use of the Kessler technique (2-strands) led to an adhesion occurrence rate of less than 57%, which is questionable, since the Kessler technique (2-strands) accounts for around 70% of all surgical tendinous repairs worldwide. Other works such as those by Smith and Evans (21), Klein (22), Cao and Tang (23), Groth (24), Lawrence et al. (25), Karjalainen et al. (26), and Lee et al. (27) show that sutures of multiple strands, designed to support higher pressures in the repaired tendons, require more training and

sophisticated skills on the part of surgeons. As a result, the difficulty in learning these techniques discourage their use worldwide, and thus, only 30% of surgeons use them. The studies done so far seek to quantify the rates of complications, but there is no study that describes a protocol that would help prevent such occurrences. Therefore, the thermographic analysis helps to establish a time at which physical therapy should be intensified in order to decrease the rate of adhesion without increasing reruptures.

The histological study showed that the behaviour of tendon healing was an appropriate cicatricial response in all groups, in which the collagen fibres presented a more exuberant thickening near the 30th day, thereby providing a higher tensile strength for early active mobilization (7). The standardization of comparison counting absolute and relative frequency of change of collagen fibers is an excellent object of study for future work. A similarity of the temperatures and of the quality of scar tissue was evident, by both thermographic and histological analysis, in the tenorrhaphy in the 3 different sutures, and was attributed to the minimization of vascular damage and uniform repairing metabolism.

Thermography is capable of measuring temperatures in real time. By observing the attenuation of local inflammation by a significant decrease in temperatures of the tenorrhaphy sites compared to the contralateral limb at different time periods, a physical therapist may be able to observe the evolution of tendon healing *in vivo*. Still, computerised infrared thermography captures the thermal differences caused by tendon microcirculation, and as a result, may be used in the protocols used for rehabilitation of the flexor tendons of the hand. The clinical relevance of this study is that physical therapists would be able to make informed decisions about the possibility of modifying the degree or intensity of movements.

References

1. Severo AL, Arenhart R, Silveira D, Ávila AOV, Berra FJ, Marcelo Barreto Lemos MB, et al. Reparo de tendões flexores da mão: análise biomecânica com diferentes técnicas de sutura. Rev Bras Ortop. 2005; 40(7):418-27.

2. Fernandes CH, Matsumoto MH, Santos JBG, Araújo PMP, Fallopa F, Albertoni WM. Resultados das tenorrafias em flexores dos dedos das mãos, na zona II, submetidos à movimentação precoce passiva assistida. *Rev Bras Ortop.* 1996;31(6):497-501.
3. Harris SB, Harris D, Foster AJ, Elliot D. The etiology of acute rupture of flexor tendon repairs in zones 1 and 2 of the fingers during early mobilization. *J Hand Surg Br.* 1999;24(3):275-80.
4. Kitsis CK, Wade PJ, Krikler SJ, Parsons NK, Nicholls LK. Controlled active motion following primary flexor tendon repair: a prospective study over 9 years. *J Hand Surg Br.* 1998;23(3):344-9.
5. Peck FH, Bücher CA, Watson JS, Roe A. A comparative study of two methods of controlled mobilization of flexor tendon repairs in zone. 2. *J Hand Surg Br.* 1998;23(1):41-5.
6. Riaz M, Hill C, Khan K, Small JO. Long term outcome of early active mobilization following flexor tendon repair in zone. 2. *J Hand Surg Br.* 1999;24(2):157-60.
7. Boyer MI, Gelberman RH, Burns ME, Dinopoulos H, Hofem R, Silva MJ. Intrasynovial flexor tendon repair. An experimental study comparing low and high levels of in vivo force during rehabilitation in canines. *J Bone Joint Surg Am.* 2001;83-A(6):891-9.
8. Kitchen S, Young S. Reparo dos tecidos. In: Kitchen S, Bazin S. *Eletroterapia de Clayton.* São Paulo: Manole; 1998.
9. Ring EFJ. Progress in the measurement of human body temperature. *IEEE Eng Med Biol Mag.* 1998;17(4):19-24.
10. Kusano N, Yoshizu T, Maki Y. Experimental study of two new flexor tendon suture techniques for postoperative early active flexion exercises. *J Hand Surg Br.* 1999;24(2):152-6.
11. Fraser I, Johnstone M. Significance of early postoperative fever in children. *Br Med J.* 1981;283(6302):1299.
12. Yeung RSW, Buck JF, Filler RM. The significance of fever following operations in children. *J Pediatr Surg.* 1982;17(4):347-9.
13. Wu YF, Zhou YL, Mao WF, Avanesian B, Liu PY, Tang JB. Cellular apoptosis and proliferation in the middle and late intrasynovial tendon healing periods. *J Hand Surg Am.* 2012;37(2):209-16.
14. Head JF, Elliot RL. Thermography: its relation to pathologic characteristics, vascularity, proliferation rate, and survival of patients with invasive ductal carcinoma of the breast. *Cancer.* 1997;79(1):186-8.
15. Koop S, Haraldson T. Skin surface temperature over the temporomandibular joint and masseter muscle in patients with craniomandibular disorder. *Swed Dent J.* 1988;12(1-2):63-7.
16. Fischer AA, Chang CH. Temperature and pressure threshold measurements in trigger points. *Thermology.* 1986, 1(4): 212-15.
17. Fischer AA. Documentation of myofascial trigger points. *Arch Phys Med Rehabil.* 1998;69(4):286-91.
18. Diakow PR. Differentiation of active and latent trigger points by thermography. *J Manipulative Physiol Ther.* 1992;15(7):439-41.
19. Brioschi ML, Yeng LT, Pastor EMH, Colman D, Silva FMRM, Teixeira MJ. Documentação da Síndrome dolorosa miofascial por imagen infravermelha. *Acta Fisiatr.* 2007;14(1):41-8.
20. Dy CJ, Hernandez-Soria A, Ma Y, Roberts TR, Daluiski A. Complications after flexor tendon repair: A systematic review and meta-analysis. *J Hand Surg Am.* 2012;37(3):543-51.
21. Smith AM, Evans DM. Biomechanical assessment of a new type of flexor tendon repair. *J Hand Surg Br.* 2001;26(3):217-9.
22. Klein L. Early active motion flexor tendon protocol using one splint. *Hand Surg.* 2003;16:199-206.
23. Cao Y, Tang JB. Biomechanical evaluation of a four-*Strand* modification of the Tang method of tendon repair. *J Hand Surg Br.* 2005;30(4):374-8.
24. Groth GN. Current practice patterns of flexor tendon rehabilitation. *J Hand Ther.* 2005;18(2):169-74.
25. Lawrence TM, Woodruff MJ, Aladin A, Davis TR. An assessment of the tensile properties and technical difficulties of two and four-*Strand* flexor tendon repairs. *J Hand Surg Br.* 2005;30(3):294-7.
26. Karjalainen T, He M, Chong AK, Lim AY, Ryhanen J. Nickel-titanium wire in circumferential suture of a flexor tendon repair: a comparison to polypropylene. *J Hand Surg Am.* 2010;35(7):1160-4.

27. Lee SK, Goldstein RY, Zingman A, Terranova C, Nasser P, Hausman MR. The effects of core suture purchase on the biomechanical characteristics of a multistrand locking flexor tendon repair: cadaveric study. *J Hand Surg Am.* 2010;35(7):1165-71.

Received: 03/12/2014

Recebido: 12/03/2014

Aprovado: 09/10/2014

Approved: 10/09/2014