

IDIOPATHIC CLUB FOOT TREATED WITH THE PONSETI METHOD. HISTOLOGICAL ANALYSIS AFTER ACHILLES TENDON TENOTOMY IN RATS WITH CLUB FOOT

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Abstract

Congenital talipes equinovarus, or clubfoot, remains one of the commonest congenital limb deformities. In most cases of clubfoot are corrected after five to six cast changes and, in many cases, an Achilles tendon tenotomy. This technique results in feet that are strong, flexible, and plantigrade.

The aim of this study is to quantify the histological modifications that occur early in the healing process of lesions made on the Achilles tendons. We made sagittal sections (without suture, with absorbable suture, with non-absorbable suture), frontal section (without suture, with absorbable suture, with non-absorbable suture), cross-section, dentate section and section into Z. For 1 month (day 7, 14, 21, 28) we studied the following histological characteristics grading it semi quantitatively: nuclear density, the amount of interstitial collagen, tendency to the parallel orientation of the nuclei of fibroblasts, granulomatous reactions.

In all evaluated sections we observed a predominantly lymphocytic and plasma cell inflammatory infiltrate, with maximum intensity on day 7 of post-intervention. The intensity of the inflammatory reaction gradually decreased to day 28. On day 7, post-intervention, we identified the presence of granulation tissue at the site of injury. From day 14 we identified a maturation of granulation tissue: the intensity of chronic inflammatory infiltrate decreases, fibroblasts becoming prevalent and producing a more obvious collagen matrix. We observed the tendency of the collagen fibers toward parallel orientation evident from day 14 for sagittal section and 21 for the frontal. By day 28, the distance between the cut ends gradually decreased with reducing the amount of fat interposed, which is absent on day 28.

Key words: clubfoot, Achilles tendon tenotomy, histological slides, section, suture, rat

Introduction

With the widespread acceptance of the Ponseti method of clubfoot treatment major surgical interventions are needed much less frequently and long-term outcomes are improved.¹ Treatment aims at correction to obtain a functional, plantigrade pain-free foot.

Long-term follow-up studies demonstrating malcorrection, overcorrection, pain, and stiffness dampened the enthusiasm for very aggressive surgery.² The main

problem with surgery is that clubfoot wounds heal by a patching up process called repair.

Long-term residual deformity and pain from surgically corrected club feet still continues to occur and presents diagnostic and therapeutic challenges for the orthopedic surgeon.³

Maintenance of function without pain has been demonstrated in a 35-year follow-up study.⁴

Objective

Is to quantify the histological modifications that occur early in the healing of lesions made on the Achilles tendons.

Material and methods

We used wild-type rats, 5 rats for each section type. All rats were housed in Thoren IVC cages maintained at positive pressure with a light cycle of 14 h light, including a dusk period, and 10 h dark. The food was administrated ad libitum in pellets. The work was carried out in accordance with the Federation of European Laboratory Animal Science Associations (FELASA).

Regarding the surgical technique it was performed a longitudinal incision of the tegument in the Achilles tendon area then we made sagittal sections (without suture, with absorbable suture, with non-absorbable suture), frontal section (without suture, with absorbable suture, with non-absorbable suture), cross-section, dentate section and section into Z.

The anesthesia was inhaled using a portable anesthetic device (Harvard Apparatus Isotec 5). Isoflurane (Forene®, Abbott, Solna, Sweden) was used as anesthetic gas (2-2.5%) and 100% oxygen (the flow rate of 1 l/min) was used as vehicle. The operations were performed under non-sterile hygienic conditions.

Laboratory rats survived after interventions and then were sacrificed with an T61 intravenous injection 0.3ml.

All procedures involving animal studies were conducted with local ethics committee opinion and under the strict supervision of a veterinarian.

The tissue material harvested by surgical technique was fixed in 10% neutral buffered formalin. Histological slides were stained with hematoxylin-eosin and Gomori trichrome stain. Stained sections were then examined with a Leica DMD108 microscope.

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For 1 month (day 7, 14, 21, 28) we studied the following histological characteristics grading them semi quantitatively:

- Nuclear density (1, 2, 3)
- The amount of interstitial collagen (1, 2, 3)
- Tendency to the parallel orientation of the nuclei of fibroblasts (1, 2, 3)
- Granulomatous reactions (+, -)

Results

The obtained histological slides were fully evaluated. To quantify the changes that occur early in the healing of lesions made on the tendons, we restricted the quantifying area to the adjacent zone of the histologically normal tendon.

In all evaluated sections we observed a predominantly lymphocytic and plasma cell inflammatory

infiltrate, with maximum intensity on day 7 of post-intervention. The intensity of the inflammatory reaction gradually decreased to day 28. Interventions which involved the suture of the tendon remains evident inflammatory infiltrate in the foreign body granulomatous reactions in the suture thread.

On day 7 post-intervention, we identified the presence of granulation tissue at the site of injury. Its quantity was reduced for interventions that involved suturing the tendon. In lesions without suture, granulation tissue occupies a large area at the interface with surrounding normal tendon histology. From day 14 we identified the maturation of granulation tissue: the intensity of chronic inflammatory infiltrate decreasing, fibroblasts that produce obvious collagen matrix becoming prevalent (Fig.1).

The amount of interstitial collagen

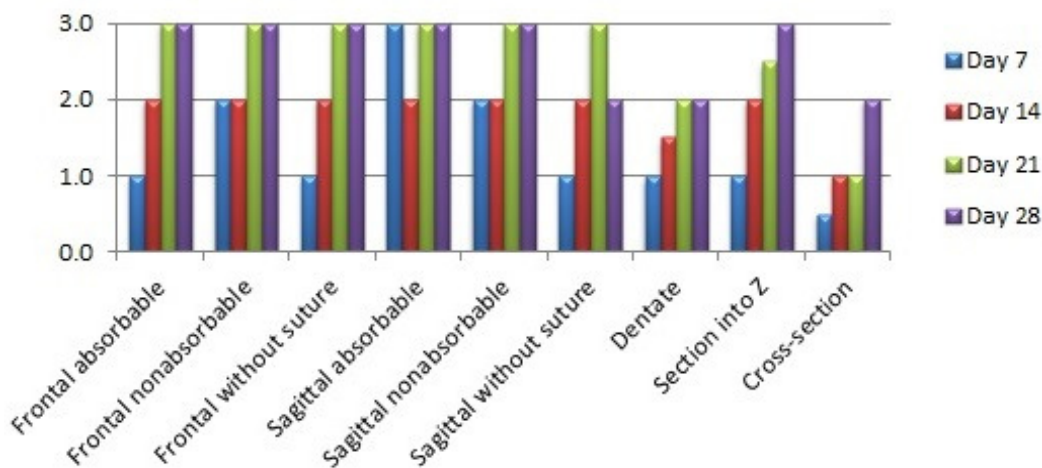


Fig.1. Variation of interstitial collagen from day 7 to day 28.

The orientation of the long axis of the nuclei of fibroblasts becomes perceptible from day 14, with progression to day 28. We observed also the collagen fibers tendency toward parallel orientation evident from day 14 for sagittal section and 21 for the frontal. The orientation in the direction of the force is reduced for the dentate section and for those sectioned into Z, in all four intervals, compared to the other interventions (Fig.2).

In cross-section we observed the adipose tissue interposition between the cut ends of the tendon. By day 28, the distance between the cut ends gradually decreased with reduction of the amount of fat interposed, which is absent on day 28. In the rats where the cross-sectioning of the tendon was performed, the scar tissue which is formed is more cellular on day 28, with a small quantity of collagen, a lack of orientation in the lines of force, which indicates an extended healing time for this type of intervention (Fig.3).

Nuclei orientation

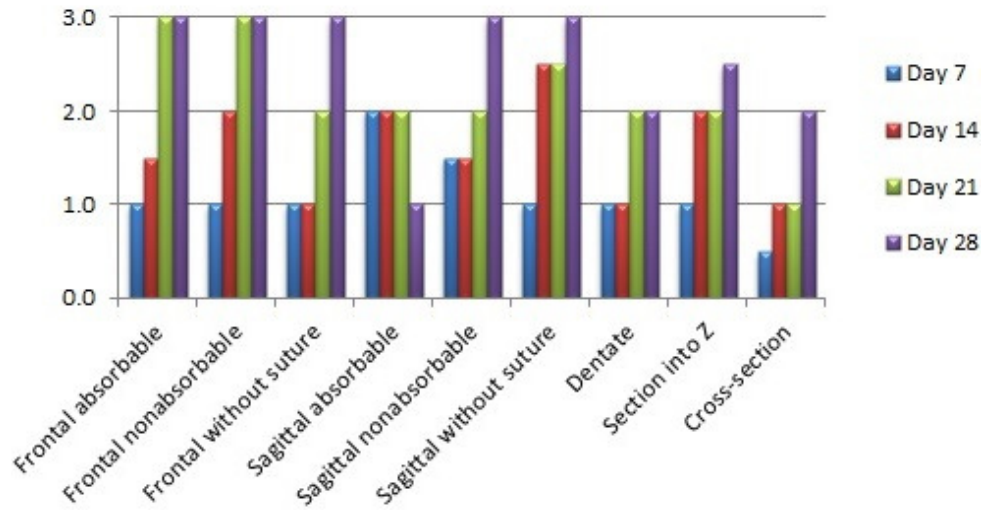


Fig. 2. Nuclei orientation from day 7 to day 28.

Nuclear density

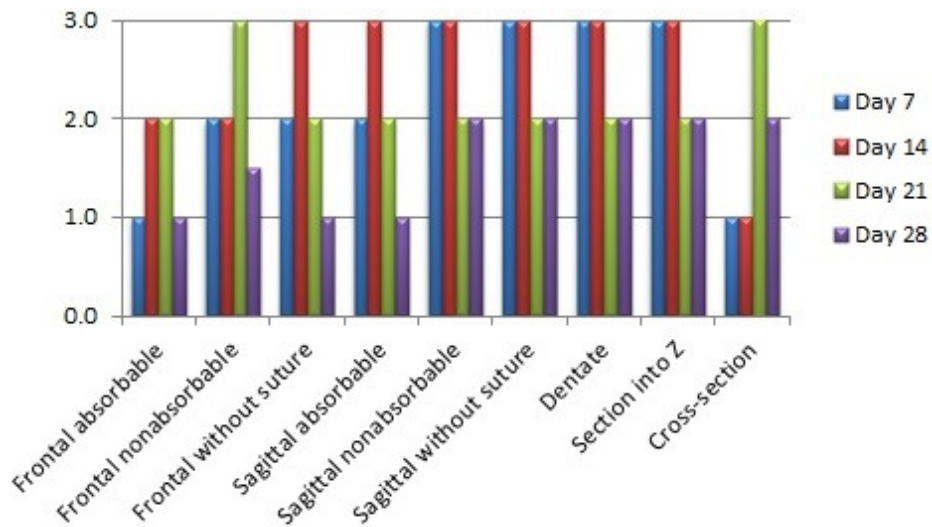


Fig. 3. Nuclear density from day 7 to day 28.

Discussions

The Ponseti method has proven to be successful around the globe, in both industrialized countries and developing nations.⁵

Radler C.⁶ thinks the Ponseti method has become the gold standard of care for the treatment of congenital club foot and despite numerous articles in MEDLINE reporting results from around the globe there are still crucial details of the Ponseti method which seem to be less commonly known or considered.

We found few studies about Achilles tendon tenotomy but not about healing time. *Maranho DA, Nogueira-Barbosa MH, Simão MN, Volpon JB*⁷ believe most cases of congenital clubfoot treated with the Ponseti technique require percutaneous Achilles tenotomy to correct the residual equinus. They performed a study to assess Achilles tendon repair after percutaneous section to correct the residual equinus of clubfoot treated with the Ponseti method. The reparative process was fast after Achilles tendon percutaneous section that reestablishes continuity between stumps. The reparative tissue evolved to tendon tissue with a normal ultrasonographic appearance except for

mild thickening, suggesting a predominantly intrinsic repair mechanism.

*Barker SL, Lavy CB*⁸ studied the correlation of clinical and ultrasonographic findings after Achilles tenotomy in idiopathic club foot. In a study of 11 tendons in eight infants, eight tendons were shown to be clinically intact and ten had ultrasonographic evidence of continuity three weeks after tenotomy. At six weeks after tenotomy all tendons had both clinical and ultrasonographic evidence of continuity.

In our study we observed that compared to the same day in the interventions with suturing, lymphocytic and plasma cell inflammatory infiltrate is evident when using non-absorbable sutures than those absorbable.

Conclusion

We observed that the healing time is better for the sutured Achilles tendons in all evaluated sections than tendons without suture which has an extended healing time.

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