



The efficacy of polypropylene mesh and synovial fluid on tendon healing in dogs (a comparative study)

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Abstract

Tendon disorders are frequent in both human and veterinary medicine with high re-injury rates and unsatisfactory therapeutic treatments. Currently, tendon injury treatments programs are mainly conservative, focused on the pain management rather than on the healing process of the damage tendon.

In the present study, we investigated the efficacy of autologous synovial fluid and polypropylene mesh to assist tendon healing in dog model. Twelve adult dogs were used in this study divided into two groups (six for each). Synovial fluid group, in which group the synovial fluid was injected in the sutured tendon area. In mesh group, the polypropylene mesh was sutured on the tenotomized tendon. Clinical observations were done for four-week post-surgery, and showed improvement in lameness disappearance, and reduce swelling and redness at the operation site in synovial fluid group better than mesh group.

Histological analysis showed that tendon defect covered with synovial fluid had a neo-tendon formation and good end-to-end healing with well organize collagen bundle and reduce gap formation. In conclusion, the histopathological and clinical observations infer that application of autologous synovial fluid on damaged tendon will improve its healing and establish a neo-tendon.

Keywords: dogs, polypropylene mesh, synovial fluid, tendon

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INTRODUCTION

Tendon is an organized connective tissue structure that transfers muscle force to the bony skeleton. Its structure and physiological function expose the higher mechanical stresses that handled by tendon tissues. These mechanical requirements remain under high clinical rates of tendon defects and present intimidating challenges for the clinical therapy of these disorders (Snedeker *et al.*, 2017).

In addition, healing after a tendon injury is usually difficult and uncertain due to the prognosis is influenced by the range of trauma, the time spent between the trauma and its treatment, and low vascularization of the tendon. However, the delayed healing of damaged tendon is due to; vascularization of the tendon is low; a healing process that leads to making adhesions with surrounding tissues, restricting mobility; tension, and active loads, which induce diastasis of the tendon ends; tendon movements (active and passive). (Spinella, *etal.* 2010).

It is important to know the study on tendon is different from the muscles, tendon tissue still under research. For example, it is impossible to get a healthy tendon tissues biopsy from patients or volunteer. Nearly, all existing data related to mechanical principles tissue physiology,

or detailed information of tendon damage or regeneration from animals studies (Snedeker *etal.*, 2017).

Tendon injuries represent considerable morbidity and disablement for several months, despite appropriate management. Healing of tendon can happen intrinsically, through tenocyte's proliferation of epitenon and endotenon, or extrinsically, via cell invasion from the surrounding sheath and synovium. In spite of remodeling, the biomechanical and chemical properties of healed tendon tissue never match those of intact tendon. Tendon basic cell biology is still not completely understood, and the management of tendon injury has a considerable challenge for clinicians (Sharma and Maffulli 2006).

A appropriate biomaterial-based –systems including implantable and injectable systems have been developed to obtain successful tendon tissue healing process. These systems can be as a promised approaches for tendon injury repair. Canine flexor tendon has been widely used for flexor tendon research

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because it has flexor tendon anatomy and functional structure similar to human and it is adaptable for therapy and surgery (Baltzer, 2011)

The bioscaffold methods to enhancement tendon healing; Methods utilized in human tendon and ligament repair may differ somewhat from those utilized in dogs in that maintenance of gliding function following healing is of less importance clinically in dogs. Materials used as a bioscaffold to support and enhance repair have included autogenous, allogeneous and synthetic materials. One described method for example involves application of a bone plate to the tendon repair; however, a second surgery must be performed to remove the plate 8 to 10 weeks postoperatively (Ratcliffe, et al. 2015).

Other synthetic implants used in dogs and humans have included carbon fiber and polypropylene mesh, however these implants may incite foreign body reactions to the materials postoperatively (Goldenberg and de Paula, 2005).

Recently, different strategies treatments are used for Achilles tendon injuries such as the use of synthetic meshes that work as a scaffold to bring the two severed ends of the tendon closer together. In addition its used to stabilizing the sites of rupture, it must be porous to facilitate cell transport, multiplication, and movement of growth factors (Ryan, et al. 2015 and Al-Asadi and Mohammad, 2018).

The adhesions around tendons after injury are a serious issue in tendon surgery. The anti-adhesive applicants are considered the most preferred methods to prevent adhesion formation. The synovial fluid could be used, which has hyaluronic acid as a natural source; it is act as vesicoelastic material. Synovial fluid will be a good alternative to other agents as an affordable and effective treatment option in tendon surgery (Kurt, et al. 2018).

The aim of the study was to compare the effects of polypropylene meshes implantation and synovial fluid in accelerating the repair of tenotomized superficial digital flexure tendons (SDFT) in dogs, depending on clinical examination and pathological evaluations.

MATERIALS AND METHODS

In this comparative study, we used twelve healthy local breed dogs, weighted between fifteen to twenty kg. The animals were put in cages have the same environment in animal house in veterinary college/ university of Basra. Dogs spent two weeks in the cages for acclimatization. We divided the animals in random way into two equal groups (six animals each), the first group was synovial treated group, and the second was polypropylene mesh treated group.

Before surgery, the animal fasted from food 12 hours and from water 5 hours. General anesthesia were done by using a mixture of xylazine 2 mg/kg B.W. and ketamine hydrochloride 10 mg/kg B.W. (Gebremedhin,



Fig. 1. The synovial fluid aspiration from carpal joint



Fig. 2. Superficial digital flexor tendon was exposed and incised transversely

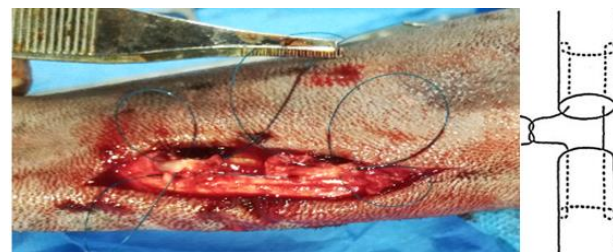


Fig. 3. The synovial fluid aspiration from carpal joint

et al. 2018). The palmar aspect of the metacarpus was prepared for surgery (clipped, shaved, and washed).

When the animal was in lateral recumbency under general anesthesia, incision was made in the palmar aspect of the metacarpus 5 cm, then subcutaneous tissue. The superficial flexor tendon (SDFT) was separated bluntly. Needles were placed on both proximal and distal end of the tendon; the SDFT was full thickness transverse incised between the two needles (**Fig. 2**). The cut ends of tendon were sutured with locking loop suture pattern (modified Kessler) by 3-0 USP non-absorbable suture material (nylon) (**Fig. 3**) (Alkhalifa, 2018).

In polypropylene treated group, a polypropylene mesh was wrapped around the sutured tendon, fixed with 3-0 nylon suture in simple interrupted pattern (**Fig. 4**). While in synovial treated group, six dogs underwent extraction of synovial fluid 0.1-0.2 ml (**Fig. 1**), the autogenous synovial fluid was applied in the site of suturing in tendon. The skin was closed by 3-0 silk with simple interrupted pattern.

The skin wound was covered with a square sterile bandage then rolled with an elastic bandage. Antibiotic was given daily Ceftriaxone 22mg/kg BW for 5 consecutive days.



Fig. 4. Nonabsorbable synthetic surgical mesh was applied around the tendon, fixed by simple interrupted suture pattern with nylon suture material

The clinical evaluations were done by reporting swelling, redness and lameness (0-4 grades) each three days until day 30.

Grade Description of lameness

0. normal (no recognizable lameness; animal weight-bears when standing)
1. lameness is barely detectable (it possible to notice lameness at walk or trot; animal weight-bears when standing)
2. Mild lameness (indistinct but definite at walk and/or trot; and when standing, animal mildly off-weights affected limb)
3. Moderate lameness (apparent at walk and/or trot; animal definitely off-weights affected limb when standing)
4. Severe lameness (carries limb when trotting but at least occasionally weight bearing walking or standing (Wendland, etal. 2016)

Samples were obtained in two different priods, after 15 and 30 days postoperatively, animals were sacrificed humanly. Samples of treated tendon were isolated, fixed in 10 % neutral buffered formalin, and were sent to laboratory in Veterinary College/ University of Basra for histopathological evaluation. Eosin and Hematoxylin stain were used.

RESULTS

Clinical result

During the 1st three days postoperatively, the animals showed the same clinical signs of lameness (between 3 and 4 grade), and swelling and redness at the site of surgery

In polypropylene mesh treated group, lameness still in grade 3 at the 15th day post-surgery (**Fig. 5**) and swelling still exist. In synovial treated groups, the animals showed disappearance of swelling and redness and grade 1 of lameness at the 15th day after surgery (**Fig. 6**) and gradually disappeared at the day 21th. While in mesh treated group, the animals showed swelling at the site of surgery and grade 2 of lameness, and



Fig. 5. Synovial treated dog at the 15th day post-surgery, animal weight-bears when standing



Fig. 6. Polypropylene mesh group at the 15th day post-surgery, animal off-weights affected limb when standing

gradually turned to grade 1 after 27 day postoperatively. At day 30th, lameness disappeared in both groups.

Histopathological Results

In polypropylene treated group after 2 week of surgery, the histopathological sections of tendons

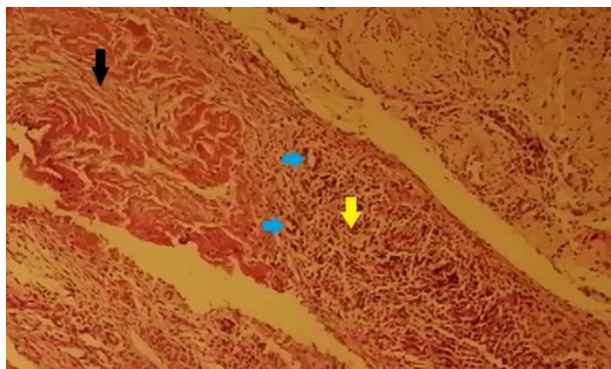


Fig. 7. Mesh treated group /2 Week after surgery. Section of tendon show a huge infiltration of lymphocytes (yellow arrow), accumulation of disorganized collagen fibers (black arrow), and angiogenesis (blue arrows). Poor tendon-to-tendon healing with obvious gap H& E, X100

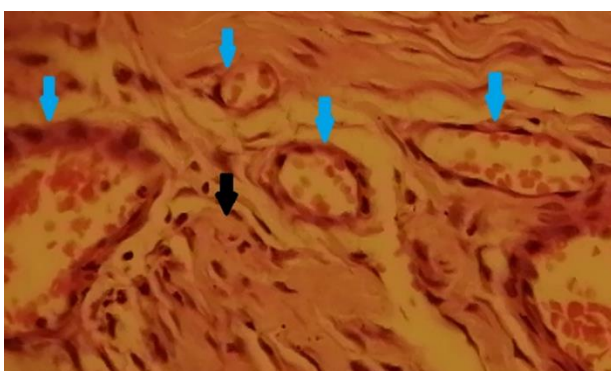


Fig. 8. Mesh treated group /2 Week after surgery, Section of tendon showed active angiogenesis (blue arrows) and disorganized of collagen and accumulation of fibroblast (black arrow) H& E, X400

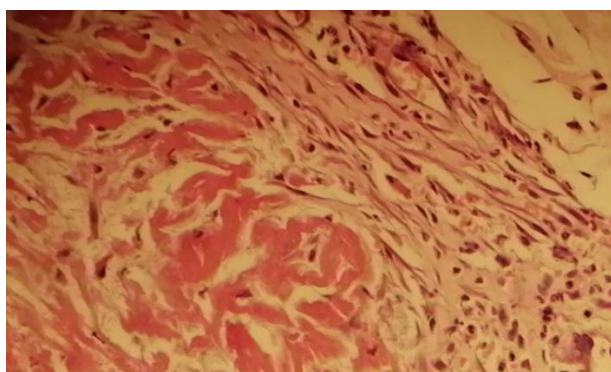


Fig. 9. Mesh treated group /2 Week after surgery. Section of tendon showed disorganized of collagen and accumulation of fibroblast H& E, X400

showed increase infiltration of lymphocytes, accumulation of disorganized collagen, accumulation of fibroblast, and increase in the active angiogenesis, poor tendon-to-tendon healing with obvious gap (Figs. 7, 8 and 9). However, area of neo-tendon after 4 week of surgery show a hug infiltration of inflammatory cells, increased angiogenesis, and disorganized collagen fibers (Figs. 10 and 11).

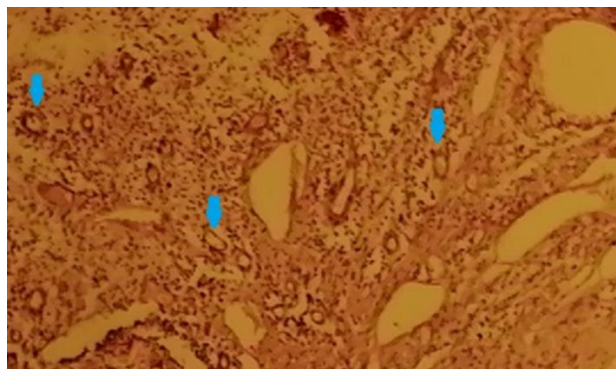


Fig. 10. Mesh treated group /4 Week after surgery: Area of neo-tendon show hug infiltration of inflammatory cells, increased angiogenesis (blue arrows), and disorganized collagen fibers. H& E, X100



Fig. 11. Mesh treated group /4 Week after surgery. Area of neo-tendon show hug infiltration of inflammatory cells (yellow arrows), increased angiogenesis (blue arrows), and few disorganized collagen fibers. H& E, X400

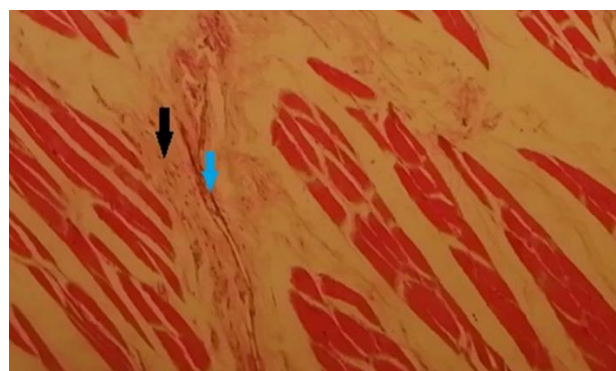


Fig. 12. Synovial treated group/2Week after surgery: Section of tendon show few newly formed blood vessels (blue arrow), accumulation of few well organized collagen fibers. Well tendon-to-tendon healing with no obvious gap. H&E, X100

After 2 weeks of surgery, the histopathological sections of tendon of synovial treated group showed few newly formed blood vessels, accumulation of few well-organized collagen fibers. The sections showed Well tendon-to-tendon healing with no obvious gap (Figs. 12 and 13). Nevertheless, after 4 week of surgery, the area of neo-tendon shows presence of newly formed blood vessels and infiltration of few inflammatory cells,

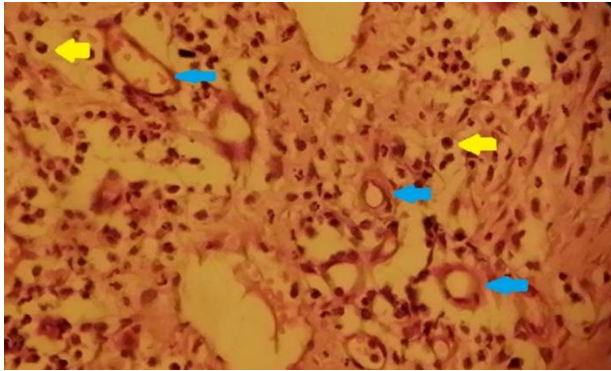


Fig. 13. Synovial treated group/2Week after surgery. Section of tendon show accumulation of few well organized collagen bundles. Well tendon to tendon healing with no obvious gap H&E, X400

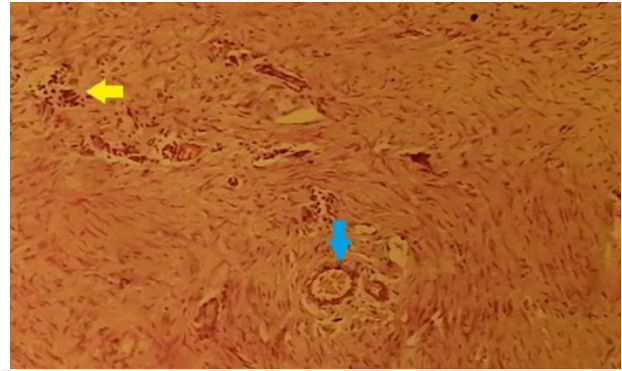


Fig. 16. Synovial treated group/4Week after surgery: Area of neo-tendon shows presence of newly formed blood vessels (blue arrow) and infiltration of few inflammatory cells. H&E, X100

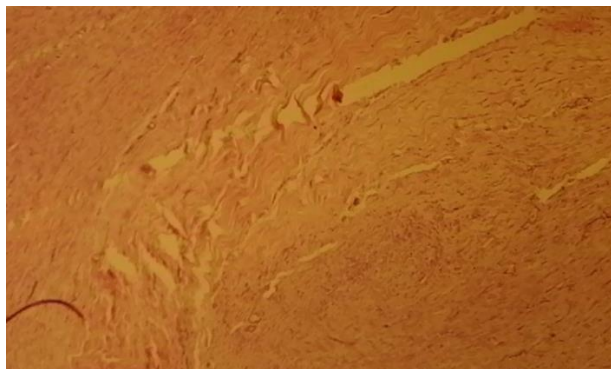


Fig. 14. Synovial treated group/4Week after surgery, area of neo tendon accumulation. Well tendon-to-tendon healing with obliteration of the gap. H&E, X40

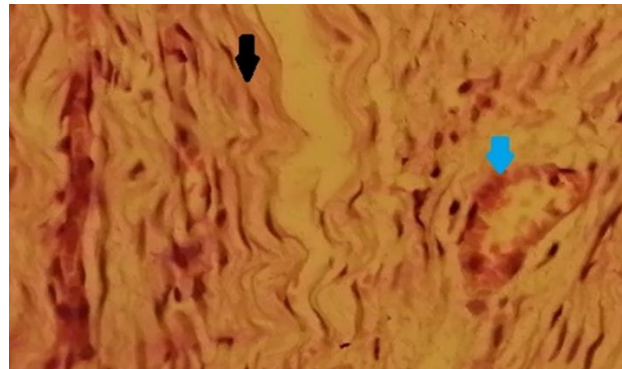


Fig. 17. Synovial treated group/4Week after surgery: Area of neo-tendon show accumulation of well-organized collagen bundle. Well tendon to tendon healing with obliteration of the gap, and newly formed blood vessels (blue arrow) H&E, X400

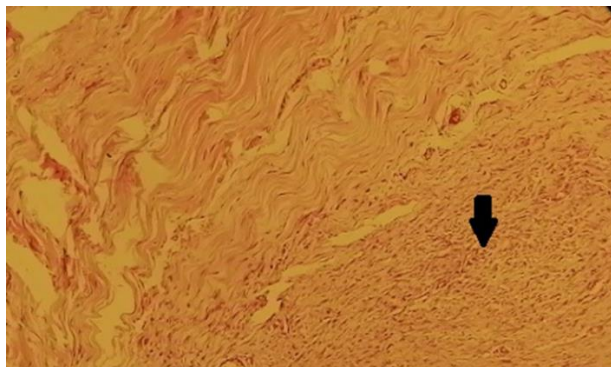


Fig. 15. Synovial treated group/4Week after surgery: area of neo-tendon (yellow arrow); accumulation of well-organized collagen bundle. Well tendon to tendon healing with obliteration of the gap. H&E, X100

accumulation of well-organized collagen bundle, well tendon-to-tendon healing with obliteration of the gap (Figs. 14-17).

DISCUSSION

The complicated recoveries of tendon after surgery are due mainly to low vascularity of the tendon; a process of healing that leads to creating adhesions with adjacent tissues, restricting of mobility; tension and

active forces, which can cause tendon ends diastasis; active and passive tendon movements (Snedeker and Foolen, in 2017)

The goal of research in tissue engineering and regenerative medicine is to design methods of treatments for tendon injury, which are able to maintain its healing for an expectable time (Spinella et al 2010). Therefore, our study was to investigate the efficacy of synovial fluid preparation compared to a commercial polypropylene mesh preparation in enhancement tenotomized tendon healing.

After surgery, the animals showed the same clinical signs, local swelling of the operative site, redness, and lameness. These results may due to hematoma, edema, and flow of inflammatory cells in the cut tendon, as a part of the inflammatory process after tendon injury (Humadi, 2019).

In the clinical monitoring of the polypropylene mesh treated group and the synovial fluid treatment group, the latter showed a significant improvement in the disappearance of swelling, redness, and lameness. Natural synovial fluid is a significant source of hyaluronic acid naturally found in the body (Kurt et al 2018). The

positive effect of hyaluronic acid was because it has anti-inflammatory activity, promotes cell proliferation, and deposition of collagen, in addition to the lubricating action on the sliding surface of the tendon. These results came in agree with Abate, et al 2014 when they used hyaluronic acid in a few of cases in clinical practice. After flexor tendon surgery, they notice greater active movement and improved function, with an early return to work and daily activities. In addition, pain was reduced and function improved in patients with tendon disorders of the elbow, patella, and shoulder when using this hyaluronic acid.

While the clinical results in the propylene network group showed delayed elimination of these clinical signs because of the long time inflammatory process.

These results were disagreed with (Al asadi and Muhammad, 2017), they stated the tenorrhaphy site wrapping with synthetic mesh speeds up the early healing response of tendon. The meshes facilitate the approximation of the ends of the tendon a few days after injury, inflammation in tendon subsides and fibroblasts proliferation, and biosynthesis of extracellular matrix and collagen fibers.

After 2 week postoperatively, tendon histopathological section of polypropylene treated group showed increase infiltration of inflammatory cells, deposition of disorganized collagen, accumulation of fibroblast, and increase angiogenesis, poor tendon-to-tendon healing with obvious gap. While after 4 week of surgery, the area of neo-tendon showed also a hug infiltration of inflammatory cells, increased angiogenesis, and disorganized collagen fibers. The mesh material is considered a foreign body adjacent to the tendon tissue, which leads to a more severe inflammatory reaction and for a longer period. These will leads to more granulation tissue than if healing were without the mesh, and this leads to a delay in the removal of the scar tissue.

The time-consuming healing process takes a long period and usually leads to a reparative scar tissue. Scars provide inferior biomechanical stability. In addition, the poor blood supply and hypo-cellular

property of the tendons are major reasons for their limited healing properties (Humadi, 2019).

In 2009, Gall et al. tested a combination of techniques (Suture and polypropylene Mesh) for the repair of distal canine Achilles' tendon ruptures (In vitro mechanical evaluation). They found a decrease in resistance to gap formation. At that time they could not recommend the Suture + Mesh techniques without further testing.

In the synovial fluid group after fifteen days postoperatively, the histopathological sections of the repaired tendon showed few newly formed blood vessels, accumulation of few well-organized collagen fibers, and well tendon ends healing with no obvious gap. After 4 weeks of surgery, the area of neo-tendon shows the presence of newly formed blood vessels and infiltration of a few inflammatory cells, accumulation of well-organized collagen bundle, and obliteration of the gap. The viscoelastic matrix of hyaluronic acid which exists in synovial fluid can act as a strong biocompatible support material and is therefore commonly used as a growth scaffold in surgery (Tamer, 2013). As we mention before, the positive effect depends on the anti-inflammatory activity of HA, promote cell proliferation, and collagen deposition, besides the lubricating action on the sliding surface of the tendon (Abate, et al. 2014).

CONCLUSION

After digital flexor tendon surgery, autogenous synovial fluid is effective clinically by reducing clinical signs of redness, swelling, and lameness and returns tendon to normal function and histologically by promoting cell proliferation, deposition of well-organized collagen and obliterating gap formation. It was defined as the best option in accelerating tendon injury healing.

The use of polypropylene mesh in flexor tendon surgery gave poor results, as it showed an increase in the period of inflammation and its clinical signs, in addition to histological signs, there was increase in blood vessels, accumulation of non-regulating collagen fibers and the appearance of the gap between the cut ends of tendon.

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