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Original Research Article

Role of Platelet Rich Plasma in Treatment of Chronic Achilles Tendinopathy

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Abstract:

Introduction: It is thought that Achilles tendopathy is related to sports activities, but it is more often found in people who do not involved in sports. The biggest cause is the excessive overburdening of the tendon. At present most of the systematic review and metanalysis shows that the role of PRP in tendinopathy is insignificant.

Aims and Objectives: This study focuses on Leucocyte-rich platelet rich plasma for treatment of chronic achilles tendinopathy in terms of patient reported VAS score, VISA-A Score. This work also measures tendon thickness and intratendinous vascularity by USG & Doppler study.

Methodology and Results: This was a Prospective longitudinal Study/case series. Patients were injected with leucocyte rich platelet rich plasma in Achilles tendon and evaluated on 6, 12, 18 weeks postinjection. In our result VISA-A score shows improvement in initial 6 weeks then slight rise upto 18 weeks with p value of <0.001. VAS score shows initial sudden decline upto 6 week then gradual decline in score with p value of <0.001. Tendon thickness decreased in significant number of patients (80.64% patients) at 18 weeks with p value <0.001. Sudden decrease in tendon vascularity (80.64% of patients) was seen at 12 weeks (p<0.001).

Conclusion: At present ongoing work in form of case series, case reports and some randomized control trials shows benificial role of PRP in Achilles tendinopathy in terms of functional / radiological / histopathological outcome indicating that further research required in field of dose and frequency of PRP administration, standardization of PRP prepration, role of adding other biologics, long term radiological / histological follow up combined with functional outcome.

Keywords: Achilles Tendinopathy, Platelet Rich Plasma, VISA-A Score, VAS Score, Achilles Tendon Thickness and Vascularity.

Introduction

Achilles tendinopathy occurs as a result of overuse injury caused by repetitive energy storage and releases with excessive compression. This may lead to a sudden injury, or in the worst scenario, can lead to a rupture of the Achilles tendon. In either cases, a lack of flexibility or a stiff Achilles tendon can increase the risk of these injuries [1] The present term that is recommended to describe this particular group of patients is 'tendinopathy'. Cook and Purdum [2] proposed a new theory when describing tendon pain termed as Tendon Continuum.

The continuum model proposed a model for staging tendinopathy based on the changes and distribution of disorganization within the tendon. Three stages are as follows:

- Reactive tendinopathy
- Tendon dysrepair
- Degenerative tendinopathy

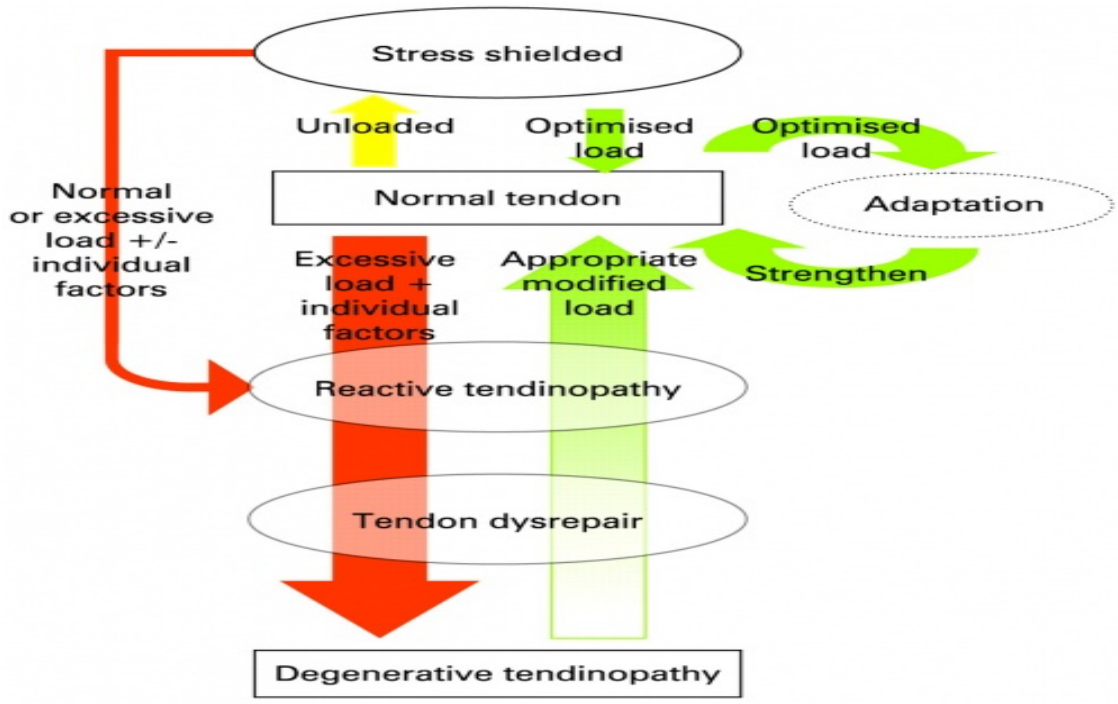

Figure 1:

It has been suggested that the tendon can move up and down this continuum and this can be achieved through adding or removing load to the tendon especially in the early stages of tendinopathy. Achilles tendinopathy can be categorised as an insertional or mid-portion, localisation being the basic difference between the two conditions. When it is at the level of transition between the Achilles tendon and the bone it is insertional form (up to 2cm from the Achilles tendon insertion to the calcaneous). When located at the level of the tendon body (2-6 cm from the insertion to the calcaneous) it is termed as mid-portion form.

Surgical specimens reveal a range of degenerative changes of the involved tendon, such as in the tendon fibre structure and arrangement as well as an increase in glycosaminoglycans, which may explain the swelling of the tendon. [3] The exact cause of tendonitis still remains unclear. Even though tendonitis of the Achilles tendon is often related to sports activities, the ailment is also often found in people who do not involve in sports. The biggest cause is the excessive overburdening of the tendon. A slight degeneration of the Achilles tendon can be latently present, but the pain only appears when the tendon is overburdened. It is also noted that the ailment is usually not preceded by trauma [4][5]

Anatomy

The Achilles tendon is not encased in a true synovial sheath but is surrounded by paratendon composed of a single layer of cells. This tissue is richly vascularized and is responsible for much of the blood supply,[6] which reaches the tendon through a series of transverse vincula that function as passageways for the vessels. The Achilles tendon also receives blood from vessels originating at the musculo-tendinous and osteo-tendinous junctions. Angiographic injection techniques have demonstrated a zone of hypo-vascularity about 2–7 cm proximal to the tendon insertion. Additionally, the number of intra-tendinous vessels and the relative area occupied by these vessels is lowest in tendon portion which is 4 cm from the calcaneal insertion.

Healthy tendons are brilliant white, with a fibroelastic texture. Within the extracellular matrix network, tenoblasts and tenocytes constitute 90–95% of the cellular elements of tendons. The remaining 5–10% consists of fibrochondrocytes, synovial cells of the tendon sheath, endothelial cells and smooth muscle cells. Collagen type 1 accounts for 65–80% and elastin accounts for about 2% of the dry mass of tendons. Tenocytes and tenoblasts lie between the collagen fibres along the long axis of the tendon.[7]

Biomechanics

Tendons transmit force generated by muscle to bone. They act as buffer by absorbing external forces to limit muscle damage, a function that demands mechanical strength, flexibility and elasticity.[8] As collagen fibres deform, they respond linearly to increasing tendon loads.[8] The original configuration is initially lost when the stretch exceeds 2% but is regained if the strain placed on the tendon remains at less than 4%; if strain exceeds 8% macroscopic rupture will

occur.[9,10] The tensile strength of tendons is related to thickness and collagen content, and a tendon with a cross-sectional area of 1 sq cm is capable of supporting 500–1000 kg. Loading of the Achilles tendon reaches up to 9 kN during running (corresponding to 12.5 times the body weight), 2.6 kN during slow walking, and less than 1 kN during cycling.[11]

Aetio-Pathology

Excessive loading of tendons during vigorous physical training is regarded as the main pathological stimulus for degeneration. Tendons respond to repetitive overload beyond physiological threshold by inflammation of their sheath, degeneration of their body, or a combination of the two. [12] The repair mechanism is probably mediated by resident tenocytes that continually monitor the extracellular matrix. Failure to adapt to recurrent excessive loads results in the release of cytokines, leading to further modulation of cell activity.[13] Tendon damage may even result from stresses within physiological limits, since frequent microtrauma may not allow enough time for repair. Microtrauma can also result from non-uniform stress within tendons, producing abnormal load concentrations and frictional forces between the fibrils, with localized fibre damage.

Achilles tendinopathy is also more common in people who have ankylosing spondylitis or psoriatic arthritis. It is also thought that genetic 'makeup' may play a part for some people. It is also more common in people who have high blood pressure, high cholesterol or diabetes.

People who are taking fluoroquinolones also have an increased risk of developing Achilles tendinopathy. Ciprofloxacin enhances interleukin-1β mediated release of matrix metalloproteinase (MMP3), inhibits tenocyte proliferation and reduces collagen and matrix synthesis.[14]

Other contributors to tendinopathy are free radical damage on reperfusion after ischaemia, hypoxia, hyperthermia and impaired tenocyte apoptosis.[15] Changes in the expression of genes regulating cell–cell and cell–matrix interactions have been reported, with down-regulation of MMP3 mRNA in tendinopathic Achilles tendon samples.[16] Type I and type III collagen mRNAs have been found at higher levels in tendinopathic samples than in normal samples.[16] In tendinopathic Achilles tendons, upregulation of MMP2 and vascular endothelial growth factor has been described, whilst MMP3 was down regulated.[17] Imbalance in MMP activity in response to repeated injury or mechanical strain may result in tendon degeneration. In addition, recent research showed an older age, higher android fat mass ratio, and waist circumference > 83cm, in men are associated with a higher chance of having Achilles Tendinopathy. The presence of COL5A1 gene variant was also found to be a possible risk factor. This gene is normally responsible for the production of tendon protein, but patients with the condition were shown to have significantly different allele frequencies of the COL5A1 BstUI RFLP compared with normal subjects. Therefore besides overuse and degeneration, Achilles Tendinopathy was proposed to have a strong metabolic influence due to poor anatomical vascularity, association with body fat and the genetic factors.

Histopathology

Histologically, tendinopathy is characterized by an absence of inflammatory cells and a poor healing response. It shows non-inflammatory intratendinous collagen degeneration, fibre disorientation and thinning, hypercellularity, scattered vascular ingrowth, and increased interfibrillar glycosaminoglycans.[8,13] Frank inflammatory lesions and granulation tissue, when they occur, are associated mainly with tendon ruptures. [18] Various types of degeneration may be seen in tendons, but in the Achilles tendon the usual types are mucoid and lipoid. [18] In mucoid degeneration (i.e. proteoglycan / glycosaminoglycan accumulation in the tendon) light microscopy reveals large mucoid patches and vacuoles between fibres. Lipoid degeneration is characterized by abnormal intratendinous accumulation of lipid, with disruption of collagen fibre structure.[8]

Paratendinopathy may occur alone or in combination with degeneration of the tendon body.[19] Histologically, mucoid degeneration, fibrosis and vascular proliferation with a slight inflammatory infiltrate have been reported.[20,21] Clinically, oedema and hyperaemia of the paratenon are seen. A fibrinous exudate accumulates within the tendon sheath.[19]

Clinical Presentation

The hallmark symptom of Achilles tendinopathy is pain. In majority of cases it occurs at the beginning and end of a training session, with a period of diminished discomfort in between. As the pathological process progresses, pain may occur during exercise, and, in severe cases, it can interfere with activities of daily living. In the acute phase, the tendon is diffusely swollen and oedematous, and on palpation tenderness is usually greatest 2–6 cm proximal to the tendon insertion. The tendon can appear to have subtle changes in outline, becoming thicker in the anterior-posterior and medial-lateral planes. The affected part of the tendon shows higher stiffness in comparison to the non-affected side. Sometimes, fibrin precipitated from the fibrinogen-rich fluid around the tendon can result in palpable crepitation. In chronic cases,

exercise-induced pain is still the cardinal symptom, while crepitations and effusions diminish. A tender, nodular swelling is usually present in chronic cases and is believed to signify tendinosis.

Imaging Methods

Ultrasonography- In acute cases, ultrasound reveals fluid accumulation around the tendon. In chronic cases, peritendinous adhesions may be shown by thickening of the hypoechoic paratenon with poorly defined borders. Hypoechoic areas within tendon can be nodular, diffuse or multifocal, and correlate well with macroscopic findings at surgery. However, it offers limited soft-tissue contrast and is less sensitive than MRI.[22]

Magnetic Resonance Imaging (MRI) provides extensive internal morphology of tendon and the surrounding structures. It is useful to evaluate various stages of chronic degeneration and for differentiation between peritendinitis and tendinosis. An excellent correlation has been reported between MRI and pathological findings at surgery.[23]

Colour Doppler demonstrates intra-tendinous vascularity in Achilles tendon. Abnormal abundance of vessels can be found on ventral aspect of the tendon.

Management

In the early phase of Achilles tendinopathy, conservative treatments are customary.24-26] Surgical management is recommended for patients who do not adequately respond to a conservative treatment programme over three to six months.[25,26,27]

The initial conservative programme is directed towards presumed etiological factors or towards relieving symptoms. The strategies include abstention from the activities that caused the symptoms and correction of training errors, foot malalignments and muscle weakness. Decreasing the intensity, frequency and duration of the activity that caused the injury, or modification of that activity, may be the only action necessary to control symptoms in the acute phase. Since collagen repair and remodelling is stimulated by tendon loading, complete rest of an injured tendon can be detrimental. Modified rest, reducing activity at the injured site but allowing normal activity elsewhere, has been recommended.

Cryotherapy has been regarded as a useful intervention in the acute phase. It has an analgesic effect, reduces the metabolic rate of the tendon and decreases extravasation from the new capillaries found in tendon injuries.[8]

Therapeutic ultrasound may reduce the swelling in the acute inflammatory phase and improve tendon healing.[28] Ultrasound stimulates collagen synthesis in tendon fibroblasts and stimulates cell division during periods of rapid cell proliferation.[29]

Deep friction massage has been advocated for tendinopathy and Paratendinopathy. In chronic cases, this should be accompanied by stretching to restore tissue elasticity and reduce the strain in the muscle–tendon unit with joint motion.

Augmented soft tissue mobilization is a new non-invasive technique that has been used with success in chronic tendinopathy, probably through controlled infliction of microtrauma with resultant fibroblast proliferation.[30]

Stretching and strengthening of the triceps surae muscle and Achilles tendon are important to preserve function of the musculotendinous unit, restore normal ankle joint mobility and decrease the strain on the Achilles tendon with normal motion. Eccentric muscle training is superior to concentric training in decreasing pain in chronic Achilles tendinopathy, and promising results have been obtained with an intensive heavy-load training regimen. [31]

If foot alignment is abnormal, orthosis that place the hindfoot in neutral may prove beneficial. A heel lift of 12–15 mm is typically used as an adjunct to management. [26] Orthotics correction can alter the biomechanics of the foot and ankle and relieve heel pain. In runners orthotics have been used with up to 75% success.[32]

Peritendinous injections of corticosteroids are controversial and there are no good scientific reasons to support their use.[35]

In 24–45.5% of patients with Achilles tendinopathy, conservative management is unsuccessful and surgery has to be considered. The objective is to excise fibrotic adhesions, remove degenerate nodules and make longitudinal incisions in the tendon so as to detect intratendinous lesions and restore vascularity (and possibly stimulate the remaining viable cells to initiate cell matrix response and healing). Multiple longitudinal tenotomies have been shown to trigger neoangiogenesis at the Achilles tendon, with increased blood flow.[36] This should improve nutrition and provide a more favourable environment for healing. Patients are encouraged to weight-bear as soon as possible after surgery.

Platelet Rich Plasma

Recently platelet rich plasma has been proposed as a useful modality in treatment of chronic achilles tendinopathy. Platelet rich plasma is autologous blood derived product principally composed of a high concentration of platelets having growth factors to accelerate healing. It is hypothesized that improvement come from growth factors present in

platelets such as- platelet derived growth factor (PDGF); transforming growth factor (TGF)-β; insulin-like growth factor (IGF); epidermal growth factor (EGF); vascular endothelial growth factor (VEGF) and fibroblast growth factor (FGF). These factors are released from the alpha granules after injury and bind to receptors on target cells like mesenchymal stem cells, osteoblasts, fibroblasts, endothelial cells, and epidermal cells.

These receptors activate an intracellular signal protein that causes the expression of a gene sequence that then directs cellular proliferation, matrix formation, osteoid production or collagen synthesis dependent on the cell activated. Specifically with regards to tendon damage / healing in Achilles tendinopathy platelet rich plasma injection would increase collagen production and cell viability and stimulate angiogenesis. Bioactive factors like serotonin, histamine, dopamine, calcium and adenosine are stored in the dense granules in platelet also promote healing. The platelets in platelet rich plasma are delivered in a clot, which contains several cell adhesion molecules including fibronectin, fibrin and vitronectin which promote cell migration. The clot itself promotes wound healing by acting as conductive matrix or scaffold upon which cells can adhere and initiate the wound healing process. In addition, platelet rich plasma has high antimicrobial potency and this property may prevent infections.

Aims and Objectives

The aim of our study was to assess the role of Leucocyte-rich platelet rich plasma in the treatment of chronic achilles tendinopathy in terms of :

Self-reported tendon pain during activity (VAS score).

Function and symptoms (VISA-A Score)

Tendon thickness and intratendinous vascularity (USG & doppler)

Material and Methods

This prospective study was conducted in the department of Orthopaedics, GSVM Medical College & LLR Hospital, Kanpur.

Participants: We included patients admitted through orthopedic OPD after proper written informed concent from participants and ethical clearance from ethics committee.

Study Design: Prospective longitudinal Study/case series.

Study Duration: 2 year, January 2020 to October 2021.

Eligible Participants: Patients having pain localized over back of ankle region about 2-6 cm from insertion of Achilles tendon.

Inclusion criteria

1. Clinical (persistent tenderness on palpation or distension in mid portion of the Achilles tendon) and Radiological (tendon thickness on USG) features of achilles tendinopathy (approx. 2-7 cm proximal to insertion on the calcaneus)
2. Symptom duration of more than 6 weeks

Exclusion criteria:

1. Age less than 18 years
2. Clinical suspicion and USG indications of other musculocutaneous injuries
3. Previous injection of steroids or any kind of blood products
4. Treatment with fluoroquinolones (known to cause tendinopathy) during last 6 months
5. Systemic disease such as diabetes, generalized inflammatory arthritis, including ankylosing spondylitis, rheumatoid arthritis or psoriatic arthritis, prior Achilles tendon tear, pregnancy, severe infection, known malignancy, bleeding disorder, nerve-related symptoms such as radiculopathy or osteoarthritis of the ankle, previous extracorporal shock wave therapy or injections or eccentric training during the 4 weeks prior to the study.

Allocation of Treatment: After detailed history (including drug and previous treatment) and clinical examination all required routine blood investigations were done (CBC/ ESR/ CRP Blood sugar Fasting & Post prandial, S. Creatinine, S. Uric acid, SGOT/ SGPT/ S. Alkaline phosphatase, Quantitative RA factor). X-ray bilateral ankle AP and Lateral views, Color Doppler USG done to confirm the diagnosis and to rule out other pathologies. In all eligible participants 3 ml autologous platelet rich plasma was injected.

Preparation of Platelet Rich Plasma:

The procedure consists of 40 ml venous blood sample drawn under aseptic conditions in a 50 ml disposable syringe with 4 ml CPD-A1 or 4 ml of 3.2% sodium citrate as anticoagulant. Then 2 centrifugations (the first at 1600 rpm for 6 minutes to separate erythrocytes and a second at 3400 rpm for 15 minutes to concentrate platelets) produced a unit (i.e. 4ml) of PRP. The unit of PRP was divided into 2 small units of 1ml and 3 ml. The sample of 1 ml was sent to the laboratory for analysis of platelet concentration and microbiological assessment. This could be considered as an advantage since it increases the safety of the procedure, the remaining 3 ml PRP was infiltrated in the symptomatic Achilles tendon region. The adequacy of the PRP to be used

for injection was based on an increase in the platelet concentration (per ml of PRP) to be 3-5 times that of the basal concentration of platelets in the whole blood.

Local Infiltration Technique: Under sterile technique 3 ml platelet rich plasma was injected using a "peppering" technique in a clock wise manner to better cover the affected area of Achilles tendon. The patient was then observed for 15 to 20 minutes and then discharged with instructions to use cold therapy / ice fomentation over the affected area and limb elevation to relieve pain. Walking was allowed but sports activities were to be avoided for 4 weeks**.** NSAIDs and other antiplatelet drugs were not allowed for 6 weeks. Tramadol orally taken on SOS basis.

Physiotherapy: After 2 weeks stretching exercises were started. These exercises were performed twice a day, every day, with precaution to stop at first sign of pain for next 3 months. In addition isotonic exercises were also performed once daily. After exercises ice fomentation done for 10-15 minutes.

Follow Up: All the patients were followed up weekly for first 6 weeks then 2 weekly for next 6 weeks and then at 18 weeks. Pain was assessed by Visual Analogue Scale (VAS) score and functional impairment by Victorian Institute of Sport activity - Achilles (VISA-A) score. In VAS score a horizontal line, 10 cm in length written with word description at each end is used to assess pain. VISA-A score contains questionnaires covering three necessary domains: Pain, Functional Status and activity thus assessing functional impairment. Colour Doppler was used to assess tendon thickness & vascularity. Final outcome was measured based on the pain reduction from the pre injection levels. Patients were also observed for any post injection complications.

Statistical Analysis: In this study all continuous data is expressed as a mean +/- standard deviation or mean and 95% confidence intervals, categorical variables are expressed as frequencies and percentages. Statistical test applied was student t test (Paired and un-paired) for assessment of results & data analysis was done using the available software (INSTAT or SPSS – 16 version). The p - value $\leq$ 0.05 was considered to be statistically significant.

Results and Statistics

In our study total 62 patients meeting inclusion / exclusion criterion of chronic achilles tendinopathy were taken in which all were selected for leukocyte rich platelet rich plasma injection therapy. The minimum follow up in our study was 6 weeks and maximum follow up period was 18 weeks.

Out of 62 patients 32 patients are Male (51.7%) and 30 are females (48.2%) indicating that incidence is slightly higher in male population with highest numbers of patients were in their 4th (50%), and 5th (29.03%) decade of life. Also the disease is more common on right side (40.32 %) than left (30.64%) and about 29.03% have bilateral involvement. The selected patients in this study had mean duration of symptoms of 3.87 months.

In our study mean age of presentation of patient is 39 years. Mean age of presentation in females is 38.31 year and 40.74 year in males. Minimum age of patient is 27 year and maximum age is 58 year. The complication was encountered in 2 patients. One patient presented with depigmentation of local skin around injection site and one presented suppuration at injection site. No other complication was observed.

The average Visual Analogue Scale (VAS) and Victorian Institute of Sport Assessment – Achilles questionnaire(VISA-A) scores for Pre Injection and 6, 12 and 18 weeks post injection were as shown in the below table:

Table 1:

	Pre injection score		**Post injection score at 6 weeks**		**Post injection score at 12 weeks**		**Post injection score at 18 weeks**	
	VAS	**VISA-A**	**VAS**	**VISA-A**	**VAS**	**VISA-A**	**VAS**	**VISA-A**
MEAN	7.59	37.70	2.41	76.93	0.83	80.83	0.75	82.25
S.D.	0.85	7.84	0.77	5.13	0.87	3.62	0.82	1.5

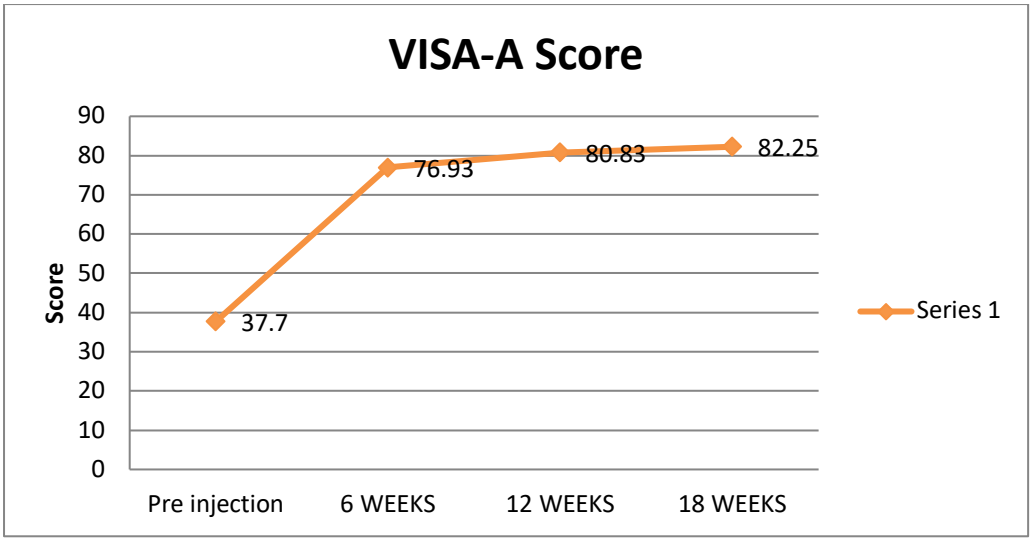

Figure 2: Difference In Visa-A Score With Time

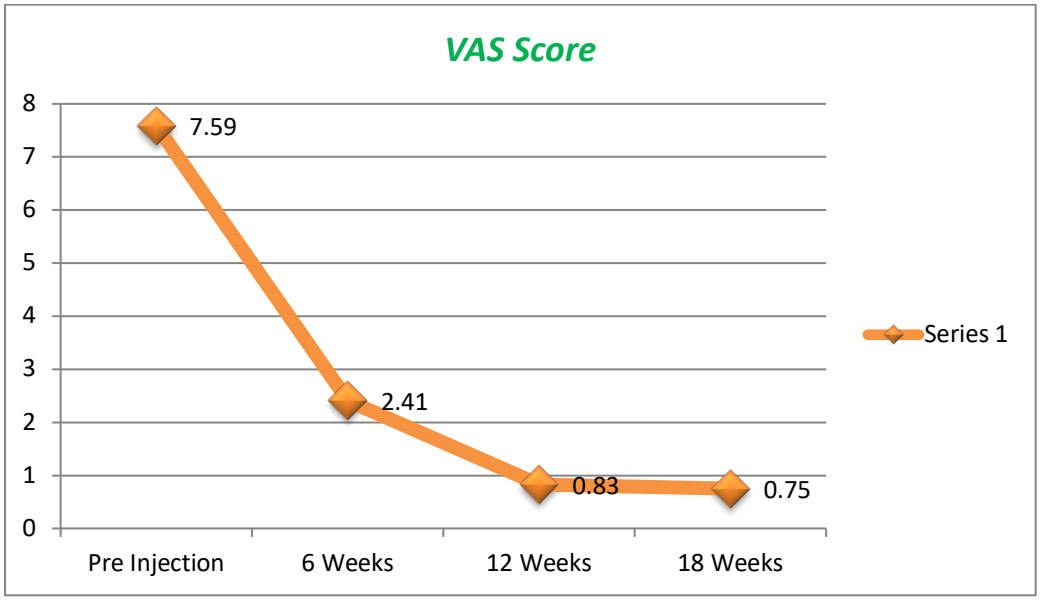

Figure 3: Difference in Vas Score with Time

Table 2: VISA-A Score Comparison

	PRP					
Time	**NO.**	**MEAN**	**S.D.**	**t value**	**p value**	**Inference**
6 Weeks	62	76.93	5.13	148.21	<0.001	Highly Significant
12 Weeks	62	80.83	3.62	69.57	<0.001	Highly Significant
18 Weeks	62	82.25	1.5	93.59	<0.001	Highly Significant

Table 3: VAS Score Comparison

	PRP GROUP					
Time	**NO.**	**MEAN**	**S.D.**	**t**	**p**	**Inference**
6 Weeks	62	2.41	0.77	34.93	<0.001	Highly Significant
12 Weeks	62	0.83	0.87	47.26	<0.001	Highly Significant
18 Weeks	62	0.75	0.82	51.37	<0.001	Highly Significant

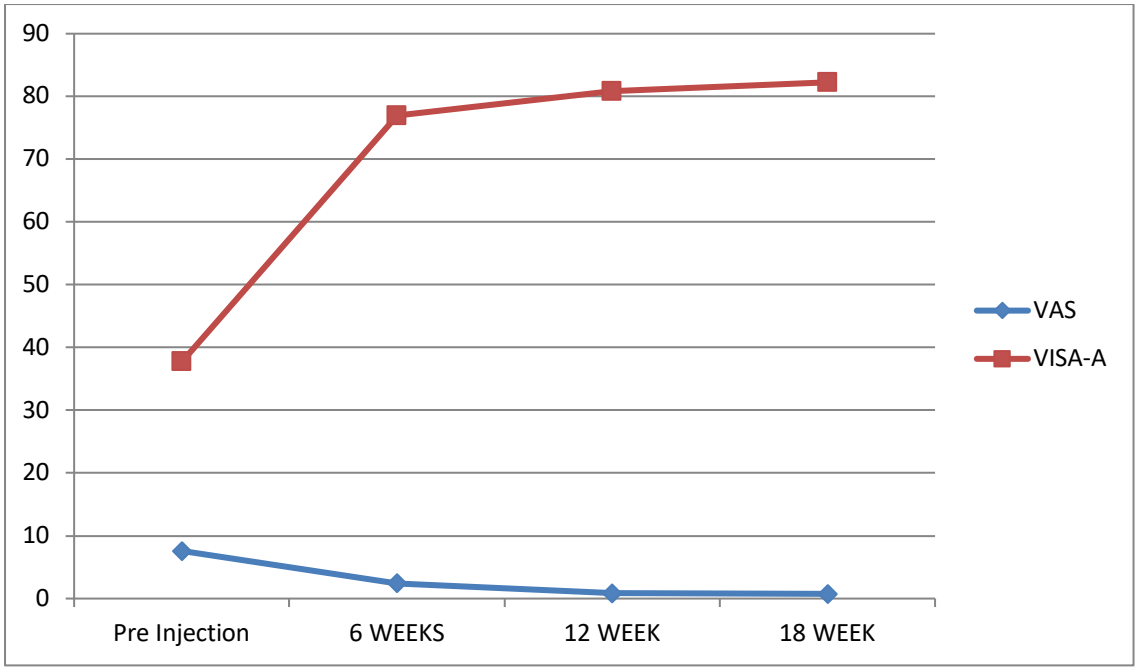

Figure 4: Comparison of VAS & VISA-A Score with time

In our result VISA-A score shows gradual improvement from pre injection score of 37.7 to 76.93 at 6 weeks, 80.83 at 12 week, 82.25 at 18 week. Sudden improvement of score is seen in initial weeks upto 6 week then slight rise upto 18 weeks with p value of < 0.001, which is highly significant. In our study VAS score shows gradual improvement from pre injection score of 7.59 to 2.41 at 6 week, 0.83 at 12 week, 0.75 at 18 week. Decline of score is seen with initial sudden decline upto 6 week then gradual decline in score is with p value of <0.001 which is highly significant.

Table 4: Difference in Tendon thickness with time (Total Patients: 62)

	Pre Injection		6 weeks		12 weeks		18 weeks	
Status	**Normal**	**Increased**	**No Change**	**Decreased**	**No Change**	**Decreased**	**No Change**	**Decreased**
Number of Patients	11	51	62	0	54	8	13	49
%	21.15	82.22	100	0	87.09	12.9	20.96	79.03
X^2	-		12.07		0.55		46.63	
P Value	-		<0.05		>0.05		<0.001	
Inference	-		Significant		Non-Significant		Highly significant	

Table 5: Difference in Tendon Vascularity with Time (Total Patients: 62)

	Pre Injection		6 weeks		12 weeks		18 weeks	
Status	**Nor-mal**	**In-creased**	**No Change**	**Decreased**	**No Change**	**De-creased**	**No Change**	**De-creased**
Number of Pa-tients	11	51	62	0	12	50	12	50
%	21.15	82.22	100	0	19.35	80.64	19.35	80.64
X^2	-		12.07		49.08		49.08	
P Value	-		<0.05		<0.001		<0.001	
Inference	-		Significant		Highly significant		Highly significant	

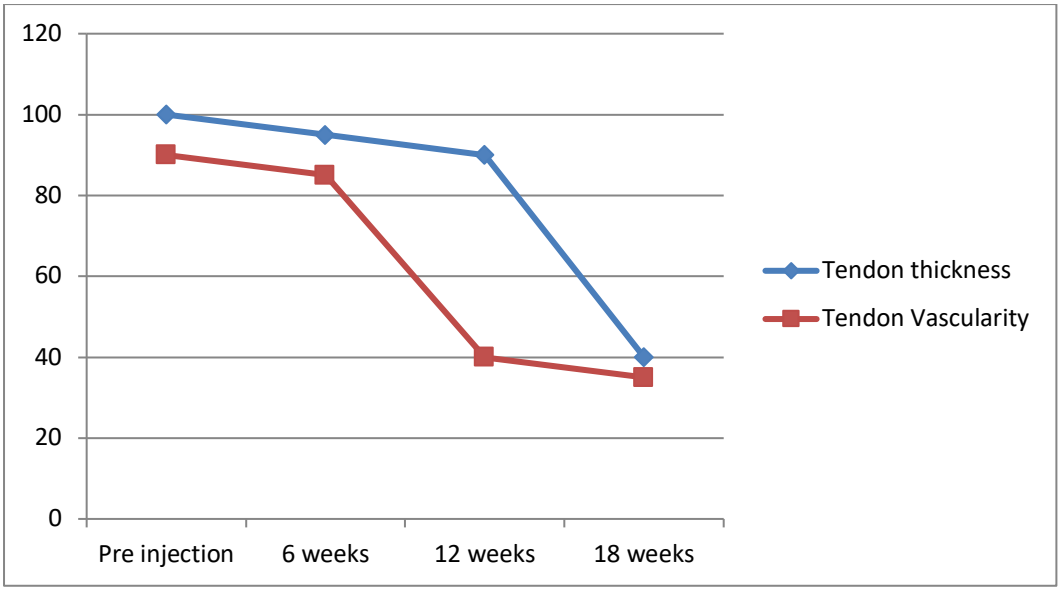

Figure 5:

In our study tendon thickness was increased in 82.22% of patients in pre-injection phase. There was no change in thickness at 6 weeks post injection as compared to pre-injection values (p value <0.05). Little or no change was observed at 12 weeks. There was decreased thickness in significant number of patients (80.64% patients) at 18 weeks post injection in comparison to pre-injection status with p value<0.001, which is highly significant. In our study tendon vascularity was increased (82.22% of patients) during the pre-injection phase. It shows no change in vascularity at 6 weeks as compared to pre-injection phase (p<0.05). There was sudden decrease in vascularity (80.64% of patients) at 12 weeks (p<0.001) followed by gradual or no change in vascularity at 18 weeks in comparison to pre-injection scores with p value<0.001 which is highly significant.

Discussion

Various modalities of treatment have been recommended for Chronic Achilles Tendinopathy. They are rest [26,27,28], activity modification, non-steroidal-anti-inflammatory drugs, counterforce braces, laser therapy, deep Friction massage, physiotherapy with stretching of triceps surae and Achilles tendon[31], extracorporeal shockwave treatment, acupuncture, ultrasound treatment[28,29] and Augmented soft tissue mobilisation[30], orthoses [32] drugs like low dose heparin and aprotinin [33, 34]. Injection of corticosteroids is controversial for treatment in Achilles tendinopathy [35] Different types of open and arthroscopic operative surgery [36] are also advised for Chronic Achilles Tendinopathy. At present, platelet rich plasma (PRP) is considered as an ideal biological autologous blood derived component beneficial for the treatment of Chronic Achilles Tendinopathy. Platelet rich plasma has been utilised and studied since 1970. It can be injected in different tissues. Our results add to the growing evidence that Platelet rich plasma injections can give long term improvement in symptoms to patients with previously intractable Achilles tendinopathy.

There are ample number of studies regarding the benefits of platelet rich plasma injection over corticosteroid injection therapy for chronic Achilles tendinopathy as seen in study conducted by Boesen AP et al(2017)[43]. The main outcome parameters considered were pain, functional activities and thickness & vascularity of Achilles tendon. Currently long term follow up data regarding the effectiveness of platelet rich plasma are lacking. Our study shows eighteen weeks (18 weeks) follow up results using VAS score, VISA-A Score, tendon thickness & tendon vascularity as parameters. In most of the studies visual analogue scores (VAS) is used as a primary outcome measure to determine pain. To evaluate function and disability Victorian Institute of Sport Assessment-Achilles Questionnaire (VISA-A) is used. Other scores available are Foot and Ankle Ability Measure (FAAM), Foot and Ankle Ability Measure-Sports (FAAMS) and Short Form health survey (SF-8) [40] as used in study conducted by Richard F Owens Jr et al (2011).

In the study by De Jonge s et al 2011, compared the effectiveness of autologous platelet rich plasma injection to Saline (Placebo) therapy in chronic Achilles tendinopathy 54 patients aged 18 to 70 were randomised and it is proved that platelet rich plasma injection is safe and beneficial with improvement in VISA-A scores (p value<0.2). Concerning functional impairment, the PRP group showed better results during the initial period. Whereas in saline group symptoms improved

progressively. There was a significant difference in decrease of pain and functional impairment after platelet rich plasma application even after one year. In present study number of patients are 62 which is similar to sample taken by De Vos RJ et al (2010), RJ Kampa DA Connel (2010). Mean age of presentation of patient is 39 years which is similar to other studies which were conducted in the recent past. Out of 62 patients 32 patients are Males (51.7%) and 30 are females (48.2%) indicating that incidence is slightly higher in males as compared to females and with highest numbers of patients were in their 4th (50%), and 5th (29.03%) decade of life.

In our study patients were symptomatic for last 6 weeks to 7 months with mean duration of symptoms of 3.87 months, it is similar to study population selected by Filardo G et al (2014). In our study population disease is more common on right side (40.32 %) than left (30.64%) and about 29.03% have bilateral involvement.

Follow up duration in our study is from 6 weeks upto 18 weeks and so forth which is similar to study duration of studies conducted by De Vos RJ et al (2010), RJ Kampa DA Connel (2010), Liu CJ et al (2019), Von wehren et al (2019), arco c Van Der Vlist et al(2021). During the follow up negligible dropouts was observed and majority of patients were satisfied with the treatment being administered to them. In this study out of 62 patients post procedure complication is negligible except for one patient presented with depigmentation of skin at injection site and one presented with suppuration at injection site.

In this study the VAS score has declined from the average pre injection score of 7.59 to 2.41 at 6 weeks, 0.83 at 12 weeks, 0.75 at 18 weeks which is almost similar to the study by Filardo G et al 2014[41] where the pre-injection VAS score shows a similar trend on follow up. In this study the VISA-A score has Improved from the average pre injection score of 37.7 to 76.93 at 6 weeks, 80.83 at 12 week, 82.25 at 18 weeks which is almost similar to the study by Filardo G et al (2014) [41], where the pre injection VISA-A score is 49.9+/-18.1 which increased to 62.9+/-19.8 at 8 weeks, 84.3+/-17.7 at 24 weeks and 90.0+/-13.9 after 4 years. Similar results were also seen in studies conducted by De Vos RJ et al (2010) [37], De Jonge et al(2011) [39], Guelfi M et al(2015) [42], Boesen AP et al (2017) [44], Liu CJ et al(2019) [45], Von Wehren et al(2019) [46].

In our study tendon thickness and vascularity which was increased in the pre injection phase has also improved. Tendon thickness shows no change at 6 weeks, little or no change at 12 weeks and decreased thickness (80.64 % of patients) at 18 weeks post injection. Tendon Vascularity which was increased (82.22 % of patients) during the pre-injection phase shows no change in vascularity at 6 weeks, sudden decrease in vascularity (80.64 % of patients) at 12 weeks followed by gradual or no change in vascularity at 18 weeks which is similar to study published by De Jonge S et al(2011)[39], Boesen AP et al(2017)[43].

A significant finding in this study is that the VAS scores have declined following administration of leucocyte rich platelet rich plasma during the course of treatment which is statistically significant (p value<0.001) . VISA-A scores has also improved after which is statistically significant (p value <0.001). Tendon thickness also shows improvement (p<0.001) and tendon vascularity also improved (p <0.001) following treatment with leucocyte rich platelet rich plasma.

This strengthens our conclusion that the platelet rich plasma injection is beneficial for chronic Achilles tendinopathy.

Table 6: Comparison with Other Studies

Author	Study Type	Sample size	Outcome variables	Follow up	Results
De Vos RJ et al (2010)[37]	RCT	27 cases vs 27 controls (saline group)	USG Evaluation	6,12,24 weeks	PRP in the treatment of chronic midportion Achilles tendinopathy does not contribute to increased tendon structure or alter the degree of neovascularisation, compared with saline group
R J Kampa et al(2010)[38]	RCT	27 cases vs 27 controls (saline group)	VISA-A , Patient satisfaction	6,12,24 weeks	VISA-A scores improved in PRP group after 24 weeks but no significant difference in patient satisfaction in comparison to saline group
De Jonge S et al(2011)[39]	RCT	27 cases vs 27 controls (saline group)	VISA-A, Patient satisfaction, USG	6,12,24 weeks & 1 Year	No clinical or ultrasonographic benefit of PRP over placebo over 1 year period

			evaluation		
Richard Owens et al(2011)[40]	Retrospective	10 cases	FAAM, SF-8,MRI	2 years	Patients receiving PRP injection demonstrated modest improvement in functional outcome measures
Filardo G et al(2014)[41]	Prospective study	27 cases	VAS Score, VISA-A	2,6 months 5 years	VISA-A score improved by 2 moths with further improvement at 6 months & stable result at 5 year. VAS score follows same trend
Guelfi M et al(2015)[42]	Prospective study	83 cases	VISA-A Score	3 weeks, 3, 6 months	Improvement in VISA-A scores seen at 3 months
Krogh TP et al(2016)[43]	Prospective study	24 cases	VISA-A, Patient satisfaction, USG evaluation	3,6 12 weeks	No Improvement in VISA-A scores, subjective symptoms(pain at rest, pain while walking) at 3 months; large dropout observed
Boesen AP et al(2017)[44]	Prospective study	27 cases vs 27 controls (High volume injections HVI)	VAS,VISA-A & USG Evaluation	6,12,24 weeks	VAS VISA-A scores improved in both groups. PRP or HVI in conjunction with with eccentric excercises improves patient satisfaction and reduces tendon thickness and vascularity.
Present study	Prospective study	62 cases	VAS, VISA-A & USG Evaluation	6,12,24 weeks	VAS & VISA-A scores improved over course of time at 6 weeks. Tendon thickness and vascularity shows no change during first 6 weeks. At 12 weeks tendon vascularity is decreased but tendon thickness shows remains at pre-injection phase. At 18 weeks tendon thickness shows a decrease as compared to baseline pre-injection values.

RCTs = Randomized control trials, PRP = Platelet-rich plasma USG: Ultrasonography MRI: Magnetic Resonance Imaging; VAS: Visual analogue scale; VISA-A: Victorian institute of sports assessment- achilles questionnaire; FAAM = Foot and ankle ability measure,

Recent Studies

Maffulli, et al [47] says that tendon repair in tendinopathy may remain altered for a long while and possibly forever, indicating a possible dissociation between morphology and symptoms. The predictive value of asymptomatic abnormal findings remains limited, and interventions in such instances are inappropriate according to current literature. Multiple risk factors and heterogeneity in population further makes difficult to draw a conclusion regarding a particular treatment modality.

In a systematic review study done by Zabrzyński, J [48] and others, Bonar score was applied for the histopathological assessment of therapeutic advances in tendinopathy, with special reference to PRP therapy. They included 21 animal studies and 1 human study (n=45). In these studies 6 studies used PRP as therapeutic intervention. They concluded that to understand the tendinopathy a link between histopathology and clinical outcome is required and they found superior tendinus tissue healing related to improved clinical results

Desouza, [49] and co-workers done a meta-analysis. Two studies in this meta-analysis included VAS scores and tendon thickness. In it 6 weeks after treatment, PRP exhibited better efficacy than the placebo treatment. There was no significant difference in VAS scores at 6 weeks and 24 weeks after treatment. However, VAS scores at 12 weeks and tendon thickness were significantly different. This study concludes that PRP injection is an effective treatment for chronic Achilles tendinopathy and has a unique potential for increasing function and reducing discomfort in these patients.

In study done by Arthur et al [50] the PRP treatment group had a slightly higher VISA–A score than the placebo group at 6 weeks and 24 weeks. At 12 weeks of treatment, the PRP injection group showed a substantial VAS improvement compared to the control group and the difference was statistically significant. Study conclusion was

that augmenting the frequency of PRP injections may boost the outcomes, and additionally, more rigorous designs and standardised clinical randomised controlled trials are needed to produce more reliable and accurate results.

Conclusion

In conclusion, our study on treatment of Chronic Achilles Tendinopathy with leucocyte rich platelet rich plasma shows that a single injection of autologous leucocyte rich platelet rich plasma improves ankle pain and functional activities more effectively. These improvements were maintained in our follow up period done at 6,12,18 weeks without any significant complications.

Leucocyte rich platelet rich plasma gives beneficial results to patients in terms of:

- Improvement in pain as evident by sudden decline of VAS score in initial 6 weeks then gradual decline in score upto 18 weeks.
- Improvement in functional status as indicated by sudden improvement in VISA-A score in initial 6 weeks then slight rise in score upto 18 weeks.
- Tendon thickness had no change in thickness at 6 weeks, little or no change at 12 weeks but decreased thickness in significant number of patients at 18 weeks post injection as assessed by colour Doppler studies.
- Tendon vascularity as assessed by colour Doppler studies had no change in vascularity at 6 weeks, sudden decrease at 12 weeks followed by gradual decrease or no change at 18 weeks follow up.

Most of the systematic review and metanalysis shows that the role of PRP in tendinopathy is insignificant and they are no more beneficial than a placebo. At present ongoing work in form of case series, case reports and some RCT shows benificial role of PRP in Achilles tendinopathy in terms of functional / radiological /histopathological outcome.

This indicates that further research required in field of dose and frequency of PRP administration ,standardization of PRP prepration , if addition of other biologics like stem cell is beneficial or not, standardized physiotherapy protocol, more rigorous outcome measurement, long term radiological/ histological follow up combined with functional outcome.

Bibliography

1. 1: Clain MR, Baxter DE. Achilles tendinitis. Foot & ankle. 1992 Oct; 13(8):482-7.
2. 2 Cook JL, Purdam CR. Is tendon pathology a continuum? A pathology model to explain the clinical presentation of load-induced tendinopathy. British journal of sports medicine. 2009 Jun 1; 43(6):409-16.
3. 3: Schubert TE, Weidler C, Lerch K, Hofstädter F, Straub RH. Achilles tendinosis is associated with sprouting of substance P positive nerve fibres. Annals of the rheumatic diseases. 2005 Jul 1; 64(7):1083-6.
4. 4: Wilson JJ, Best TM. Common overuse tendon problems: a review and recommendations for treatment. American family physician. 2005 Sep 1; 72(5):811-8.
5. Paavola M, Kannus P, Järvinen TA, Khan K, Józsa L, Järvinen M. Achilles tendinopathy. JBJS. 2002 Nov 1;84(11):2062-76
6. Carr AJ, Norris SH. The blood supply of the calcaneal tendon. Bone Joint Surg 1989;71: 100–1 [PubMed] [Google Scholar
7. Kirkendall DT, Garrett WE. Function and biomechanics of tendons. Scand J Med Sports 1997;7: 62–6
8. Jozsa LKP. Human Tendon: Anatomy, Physiology and Pathology. Champaign: Human Kinetics, 1997
9. Kvist M. Achilles tendon injuries in athletes. Ann Chir Gynaecol 1991;80: 188–201]
10. Leppilahti JOS, Karpakka J, et al. Overuse injuries of the Achilles tendon. Ann Chir Gynaecol 1991;80: 202–7
11. Komi PV, Fukashiro S, Jarvinen M. Biomechanical loading of Achilles tendon during normal locomotion. Clin Sports Med 1992;11: 521–31
12. Benazzo FMN. An operative approach to Achilles tendinopathy. Sports Med Arthrosc Rev 2000;8: 96–1001.
13. Leadbetter WB. Cell-matrix response in tendon injury. Clin Sports Med 1992;11: 533–78.
14. Corps AN, Curry VA, Harrall RI, Dutt D, Hazleman BL, Riley GP. Ciprofloxacin reduces the stimulation of prostaglandin E (2) output by interleukin-1beta in human tendon-derived cells. Rheumatology (Oxford) 2003;42: 1306–10.
15. Bestwick CSMN. Reactive oxygen species and tendon problems: review and hypothesis. Sports Med Arthrosc Rev 2000;8: 6–16
16. Ireland D, Harrall R, Curry V, et al. Multiple changes in gene expression in chronic human Achilles tendinopathy. Matrix Biol 2001;20: 159–69 [PubMed] [Google Scholar]
17. Alfredson H, Lorentzon M, Backman S, Backman A, Lerner UH. CDNA-arrays and real-time quantitative PCR techniques in the investigation of chronic Achilles tendinosis. J Orthopaed Res 2003;21: 970–5
18. Maffulli N, Barrass V, Ewen SW. Light microscopic histology of achilles tendon ruptures. A comparison with unruptured tendons. Sports Med 2000;28: 857–63

19. Almekinders LC, Temple JD. Etiology, diagnosis, and treatment of tendonitis: an analysis of the literature. Medicine Sci Sports Exercise 1998;30: 1183–90.
20. Nelen G, Burssens A. Surgical treatment of chronic Achilles tendinitis. Am J Sports Med 1989;17: 754–9.
21. Clancy WGJ, Brand RL. Achilles tendonitis in runners: a report of five cases. Am J Sports Med 1976;4: 46–57.
22. Maffulli N, Regine R, Angelillo M, Capasso G, Filice S. Ultrasound diagnosis of Achilles tendon pathology in runners. Br J Sports Med 1987;21: 158–62.
23. Schepsis AA, Leach RE. Surgical management of Achilles tendon overuses injuries: a long-term follow-up study. Am J Sports Med 1994;22: 611–19.
24. Paavola MKP, Paakkala T, Pasanen M, Järvinen M. Long-term prognosis of patients with Achilles tendinopathy. An observational 8-year follow-up study. Am J Sports Med 2000;28: 634–42 [PubMed] [Google Scholar]
25. Kvist H, Kvist M. The operative treatment of chronic calcaneal paratenonitis. J Bone Joint Surg 1980;62B: 353–7.
26. Clement DB, Taunton J, Smart GW. Achilles tendinitis and peritendinitis: etiology and treatment. Am J Sports Med 1984;12: 179–84
27. Nelen G, Burssens A. Surgical treatment of chronic Achilles tendinitis. Am J Sports Med 1989;17: 754–9.
28. Jackson BA, Schwane JA, Starcher BC. Effect of ultrasound therapy on the repair of Achilles tendon injuries in rats. Med Sci Sports Exercise 1991;23: 171–6.
29. Ramirez A, Schwane JA, McFarland C, Starcher B. The effect of ultrasound on collagen synthesis and fibroblast proliferation in vitro. Med Sci Sports Exercise 1997;29: 326–32.
30. Gehlsen GM, Ganion LR, Helfst R. Fibroblast responses to variation in soft tissue mobilization pressure. Med Sci Sports Exercise 1999;31: 531–5.
31. Silbernagel KG, Thomee R, Thomee P, Karlsson J. Eccentric overload training for patients with chronic Achilles tendon pain—a randomized controlled study with reliable testing of the evaluating methods. Scand J Sports Med 2001; 11: 197–2066.
32. Gross ML, Davlin L, Evanski PM. Effectiveness of orthotic shoe inserts in the long-distance runner. Am J Sports Med 1991;19: 409–12.
33. Capasso G, Maffulli N, Testa V, Sgambato A. Preliminary results with peritendinous protease inhibitor injections in the management of Achilles tendinitis. J Sports Traumatol Rel Res 1993;15: 37–43.
34. Sundqvist H, Forsskahl B, Kvist M. A promising novel therapy for Achilles peritendinitis: double-blind comparison of glycosaminoglycan polysulfate and high-dose indomethacin. Int J Sports Med 1987;8: 298–303.
35. Speed CA. Corticosteroid injections in tendon lesions. BMJ 2001;323: 82–6.
36. Friedrich T, Schmidt W, Jungmichel D, Horn LC, Josten C. Histopathology in rabbit Achilles tendon after operative tenolysis (longitudinal fiber incisions). Scand J Med Sci Sports 2001; 11: 4–81.
37. Robert J de Vos 1, Adam Weir, Hans T M van Schie, Sita M A Bierma-Zeinstra, Jan A N Verhaar, Harrie Weinans, Johannes L Tol PMID: 20068208.
38. R J Kampa D A Connell PMID: 21070532.
39. Suzan de Jonge 1, Robert J de Vos, Adam Weir, Hans T M van Schie, Sita M A Bierma-Zeinstra, Jan A N Verhaar, Harrie Weinans, Johannes L Tol PMID: 21602565.
40. Richard F Owens Jr 1, John Ginnetti, Stephen F Conti, Carmen Latona; PMID: 22338951.
41. Giuseppe Filardo 1, Elizaveta Kon 2, Berardo Di Matteo 1, Alessandro Di Martino 1, Giulia Teei 1, Patrizia Pelotti 3, Annarita Cenacchi 4, Maurilio Marcacci 1 PMID: 24960641.
42. Matteo Guelfi 1, Andrea Pantalone 2, Daniele Vanni 2, Michele Abate 2, Marco G B Guelfi 3, Vincenzo Salini 2 PMID: 26235856.
43. Thøger P Krogh 1, Torkell Ellingsen 2, Robin Christensen 3, Pia Jensen 4, Ulrich Fredberg 2 PMID: 27257167.
44. Anders Ploug Boesen 1 2, Rudi Hansen 1, Morten Ilum Boesen 3, Peter Malliaras 4, Henning Langberg 5 PMID: 28530451.
45. Chun-Jie Liu 1, Kun-Lun Yu 2, Jiang-Bo Bai 2, De-Hu Tian 2, Guo-Li Liu 3.
46. Lutz von Wehren 1, Kerstin Pokorny 2, Fabian Blanke 3, Jannis Sailer 4, Martin Majewski 5.
47. Maffulli, N., Nilsson Helander, K. & Migliorini, F. Tendon appearance at imaging may be altered, but it may not indicate pathology. Knee Surg Sports Traumatol Arthrosc. 2023; 31: 1625–1628.
48. Zabrzyński, J.; Gagat, M.; Huri, G.; Łapaj, Ł.; Paczesny, Ł.; Zielińska, W.; Zabrzyńska, M.; Szwedowski, D.; Kruczyński, J. Therapeutic Advances in Tendinopathy Quantified Microscopically Using Bonar Score, with a Special Reference to PRP Therapy—A Systematic Review of Experimental Studies. Appl. Sci. 2021, 11, 4973.
49. Desouza, C., Dubey, R. & Shetty, V. Platelet-rich plasma in chronic Achilles tendinopathy. Eur J Orthop Surg Traumatol. 2023; 33: 3255–3265.
50. Arthur Vithran, D.T.; Xie, W.; Opoku, M.; Essien, A.E.; He, M.; Li, Y. The Efficacy of Platelet-Rich Plasma Injection Therapy in the

Treatment of Patients with Achilles Tendinopathy: A Systematic Review and Meta-Analysis. J. Clin. Med. 2023; 12: 995.