

# Efficacy of ultrasound-guided hyaluronic acid injections in achilles and patellar tendinopathies: a prospective multicentric clinical trial

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## SUMMARY

**Background.** Achilles and patellar tendinopathies are common causes of chronic pain and functional impairment. Conservative management can be effective, but it is time-consuming and it requires intensive patient compliance.

Hyaluronic acid (HA) is a key component of the extracellular matrix and its anti-inflammatory, lubricant and analgesic properties are well documented in osteoarthritis. Growing pre-clinical studies indicate a possible role of HA in tendon pathology, while clinical evidences are still lacking.

**Objectives.** The primary objective was to assess the efficacy of a new formulation of HA obtained from biofermentation (Hyalotend® Fidia Farmaceutici, Italy) to improve the clinical symptoms assessed by the VISA-A and VISA-P questionnaires at 90 Days of follow-up.

Secondary objectives were to evaluate improvement in pain (NRS-11), US parameters (tendon appearance and neovascularization), and quality of life (EQ-5D). Safety was also evaluated.

**Methods.** This was a prospective, open-label, multicenter clinical trial. Thirty-five patients (26 in AT group; 9 patients in PT group), who satisfied inclusion and exclusion criteria, were recruited during a 4-month period. Each patient received 1 injection weekly for 3 weeks under US-guidance at the painful site, and was evaluated at 14, 45 and 90 days after the procedure. Subjects were instructed to record on a diary any intakes of the permitted rescue medication for pain relief oral paracetamol, up to the "rescue dose" of 3 g/Day (i.e. 6 tablets/Day) throughout the study period and to interrupt treatment at least 24 hours prior to each visit.

**Results.** Significant improvement occurred in both VISA-A (23.22±23.17; 95% CI: 13.20; 33.24; p<.0001) and VISA-P (19.25±11.61; 95% CI: 9.54; 28.96; p=0.0022). NRS-11 score significantly decreased in subjects with AT or PT during the study (p<0.0001 and p=0.0040 respectively). Significant improvement of swelling and tenderness evaluated with US in the AT group was revealed (McNemar's test; p=0.0016 and p=0.0114, respectively), while in the PT group the presence of these clinical symptoms showed only a non-significant tendency to decrease (p=0.3173 for both). The EQ-5D-5L total score increased in both the AT and PT groups (mean change vs. baseline at day 90 equal to 15.96±18.95 (range: -50; 45) and 15.50±32.42 (range: -40; 71), respectively. Seven patients (20.59%) experienced at least one adverse event (AE), all AEs had mild severity. No serious AE or other significant AE leading to study treatment discontinuation or temporary interruption was reported.

**Conclusions.** Three US-guided HA injections may induce prompt improvement in pain and function in mid-portion Achilles and patellar tendinopathies that last until 90 days of follow-up. Amelioration in tendon structure, neovascularization and clinical parameters may also be achieved. The treatment is also safe and well-tolerated.

## KEY WORDS

hyaluronic acid; injection; Achilles tendon; patellar tendon; tendinopathy; ultrasonography

## INTRODUCTION

Intra-articular hyaluronic acid (HA) injections are widely used to treat degenerative joint pathologies like osteoarthritis, while the role of HA in tendinopathies is still debated. Nevertheless, increasing interest in this field is leading to growing preclinical and clinical studies (1). Tendinopathies are characterized by decreased collagen I production, enhanced collagen III, tendon structure disruption and ineffective neovascularization (2).

HA could have disease-modifying effects on chronic tendinopathy through several mechanisms, leading to increase tenocyte viability and proliferation, increase in collagen I production, and reducing apoptosis in a dose-dependent manner (3-7). Furthermore, HA could be partially involved in neovascularization regulating VEGF (Vascular Endothelial Growth Factor) and collagen IV (8).

In tendon disorders, ultrasonography (US) guidance permits accurate HA injection in the target site with several intrinsic advantages (real-time execution, absence of ionizing radiation, low cost and availability of device) (1).

However, there is still insufficient evidence to clearly recommend the routine use of HA in Achilles and patellar tendinopathies (9).

This prospective study evaluated the efficacy and safety of 3 US-guided peritendinous injections of a new formulation of 500-730 kDa HA (Hyalotend®, Fidia Farmaceutici SpA, Italy) in mid-portion Achilles and patellar tendinopathies.

## METHODS

### Study design

This was a pre-market, prospective, open-label, multicenter clinical study conducted in Italy. Patients were enrolled from January 2017 to May 2018 in 5 different sites. The study was conducted in accordance with the principles of the Declaration of Helsinki and the International Conference on Harmonization (ICH) Consolidated Guideline on Good Clinical Practice (GCP). All patients provided informed consent prior to entry into the study and were able to understand the nature and aims of the study, including possible risks and side effects, to cooperate with the Investigator, to comply with the study requirements, and to attend all planned study visits.

The study protocol, patient information leaflet and the informed consent document were submitted to the Italian Ministry of Health (IMH) and to the reference Independent Ethics Committee (IEC) of each center participating in the study before any study-related procedure was started, and approval obtained from the IMH and IEC of each participating centre before the beginning of the study.

### Patients

Male and female patients aged between 18 and 65 years presenting with a symptomatic Achilles or patellar mid-portion tendinopathy for  $\geq 6$  weeks duration and confirmed by ultrasound evaluation were enrolled in the study. Symptomatic phase was defined as a pain score  $\geq 5$  on NRS-11 on the target tendon at inclusion. The main exclusion criteria were suspected tendon rupture or insertional tendinopathy, general, severe intercurrent illnesses (eg, uncontrolled diabetes mellitus, peripheral neuropathy), known hypersensitivity to the product (active compound and excipients) or any component or procedure used in the study. Smokers and patients with BMI  $\geq 35$ , pregnant or breast-feeding women were also excluded. Females of childbearing age were required to be using an appropriate method of contraception. Patients who received an injection of hyaluronic acid or of Platelet-Rich Plasma (PRP) in the target tendon within the last 3 months or had been treated with systemic and/or local steroids within the last 4 weeks, immunosuppressive drugs within the last 3 months, repeated use of non-steroidal anti-inflammatory drugs (NSAIDs) within the last week or any use within the last 24 hours, or fluoroquinolones within the last 4 weeks were also excluded.

### Device characteristics

Hyalotend® (Fidia Farmaceutici SpA, Padua, Italy) is formulated as a highly viscous solution of sodium hyaluronate obtained from biofermentation with a medium molecular weight (MMW) in buffered physiological sodium chloride, pH 6.8-7.5. The solution (20 mg/2 ml) is contained in sterile, single-use, prefilled syringes for peritendinous injection.

### Study treatment and injection technique

HA was administered as a peritendinous ultrasound-guided injection between paratenon and tendon. Each patient received one 2 ml weekly injection for three consecutive weeks. US evaluations were performed by one full-trained investigator in each center, using a 5-12 MHz linear probe and PRF set at 0.5 kHz. The contralateral non-painful tendon was also examined as intra-patient comparison.

All Achilles tendon injections were performed with the patients prone. A 22-gauge needle was introduced at a 30-degree angle using a dorsolateral approach, with the probe in a transverse plane (**Figure 1**).

All patellar tendon injections were performed with the patients in supine position with knee flexed at 20° with a support. A 22-gauge needle was inserted immediately posterior to the patellar tendon, at the interface between the

tendon and paratenon, using a dorsolateral approach, with the probe positioned in transverse plane.

Patients will be allowed to walk immediately but were advised to refrain from strenuous activity for 3-4 Days.

## Outcomes

The primary outcome was change in the Italian version of the Victorian Institute of Sports Assessment-Achilles' questionnaire (VISA-A) and the Victorian Institute of Sports Assessment-Patellar (VISA-P) at 90 days of follow-up(10,11). The functional improvement after 90 Days was determined as the mean change from baseline in the VISA-A and VISA-P total scores, adopting either the prevalence or the Last Observation Carried Forward (LOCF) approach.

The secondary outcomes were: 1) change in pain on NRS-11 2) the intensity of clinical parameters (redness, warmth, swelling, tenderness on palpation, crepitus on motion, accu-

mulation of tissue fluid), evaluated as presence/absence; 3) improvement in US parameters (axial and sagittal thickness of the target tendon) and neovascularization on power Doppler using a 4-points scale(12) 4), Patient Global Assessment (PGA), Clinical Observer Global Assessment (COGA) 5), rescue medication consumption (paracetamol) 6) and Health-related Quality of Life (EuroQoL EQ-5D-5L questionnaire).

## Statistical analysis

The primary efficacy analysis was performed on the Full Analysis Set (FAS) population (i.e. all subjects who signed the informed consent and received any dose of study treatment), and on the Per Protocol (PP) population (i.e. all subjects in the FAS without major protocol and non-protocol deviations) as supportive. The secondary efficacy analyses were performed on the FAS population only. The analyses on safety parameters, prior and concomitant medications and drug exposure were conducted in the Safety set (i.e. all subjects who signed the informed consent, received any dose of study treatment and had at least one post-baseline safety assessment).

Results were based on non-missing data (i.e. prevalence approach), except for the primary endpoint (i.e. LOCF approach).

In case of non-normal distribution of data, a rank transformation of the original values was applied.

Descriptive statistics were provided for all variables overall and by treatment group. Continuous variables were summarized by using number, mean, median, standard deviation (SD) and range. Categorical variables were summarized by using frequency distributions and percentages. Results were considered statistically significant if the p-value was  $\leq 0.05$ .

A paired t-test or a Wilcoxon signed rank sum test was applied for primary and secondary outcome measures, except for clinical parameters evaluation in which McNemar's test was applied for each parameter.

Adverse Events (AEs) were classified into Treatment-Emergent AEs (TEAEs) and Non-TEAEs. The number of total TEAEs and the absolute and relative frequency of subjects with TEAEs, serious TEAEs or TEAEs leading to study treatment discontinuation are provided overall and by tendinopathy. TEAEs are summarized by System Organ Class and Preferred Term, overall and by tendinopathy type. Suspected drug-related TEAEs and TEAEs at the injection site are presented as well.

All statistical tables, listings and analyses were produced using SAS® release 9.4 64-bit or later (SAS Institute, Inc., Cary, NC, USA).



**Figure 1.** a) the needle is insert in a dorsolateral approach, with the probe in a transverse plane; b) US visualization of the needle during injection

Data from all investigational sites were pooled and summarized. Continuous data were summarized by means of common summary statistics (i.e. number of observations (n), mean, standard deviation (SD), median, first and third quartiles, minimum and maximum). Categorical data are presented by absolute and relative frequencies (n and %). The percentage calculation was based on total values (i.e. the count was shown also in the missing category, unless otherwise specified).

Prior to any analyses on the efficacy parameters, normality was assessed by means of statistical tests (i.e. Shapiro-Wilk test, Kolmogorov-Smirnov test). A paired t-test or a Wilcoxon signed rank sum test (in case of non-normally distributed data) was used for the analyses on the efficacy parameters. For all inferential analyses, two-sided p-values  $\leq 0.05$  were considered statistically significant.

Statistical significance was required for both VISA-A and VISA-P questionnaire improvement at Day 90 (i.e. primary variable), thus no alpha adjustment was applied.

## RESULTS

### Distribution of patients (patients' population)

Thirty-five patients were enrolled, after signed written informed consent, 26 patients with Achilles tendinopathy (AT) and 9 with patellar tendinopathy (PT).

Thirty-one enrolled patients (88.57%) completed the study, 23 patients (88.46%) with AT and 8 (88.89%) with PT, 4 patients (11.43%) did not complete the study. Among subjects with AT, 3 patients did not complete the study due to withdrawal after informed consent, patient non-compliance, or loss to follow-up. One patient with PT discontinued due to insertional tendinopathy, not diagnosed at patients screening and inclusion visit.

Thirty-four of 35 enrolled patients (97.14%) received at least one injection and had at least one post-baseline safety assessment, and thus were included in the Full Analysis Set (FAS) and in the Safety population. One patient with patellar tendinopathy was excluded from the FAS and Safety sets.

Twenty-nine enrolled patients (82.86%), i.e. 22 out of 26 patients in the AT group and 7 out of 9 subjects in the PT group, completed the study without any significant protocol/non-protocol deviations and were thus included in the Per Protocol (PP) analysis.

An overview of demographic data and baseline characteristics of the FAS population is provided in **Table I**.

Mean age at informed consent was  $52.27 \pm 7.44$  years (range: 34; 65 years) in the AT group and  $31.75 \pm 9.18$  years (range: 20; 46 years) in the PT group.

All patients were Caucasian, and most were male (19 out of 26 subjects with AT and all subjects with PT except one), and well matched at baseline for height, weight and BMI and perform physical activity regularly (**Table I**).

### Primary efficacy endpoint

Results were positive in both AT and PT group, respectively in VISA-A and VISA-P questionnaires.

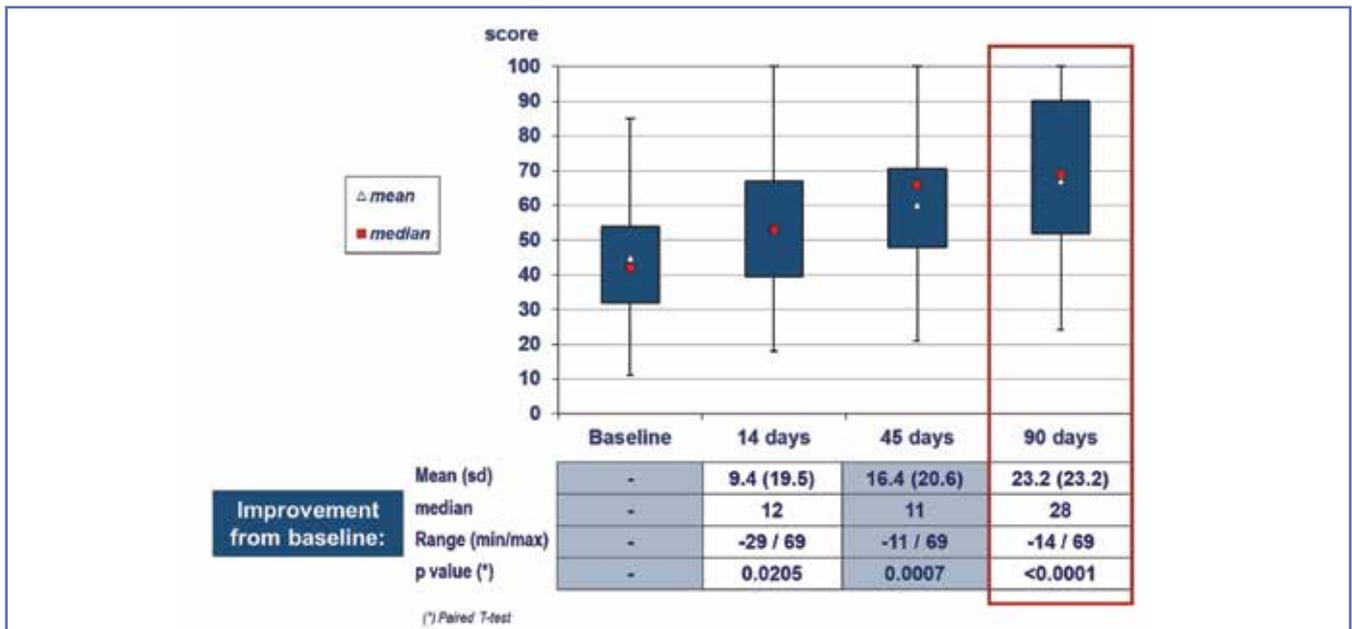
The VISA-A total score was  $44.88 \pm 17.37$  at baseline and  $67.17 \pm 22.66$  at Day 90. The mean change from baseline was  $23.22 \pm 23.17$  (95% CI: 13.20; 33.24;  $p < 0.0001$ ), indicating a statistically significant and clinically relevant increase in the VISA-A total score after 90 Days.

A statistically significant and clinically relevant increase in the VISA-A total score at Day 90 was obtained also adopting the LOCF approach, i.e. the VISA-A total score was  $44.88 \pm 17.37$  at baseline and  $66.24 \pm 22.45$  at Day 90 and a mean change from baseline of  $21.36 \pm 23.10$  (95% CI: 11.83; 30.89;  $p = 0.0001$ ) (**Figure 2**).

Results for the PP population were similar. The mean VISA-A scores for each single item of the VISA-A questionnaire increased from baseline to Day 90. The average VISA-A total score was  $43.50 \pm 17.39$  at baseline and  $67.82 \pm 22.98$  at Day 90. Mean change from baseline was equal to  $24.32 \pm 23.09$  (95% CI: 14.08; 34.56;  $p < 0.0001$ ), indicating a statistically significant increase in the VISA-A total score at 90 Days. The mean VISA-A score for each questionnaire item increased from baseline to 14 or 45 Days. The average VISA-A total score was  $44.88 \pm 17.37$  at baseline,  $53.17 \pm 19.67$  at Day 14, and  $60.17 \pm 19.40$  at Day 45. Mean change from baseline was equal to  $9.42 \pm 19.54$  (95% CI: 1.17; 17.67;  $p = 0.0205$ ) at Day 14 and to  $16.42 \pm 20.60$  (95% CI: 7.72; 25.12;  $p = 0.0007$ ) at Day 45, indicating a

**Table I.** Demographic and clinical characteristics of FAS population

	AT group	PT group
Gender, n		
Male	19	7
Female	7	1
Age, years	$52.27 \pm 7.44$	$31.75 \pm 9.18$
Weight, kg	$77.27 \pm 15.11$	$76.25 \pm 14.80$
Height, m	$174.46 \pm 9.21$	$179.50 \pm 9.70$
Body Mass Index	$25.23 \pm 3.53$	$23.51 \pm 2.72$
Duration of tendinopathy, weeks	$29.50 \pm 24.06$	$158.13 \pm 256.66$
Regular sport practice, %	73.08%	87.5%
Hours of training per week	$4.28 \pm 2.14$	$8.71 \pm 2.50$

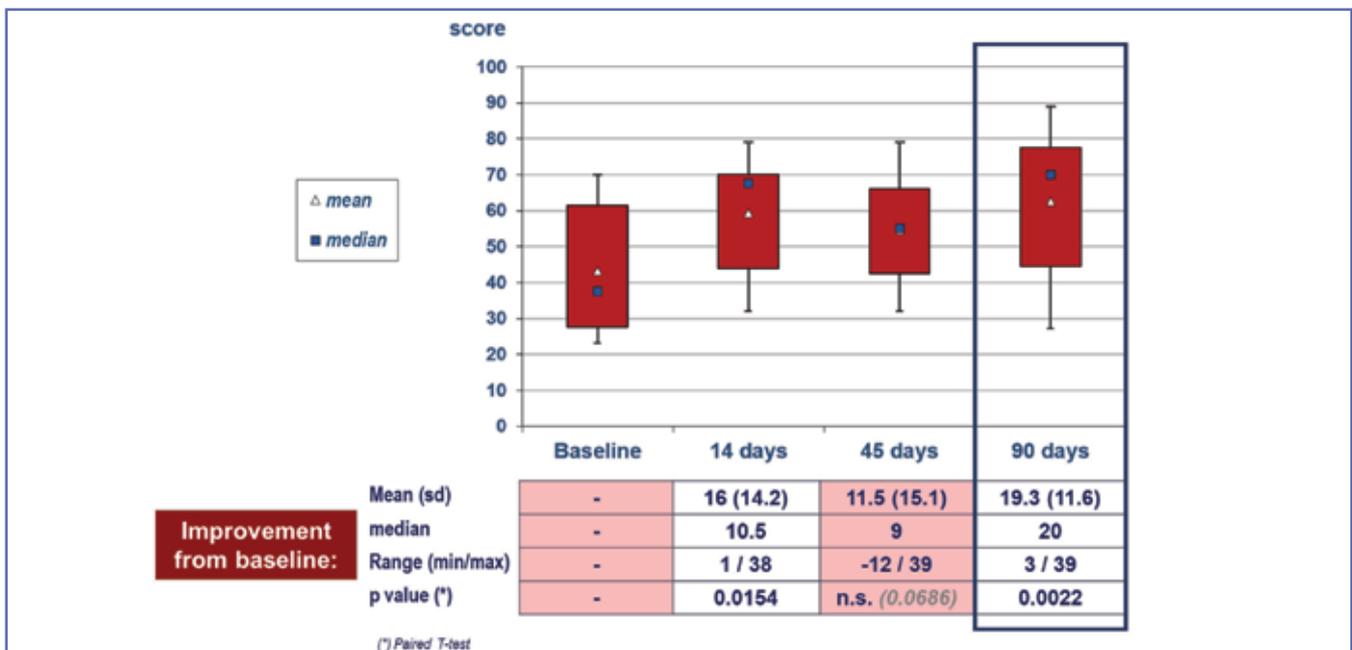


**Figure 2.** VISA-A questionnaire total score - change vs. baseline (prevalence approach) FAS population - Achilles tendinopathy (N=26)

statistically significant increase in the VISA-A total score at both visits.

The average VISA-P total score was 43.25±18.68 at baseline and 62.50±21.35 at Day 90 (Figure 3). The average VISA-P total score was 59.25±16.81 at Day 14, and

54.75±15.85 at Day 45. Mean change from baseline was equal to 16±14.20 (95% CI: 4.13; 27.87; p= 0.0154) at Day 14 and to 11.50±15.13 (95% CI: -1.15; 24.15; p= 0.0686) at Day 45, suggesting a statistically significant increase in the VISA-P total score after 14 Days. Mean change



**Figure 3.** VISA-P questionnaire total score - change vs. baseline (prevalence approach) FAS population - patellar tendinopathy (N=8)

from baseline was equal to  $19.25 \pm 11.61$  (95% CI: 9.54; 28.96;  $p=0.0022$ ), thus suggesting a statistically significant increase in the VISA-P total score at Day 90. No missing data of VISA-P total score are present in the PT group (FAS population), thus the LOCF approach led to the same results as the prevalence approach. Results for the PP population were very similar. The average VISA-P total score was  $39.43 \pm 16.46$  at baseline and  $60 \pm 21.76$  at Day 90. Mean change from baseline was equal to  $20.57 \pm 11.87$  (95% CI: 9.59; 31.55;  $p=0.0038$ ), suggesting a statistically significant increase in the VISA-P total score at 90 Days.

### Secondary efficacy endpoints

#### NRS-11

NRS-11 scores progressively decreased from baseline to each subsequent evaluation in both groups. The mean changes from baseline were equal to  $-2.38 \pm 2.43$  (95% CI: -3.40; -1.35;  $p<.0001$ ) in the AT group and  $-3.88 \pm 2.75$  (95% CI: -6.17; -1.58;  $p=0.0053$ ) in the PT group at Day 14;  $-4.17 \pm 3.02$  (95% CI: -5.44; -2.89;  $p<.0001$ ) and  $-4.00 \pm 3.51$  (95% CI: -6.93; -1.07;  $p=0.0145$ ) at Day 45;  $-4.52 \pm 3.25$  (95% CI: -5.93; -3.12;  $p<.0001$ ) and  $-4.75 \pm 3.20$  (95% CI: -7.42; -2.08;  $p=0.0040$ ) at Day 90. Thus, suggesting statistically significant pain reduction at each evaluation in both groups (Figure 4 and Figure 5).

### Clinical parameters

The proportion of patients with AT who presented swelling of the target tendon tended to progressively decrease during the study from 73.08% (19 subjects out of 26) at baseline to 34.78% (8 subjects out of 23) at Day 90. Similarly, the presence of swelling tended to decrease from 3 patients out of 8 (37.50%) at baseline to only one of 8 patients (12.50%) at Day 90 in the PT subset.

Likewise, the number of patients with tenderness in both the AT and PT groups tended to decrease over time: 25 AT patients out of 26 (96.15%) and 5 PT patients out of 8 (62.50%) presented tenderness at baseline, while only 14 AT patients out of 23 (60.87%) and 3 PT patients out of 8 (37.50%) showed this symptom at Day 90. In particular, 9 patients with AT out of 23 (39.13%) who reported tenderness at baseline no longer presented it after 90 Days.

Only 1 patient with AT out of 26 (3.85%) presented redness at baseline, while the patient no longer reported this symptom at Day 90. No patients with patellar tendinopathy reported redness at any visit.

Two (2) patients out of 26 (7.69%) in the AT group and only 1 patient out of 8 (12.50%) in the PT group presented effusion at baseline. No patient presented effusions after 90 Days.

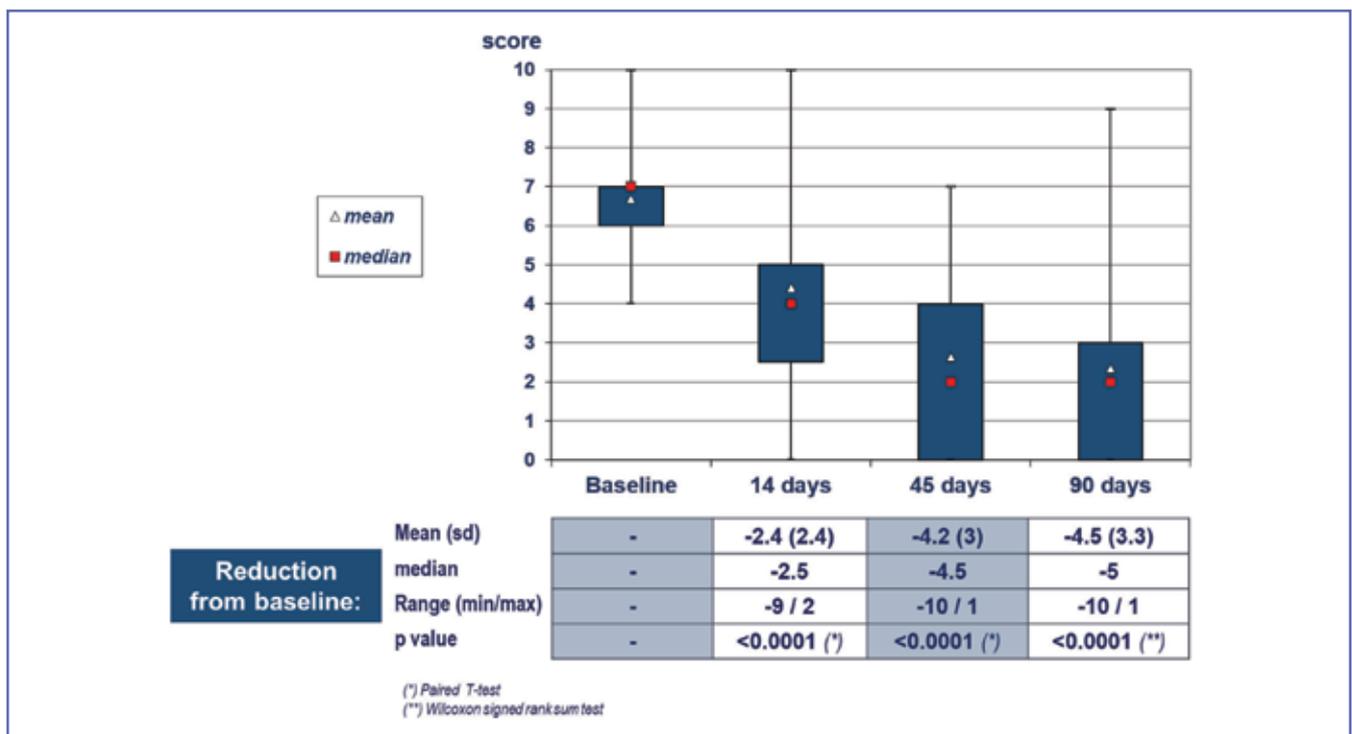
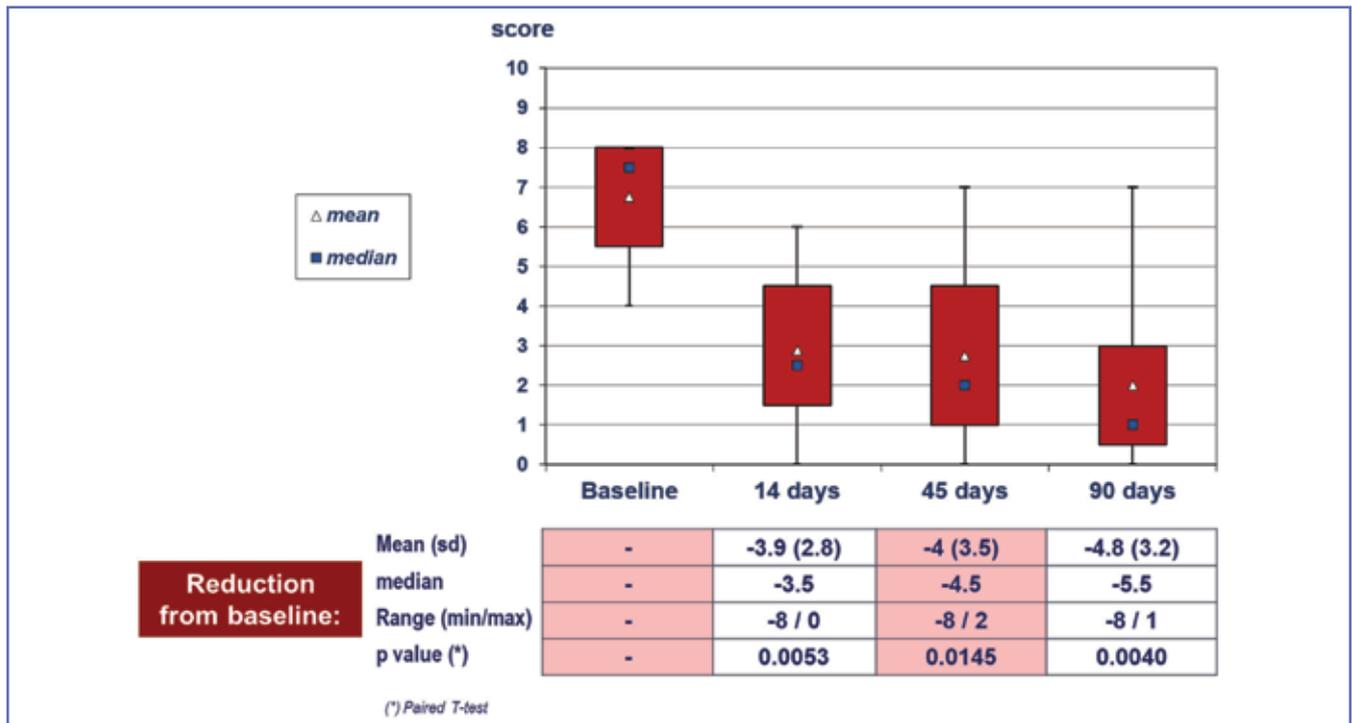


Figure 4. Pain evaluation (NRS-11 score) – change vs. baseline (FAS population) Achilles tendinopathy (N=26)



**Figure 5.** Pain evaluation (NRS-11 score) – change vs. baseline (FAS population) patellar tendinopathy (N=8)

### US parameters

The axial thickness of the target tendon remained stable up to 90 Days in both the AT and PT groups, while the sagittal thickness of the target tendon tended to decrease throughout the study especially among patients with patellar tendinopathy. The mean changes from baseline to Day 90 were equal to  $-0.08 \pm 1.79$  (range: -3.40; 3.70) and  $0.61 \pm 2.99$  (range: -3.00; 6.00) for axial thickness and  $-0.23 \pm 1.39$  (range: -2.90; 2.70) and  $-2.76 \pm 4.40$  (range: -13; 0.10) for sagittal thickness, in the AT and PT subsets, respectively.

Among patients with Achilles tendinopathy, both the axial and sagittal thickness of the contralateral tendon remained essentially stable from baseline. After 90 Days, the mean change from baseline was equal to  $-0.21 \pm 0.90$  (range: -1.90; 2.10) for axial thickness and  $-0.13 \pm 1.13$  (range: -3.10; 1.00) for sagittal thickness.

Conversely, patients with patellar tendinopathy presented a slight increase of the thickness of the contralateral tendon at each visit from Day 7 considering both the axial and the sagittal image. The mean change from baseline to Day 90 was equal to  $1.11 \pm 2.18$  (range: -0.80; 6.00) for axial thickness and  $1.26 \pm 2.83$  (range: -1.20; 8.00) for sagittal thickness. In both AT and PT groups, vascularization on Power Doppler progressively decreased. No visible blood vessels (grade=0) ranged from 8% and 12.50% at baseline, to

26.09% and 62.50% at Day 14, and to 25% and 62.50% at Day 45 in the AT and PT subsets, respectively. At Day 90, 10 out of 23 patients with AT (43.48%) and 5 out of 8 patients with PT (62.50%) presented no visible blood vessels in the target tendon.

### Patient Global Assessment (PGA) and Clinical Observer Global Assessment (COGA)

The PGA score tended to decrease from baseline to each subsequent visit in both groups. The mean change vs. baseline at Day 90 was equal to  $-0.57 \pm 1.12$  (range: -2; 1) in the AT group and  $-0.88 \pm 1.25$  (range: -3; 1) in the PT group, indicating improvement in clinical condition and global health. For exploratory purposes, a paired t test or a Wilcoxon signed rank sum test (if data were not normally distributed) was applied to the mean change from baseline of PGA score, overall and by tendinopathy type. The resulting p-value at Day 90 was equal to 0.0220 and 0.0875 for patients within the AT and PT groups, respectively.

The COGA score tended to decrease from baseline to each subsequent visit in both groups. The mean change from baseline after 90 Days was equal to  $-1.13 \pm 1.46$  (range: -3; 2) in the AT subset and  $-0.75 \pm 0.46$  (range: -1; 0) in the PT subset.

For exploratory purposes, a paired t test or a Wilcoxon signed rank sum test was applied to the mean change from baseline of COGA score, overall and by tendinopathy type. Among patients with AT, the estimated p-value was lower than 0.05 at each visit and the resulting p-value at Day 90 was equal to 0.0018 and 0.0313 for patients within the AT and PT groups, respectively.

### Health-related quality of life (EQ-5D-5L)

The mean scores for mobility, self-care, usual activities and pain/discomfort tended to decrease from baseline to Day 90 in both Achilles and patellar tendinopathy groups, while the mean scores for anxiety/depression remained essentially stable after 90 Days in both tendinopathy subsets.

For exploratory purposes, a paired t test or a Wilcoxon signed rank sum test (if data were not normally distributed) was applied to the mean change from baseline of each EQ-5D-5L item score, overall and by tendinopathy type. The resulting p-value at Day 90 was equal to 0.0112 for usual activities and pain/discomfort scores in patients with PT, while it was equal to 0.0006 for usual activities and lower than 0.0001 for mobility and pain/discomfort scores in the AT group.

The average EQ-5D-5L total score increased after 90 Days in both the Achilles and patellar tendinopathy groups. The mean total score was equal to  $63.96 \pm 15.29$  (range: 28; 90) and  $64.25 \pm 33.34$  (range: 9; 95) at baseline, and  $80.00 \pm 21.37$  (range: 0; 100) and  $79.75 \pm 23.52$  (range: 30; 100) at Day 90 in patients with AT and PT, respectively. Mean change from baseline at Day 90 was equal to  $15.96 \pm 18.95$  (range: -50; 45) in the AT group and  $15.50 \pm 32.42$  (range: -40; 71) in the PT group.

Exploratory results of a paired t test or a Wilcoxon signed rank sum test applied to the mean change from baseline at Day 90 of EQ-5D-5L total score reported a p-value lower than 0.05 in the AT group only.

## DISCUSSION

The present management of tendinopathies is based on early mobilization with eccentric exercises as the cornerstone of conservative treatment (12). Nevertheless, the treatment period may last several months and may be discouraging, especially in athletes (13,14). Growing clinical evidences suggested that HA may induce prompt relief in different painful tendinopathies (15-17).

The primary objective of this study was to evaluate the efficacy of three weekly US-guided peritendinous injections of MMW-HA in midportion Achilles and patellar tendinopathies at 90 Days of follow-up. The increase of the

VISA-A and VISA-P total scores from baseline to Day 90 was significant, thus indicating a significative progressive functional improvement with HA injections after 14, 30 and 90 Days in both groups, though we acknowledge that in the PT group the planned sample size target was not achieved. Our findings point toward short-term efficacy of three consecutive HA injections and confirmed the observations of other Authors who reported progressive pain relief after three consecutive 500-730 kDa injections to 56 Days of follow-up (18). However, in the study of *Fogli et al* (18), both insertional and mid-portion tendinopathies, which are distinct pathological entities, were considered (19,20).

Furthermore, prompt significant improvement at 14 Days of follow-up may suggest a possible role in sporting populations, where a rapid clinical benefit is often extremely important. Prompt efficacy is in accordance with *Lynen et al.* who proposed 2 injections of a combination of HA and mannitol in mid-portion Achilles tendinopathy (21). Three consecutive injections of 500-730 kDa HA determined rapid symptoms improvement, compared to ESWT, also in non-calcific rotator cuff tendinopathy (22).

Peritendinous injections are widely used in clinical practice and this is strongly linked to the increased use of US. Indeed, US allows better accuracy, helping to achieve better outcomes also for less approachable locations (22-24). Our US-guided technique permitted a real-time visualization of the needle during the procedure, ensuring the correct distribution of the product, with low-risk of failure to inject in the desired area.

In both AT and PT groups, neovascularization on Power-Doppler progressively decreased, as sagittal tendon thickness. These improvements in tendon characteristics could be linked to the role of HA to promote tendon gliding and reduction in friction and tendon adhesions, possibly a disease-modifying effect (18,25). Ineffective neovascularization plays a central role in pain development (26-28), therefore neovascularization decrease may at least partially explain pain relief.

The treatment was well tolerated with only one TEAE in AT group, probably related to the injection procedure. Compliance was satisfactory with 88.57% of patients who completed the follow-up and the proportion of patients who needed rescue medication for pain relief was limited, ranging from 12% and 12.50% at Day 7 to 13.04% and 0% at Day 90 in the AT and PT subset, respectively, without relevance for outcome.

Our study presented 2 main limitations: the absence of a control group and the low number of patients included in the PT group, despite the extension of the enrollment period and the add-on of new investigational centers.

## CONCLUSION

Three US-guided HA injections may induce prompt improvement in pain and function in mid-portion Achilles and patellar tendinopathies that last until 90 Days of follow-up. Amelioration in tendon structure, neovascularization and clinical parameters may also be achieved. The treatment is also safe and well-tolerated.

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