



## AXIAL SPEED OF SOUND IN INJURED TENDONS: A PRELIMINARY STUDY

<sup>1</sup> Claudio Vergari, <sup>1</sup> Philippe Pourcelot, <sup>1</sup> Bérangère Ravary-Plumioën, <sup>1</sup> Anne-Gaëlle Dupays, <sup>1</sup> Fabrice Audigie,

<sup>1</sup> Jean-Marie Denoix, <sup>2</sup> David Mitton, <sup>3</sup> Pascal Laugier and <sup>1</sup> Nathalie Crevier-Denoix

<sup>1</sup> USC INRA ENVA BPLC, Ecole Nationale Vétérinaire d'Alfort, Maisons-Alfort, France; email: c.vergari@gmail.com

<sup>2</sup> Université de Lyon, F-69622, Lyon, France; Ifsttar, UMR\_T9406, LBMC; Université Lyon 1.

<sup>3</sup> UPMC Univ Paris 6, UMR CNRS7623, LIP, Paris, F-75005 France.

### SUMMARY

The objective of this study was to determine if axial quantitative ultrasound in tendon can be applied to evaluate the biomechanical consequences of tendon lesions.

Tendon axial speed of sound was measured in-vivo on 12 horses, before and after the surgical induction of a core lesion, and compared to the tendons' mechanical parameters measured in-vitro after euthanasia of the horses.

A correlation was found between the variation of speed of sound, between healthy and injured tendon, and tendon's instantaneous elastic modulus, confirming that axial quantitative ultrasound is a promising technique to evaluate tendon's health.

### INTRODUCTION

Injuries of the superficial digital flexor tendon (SDFT) are a common problem in racing horses. Lesions are often accompanied by hypertrophy of the affected area and a decrease in tendon elastic modulus [1].

Ultrasonography is the preferred technique to evaluate the status of tendon acute lesions. However, chronic lesions often present a normal mean echogenicity, while other more complex techniques, such as magnetic resonance imaging, still detect tissue alterations. Tendon axial quantitative ultrasound, initially designed for non-invasive measure of tendon force [2], is a good candidate to characterize chronic tendon lesions. The theory of ultrasound wave propagation predicts that the speed of sound (SOS) depends on the elastic properties. For example, the velocity of bulk compression wave is proportional to the square root of the bulk modulus. Besides, it has been demonstrated that tendon elastic properties are correlated with SOS in-vitro [3]. However, no study on this application has been conducted so far.

If the tendon load is standardized, for example at the time of the load peak observed in the stance phase at the walk, axial SOS can be exploited to evaluate the tendon elastic modulus in-vivo, which, in turn, can give information on tendon health and quality, especially when a lesion is present.

The objective of this preliminary study was to evaluate if axial quantitative ultrasound can be applied to evaluate tendon lesions. Axial SOS was measured in-vivo in 12 superficial digital flexor tendons (SDFTs), before and about

4 months after bilateral surgical induction of a core lesion, and compared with the tendons' elastic properties which were determined through tensile tests on isolated tendons.

### METHODS

Measures were performed on the SDFTs of 12 French Trotters (2-4 years old,  $429 \pm 39$  Kg average body mass), who participated in a clinical trial testing a regenerating agent on tendon lesions.

The SDFT axial SOS (defined as the velocity of the first arriving signal, FAS, with signals at 1 MHz) was measured in the metacarpal area of the right limb of each horse, with the probe and fixation protocol described in [4], during 6 series of straight line walk on an asphalt pavement. Measurements were performed before ( $SOS_0$ ) and about 4 months after ( $SOS_4$ ) the bilateral surgical induction of a core lesion in the tendon (as approved by an ethical committee). SOS values ( $SOS_0$ ,  $SOS_4$ ) of each horse were calculated by averaging the maximal SOS values (corresponding to the maximal SDFT load at the walk) measured in 30 strides (finding a coefficient of variation inferior to 0.8%).

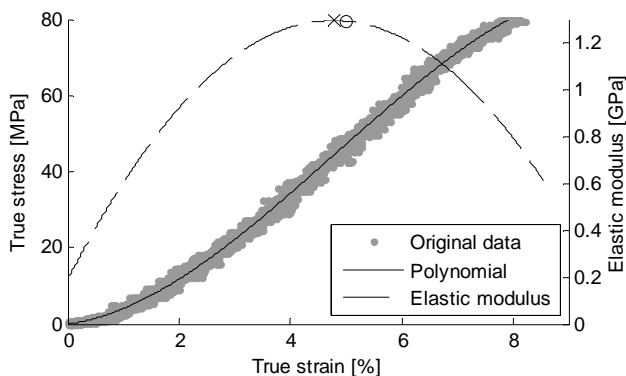
The horses were then euthanized; each right SDFT was isolated (keeping the distal insertion on the middle phalanx intact) and tested in tension, according to the protocol described in [1], at a constant speed of 60 mm/min, in order to measure the instantaneous elastic modulus and the true stress and strain at tendon failure. Each tendon cross-sectional area was determined at rest by ultrasonography, then the tendon was installed in the testing machine by clamping its free end in a *cryo-jaw* and blocking the phalanx with metal rods. Two needles were transversely inserted in each tendon, delimiting a tendon segment (3.4 cm long in average) corresponding to the SOS measuring zone, and equipped with reflective markers on both ends. Those markers were filmed with a high definition camera (Casio Exilim EX-F1) during the test and tracked with a custom made software. The center of gravity of each pair of markers, corresponding to the tendon axis, was calculated in order to determine the tendon true strain as  $\ln(L/L_0)$ , where  $L$  and  $L_0$  are, respectively, the instantaneous and initial distances between the 2 centers of gravity of the 2 pairs of markers. True stress (the ratio of instantaneous force to instantaneous cross-sectional area) was approximated assuming that tendon is incompressible [5]. Instantaneous elastic modulus ( $E_{INST}$ )

was then evaluated as the maximum of the derivative of a 3<sup>rd</sup> degree polynomial approximating the true stress/true strain curve [1] (Figure 1). Elastic modulus approximation for small deformations ( $E_{MAX}$ ) was evaluated as well, in order to be compared to other values found in the literature.

## RESULTS AND DISCUSSION

Four months after lesion induction, all tendons presented a decrease between 1.8 and 10% of the initial SOS. Average  $SOS_0$  was  $2179 \pm 31$  m/s (mean  $\pm$  SD) while  $SOS_4$  was found about 113 m/s lower ( $2067 \pm 67$  m/s). Average true stress at tendon failure was  $72.3 \pm 8.7$  MPa, while average true strain at failure was  $11.3 \pm 1.8$  %. As the stress/strain curve of 2 tendons out of 12 did not reach an inflexion point before failure, they could not be used to determine  $E_{INST}$  and  $E_{MAX}$ . On the remaining 10 tendons, average  $E_{INST}$  and  $E_{MAX}$  were  $1.04 \pm 0.18$  and  $0.90 \pm 0.17$  GPa, respectively. In order to compare the properties of all 12 tendons on standard conditions, the elastic modulus at 5% strain ( $E_{5\%}$ , Figure 1) was also calculated.

A statistically significant correlation ( $p < 0.1$ ) was found between SOS ratio in percentage ( $SOS_4 / SOS_0 * 100$ , Figure 2) and  $E_{INST}$ ,  $E_{MAX}$  and  $E_{5\%}$ .  $E_{MAX}$  was slightly higher than the average values previously found on spontaneous lesions ( $0.74 \pm 0.33$  GPa [1]). No correlation was found with true stress or strain at tendon failure.

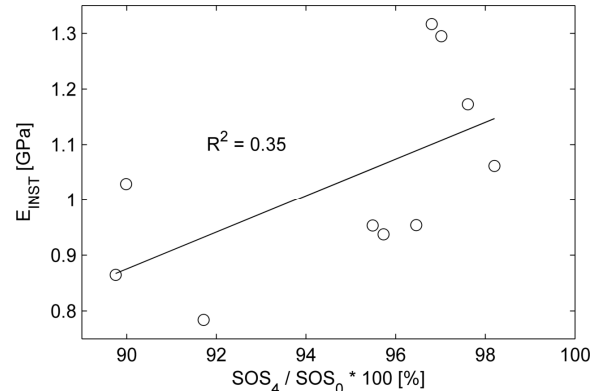


**Figure 1:** True stress vs. true strain curve of tendon #2. The polynomial approximation is reported (solid line) as well as its derivative (dashed line), the instantaneous elastic modulus. The cross and the circle represent, respectively,  $E_{INST}$  and  $E_{5\%}$ .

Since the average wave velocity in equine SDFT tendon at maximal loading is about 2000 m/s [2], the wavelength ( $\lambda$ ) of axially propagating ultrasonic waves (at 1 MHz) is about 2 mm ( $\lambda = SOS / \text{frequency}$ ). In theory, the measured axial SOS is only affected by the mechanical properties of a superficial layer with a thickness comparable to the wavelength (i.e., 2 mm). The tissue's heterogeneities introduced by the chronic core lesion could explain the relatively poor correlation between the tendons' global elasticity and SOS, because the thin superficial layer

explored by ultrasound waves might mechanically differ from the global properties which were measured.

Moreover, although SOS measurements were performed in-vivo during walk, in order to standardize each horse's tendon load, differences in locomotion and limb conformation between horses may have introduced a variability in tendon loading which is hard to account for.



**Figure 2:**  $E_{INST}$  versus ratio of SOS (expressed in percentage) before and 4 months after the surgical induction of a lesion. The correlation between the two variables is statistically significant ( $p < 0.1$ ).

## CONCLUSIONS

These preliminary results suggest that the axial SOS in tendon is affected by the tendon's health, so axial ultrasound transmission is indeed a promising candidate to evaluate chronic lesions. The architecture and histology of the superficial layer of tissue of these tendons, which is explored by the ultrasound waves, has to be further analyzed in order to define how they affect the SOS in injured tendon. Besides, given the dorso-palmar thickness of injured SDFTs (up to, and sometimes above, 1 cm), a probe working at lower frequencies ( $\sim 0.5$  MHz) has now to be tested in order to extend the area explored by the ultrasound technique.

## ACKNOWLEDGEMENTS

The authors are grateful to the Direction Générale de l'Enseignement et de la Recherche, the Région Basse-Normandie and the Institut National de la Recherche Agronomique for financial support. This study was funded by the Agence Nationale de la Recherche.

## REFERENCES

1. Crevier-Denoix N, et al. *Journal of Biomechanics*. **38**:2212-2220, 2005
2. Pourcelot P, et al. *Journal of Biomechanics*. **38**:2124-2129, 2005.
3. Kuo PL, et al. *Ultrasound in Medicine & Biology*. **27**:1275-1284.
4. Crevier-Denoix N, et al. *Equine Veterinary Journal*. **41**: 257-261, 2009.
5. Vergari C, et al. *Journal of Biomechanics*. **Article in press**.