

STRENGTH AND STIFFNESS OF REPAIRED TENDONS

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ABSTRACT

Rotator cuff tears of the shoulder are a common cause of pain and disability. Although surgery is frequently beneficial, re-tearing of the tendons is likely to re-occur. In many cases even if the reparation is successful it will still generate discomfort, problems with mobility, as well as a sharp pain. This project is funded in the cooperation with the Hospital Clínico San Carlos de Madrid. The purpose of this work is to analyze the effect of the surgical repair and the application of different therapies, including mesenchymal stem cell therapy on the biomechanical properties (strength and stiffness) of the repaired tendon. An animal model of rotator cuff tendon reparations has been developed on laboratory rats. To obtain the mechanical response of the healthy and repaired tendons, it was necessary to develop an experimental set up to reproduce the in-vivo working conditions of the tendons (37 °C, immersed in physiological serum), and especially the load transfer. The biomechanical properties (maximum load and stiffness) have been measured in healthy and repaired tendons. A total of 70 rats are used in this particular study. It has been found that the repaired tendon is stronger than the original one. However, the repaired tendons demonstrate less flexibility than the healthy (original) ones prior to the damage.

RESUMEN

La rotura del manguito rotador del hombro es un problema clínico común, especialmente en los ancianos. Aunque la cirugía reparadora suele resultar exitosa, es bastante común que aparezcan problemas de movilidad, molestias e incluso dolores agudos. En este trabajo, realizado en colaboración con el Hospital Clínico San Carlos de Madrid, se ha analizado el efecto de diferentes técnicas de reparación sobre las propiedades biomecánicas (resistencia y flexibilidad) del tendón reparado, comparándolas con muestras control de tendones sanos. Para ello se ha utilizado un modelo animal (ratas de laboratorio) y se ha desarrollado un dispositivo experimental que reproduce las condiciones de trabajo in-vivo de los tendones. En total se han estudiado 70 tendones sanos y 70 reparados mediante 5 técnicas diferentes. Los resultados muestran que en general los tendones reparados poseen mayor resistencia que los originales, pero pueden haber visto reducida su flexibilidad, lo cual puede tener repercusión de forma importante en su funcionamiento.

PALABRAS CLAVE: Supraspinatus tendon, strength, stiffness

1. INTRODUCTION

Rotator cuff tear is a common clinical problem. This particular injury affects 40 per cent (or more) of people over the age of 60, and is a frequent cause of a debilitating pain, such as reduced shoulder function and weakness [1]. Although surgery is frequently beneficial, re-tearing of the tendons is likely to re-occur. In many cases, even if the reparation is successful, it will still

generate discomfort, problems with mobility, as well as a sharp pain. At present, the challenge is to develop a reparation technique able to avoid all these problems. It is accepted that there are many factors responsible for a successful repair of the fractures in a rotator cuff, mainly the size of the tear [3], time from injury until the repair [4], quality of tendon [5], biological response [6] and surgical technique [7]. Previous studies demonstrated that traditional repairing methods have

made an enormous progress since McLaughlin's description of a surgical method to reattach tendon to humeral bone. This study was the basis for other numerous analytical approaches to fixation techniques. Their aim was to find a greater strength of fixation or to define a technique that would be less invasive [8,9,10].

The ideal repair should have high initial fixation strength and maintain mechanical stability until the healing process is finalized [11]. Current studies focus on cognition of a mechanical environment that would considerably increase the healing of rotator cuff. Although the number of investigations was made for animal model of rotator cuff, knowledge of mechanical properties of supraspinatus tendon and technique of their repair required developing [12, 13, 14]. Previous results show that strength of repaired tendons depend on the repairing technique.

In last years, mesenchymal stem cell therapy has emerged as a promising technique. Mesenchymal stem cells are of stromal origin and may differentiate into a variety of tissues. MSCs are multipotent stem cells that can also differentiate into a variety of cell types, including: osteoblasts (bone cells), chondrocytes (cartilage cells) and adipocytes (fat cells) [15]. While this is an accurate description for one function of MSCs, the term fails to convey the relatively recently-discovered roles of MSCs in the repair of tissue. Heretofore there are not many information in current publishing dealing with the healing response after the application of mesenchymal stem cell therapy (MSCs).

The purpose of this work is to analyze the effect of the surgical repair and the application of different therapies, including mesenchymal stem cell therapy on the biomechanical properties (strength and stiffness) of the repaired tendon. An animal model of rotator cuff tendon reparations has been developed on laboratory rats in the Hospital Clinico San Carlos de Madrid. To obtain the mechanical response of the healthy and repaired tendons, it was necessary to develop an experimental set up to reproduce the in-vivo working conditions of the tendons (37 °C, immersed in physiological serum), and especially the load transfer. The biomechanical properties (maximum load and stiffness) have been measured in healthy and repaired tendons. Right tendons of each rodent are used as 'control' samples, left ones are injured and repaired. There are five groups for repaired tendons depending on the type of repaired.

2. MATERIAL AND METHODS

Experiments were conducted in accordance with the "European Convention on Animal Care". As it was already mentioned in the introduction, this specific project was funded with the support of Hospital Clinico San Carlos de Madrid.

An animal model of rotator cuff tendon repairs was developed on rats. A total of 70 rats were used in this study. Both the right and left supraspinatus tendons

were tested from every rodent. Left tendon was surgically detached and repaired after the damage. Right tendons were left unaffected and treated later as 'control' tendons.

All tendons used in this study were divided into 6 groups depending on the repairing type [Table 1]. Tendons belonging to Group 0 (n=9) were injured and left for natural repair without applying any treatment. Samples in Group 1 (n=12) were repaired with prolene. Group 2 (n=13) and Group 3 (n=13) included tendons, which were repaired using gel col 1. Furthermore, in Group 4, mesenchymal stem cell therapy was used in addition to gel col 1. Group 4 (n=10) and Group 5 (n=10) were repaired in similar way as Group 2 and Group 3, however, different kind of gel was applied, OrthAdapt- highly organized Type 1 collagen which reinforce the repair or reconstruction of soft tissues. Group 5 was treated by mesenchymal stem cell therapy as well.

Additionally all the tested rotator cuffs were divided into two groups, depending on the time between repairs and excision from animals for the tests. Rats in all repairing groups were sacrificed at 4weeks, 8weeks after repairing.

Group	Number of samples	
	After one month	After two months
0- natural repaired	6	3
1-with prolene	8	4
2-with Gel col 1	8	5
3-with Gel col1+MES	10	3
4- with Gel OrthADAPT	7	3
5-With Gel OrthADAPT+MES	5	5

Table 1. Number of tendons tested for each groups .Tendons were divided into 6 groups depending on the repairing technique.

Rotator cuff tendons with humerus and scapula complex were used for biomechanical test. Rotator cuff muscles, and other associated muscles and tissues were involved in the examination. Special technique was applied in order to introduce the specimens into the tensile machine. The humerus was fixed on silicon tube diameter 15mm in range, filled up with synthetic resin. The scapula of the specimen was fixed on a polypropylene's tube, 25mm in diameter, also filled up

with synthetic resin. Subsequently after approximately 24h hours, pre-prepared samples were attached to the tensile machine. An experimental set-up reproduced the in-vivo working conditions of the tendons (37°C, immersed in physiological serum).

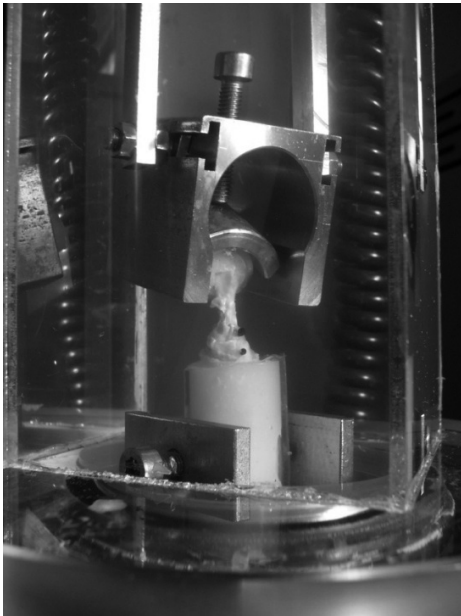


Figure 1. The experimental set up used to reproduce the load transfer in the tendons.

An experimental set-up was developed to reproduce the load transfer in central working condition of the tendons [Figure 1]. The rotator cuff tendons were tested in parallel to the long axis of the supraspinatus in their physiological position. Velocity during the testing amounted to 1,8 mm/min. During the test, the samples were loaded up to 10N and then unloaded to the initial point and loading again until they reached the failure point [Figure 2].

To obtain information about an estimation of stiffness of the tendons, tensile machine registered load [N] and displacement [mm]. Repaired stiffness was defined as the slope of the load divided by displacement in failure point. Failure was defined, as the maximum value of the load needed for damaging the sample permanently. Maximum load for repaired tendons ranged from 15 to 40N.

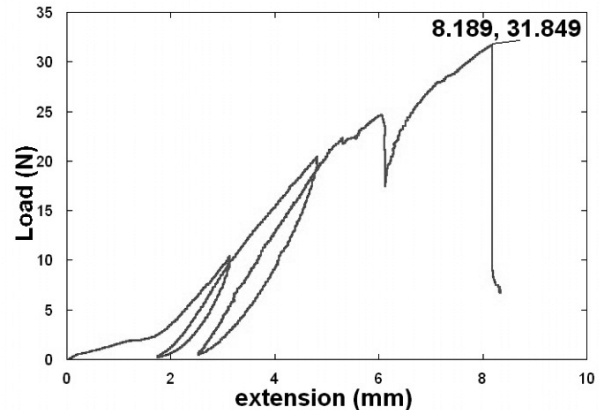


Figure 2 Experimental load-displacement curve recorded by tensile machine .

3. RESULTS

Experimental results have shown that repaired tendon is stronger than the healthy one. In all of the groups the failure loads of repaired tendons were significantly higher than in the corresponding 'control' groups. Mechanical properties of repaired tendons depended on the technique of repair and the time between the damage and repair.

Figure 3 shows the difference between the maximum load of repair tendon and the 'control' tendons in all repaired groups. Results were expressed in terms of % of strength of healthy tendon. Contrast is evident between Group 0, with no treatment applied, repaired naturally, and other groups, which were surgically repaired. Differences between 'control' sample and naturally repaired sample was the highest comparison to other groups. In the case when tension was tested after one month from the tear equals around 50% (Figure 3). In the case of Group 1 where samples were repaired with prolene that difference was nearly halved 20%. It was observed similar value of maximum load accordingly between Group 2 and Group 4 as well as between Group 3 and Group 5. In these pairs of groups, repairing techniques were similar. Group 2 and Group 4 were repaired using medical repairing gel col 1 and in Group 3 and Group 5 mensynchynel stem cell therapy was additionally applied. Mechanical properties after 4 weeks of injury resembled the 'control' sample more in the repairing technique with the use of medical gel coll.

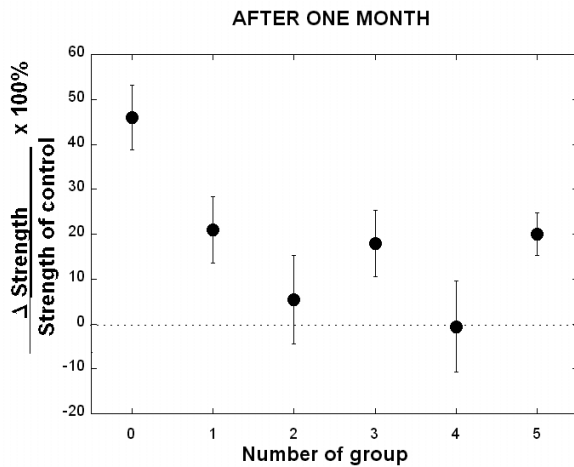


Figure 3 Difference between maximum load in repaired tendon and 'control' tendon in each one of the six repaired groups after 4 weeks of injury. Results were expressed in terms of % of strength of healthy tendon.

After eight weeks from the repair [Figure 4] difference of maximum load between each group is smaller comparing to results after four weeks, and their value was less than 30% in almost all cases. Similarly as in the case of previous samples, after 4 weeks, results in groups cured by medical gel shown akin mechanical properties. After eight weeks, a greater contrast of tensile value was observed in a group where mensynchymel stem cell therapy was applied. Tendons treated with Orthopal gel (Group 5) and mansynachymel stem cell therapy reached values of failure force closer to 'control' tendons than the samples from Group 3 were treated with the same technique, however with a different gel.

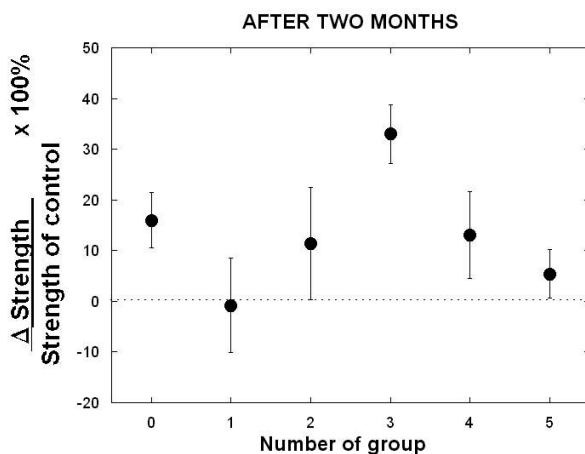


Figure 4 Difference between maximum load in repaired tendon and control tendon in each one of the six repaired groups after 8 weeks of injury.

Repairing technique has also influence on flexibility of repaired tendons. The highest value of stiffness the range of movements are reduced. The results for stiffness [Figure 5] in every repairing group after 4 weeks of injury were appropriate for value of maximum load in that group. Tendon without any treatment (Group 0) was stronger but value of stiffness was also the higher. These results allow this particular research to conclude that flexibility in these cases is considerably low. In other groups, in medically repaired tendons, stiffness resembled the healthy tendons (control samples). Observed similarities in Groups 2 and 4 were due to the application of a similar technique of treatment.

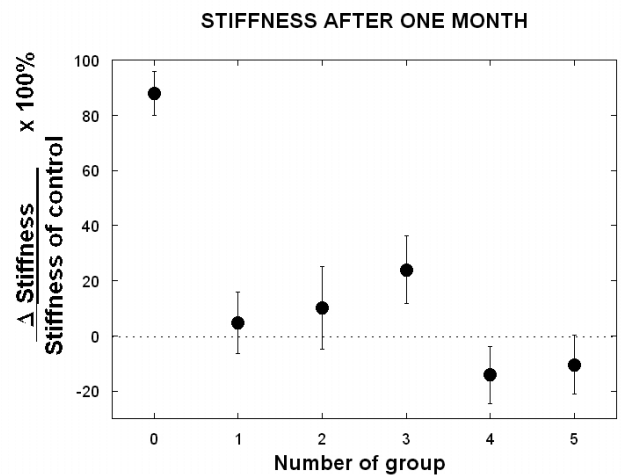


Figure 5 Difference between estimated stiffness of repaired tendon and 'control' tendon in every of six repaired groups after 4 weeks of injury.

4. DISCUSSION

The rat model was already used by others researchers in order to investigate the mechanical strength of different rotator cuff techniques. It was found before that repair tension is related to healing. Tension significantly increased early after detachment and progressively increased with additional time. Previous studies also prove that an increase in tension can have a detrimental effect on the failure properties of healing tissues, and stimulate the production of scar tissues [17].

The biomechanical study on rotator cuff confirmed that biomechanical properties of repaired supraspinatus tendon depend on the technique of repair. It was found that the repaired tendons are generally stronger than the healthy ones and that difference was greater after four weeks from the injury than after the period of eight weeks from injury. Largely caused by accumulation of tissues around the tear. When tendon is injured the healing process continues through an inflammatory

phase, followed by migration of the cells to the injured area. These cells fill the wound with a disorganized collagen matrix that is eventually remodeled to restore the functional linear collagen bundles and tensile strength of the original tissue [16]. Accumulated cells make tendons stronger but at the same time they constrict a range of movements, which makes tendon less flexible. Our study shows that tendons left without treatment (Group 0) needed to reach a higher value of load for the failure to occur; but concurrently the stiffness of supraspinatus was higher. Limitation in the range of movement within that group was noticeably lower.

Current studies show that the failure load is dependant on the structure type and the applied treatment therapy. Early results (after four weeks) affirm that properties were much different to those, which had undergone the treatment twice in length. The results from Group 5 appear promising after 8 weeks from the fracture. In this particular group apart of OrthADAPT, mesenchymal stem cells therapy was also applied. Several animal case studies were carried out in order to evaluate the efficiency of MSC local injection in tendon repair. Biomechanical analysis revealed an improvement of biomechanical properties, tissue architecture and functionality of the tendons after injury where the treatment was supported with the use of MSCs-collagen composites. After 12 weeks of injury, maximum stress for the repaired tissues was inconspicuously higher than the value of the healthy tendons [18].

To summarize, this work shows that an estimate strength and flexibility of tendons depends on a number of significant factors, which have an influence on the results of repair. The greatest influence however, is that of the technique of repair as well as the time from injury until treatment, quality of tendons and biological healing response. These particular parameters should be considered to a greater extent in the future research. In the outcome gave the results of highest estimate stiffness but lower range of flexibility which reduce shoulder function and decreasing limits of movements.

5. ACKNOWLEDGMENTS

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