# Stem cells and tendinopathy: state of the art from the basic science to clinic application

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## Summary

**Management of tendinopathies and tendon rupture is challenging.**
**In the last few decades, several emerging strategies including tissue engineering with mesenchymal stem cells have been proposed to enhance tendon healing. They hold the promise to yield more successful outcomes for the management of patients with tendon pathology.**
**Current *in vitro* studies support the application of these cell-based therapies for the regeneration of tendon tissues. However, these cell-based strategies have been investigated only in pre-clinical studies and the role of stem cells needs to be confirmed. We performed a review of the literature to focus on actual knowledge and the future perspectives of stem cells for tendon regeneration and tendon engineering.**

*Key words: tendon, mesenchymal stem cells, tendon stem cells, tendinopathy.*

## Introduction

The aetiology of tendinopathy remains unclear, and many causes have been theorised. Hypoxia, ischaemic damage, oxidative stress, hyperthermia, impaired apoptosis, inflammatory mediators, fluoroquinolones, and matrix metalloproteinase imbalance have all been implicated as mechanisms of tendon degeneration[1, 2].
It remains unclear whether different stresses induce different responses. Active repair of fatigue damage must occur, or tendons would weaken and eventually rupture. The repair mechanism is probably mediated by resident tenocytes, which 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[3, 4]. Tendon damage may even occur from stresses within physiological limits, as frequent cumulative microtrauma may not allow enough time for repair[5, 6]. Microtrauma can also result from non-uniform stress within tendons, producing abnormal load concentrations and frictional forces between the fibrils, with localised fibre damage[3, 7].
In the last few decades, several emerging strategies including tissue engineering with mesenchymal stem cells (MSC) have been proposed to enhance tendon healing. They hold the promise to yield more successful outcomes for the management of patients with tendon pathology. MSCs represent an archetype of multipotent somatic stem cells with ability to differentiate along a variety of cell lineage[8]. They could be used for tissue engineering and regenerative medicine[9].
The notion of stem cell is a developing concept[10]. Stem cells are undifferentiated cells with ability of self-renewing and differentiating in progenitor or precursor cells. The latter are committed cells for a specific cell lineage, but are not able to self-renew[11, 12].
Although MSCs were originally isolated from bone marrow[13, 14], similar population have been reported in other tissues. Human MSCs have been isolated from adipose tissue[15], umbilical cord[16-18], placenta[19], peripheral blood[20, 21], connective tissues of the dermis and skeletal muscle[22]. MSCs from different sources have shown to express similar surface markers, self-renewal capacity and multipotent differentiation properties[23, 24].
MSCs reside in practically all organs and tissues[25] in a specialized microenvironment composed of extracellular matrix (ECM), cells and cytokines called "stem-cell niche"[26]. The niche maintains a balance of quiescence, self-renewal and cell-fate commitment[27].
A stem cell population has been recently identified in human tendons, residing in a unique tendon ECM niche[27]. Tendon stem cells (TSCs) have multi-differentiation and self renewal potential[27-29]. Such a stem cell population could be involved in tendon homeostasis, remodelling, and repair, by ensuring replacement of mature cells lost, or in the pathogenesis of tendinopathy, as this tendon disorder is associated with chondroid and fatty degeneration, and ossification[1, 30-32].
Regenerative medicine is a transdisciplinary field that combines advances in biology, chemistry, clinical medicine, engineering, and material sciences[33]. The ultimate goal consists in restoring the natural healing process that eventually leads to regeneration of damaged tissues and organs[33].
The rising number of investigations on stem cells and tissue engineering for the regeneration of musculoskeletal tissues

had opened new perspectives also in the field of stem cells and tendon regeneration. We performed a review of literature to focus on actual knowledge and the future perspectives of the stem cells for the management of tendinopathies.

### Basic science

TSCs were recently demonstrated to answer differently to higher and lower mechanical stresses. In fact low mechanical stretching promote differentiation of TSCs into tenocytes while high mechanical stretching promote differentiation of TSCs into other lineages (adipogenic, chodrogenic and ostegenic). This could explain because high mechanical strains are associated with tendinopathies[29].
The use of stem cells for the management of tendinopathy and tendon repair has been recently focused on showing positive results.
*In vitro* studies have shown the likely for stem cell therapies to provide tendon regeneration rather than repair of tendon tissue[34]. Regeneration involves slow replacement of tissues with identical tissue. It occurs readily in the embryo, hardly at all in the neonates and is never observed in adults[35]. In contrast, repair is a more rapid process, involving the inflammatory cell cascade, followed by matrix deposition and then a remodeling process which attempts, in part, to regenerate damaged tissue in the adult[36].
Injected MSCs were demonstrated to provide good histological scores in the management of collagenase inducted tendinitis lesions in equine flexor digitorum superficialis tendons in horses[37].
Bone marrow mesenchymal cells (cBMSC) and bone marrow mononuclear cells (BMMNC) showed to be efficacious in regenerating tendon tissue after collagenase-induced tendinitis in a sheep model, demonstrating a higher type I collagen expression compared with control tendons[38].
However there is no consensus on which kind of stem cells provide the best results in tendon repair. In fact the differentiation and proliferation potential of stem cells can vary depending on their origins[34]. For example embryonic stem-cells could result in a teratoma formation, while bone marrow stem-cells may form ectopic bone. For this reason the source from which the stem cell are isolated is important and should be accurately selected[39].
Tendon stem cells have been hypothesized to have a crucial role in the development of calcifying tendinopathy due to the erroneous differentiation of TSCs to chondrocytes or osteoblasts. For this reason it was hypothesized that the re-direction of the differentiation of resident TSCs or supplementation of MSCs programmed for tenogenic differentiation may be appealing targets for the treatment of tendinopathy in the future[40].
These results thus suggest that TSCs may be a promising therapeutic cell source for tendon regeneration and tendon tissue engineering.

### Clinical application

In literature there is a lack of clinical studies on the management of tendon ruptures or tendinopathies with stem cells. In the cellular treatment of tendon disorders, a small number of clinical trials are being currently undertaken to assess the safety and efficacy of differing cell lines to treat tendinopathy.
BMSCs have been shown effective in the management of superficial digital flexor tendon injuries in horses; BMSCs were inoculated in the injured tendons leading to lower of re-injury rate compared with the re-injury rate obtained with the conventional non cellular based management[41, 42].
Lacitignola et al.[43] showed in an *in vivo* collagenase-induced tendinopathy study that autologous BMSCs could be injected intratendinously producing effective tendon regeneration.
Pacini et al.[44] showed recovering of normal activity in horses affected by superficial digital flexor tendinopathy managed with targeted intralesional injection of BMSCs.
Also adipose derived stem cells were showed to be effective in the treatment of equine tendinopathies leading to normal horse activity recovery[45].
As MSCs are now used as a therapeutic strategy in race horses to treat flexor digitorum superficialis tendinopathy it should be interesting to translate it in human tendon patology.
There is only one clinical study performed on human subjects showing that inoculation of bone marrow mononuclear cells in tendinopathic patellar tendons has good mid-term clinically and ultrasound results[46].
As demonstrated by these preliminary studies, management of tendinopathies with stem cells is promising even though more clinical studies are needed to validate this treatment approach.

## Conclusion

Stem cells are promising candidate for the management of tendinopathies and tendon rupture. However, these cell-based strategies have been investigated only in preclinical studies and the role of stem cells needs to be confirmed. Further research is required to identify mechanisms involved in tendon regeneration and in survival, proliferation, and differentiation of stem cells. Tendon tissue regeneration represents a biological alternative for replacement of large tissue loss after severe damage, based on combination of adult or embryonic stem cells, factors or stimuli, and biomaterials. This technology appears to be promising and will probably grow to be an important therapeutic option in musculoskeletal regenerative medicine.

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