A NEW GENERATION OF TENOCYTES GROWN IN-VITRO TO EFFECT TENDON REPAIR AND REGENERATION

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Short Abstract
Healing of tendon injuries in the horse is achieved by using a unique, non drug based, culture system, using New Generation/Therapy Cells grown from tenocytes taken from the nuchal ligament of the affected horse. These biopsied cells are multiplied in the laboratory and then these New Generation/Therapy Cells are transplanted back into the damaged tendon to effect repair and regeneration.

Introduction
Tendon injuries can severely limit a horse's performance, ending careers and sometimes cutting short a horse's life. To date, no really effective treatment has been available to treat, what is, an all too common injury.

Limited success has been obtained through the use of mesenchymal "stem cells". Although this technology is promising, scientists are still faced with the problem of imparting cellular memory to these cells.

In recent years our collaborative research team has established a unique, non drug based methodology that enables cells to re-enter the mitotic cell cycle. With this treatment around 100 tenocytes harvested from the affected individual are grown to 20 million tenocytes in vitro for transplantation back to the donor (auto transplantation). These cells (New Generation/Therapy Cells) are dedicated to their role as tenocytes and, as such, repair the tendon rupture through regeneration of the tendon within the lesion.

To date, all lesions treated have been fully in-filled and clinical results have been very encouraging.

Materials & Method
The method we have developed uses a biopsy taken from a horse with a ruptured tendon (discrete core lesion). To ensure there is no further damage caused to the tendon, samples are taken from the nuchal ligament using a specialized spring loaded biopsy technique under local anaesthesia. Two samples are taken. The samples are then transported to the laboratory where cellular growth is stimulated. In short, cells are encouraged to adhere to the side of a culture dish and are then placed in a modified "hanging drop" culture system, leading to the division/multiplication of adult tenocytes. After approximately 28 days the tenocytes are harvested using a Trypsin/EDTA incubation technique. On average, approximately 20 million cells are harvested and these are injected back into the core tendon lesion.

Results
To date, 45 horses with discreet core tendon lesions have been treated in Australasia and North America. No negative effects have been observed in any horse, at the donor site, post harvest. Using the technique described, tenocytes have been successfully harvested and grown from all patients. Of considerable interest to the authors is that tenocytes grown in laboratory conditions show strong linear cellular alignment in a tendon like fashion.

In all transplants, tenocytes have been introduced into the core lesion, also with no untoward sequelae. The reintroduction procedure is normally achieved standing, using sedation and a ring block, although sometimes general anaesthesia is necessary. All lesions are monitored via ultrasound and all have shown complete infilling. No overgrowth of cells/tendon has been observed in any clinical case. It is the authors' belief that cellular growth is limited by spatial compaction of the lesion.

Of the 45 horses treated thus far (all of whom had previously been in full competition) 32 have been fully rehabilitated, enabling them to compete again. Of this 32, 27 have resumed full competition (84%) without further injury. 2 of the remaining 5 have not raced due to tearing of an area adjacent to the original lesion and 3 were unable to compete due to an unrelated complaint. The other 13 horses are still being rehabilitated. All treated horses have tenocytes stored in liquid

nitrogen in case any further treatment is needed for that individual.

Discussion
Post treatment results, expressed as return to full athletic performance, have been extremely encouraging. We have not rushed rehabilitation and we feel this has assisted the positive clinical outcome.

To the authors' knowledge this is the first time auto transplantation has been performed in any species using dedicated adult cells that have been re entered into mitosis and created a New Generation/Therapy Cell. We believe that this represents an exciting development in regenerative medicine with implications that could be very wide ranging.

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