OVEREXPRESSION OF NITRIC OXIDE SYNTHASES IN TENDON OVERUSE

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Introduction:
Tendon disorders, including rotator cuff, are extremely common and represent a major clinical problem. Our understanding of the etiology and pathomechanism of this condition is limited and current treatment strategies are not particularly effective. Mechanical overload of rotator cuff tendons, as a result of repetitive microtrauma from overuse, has been proposed as the primary etiologic factor in rotator cuff tendinopathy. Evidence of two contrasting processes, tissue degeneration and repair, are present in pathological samples of rotator cuff tendons when compared to normal tendons. Nitric oxide (NO), a free radical produced by nitric oxide synthases (inducible-, endothelial – and neuronal-NOS) is a potent regulator and stimulator of biological processes. Overproduction of NO can trigger destructive processes, while under other circumstances NO may play a positive role in tissue healing. The aim of our study was to determine whether NOS isoforms are expressed in rotator cuff tendon as a result of chronic overuse.

Methods:
Supraspinatus tendon overuse was modelled by using an established repetitive exercise protocol of rats, which consists of treadmill running. Twelve animals were subjected subjected to chronic exercise running at 17 m/min, on a 10 degree decline, for one hour per day, five days a week for four weeks. A group of twelve rats of the same strain, gender, age and weight subjected to normal cage activity were used as controls. Supraspinatus tendons from both groups of animals were collected, RNA was extracted and processed for competitive RT-PCR using NOS isoform specific primers. PCR products were fractionated and subjected to densitometric analysis, which allowed semiquantitative estimation of the mRNA expression of all three NOS isoforms. Data are presented as mean +/- SEM. Differences among experimental groups were assessed by un-paired Student’s t test. The level of statistical significance was set at p < 0.01.

Results:
The mRNA expression of all three NOS isoforms increased in the supraspinatus tendons as a result of its overuse (Figure 1.). iNOS mRNA expression demonstrated a four fold (p < 0.01) increase in the overused supraspinatus tendons (4.2x10^-3 +/- 1.3x10^-3 attomol) when compared with the cage activity controls (1.1x10^-3 +/- 0.5x10^-3 attomol). eNOS mRNA expression also increased by nearly four fold in the overused supraspinatus tendons (152x10^-6 +/- 33x10^-6 attomol) when compared with supraspinatus tendons from the cage-confined controls (p < 0.01). There was an increase in nNOS mRNA expression in the overused rat tendons (6.0x10^-3 +/- 3.8x10^-3 attomol) compared with the controls (2.0x10^-3 +/- 0.4x10^-3 attomol), which increase did not reach statistically significant level (p = 0.08).

Discussion:
1. This study is the first to show that NOS isoforms are upregulated in rotator cuff tendon as a result of chronic overuse.

2. It is interesting to note that very low, but detectable amounts of all three NOS isoform mRNAs are present in the normal supraspinatus tendon.

These data suggest the involvement of nitric oxide in the response of tendon tissue to increased mechanical stress. Therapeutic strategies for modulating nitric oxide pathways may provide potential novel approaches for the treatment of rotator cuff tendon disorders.

References:

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