The effects of the chemo-protective agent 3H-1,2-dithiole-thone (D3T) on dystrophic pathology in an animal model of Duchenne muscular dystrophy

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Duchenne Muscular Dystrophy (DMD) has a prevalence of 1~3500 live male births globally, with affected patients prematurely dying in their early 30s due to respiratory and cardiac failure. The primary pathogenesis of DMD is due to the absence of the structural protein dystrophin from the muscle surface membrane (sarcolemma) (Emery, 1993). The absence of dystrophin leads to the loss of membrane stability causing muscle fibres to become more susceptibility to contraction-induced damage (Danowski *et al.*, 1992), resulting in a chronic inflammatory state accompanied by a high level of oxidative stress (Tidball & Wehling-Henricks, 2005). 3H-1,2-dithiole-thone (D3T), currently in clinical trials for the treatment of cancer, activates the transcription factor Nuclear Factor-Erythroid 2-related factor 2 (Nrf2), which regulates the induction of antioxidant genes in a number of tissues including skeletal muscle (Karuri *et al.*, 2006). D3T has been shown to improve the pathology of a number of chronic inflammatory conditions such as sepsis (Thimmulappa *et al.*, 2006) and chronic obstructive pulmonary disease (Hart *et al.*, 1998). We tested the hypothesis that D3T would ameliorate the dystrophic pathology in mdx mice. D3T was administered to 12 week old mdx mice *via* oral gavage, (6μL/g in sucrose vehicle) 3 times a week for 4 weeks. At the end of the treatment period, mice were anaesthetized (60 mg/kg sodium pentobarbitone), and muscles were excised for biochemical and histological analysis.

In dystrophic muscle, D3T resulted in the up regulation of mRNA expression of Glutathione reductase and Aconitase, which are key enzymes involved in the production of glutathione (GSH). D3T also increased the active form of GSH in dystrophic *tibialis anterior* (TA), diaphragm and *soleus* muscles as measured by a GSH assay (Cayman Chemical, Ann Arbor, Michican, USA). D3T treatment resulted in a significant reduction in the mRNA transcript levels of pro-inflammatory cytokines TNF-α and IL-1β in the diaphragm and TA muscles of mdx mice. Despite the significant reduction in markers of inflammation and upregulation of the antioxidant system, histological analysis revealed no significant effect on gross cell morphology and no reduction of pathological collagen formation.

In conclusion D3T, already in phase II clinical trials in humans for the treatment of cancer, significantly increases the antioxidant defence system and reduces key markers of inflammation in dystrophic mice, indicating that targeting Nrf2 is a potential therapeutic approach for the treatment of DMD.

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