USING LABORATORY MODELS TO DEVELOP MOLECULAR MECHANISTIC TREATMENTS FOR DUPUYTREN'S DISEASE

Martin C. Robson, M.D. and Wyatt G. Payne, M.D.
Institute for Tissue Regeneration, Repair, and Rehabilitation, Bay Pines VAHC, Bay Pines, FL
Division of Plastic Surgery, University of South Florida, Tampa, FL

HYPOTHESIS: Transforming growth factor beta (TGF-B) has been implicated in the pathobiology of progressive fibrotic disorders. Modern laboratory models can demonstrate the role of TGF-B in Dupuytren's Disease and molecular mechanistic treatments proposed.

METHODS: An *in vitro* model, the fibroblast-populated collagen lattice (FPCL), and an *in vivo* model, Dupuytren's affected palmar fascia explanted onto an athymic "nude" rat, were used to evaluate the hypothesis. Collagen lattices were populated with fibroblasts harvested from Dupuytren's affected palmar fascia or with normal palmar fascia fibroblasts. Gel contraction was measured daily for five days. The supernatant obtained from the culture medium was analyzed by human TGF-B2 immunoassay. Identical FPCLs were established and had tamoxifen 8umol/ml added.

Specimens of Dupuytren's affected palmar fascia were explanted onto the "nude" rats and perfused with either TGF-B2, TGF-B2 antibody, or saline as a control. Biopsies were harvested from the explanted tissues at 30 and 60 days. Part of the biopsies were used for immunohisto-chemistry and part placed in tissue culture for cell kinetics. Histology, trichrome staining, and immunohistochemistry for TGF-B2, collagen I, and collagen III were performed on the biopsy sections. Total protein, DNA synthesis, and cell counts were evaluated from the cell cultures.

The numerical data between the groups in the various experiments were compared using a one-way analysis of variance. An all pairwise comparison procedure (Tukey's test) was performed for statistically significant differences identified by ANOVA. Significant differences were determined to have a P value <0.05.

RESULTS: FPCLs populated with fibroblasts from Dupuytren's affected palmar fascia contracted significantly more than FPCLs populated with normal palmar fascia fibroblasts ($P<0.001$). In tamoxifen-spiked FPCLs, lattice contraction was less than in untreated FPCLs ($P<0.05$). The amounts of TGF-B2 measured in the FPCL supernatants were also less when fibroblasts were exposed to tamoxifen ($P<0.05$).

Perfusion of explanted Dupuytren's tissue by TGF-B2 upregulated collagen I and collagen III from the explants when compared to vehicle control ($P<0.001$). Perfusion with antibody prevented this upregulation when compared to vehicle control ($P<0.001$). Cell cultures derived from fibroblasts obtained from the explants perfused with TGF-B2 increased DNA synthesis, protein production, and fibroblast kinetics. Apoptotic protein assessment suggested that TGF-B2 decreases apoptosis, allowing a decrease in programmed cell death.

SUMMARY:

- Etiology of Dupuytren's Disease is unclear.
- Dupuytren's Disease is pathobiologically related to other progressive fibrosing disorders.
- TGF-B plays a role in the pathogenesis of Dupuytren's Disease.
- Agents that abrogate or neutralize TGF-B may provide novel treatment for Dupuytren's Disease.