MESSAGE FROM THE GRAYSON-JOCKEY CLUB RESEARCH FOUNDATION

RESOLVING JOINT INFLAMMATION FROM INSIDE

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OSTEOARTHRITIS is a major cause of joint disease, leading to poor performance and early retirement of horses; a heavy economic and emotional burden to the equine industry. Current treatments provide limited recovery of joint function, creating an urgent need for more efficient therapies. Development of new treatments requires a more comprehensive understanding of the mechanisms causing OA.

One fundamental characteristic of joint disease is sustained, low-grade inflammation. Cells called macrophages are the main drivers of joint inflammation; however, these complex cells can both incite and resolve inflammation, depending on the circumstances. Macrophages in the synovial (joint) membrane and fluid are essential in promoting joint health by clearing aggressors, by secreting key molecules required for optimal joint function, and by forming a shield that protects tissues undergoing repair, similar to a wound scab.

However, when the amount of tissue damage overwhelms these housekeeping functions, macrophages stimulate inflammation as a means of recruiting more cells to cope with increased demands for repair. If this response is efficiently accomplished, macrophages then resolve the inflammatory process, returning the joint to a healthy state.

The sequence of studies described herein focuses on understanding the dual function of macrophages in driving and resolving joint inflammation, and ultimately in harnessing their therapeutic potential to treat joint disease.

The first study, funded by the American College of Veterinary Surgeons Research Foundation, compared the response of macrophages in normal and OA-affected joints. We observed that macrophages had a similar response in both healthy and OA-affected joints but were markedly activated in arthritic joints showing obvious, severe inflammation. Synovial fluid from OA joints had lower levels of a protein that recruits macrophages from the bone marrow following injury (SDF-1), and of a macrophage-derived anti-inflammatory protein called interleukin (IL)-10.

Our results suggest macrophage recruitment and associated anti-inflammatory mechanisms are impaired in OA-affected joints, preventing joint inflammation from being resolved, and that re-establishment of these functions could greatly aid in the treatment of OA.

Macrophages in the bone marrow (BMNC or bone marrow mononuclear cells) are used in people to treat inflammation in several chronically inflamed tissues and produce molecules essential for joint health, such as IL-10. Our second study, funded by the Grayson-Jockey Club Research Foundation, investigated the effects of injecting each horse's own (autologous) BMNC into normal and inflamed joints using an experimental model of joint inflammation in six Thoroughbred horses.

Inflamed joints treated with BMNC showed dramatic visual and measurable markers of improvement, with increasing macrophages and IL-10 in the joint fluid, which remained low in joints treated with placebo.

The results of this study confirmed that increasing macrophages in inflamed joints by BMNC injection recovered the macrophage- and IL-10-associated mechanisms required to resolve joint inflammation that are impaired during OA.

We conducted a third study, also funded by the GJCRF, that further investigated how BMNC (same group of horses as above) responded to laboratory culture in fluid from their normal and inflamed joints. Normal and inflamed joint fluid induced macrophages from BMNC to develop similar responses that precisely combined pro- and anti-inflammatory mechanisms, both essentially required for tissue repair and recovery of joint health. Such a balanced response was proportional to the inflammatory challenge. Macrophage proliferation and secretion of IL-10 and IGF-1 (both essential for cartilage metabolism) were highest for cultures in fluid from inflamed joints. BMNC cultured in normal or inflamed synovial fluid were ultimately comparable to cells native to normal joints, suggesting that the healthy state was recovered in inflamed joint fluid.

These observations suggest robust proliferation of BMNC in inflamed joint fluid provides a "bigger army"

to cope with the intensity of inflammation, allowing inflammation to be resolved and homeostasis (health) to be recovered.

Finally, a pre-clinical pilot study supported by the Virginia Horse Industry Board and the Veterinary Memorial Fund followed 18 Thoroughbred horses with naturally occurring OA treated with either BMNC, triamcinolone (commonly used corticosteroid), or placebo. Horses were evaluated for lameness (subjectively and using Lameness Locator®) and synovial fluid analysis at 0, 7, and 21 days post-injection. BMNC injection was performed without adverse effects, with BMNC-treated horses being the only ones to show improved lameness during the study period.

In summary, BMNC injection is a safe, natural, point-of-care therapy that provides a promising means to recover and sustain macrophage-associated effects required for joint integrity while preserving mediators of joint health often impaired by conventional therapies.

Currently, we are delving deeper, using cutting-edge "omics" techniques to define the molecular products of macrophages that drive resolution of inflammation and recovery of joint health.

Our long-term goal is to harness these natural processes to develop new treatments that stand to benefit thousands of OA patients of all species. BH