# Efficacy of Low Doses of Type II Collagen in a Rat Rheumathoid Arthritis Model: Relevance of Oral Tolerance

L. Di Cesare Mannelli[1], L. Micheli[1], D. Baptista De Souza[1], L. Cinci[1], M. Zanardelli[1], C. Ghelardini[1]

[1]Dept. of Neuroscience, Psychology, Drug Research and Child Health - Neurofarba - Pharmacology and Toxicology Section, University of Florence, Florence, Italy

A key feature of the intestinal immune system is its ability to protect against infection while avoiding the development of destructive inflammatory responses to the normal microbiota. Furthermore, tolerance generated at mucosal surfaces can translate to a more generalized systemic tolerance - a characteristic of great therapeutic potential. Oral tolerance is a form of peripheral immune tolerance in which mature lymphocytes in the peripheral lymphoid tissues are rendered non functional or hyporesponsive by prior oral administration of antigens. The immunologic response to orally administered antigens occurs in the gut-associated lymphoid tissue (GALT) where dendritic cells take up antigens and present them to T cells to generate regulatory T cells (Treg), which induce systemic immune tolerance.

This therapeutic concept may be applied to painful joint diseases as rheumatoid arthritis characterized by immune disregulation or tissue degeneration. For these purposes the effect of low doses of native porcine collagen Type II was evaluated in the Complete Freund Adjuvant (CFA) rat model related to rheumatoid arthritis. 1 mg $kg^{-1}$ collagen per os daily administered for 14 days after injury was able to fully prevent CFA-induced pain threshold alteration measured by a mechanical noxius (Paw pressure and Pam test) or non-noxious (Electronic Von Frey test) stimulus. Collagen treatment was also able to improve hind limb weight bearing alterations (Incapacitance test).

Collagen treatment was able to decrease the plasmatic levels of the inflammatory cytokines IL-1, IL-6 and TNF-α. Moreover inhibitory cytokines, such as transforming growth factor (TGF)-β and IL-10, which play an important role in Treg generation, were modulated both in the plasma and in the intestinal tissue in a time-dependent manner. Finally, a decrease of C2C (a fragment of degraded type II collagen) and CPII (type II procollagen carboxy-propeptide) level ratio and morphological measures, suggest a collagen-dependent decrease of structural joint damage.

The present data propose the role of low dosed collagen in articular pain treatment and deep inside the mechanism of oral tolerance. It is hoped that uncovering the pathways used by the intestinal immune system to prevent immune pathology may direct therapeutic approaches to a broad range of autoimmune and inflammatory conditions.