Internship proposition

(One-page max)

# Master 2 GP Medicine 4R (Repair, Replace, Regenerate, Reprogram)

**Lab:** Inserm UMRS 1229-RMeS Regenerative Medicine & Skeleton

**Team:** REJOINT

**Name and position of the supervisor:** GUIHO Romain, Prof. associé

**Email of the supervisor:** romain.guiho@univ-nantes.fr

**Candidate (if internship filled):** Liane Fontaine

**Title of the internship: Deciphering Senescence-associated cellular Communications in an in vitro murine model of intervertebral disc degeneration**

**Summary of the internship proposal:**

Intervertebral Disc Degeneration (IVDD) is characterized by the degradation of the intervertebral disc, associated with loss of functions, disability, and ultimately pain. The pathophysiology of IVDD remains not yet fully understood and only palliative treatments exist to limit pain. In the past decade, several studies have suggested that the senescence of the Nucleus Pulposus (NP) and the Annulus Fibrosus (AF) cells could play a possible role in the pathogenesis of IVDD, linking their senescent-associated secretory phenotype (SASP) to the onset and perpetuation of the disease.

Senescent cells (SnCs) seem to accumulate in all tissues during aging, while maintaining context-dependent specificities related to their original cell type. Latest evidence indicates that a systemic elimination of SnCs (a process so-called 'senolysis') could delay the spontaneous disc degradation in aged mice. However, this senolysis is only an indirect proof of disc cells senescence contribution to the degeneration because the systemic effect of the approaches used cannot rule out the involvement non-disc SnCs (e.g., immune system, ligaments, bone). In this context, our preliminary data highlight the possible role of SnCs within the chondral and subchondral endplates as early players of the age-related disc degeneration. These observations challenge the established disc-autonomous model of degeneration, also suggesting a potential senescence propagation between the different cellular compartments of the disc and its surrounding environment. Deciphering how all the different cellular compartments of the disc and its environment communicate in the context of SnCs occurrence would help determine potential cellular and molecular initiators and could uncover soluble agents that can be specifically targeted as a new therapeutic option to delay global disc degeneration.

Therefore, this internship project aims to develop an in vitro platform to study these inter-compartment SnCs-dependent communications, in order to unravel key cellular partners and connection mechanisms. We propose to:

1. Establish a robust in vitro model of cellular senescence for disc cells. Nucleopulpocytes, AF cells, chondrocytes from endplates and mesenchymal stem cells differentiated into the osteoblastic lineage will be isolated from rat tails. Cellular senescence will be induced and evaluated (e.g., immunostainings, flow cytometry for cell cycle analysis, SA-β-Gal staining, qRT-PCR).
2. Develop a mini-coculture platform for exploring senescence propagation in vitro. SnCs will be comprehensively characterized in co-culture. Senescence phenotype will be confirmed, and relevant activated pathways identified using RNAseq. Related analysis will be performed using Rstudio.

**Profile(s) linked to the project:**

■ Experimental Biology *(Recherche expérimentale)*

☐ Clinical Research *(Recherche clinique)*