INVESTIGATING THE DYNAMICS OF THERMOGENIC PROCESSES IN AGING

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XXXIXth Cycle - A.Y. 2023/2024

Endothermy is one of the most significant events in vertebrate evolution: unlike reptiles, amphibians and fishes, which rely on environment temperature, mammals and birds can generate heat by endogenous mechanisms. Avian and placental mammalian lineages independently developed the ability to produce heat energy, or *thermogenesis*, through separate evolutionary processes. Notably, mammals developed a specific mechanism to perform non-shivering thermogenesis by synthesizing the mitochondrial uncoupling protein 1 (UCP1) which exploit electron transport chain reactions. The dissipation of energy through heat production occurs in specialized thermogenic adipose tissues (TAT) such as subcutaneous white adipose tissue and interscapular brown adipose tissue. TAT are composed of an intricate network of cells including mature adipocytes, fibroblasts and immune cells. Interactions between adipocytes and immune cells are essential to maintain metabolic homeostasis. Of note, TAT activity progressively declines during aging and type 2 diabetes. In parallel, progressive low-grade inflammation is a pervasive risk factor for developing age-associated diseases. This leads to speculate that TAT might represent pharmacological targets to fight the metabolic and inflammatory status of aging. However, it is still unclear how environmental stresses and immune failure accelerate the loss of thermogenic potential, as well as little is known about how TAT reduction contributes to systemic decay and whether limiting the thermogenic tissues contraction can prevent age-related diseases.

The *single-cell RNA transcriptomics*, a novel Next Generation Sequencing technology, will be used to unravel the complexity of brown adipose tissue. Through the quantification of the messenger-RNAs transcribed, this approach makes it possible to measure the genes expression at the individual cell level, reconstruct heterogeneous cell communities, compare transcriptional profiles and identify cell-type specific response to perturbations. This technique will be used to characterize the transcriptome of cells involved in thermogenic processes, both in aging and under environmental stressors (cold exposure, food intake, pollutants). scRNAseq analysis will focus on metabolic pathways activation (or repression) in aging adipocytes, highlight the role of immune cells in pathophysiological dynamics and also identify pivotal aging-related genes.

The study timeline can be summarized as follows:

- The first year will be mainly dedicated to preliminary data-mining and in-silico analysis of studies available from many biological databases such as NCBI Gene Expression Omnibus;
- Single-Cell RNA sequencing of original data from elderly subjects and murine models of aging will be performed throughout the second year;
- In the third year, complementary analyses and wet-laboratory experiments will be conducted for evidence validation.