Burning fat to heat

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Global prevalence of obesity has nearly tripled since the 1970s. According to the latest WHO Global Health Observatory data, over 39% of adults, amounting to 1.9 billion people, were overweight, with at least of 2.8 million people dying as a result of obesity (*1*–*3*). Like COVID-19, the global spread of obesity is an ongoing pandemic, albeit a non-communicable one. Overabundance of adipose tissue is the direct clinical symptom of obesity. Contrary to popular belief, adipose tissue is not only a simple organ for energy storage, but also an important thermogenic and endocrine organ, therefore, a functional adipose organ is key to metabolic health (*4*). My research aims to systemically study the function and regulatory circuits of adipose tissue from a health perspective.

Adipose tissue can be divided into functionally and morphologically distinct white adipose tissue (WAT) and brown adipose tissue (BAT). Unlike lipid storage in WAT, BAT generates heat via non-shivering thermogenesis. Recent studies have demonstrated the presence of active BAT in adult humans and can be activated by cold exposure or adrenergic receptor agonists (*5*–*7*), which ignite the possibility to restore energy balance in overweight individuals (*4*). However, only a small portion of adults possess active BAT, restricted this potential therapeutic approach from being widely applicable.

To gain insights of the regulatory circuits of human brown fat, we reasoned that the certain clinical parameters are distinct among individuals with active BAT. Since ^{18}F-FDG PET/CT scans have been well established to detect active BAT in humans (*5*–*7*), we conducted a retrospective analysis of PET/CT scans from 2007 – 2015 (*8*).

Identifying an epigenetic programming of brown adipose tissue

Clinical PET/CT scans of 8,440 individuals revealed an intriguing correlation between the season of conception and the propensity to form active BAT, for example, individuals with active BAT were more likely to have been conceived in the colder period of the year (*8*). Given the retrospective nature and the potential confounders, causality could not be inferred.

To verify such a correlation and find possible causality, we employed mouse model systems to investigate the effect of parental cold exposure (CE) in their offspring's BAT activity. Strikingly, CE before conception or during gestation resulted in higher thermogenic activity in the offspring. Further investigation found the effects of parental CE is transmitted though the paternal lineage via sperm revealed by an in vitro fertilization system, suggesting an intergenerational regulation axis connecting ambient temperature – germline - BAT. Our focus subsequently shifted to studying this unknown mechanism of cold induced epigenetic programming in BAT.

Using a series of adrenergic agonists and antagonists analyses as well as a brown cell depletion mouse model, we identified that either genetically depleting brown adipocytes or pharmacologically inhibiting ADRB3 (β3-adrenergic receptor) erased the impact of paternal CE in the offspring, which demonstrated the effect of paternal CE on thermogenesis was mediated through the activation of brown adipocytes through ADRB3. Because the phenotypic changes of paternal CE offspring are mediated through the sperm, we performed whole-genome bisulphite sequencing of sperm and transcriptomic analyses of BAT. Besides distinct clustering of each group in principal component analysis at global level, integrated analysis of DNA methylation and RNA sequencing data suggested enhanced neuronal development in BAT, which was functionally confirmed by increased release of norepinephrine in paternal CE offspring. These results indicate that CE induces an epigenetic programming of the sperm such that the offspring harbor hyperactive BAT and an improved adaptation to hypothermia. Such mechanisms formed the rationale designing therapies and personalized strategies to induce BAT functionality to counteract obesity and comorbidity diseases (*9*).

A novel type of adipocyte and communication

Radical morphological and functional changes of BAT observed under different conditions (*10*) suggest that adipocytes can interconvert from one phenotype to the other (*11*). To understand such cellular dynamics and heterogeneity, analyzing adipose tissue at a single cell resolution presents a straightforward approach to address this biological question (*12*, *13*). On the flip side this approach is technically challenging for the lipid laden adipocytes, which are highly variable in size, of low density and delicate nature (*14*). To overcome such limitations, I developed a fat specific single nucleus RNAseq method that is compatible with frozen archived adipose tissue samples in a clinical setting (*15*).

By resolving cellular compositions and dynamics of brown adipose tissue in mouse and human, we identified a rare adipocyte population (P4 cells) that increases in abundance at higher temperatures and decreases at cold conditions. Using immunostaining and electrical microscope imaging, we confirmed the transcriptionally defined distinct P4 cells type *in vivo*, which exhibited both multilocular and unilocular morphology with randomly oriented cristae in their mitochondria (*15*).

As alterations in the structure of mitochondria are likely to compromise their function, we hypothesized that the P4 cells are associated with changes in BAT activity. We found that loss of ALDH1A1 expression (aldehyde dehydrogenase 1A1, exclusively expressed in P4 cells) promoted BAT function and protected animals against obesity. In light of the small number of P4 cells, this finding suggests a regulatory role of P4 cells in modulating whole-organ thermogenesis. Indeed, we observed acetate level is reduced in cells loss of ALDH1A1, a molecular that inhibits brown adipocytes thermogenesis through the action of GPR43 (*15*).

Together, this work presents the first single cell map for human brown adipose tissue, identifies novel regulatory circuits in the adipose tissue microenvironment that modulates adipocyte thermogenesis, opens a new avenue to harness brown fat fighting against obesity (see the figure).

Novel obesity treatment strategies

The study of brown adipose tissue is an expanding field that focusses on the thermogenic tissue as a weapon against obesity. Unveiling the critical regulatory elements in adipose tissue will help us make the best use of BAT or even igniting dormant inactive adipocytes throughout the body to produce heat. My work identifies epigenetic regulations and local modulations of BAT as previously unknown mechanisms controlling thermogenesis. Looking ahead, I hope that we and others can demonstrate that targeting thermogenic adipose tissue is a promising therapeutic intervention for obesity.

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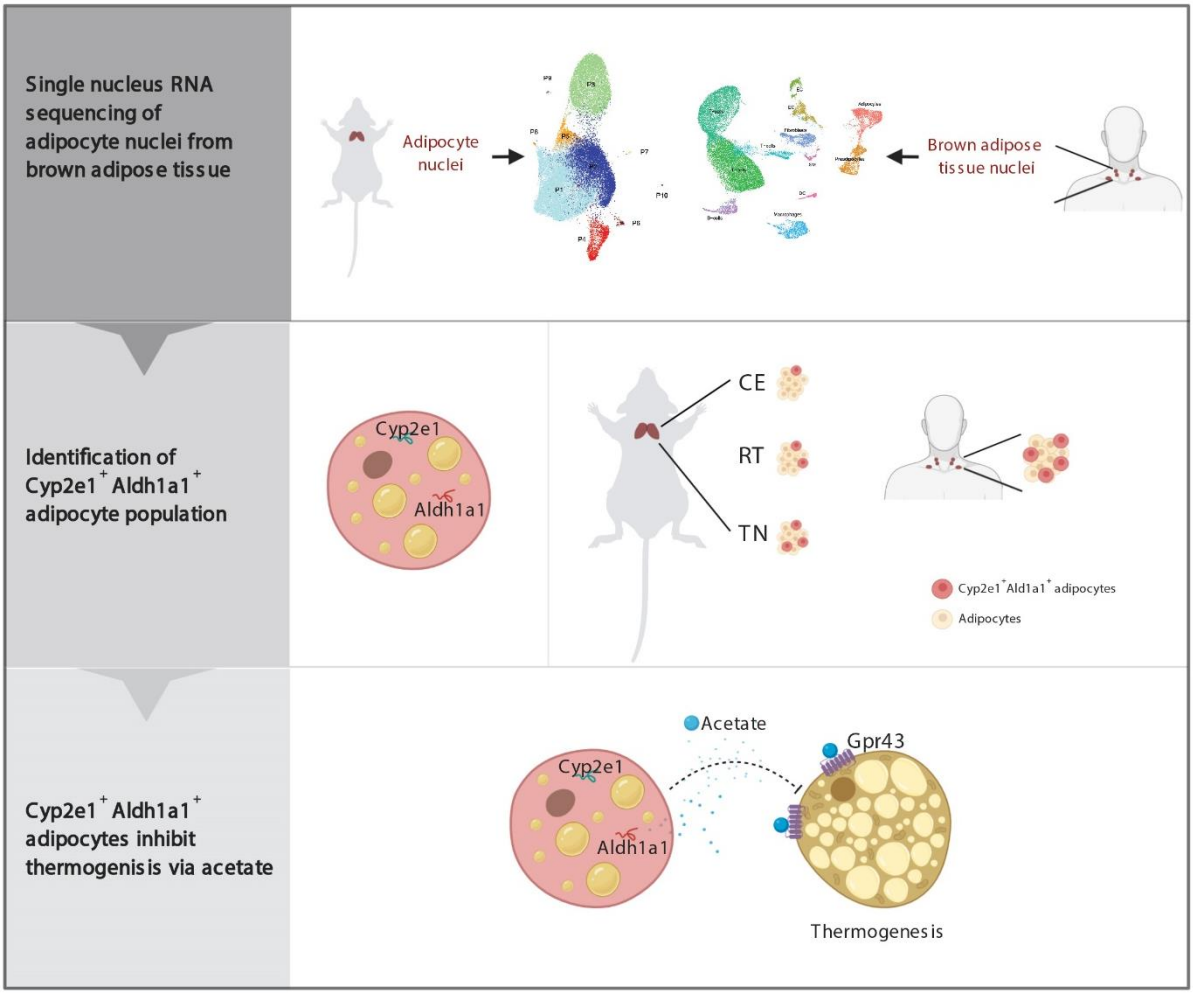

Figure legend

Single nucleus RNA sequencing of mouse brown adipocytes and human deep neck adipose tissue reveal the landscape of cellular composition of brown adipose tissue. A distinct adipocyte population that expresses Aldh1a1 and Cyp2e1 is consistently observed in human and mouse at all conditions, with more abundant proportion at higher temperature. This distinct type of adipocytes release acetate and in turn inhibit the thermogenic activity of neighboring brown adipocyte, through the action of Gpr43.