Abstract

Metabolic diseases are a growing public health concern, with obesity and diabetes affecting millions worldwide. Beige adipocytes, which arise from adipose tissue under specific stimuli, possess thermogenic capabilities similar to brown adipocytes. This thermogenic function is primarily mediated through mitochondrial uncoupled respiration. MTHFD2, an enzyme involved in folate and one-carbon metabolism, plays a key role in mitochondrial redox homeostasis. Although the link between MTHFD2 and mitochondrial function is not fully explored, its dysregulation in metabolic disorders suggests it may be a promising therapeutic target. To investigate this, primary preadipocytes were isolated from inguinal white adipose tissue (iWAT), differentiated into beige adipocytes, and treated with isoproterenol and/or the MTHFD2 inhibitor DS18561882. Gene expression of thermogenic and mitochondrial markers was quantified using qPCR, and mitochondrial respiration was assessed with the Seahorse analysis. Isoproterenol treatment upregulated thermogenic and mitochondrial gene expression and enhanced mitochondrial respiration, particularly at 1 μM for 6 hours. Inhibition of MTHFD2 significantly reduced isoproterenol-induced thermogenic gene expression. Functionally, isoproterenol increased basal respiration and decreased coupling efficiency, a hallmark of thermogenic activation. In contrast, MTHFD2 inhibition under thermogenic activation reduced mitochondrial respiration, decreased uncoupled respiration, and increased coupling efficiency, suggesting a metabolic shift from heat production to ATP synthesis. These findings underscore the crucial role of MTHFD2 in supporting mitochondrial activity during thermogenesis in beige adipocytes and highlight its potential as a target for treating metabolic diseases.

Keywords: *Metabolic disease, beige adipocyte, Isoproterenol, MTHFD2, mitochondrial respiration*