

**Title :** Analysis of Gene Variation in the Middle Region of Melanocortin 4 Receptor Gene in Normal Weight Thai Individuals

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## ABSTRACT

Nowadays, obesity is a major global health issue, with the number of obese patients rapidly increasing in many countries, including Thailand, where obesity rates have been continuously rising. This study investigates the mid-region variants of the melanocortin 4 receptor (*MC4R*) gene in Thai volunteers with normal body weight, to be used as a database for the prognosis of obesity. *MC4R* gene plays a crucial role in regulating body weight and energy balance through the leptin-melanocortin signaling pathway. Nucleotide variations (Single Nucleotide Polymorphisms; SNPs) in this gene affect the response to appetite regulation. Firstly, DNA was extracted from hair roots of 48 Thai volunteers, aged 18-60, with a Body Mass Index (BMI) between 18.50 and 22.99. The extracted DNA was then used to amplify the mid-region of the *MC4R* gene (position 5,555-6,166) using Polymerase Chain Reaction (PCR). The PCR products were analyzed by agarose gel electrophoresis, which revealed a single, specific PCR product of 612 base pairs. The concentration of the PCR product was sufficient for nucleotide sequencing using the Big-Dye Terminator technique. All PCR products were sent to APICAL SCIENTIFIC SDN. BHD. (Malaysia) for nucleotide sequence analysis with the MC4R-F1 primer. In the final step, the nucleotide sequence of the *MC4R* gene mid-region in the volunteers were compared with the reference sequence from the National Center for Biotechnology Information (NCBI) to identify SNPs. The middle region of the *MC4R* gene from 48 DNA samples were analyzed and no SNP was observed. Although nucleotide variation was absent in the analyzed region, this result provided a valuable reference for establishing a control group for comparison with individuals with obesity. Moreover, it could help improving the ability to distinguish obesity-related SNPs from nucleotide variations found in normal weight individuals, thereby contributing to more accurate genetic screening. These findings support the development of more effective obesity screening models, particularly within the Thai population, ultimately improving the quality of life for individuals with obesity and reducing the overall public health burden in Thailand.

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