UNIVERSITI TEKNOLOGI MARA

IDENTIFYING ADIPOSE-SPECIFIC AMP-ACTIVATED PROTEIN KINASE ISOFORMS VIA GENE EXPRESSIONS OF ADULT MALE POSTMORTEM CASES

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ABSTRACT

About 2.5 billion are overweight of which 890 million of people are obese. In Malaysia, a survey by The National Health Screening Initiative 2023 found that more than half, or 53.5% of Malaysians screened were overweight or obese (31.3% overweight and 22.2% obese). Adenosine monophosphate-activated protein kinase (AMPK) is an enzyme that plays an important role as a cellular energy sensor and has been used for the treatment of metabolic diseases, including obesity. Activation of AMPK in hypothalamus can influence appetite, while its activation in liver and adipose tissue inhibits fatty acid synthesis. Studies have shown that different AMPK isomers are predominant in different tissues with 12 possible complexes. However, to date, there have been few studies on the determination of the predominant isomer in adipose tissue especially in humans. The main objective of this study therefore was to determine the different types of AMPK isoforms present in human subcutaneous and visceral (perihepatic and pericardiac) adipose tissue in both underweight/ lean and overweight/ obese adult male. About 500mg to 1000mg adipose tissue were harvested from 96 adult male postmortem cases from Institut Perubatan Forensik Negara, Hospital Kuala Lumpur. RNA was extracted before proceeding with the qPCR analysis to measure the mRNA level of the seven AMPK subunits. There are different predominant isoforms expressed in adipose tissues from the different sites. Findings from this study showed that in subcutaneous adipose tissue, $\alpha 1\beta 1\gamma 1$ AMPK complex was predominantly expressed in overweight/ obese subjects whilst $\alpha\beta 2\gamma 3$ AMPK complex was predominantly expressed in underweight/ lean individuals. Different finding was observed in visceral adipose tissue where in both sites, $\alpha 2$ isoform is predominantly expressed. In perihepatic adipose tissue, $\alpha 2\beta 1\gamma 1$ is predominantly expressed in overweight/ obese while $\alpha 2\beta 1\gamma 3$ is predominantly expressed in underweight/ lean group. Meanwhile in pericardiac adipose tissue $\alpha 2\beta 2\gamma 3$ was upregulated in overweight/ obese whilst in underweight/ lean group $\alpha 2\beta\gamma 3$ was predominantly expressed. In conclusion, this study has identified AMPK complexes that is predominantly expressed in both subcutaneous ($\alpha 1\beta 1\gamma 1$) and visceral (perihepatic, $\alpha 2\beta 1\gamma 1$; pericardiac, $\alpha 2\beta 2\gamma 3$) adipose tissue of overweight/ obese individuals to facilitate future studies in targeting obesity using AMPK as a therapeutic target. Future studies should explore the correlation between the different complexes present and its effects on adipose tissue.

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CHAPTER 1
INTRODUCTION

1.1 Background of Study

AMP-activated protein kinase (AMPK), the downstream component of a protein kinase cascade, is a heterotrimeric complex, composed of a catalytic α and two regulatory subunits, β and γ.

In mammals, the α-subunit and β-subunit exist in two isoforms (α1 & α2; β1 & β2) while the γ-subunit exists in three isoforms (γ1, γ2, & γ3), such that 12 possible complexes may be formed exhibiting differential tissue specificity. Each combination complex, however, has a different function under different physiological conditions and in order to fully activate AMPK, all three subunits must interact together to form a complex. AMPK is activated both by an increase in the intracellular AMP:ATP ratio and phosphorylation of Thr-172 on the α-subunit (Simon A. Hawley et al., 1996).

AMPK acts by phosphorylating and regulating proteins concerned with nutrient metabolism, largely acting to stimulate catabolic ATP-generating pathways whilst suppressing anabolic ATP-consuming pathways. It has core roles in controlling energy balance at both cellular and whole-body levels (Bijland, Mancini, & Salt, 2013) and because of its metabolic actions it is continuously being studied as a therapeutic target in the treatment of obesity.

AMPK is found in skeletal muscle, liver and adipose tissue, however only the role of AMPK in liver and skeletal muscle of animals have been widely studied (Carling, Thornton, Woods, & Sanders, 2012; D. G. Hardie, Ross, & Hawley, 2012; Long & Zierath, 2006). Information on tissue-specific AMPK isoforms in humans, specifically its role in adipose tissue remains lacking. Thus, several knowledge deficiencies remain which needs to be addressed for further understanding of the role of AMPK in regulating metabolic processes such as: 1) although AMPK isoforms have been identified in cell culture and animal studies, there is still lack of data on tissue-specific AMPK isoforms in humans, 2) to the best of our knowledge, there has been very limited study on determination of AMPK isoforms in adipose tissues which limits