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Activities of catabolic enzymes of the endocannabinoid system in adipocytes from metabolically healthy humans

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The peripheral endocannabinoid system (ECS) is upregulated in obesity, as shown by elevated blood concentrations of the endocannabinoids anandamide and 2-arachidonoyl glycerol (2-AG) (Blüher *et al.* 2006). The two principal enzymes involved in anandamide and 2-AG catabolism are fatty acid amide hydrolase (FAAH) and monoacylglycerol lipase (MGL) respectively. Previous studies have reported FAAH and MGL mRNA to be upregulated, downregulated or not different in the subcutaneous adipose tissue of obese humans compared to lean (Engeli *et al.*, 2005; Pagano *et al.* 2007). However, no study has investigated the effects of obesity on the activities of these two enzymes in mature adipocytes. The overall aim was to investigate whether there is any correlation between these enzymes and markers of adiposity and metabolism in metabolically healthy humans over a range of body mass indices (BMIs).

Ethical approval was granted by the University of Nottingham Medical School Ethics Committee, and volunteers were recruited from staff and students (n=28; BMI range 19-34; age 31±8.7). Exclusion criteria included type 2 diabetes and hypertension. Anthropometric measurements, such as height, weight and various skinfold thicknesses, were taken on all volunteers. Following an overnight fast, a venous blood sample was taken and a subcutaneous abdominal adipose sample obtained via a needle biopsy under local anaesthetic. Blood serum was separated and stored at -80^{o}C for later analysis of glucose, insulin and adipokines. Adipose tissue was immediately digested with collagenase to release mature adipocytes, which were stored at -80^{o}C and later thawed, homogenised and centrifuged (20,000 *g*, 20 minutes). The particulate and cytosolic fractions were stored at -80^{o}C until assay of FAAH activity using 2 μM *N*-arachidonoyl-[^{3}H]-ethanolamine as substrate or MGL activity using 100 μM 2-oleoyl-[^{3}H]-glycerol.

In linear regression analyses, FAAH activity positively correlated with BMI (r^2=0.14, P<0.05) and waist circumference (r^2=0.18, P<0.05), but showed no significant relationship with any other anthropometric or metabolic markers measured in this study. However, FAAH activity did show non-significant positive correlation trends with several adiposity markers, such as neck circumference, hip circumference and abdominal skinfold thickness (P<0.1). MGL activity correlated only with diastolic blood pressure (r^2=0.18, P<0.05) and mean arterial pressure (MAP; r^2=0.16, P<0.05) and no other measurements.

In this pilot study, with a sample of metabolically healthy subjects, FAAH activity significantly increases with BMI, but not with many other estimations of adiposity. This increase may be a compensatory mechanism for elevated anandamide levels in the circulation, or possibly a reflection of generalised upregulation of the ECS. MGL activity has no relationship with any anthropometric or metabolic markers, but does correlate with MAP. Further work in this research will focus on increasing the sample size and including a larger number of obese and morbidly obese subjects to further assess the non-significant trends between FAAH activity and adiposity identified in this study.

Blüher, M., *et al.* (2006). *Diabetes* **55**: 3053-3060.

Engeli, S *et al.* (2005). *Diabetes* **54**: 2838-2834.

Pagano, C *et al.* (2007). *J. Clin. Endocrinol Metab* **92**(12): 4810-4819.