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Regulation of lipid metabolism in skeletal muscle by IDOL



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Inducible Degradation of the Low-density lipoprotein Receptor (LDLR), IDOL, is an E3 ligase that targets LDLR, very low-density lipoprotein receptor (VLDLR) and apolipoprotein E receptor 2 (apoE2) for degradation. We have previously demonstrated that IDOL is an evolutionarily conserved regulator for lipid uptake. Furthermore, single nucleotide polymorphisms in IDOL are associated with altered plasma cholesterol levels in humans. Studies have demonstrated a role for IDOL in the brain and liver; however, the importance of IDOL in the regulation of skeletal muscle lipid metabolism remains to be determined.

Using a range of *in vitro*, *ex vivo* and *in vivo* approaches, we aimed to determine the role of IDOL in the regulation of lipid metabolism in skeletal muscle. Ectopic expression of IDOL in C2C12 myotubes was associated with an increased extracellular acidification rate. Interestingly, it was also associated with a 10-fold increase in fibroblast growth factor (FGF)-21. Similarly, administration of IDOL adeno-associated virus (AAV; 1E10) to the tibialis anterior (TA) was associated with a marked increase in FGF-21 expression compared to the contralateral control-treated TA muscle in mice. A significant reduction in a range of lipid species including several ceramide species was also observed in muscle of IDOL-AAV treated mice. In contrast, comparison of IDOL fl/fl and IDOL fl/fl MCK^{Cre}/- mice demonstrated that skeletal muscle specific deletion of IDOL was associated with poorer glucose handling as indicated by a glucose tolerance test following either chow or high fat diet, in the absence of a phenotype in body weight or percent fat.

Together, these studies establish a role for IDOL in the regulation of skeletal muscle lipid metabolism. Further studies are warranted to understand the underlying mechanisms by which IDOL mediates these effects.

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Combining intermittent fasting with high intensity interval training reduces fat mass by increasing fat oxidation and mobilization



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Combining lifestyle interventions such as intermittent fasting (IF) and high intensity interval training (HIIT) result in higher fat loss than IF and HIIT alone. However, the molecular pathways that underpin these changes are unclear. The present study investigated the effects of IF and/or HIIT on molecular markers of epididymal adipose tissue function. Eight-week-old male mice (C57BL/6) were initially fed a high fat (HF, 60%) and sugar (S, 30% w/v) diet for 12 weeks to induce obesity. They were then randomly allocated to four

groups: IF (fasting for 2 alternate days/week), HIIT (3 days/week), combined IF + HIIT (2 alternate fasting days and 3 days HIIT) and CON (*ad libitum* HF/S diet) for 12 weeks. Body weight, fat mass (Echo MRI) and epididymal adipose tissue gene expression (qRT-PCR) were measured at the end of intervention period. At the end of intervention period, IF and IF + HIIT groups displayed significantly lower body weights ($p < 0.05$), fat mass ($p < 0.05$) and expression of the leptin gene ($p < 0.05$) compared to both CON and HIIT groups. The IF + HIIT group displayed increased expression of genes related to fatty acid oxidation (Hydroxyacyl-coenzyme A dehydrogenase; (HADH), $p < 0.05$) and intracellular trafficking (Fatty acid binding protein 4; FABP4), $p < 0.01$) compared to IF group only. A significant positive correlation ($R = 0.66$, $p < 0.05$) between HADH and FABP4 was also observed in IF + HIIT group. Simultaneous higher expression of both HADH and FABP4 suggests that combination of IF and HIIT has a synergistic effect on reduction in fat mass possibly due to increased fatty acid oxidation and mobilization. In summary, IF with or without HIIT are effective strategies for reducing fat mass accumulation potentially via different molecular signaling pathways within adipose tissue.

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Does activation of TRPM8 with menthol or icilin reduce blood glucose and body weight?



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Brown adipose tissue (BAT) is a potential target against obesity and T2D, because activation of BAT induces oxidation of fatty acids and glucose and increases energy expenditure. Theoretically, BAT is activated by beta 3 adrenergic receptors ($\beta 3AR$). However, $\beta 3AR$ agonists have been unsuccessful in reducing weight and blood glucose in humans. Cold temperatures can also activate BAT and increase the metabolism of fatty acids and glucose. Transient receptor potential cation channel subfamily M member 8 (TRPM8) is a cold-sensing cation channel expressed on BAT and is activated by menthol and icilin, with icilin being more potent.

Diet-induced C57Bl/6J mice ($n = 17$) were divided into four treatment groups: vehicle (100 L.s.c); menthol (40 mg/kg s.c, 100 L.); low dose of icilin (2 mg/kg s.c, 100 L) and high dose of icilin (5 mg/kg s.c, 100 L) and treated daily for 1 week. Mice were implanted with biotelemetry devices (G2 E-mitter, STARR Life Sciences Corp, PA, USA). Food intake and body weight were recorded daily. BAT temperature and activity were recorded every minute. Glucose tolerance was assessed by ipGTT before and after 7-days of drug treatment. Body composition was assessed by DEXA (Lunar PIXI densitometer, PIXImus, WI USA) immediately after GTT.

Food Intake

Mice treated with low dose of icilin had significantly reduced food intake on D5-7 ($p < 0.05$), menthol did not change food intake.

Body Weight and Composition

Low dose icilin reduced body weight on D6-7 ($p < 0.05$). Menthol treatment did not change body weight.

BAT Temperature and Activity

Still being analysed.

Glucose Tolerance

Low dose icilin treatment reduced glucose throughout the GTT ($p < 0.05$) but did not change baseline or resting blood glucose. Menthol did cause any significant changes to blood glucose.