Lipid regulation at the intersection between isoleucine and rapamycin

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Summary

Metabolic dysfunction underlies several age-related diseases, making an energetic-focused geroprotective treatment an ideal approach to extend healthspan. Dietary isoleucine restriction and rapamycin, two distinct health- and life-extending treatments with opposing effects on metabolic physiology. This research project utilizes a diet x drug combinatorial regimen to determine whether the two treatments can act synergistically and to unveil the role of rapamycin targets in isoleucine restriction.

- Surprisingly, rapamycin blocks the physiological effects of isoleucine restriction’s in improving body composition and energy expenditure.
- Isoleucine restriction is unable to improve glucose clearance in animals with rapamycin.
- Rapamycin blocks the induction of FGF21 from the liver by isoleucine restriction.
- Rapamycin specifically blocks the upregulation of lipolytic genes in the iWAT by isoleucine restriction.

These results showed that simultaneous initiation of both of these treatments resulted in a gross phenotype resembling a rapamycin-treated animal. This identification of lipolytic genes as a key molecular mechanism represents an important step in further understanding isoleucine restriction and the development of an effective anti-aging therapy.

Dietary compositions and experimental groups. A) Two different amino acids defined diets were utilized. The low isoleucine diet is composed of 1/3 of the isoleucine in the control diet. B) Four groups of male C57BL/6J mice at 9 weeks of age will be given each indicated treatment. The rapamycin injections were given intraperitoneal daily at 4 mg/kg.

Rapamycin prevents weight loss by the low ile diet. A) Mice fed the low ile diet experienced rapid weight loss that plateaus after 4 weeks. In contrast, mice that were simultaneously injected with rapamycin did not experience any weight loss compared to the rapa + control diet animals. B) As we have previously shown, animals fed low ile diet exhibit compensatory overeating despite the drastic weight loss. This increase is slightly blunted in the ile animals that were also injected with rapa.

Rapamycin injection prevents the loss of fat mass during low ile dietary intervention. A) Body composition percentage of animals, as determined by EchoMRI non-invasive body composition monitor, revealed that rapa injection prevents the learning effect of isoleucine restriction. B) Evaluation of lean mass over time found low ile induces significant loss of lean mass. C) Evaluation of absolute fat mass revealed that simultaneous treatment with low ile and rapa injection prevents the loss of fat mass induced by the diet. In the combination group as the most fat mass.

FGF21 fasted blood serum

The induction of FGF21 from the liver by low ile is potentily inhibited by rapamycin. A) As determined by ELISA assay, circulating FGF21 is highly induced by a low ile diet but this increase is completely blocked by rapa treatment. B) rt-PCR evaluation of major tissues that generate FGF21 after low ile found that rapa blocks the induction of FGF21 expression in the liver (B) and the quad muscle (C), but not in the iWAT (D).

The induction of FGF21 from the liver by low ile is unaffected by rapa in the iWAT. A) UCP-1, a mitochondrial proton unipporter that uncouples the respiration from ATP synthesis, is highly induced by the low ile diet and unchanged by rapa. B) Cidea, a transcriptional regulator of UCP-1 that regulates fat browning, is highly induced by low ile and unchanged by rapa. C) BclxL, a lipid estrogen that is known to be associated with thermogenesis, is induced by low ile and unchanged by rapa.

The induction of lipogenic genes by low ile is unaffected by rapa in the iWAT. A) Acc1, a cytoplasmic enzyme that catalyzes acetyl-CoA into malonyl-CoA, is induced by low ile diet and unchanged by rapa. B) Acc2, the mitochondrial counterpart of Acc1, is also induced by low ile diet and unchanged by rapa. C) Fasn, the fatty acid synthase that generates palmitate from acetyl-CoA and malonyl-CoA is induced by low ile diet and unchanged by rapa.

Conclusions: Isoleucine restriction and rapamycin treatment interacts unexpectedly at the transcriptional level of lipolytic genes. The regulation of lipolysis is evidently essential for the gross metabolic health improvements seen in this dietary intervention.

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