FoxO1 does not participate in the hepatic sympathetic modulation of gluconeogenesis in mice under cold stress

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Background: Although it is well-established, CREB (cAMP-responsive element-binding protein) and FoxO1 (Forkhead box O1) are the main gluconeogenesis transcriptional factors. However, the role of noradrenergic fibers in these factors modulation and its physiological effects on gluconeogenesis is still unknow. Previously, we found that CREB and its co-activator 2 (CRTC2) is necessary for gluconeogenic genes activation in mice under cold stress by hepatic sympathetic innervation. Therefore, we aimed to investigate if FoxO is also involved in this response.

Methods: The architecture of hepatic noradrenergic innervation in male mice (C57Bl6J; CEUA nº183/2015) was investigated by the 3DISCO (3D imaging of solvent-cleared organs) technique. We found sympathetic nerves do not make direct contact with hepatocytes and are restricted to the vasculature. Neonate mice were sympathectomized (6-OH-Dopamine) and 8-10 weeks later were exposed to cold (4ºC) for 3-6h and the liver was harvest for enzymatic activity, western blot, and Rt-PCR analysis. Data were expressed as means ± SEM ($p<0.05$).

Results: In innervated mice, cold exposure (6h) increased plasma levels of glucose, corticosterone and glucagon but suppressed insulinemia. Cold also increased activity and mRNA levels of PEPCK and G6Pase in the liver. These effects were associated with a decrease of AKT signaling and an increase in the levels of hepatic norepinephrine, Ser133CREB phosphorylation, FoxO1 deacetylation, FoxO1/3 and CRTC2 dephosphorylation. Sympathectomy abolished the activation of CREB/CRTC2 but did not interfere with FoxO status. Moreover, AMPc and Ca2+ pathways were stimulated during the acute cold stress and the sympathectomy selectively modulated the Ca2+ signaling.

Conclusion: Data indicate CREB/CRTC2 are recruited by noradrenergic fibers in response to cold, probably by Ca^{2+}-dependent signaling, without FoxO participation, leading to gluconeogenic gene transcription and glucose production.

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