A Sweet New Role for Ubiquitin-Specific Protease 2 in Controlling Hepatic Gluconeogenesis

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Hepatic glucose production is critical for maintaining normoglycemia during periods of nutrient deprivation, and dysregulation of hepatic gluconeogenesis is integral to the development of diabetic hyperglycemia. In recent decades, the signaling cascades downstream of the principal physiologic stimuli that suppress (insulin) or induce (glucagon and glucocorticoids) gluconeogenesis have been examined in detail. These signaling pathways can affect glucose production at multiple regulatory levels, including by covalent modification of enzymes involved in glucose and glycogen metabolism, alterations in regulatory metabolite concentrations, and by transcriptional regulation of gluconeogenic enzymes to control the capacity for glucose production. Given that hepatic glucose metabolism is tightly regulated and important for the survival of the organism, it is not surprising that several complex and elegant mechanisms of gluconeogenic control have emerged.

It has been known for some time that attachment of several molecules of ubiquitin, a small 8.5 kDa protein, to other proteins regulates protein activity and stability, often by targeting the protein for proteasomal degradation (1). Ubiquitination of proteins is mediated by E3 ubiquitin ligases, while a large number of ubiquitin-specific proteases are known to act as deubiquinases. Recent studies have suggested that the process of ubiquitin-mediated proteasomal degradation is a regulated step in controlling gluconeogenesis. The cAMP response element-binding protein–regulated transcriptional coactivator 2 (CRTC2) is a critical regulator of gluconeogenic enzyme expression (2). CRTC2 phosphorylation by the insulin signaling cascade leads to its ubiquitination, which targets the coactivator for degradation (3). Insulin also simulates the ubiquitination and degradation of the forkhead box family member, FOXO1, which is another important regulator of gluconeogenesis (4). Ubiquitin-mediated degradation of Krüppel-like factor 15 (KLF15), in response to treatment with the antidiabetic drug metformin, also inhibits the effects of KLF15 on gluconeogenesis (5). Finally, there is also evidence that the gluconeogenic enzyme phosphoenolpyruvate carboxykinase (PEPCK) is targeted for degradation by ubiquitin in response to high glucose levels (6).

Whereas previous work has focused on the process of ubiquitination of these factors, in this issue of Diabetes, Molusky et al. (7) discovered an important role for a deubiquitinating enzyme in the transcriptional regulation of gluconeogenesis. The ubiquitin-specific protease 2 (USP2) is one of a large family of proteins that cleave ubiquitin from ubiquitinated proteins. Molusky et al. profiled the expression of USP family members in mouse liver and found that a specific transcriptional start site variant of USP2, USP2-45, is robustly induced by fasting when gluconeogenesis is induced. USP2 was also regulated in a circadian fashion with its expression being highest during the light phase when rodents self-limit food intake. USP2 expression was induced by counter-regulatory hormones and the peroxisome proliferator–activated receptor coactivator 1α (PGC-1α), which all strongly activate gluconeogenesis (Fig. 1). In contrast, USP2 expression was suppressed by insulin treatment. The authors went on to demonstrate in lean mice that liver-specific overexpression of USP2 activates, while RNAi-mediated knockdown of USP2 reduces, hepatic gluconeogenic gene expression and plasma glucose concentration. These effects were also observed when similar studies were performed in mice rendered obese by feeding a high-fat diet.

Molecularly, these effects on glucose metabolism were attributed to increased expression of the 11β-hydroxysteroid dehydrogenase 1 (HSD1) (Fig. 1), which is involved in conversion of the glucocorticoid prohormone to the active hormone (cortisol in humans or corticosterone in rodents). Heightened HSD1 activity is involved in local tissue hypercortisolemia that is observed in, and contributes to, insulin resistance in adipose tissue and liver of obese individuals (8). In the present work, the increase in HSD1 was linked to deubiquitination of transcription factor CCAAT/enhancer-binding protein α (C/EBPα), which is known to regulate HSD1 expression. C/EBPα was shown to be a target of the ubiquitin protease activity of USP2. However, whether C/EBPα is required for the effects of USP2 on HSD1 expression and gluconeogenesis was not determined.

The circadian signaling effects on hepatic USP2-45 expression fit well with another very recent publication (9) showing that USP2 is regulated in a variety of tissues in a circadian pattern. The expression of several clock components was altered in the suprachiasmatic nucleus of brains of USP2 null mice, and USP2 was found to form a complex with several clock components. USP2 increased the stability of BMAL1, an important regulator of circadian rhythms that was linked to regulating hepatic USP2 expression in the study by Molusky et al. In addition, this study by Scoma et al. (9) showed that USP2 nullizygous mice displayed increased phase delays after exposure to low irradiance light. Together these studies raise the intriguing possibility that USP2 is a component of the circadian clock that influences rhythmicity by stabilizing transcriptional regulators of gluconeogenesis. Given that metabolism is closely linked to the circadian clock, regulation of hepatic gluconeogenesis by USP2 fits quite well with these findings. It will also be of interest to determine whether USP2 null mice have alterations...
in glucose homeostasis or the circadian fluctuations in gluconeogenic rates.

Molusky et al. (7) also identified C/EBPα as a novel target of USP2-mediated deubiquitination. It is likely that other transcription factor targets of USP2 that regulate gluconeogenesis will be discovered in future studies. For example, the stability of CRTC2, FOXO1, PGC-1α, and KLF15 is influenced by ubiquitination in response to insulin or metformin (3–5). Could these transcriptional regulators or the gluconeogenic enzymes themselves also be targets for USP2 (Fig. 1)? Several other hepatic pathways that control intermediary metabolism (fatty acid oxidation, triglyceride metabolism, and de novo lipogenesis) are also strongly influenced by insulin, glucocorticoids, and circadian signaling. It remains to be determined whether USP2 also influences these pathways.

Finally, are the current findings in mice likely applicable to humans given the differences in human and mouse physiology? If USP2 also is a central regulator of hepatic intermediary and glucocorticoid metabolism in humans, this could be an important drug target for treatment of insulin resistance and diabetes, as well as a novel way to intervene in subjects with uncontrolled glucocorticoid production such as Cushing syndrome. The present studies open the door to this line of investigation and are likely just the first step toward a better understanding of this regulated protease.

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FIG. 1. USP2-45 and the regulation of hepatic metabolism. The figure depicts the known and speculative effects of USP2-45 on hepatic energy metabolism and invokes the potential mechanisms for these effects. Dashed lines or boxes indicate speculative effects or mechanisms that remain to be shown scientifically.