Analysis of the serum proteome of diet induced Type 2 diabetic mice

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In this study we used a set of 30 diet induced type 2 diabetic mice to evaluate serum proteomic changes during the progression of animals from normal, to obese, to hyper-insulinemic, and finally to a type 2 diabetic state. C57Bl/6J male mice were reared on a normal or high-fat diet at weaning and their physiological responses (i.e. age, weight, fasting plasma glucose and insulin, and glucose tolerance response) were monitored at regular time intervals. Onset of hyper-insulinemia could be observed as early as 2 weeks on a high fat diet with corresponding glucose intolerance. Fasting blood glucose levels rose significantly after 4 weeks on the high-fat diet. During the development of diabetes, serum was harvested at different time points and used for proteomic analysis employing 2D gel electrophoresis followed by mass spectrometry. Many serum proteins showed significant changes as early as 4 weeks with 32 increasing and 47 decreasing by more than 2-fold. We identified multiple modified forms of proteins some of which were distributed over 2 isoelectric point (pI) units. The amount of each modified protein changed differently in relation to the development of type 2 diabetes. For example, 6 protein spots were identified as transthyretin all with a molecular weight (MW) of approximately 15 kD, but the pI's ranged from 5.1 to 6.7. One form of transthyretin with a pI of 5.7 gradually increased whereas another form with a pI of 6.5 gradually decreased in high-fat fed mice as they developed diabetes. However, the pI 6.0 form increased before mice became hyper-glycemic while this form decreased in control mice. Similarly, contrapsin was found in two forms with a MW of approximately 70kD. The lower pI form was up-regulated by 4-fold within 4 weeks on high-fat diet. This form of contrapsin did not change in control mice. However, the higher pI form of contrapsin was not up regulated until a much later time point as the disease progressed. We observed similar up- or down-regulations specific to certain modified forms for the following proteins; apolipoproteins, alpha-1 antitrypsin, plasma retinol binding proteins and transferrin. All of these proteins have been previously shown to be associated with diabetic conditions. These results show that the proteomic approach to study the development of type 2 diabetes may uncover currently unknown post-translationally modified diagnostic and/or therapeutic protein targets.

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