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Relative quantitative proteomic analysis reveals wound response proteins correlated with after-cooking darkening

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Abstract

Many common potato tuber defects are difficult to elucidate because of the degree of genetic complexity involved, making systems biology approaches necessary. Interaction between chlorogenic acid and iron is responsible for the darkening of potato tuber tissues upon heating - termed after-cooking darkening (ACD). To explore mechanisms of darkening severity in tuber tissues, we have employed relative quantitative proteomics to discover differentially expressed proteins involved in ACD. Tuber tissue samples were collected from a family of diploid clones which possess a highly segregated degree of the darkening. Exploiting this segregation, as well as the observation that darkening is more prevalent in the stem end of the tuber than the apical end, three sample groups were formed: (i) stem ends of three high-ACD clones, (ii) stem ends of three low-ACD clones, and (iii) apical ends of three low-ACD clones. Protein samples were digested and differentially labeled using isotopic reductive methylation, allowing for an orthogonal two-way comparison of protein profiles of the sample groups using 2-D-LC-MS/MS. Using a cutoff fold change of 2 between the high- and the low-ACD sample groups, 30 proteins showed a correlation with tissue darkening. Overall, we observed changes in relative protein abundance that showed an enhanced wound-response program in high-ACD tissues. Among these proteins, five proteins were further validated at the transcript level using qRT-PCR. These proteins may be incorporated into design strategies to create potato cultivars with low levels of ACD. Copyright © 2010 WILEY-VCH Verlag GmbH & Co. KGaA, Weinheim.

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