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Characterizing the Seed Properties of a High-lysine Barley Mutant (Hordium vulgare, cv. M98) using Amino Acid Analysis and Proteomic Approach

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Lysine is one of the most essential amino acids for humans and animals alike. However, its concentration in cereals that constitute an important component of our diet, is low. In the long-term, we are focused on developing a new high-lysine barley line, which has a normal (plump) endosperm. Due to its nutritional relevance, a high-lysine mutant of barley, M98, has been developed at NICS. Unfortunately, the chemically-induced mutant has an undesirable agronomic trait: a shrunken endosperm. As a biochemical characteristics of M98, the lysine content in the endosperm of M98 was higher (44.9 mg/100g) than its parent cultivar Chalssalbori (29.5 mg/100g), while the weight of M98 endosperm was lower than Chalssalbori. Moreover, we found a higher proportion of lysine in the total amino acid content in M98 (0.54 %) compared to Chalssalbori (0.37 %). In order to obtain detailed insight into the protein components of the mutant endosperm, we have embarked on a gel-based proteomics approach for comprehensive proteomics analysis of cereal crop seed proteomes. For this, we have optimized an improved protein extraction protocol, and an immobilized pH gradient (IPG)-based two dimensional (2D)gel electrophoresis-based separation of the extracted proteins solubilized in an modified lyss buffer (LB-TT). Results reveal a dramatic increase in the solubility of proteins from barley endosperm and increased spot resolution on 2D gels. Further analysis on proteomic characteristics of endosperm in M98 compared to Chalssalbori will be discussed.