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Determination of soluble wheat protein fractions using the Bradford assay

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Determination of different grain protein fractions in wheat cultivars is an important task in analyzing bread baking quality. In many laboratories the Bradford assay is used to determine protein concentrations in solutions. In any protein assay (including Bradford), the ideal protein to use as a standard is the purified protein being assayed. In the absence of such an absolute reference protein another protein must be selected as a relative standard such as bovine serum albumin (BSA) which is widely used. In case of BSA calibration, gluten concentration was underestimated (50-54%) compared to calibration with the respective purified wheat proteins (65-70%) in extracts of wheat grain samples. This result is explained with the different amino acid composition of BSA and wheat protein fractions leading to a more intense signal with BSA in the Bradford assay. The aim of this work was to find conversion factors for BSA to determine correct albumin-globulin, gliadin, and glutenin concentrations, because these purified wheat grain protein fractions are mostly not available to be used for calibration purposes. Calibration of the Bradford assay using BSA as well as purified wheat protein fractions allowed to calculate the conversion factors of 2.11 for BSA/albumin-globulin, 4.24 for BSA/gliadin and 3.42 for BSA/glutenin. Application of these conversion factors proved to accurately adjust protein concentrations of wheat fractions originating from ten cultivars, determined with BSA calibration of the Bradford assay. Thus, BSA calibration of the Bradford assay in combination with the conversion factors can be used to determine protein concentration of wheat grain fractions.