Managing variance in order to assess the relationship between gluten concentration and visual assessment of gluten-containing grains in oats, oilseeds, and pulses

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Celiac disease is a lifelong medical condition observed in genetically susceptible individuals. Immune-mediated adverse reactions and progressive deterioration of the lining of the small intestine occur in response to the ingestion of specific gluten proteins fractions found in wheat, rye, barley and triticale, as well as their hybridized strains. There is no cure and the only treatment is a gluten-free diet not exceeding 20 ppm (mg/kg). As a tool to assist in developing best practices, the relationship between the visually-assessed contamination of non-gluten containing grains (NGCG; oats, pulses, oilseeds) with gluten containing grains (GCG) was investigated. Contamination of NGCG occurs due to the presence of GCG kernels, or fragments of kernels. This results in a heterogeneous sample, which is a particular challenge to sample and analyze, particularly when the GCG may be a different size and/or density than the NGCG. In addition, the low limit of 20 mg/kg for "gluten-free" implies that it may take as little as 4-5 kernels of wheat to contaminate a 1 kg sample of NGCG. In this work, the variance in gluten measurements obtained from the analysis of NGCG processed using two sample preparation schemes was evaluated with the aim of minimizing the effects of sample heterogeneity on gluten measurements. The R-Biopharm Ridascreen Gliadin ELISA was used to determine gluten concentrations. The low variability between duplicate aliquots taken from test portions (ranging from 0-30.6% relative standard deviation [RSD], with over three quarters in the 0 to 9% range) demonstrated that the ELISA itself was precise and contributed a low amount to the overall variability of gluten results. The processing of ground samples using rotary sample division and the use of a 1 g test portion for all grains decreased the variability of gluten results for most samples. Using the improved sample preparation scheme, the variability in gluten amongst test portions ranged from 1 to 85% RSD, with more than three quarters in the range of 1-50%. In the original sample preparation scheme, the variability amongst test portions ranged from 1 to 143% RSD, with only slightly over half in the range of 1-50%. The high lipid content hemp seed was a particular challenge to grind, and this was reflected in the higher variability in gluten measurements between test portions (mean RSD = 61%). At concentrations relevant to existing thresholds of gluten contamination, there was no relationship between gluten concentration in NGCG and cereal contamination as determined by visual inspection.