





Abstract

Gluten in Beers: Evaluation of Reproducibility of the R5-Based Competitive Enzyme-Linked Immunosorbent Assay Method Using Real Samples [†]

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Abstract: Beer is the most widely consumed alcoholic beverage in Europe. In many occasions, its consumption is linked to social relations and a fruitive use. To comply with this, the market should offer gluten-free beers that are safe to be consumed by people with celiac disease or those who need to avoid gluten. Brewing hydrolyzes gluten, and this compels the analytical determination of this hydrolyzed protein to be carried out using a competitive ELISA method. The most commonly used competitive ELISA for this purpose is based on the R5 antibody, which has some disadvantages, such as less robustness compared to the homologous sandwich ELISA. The aim of this study was to evaluate the reproducibility of the R5-based competitive ELISA through detecting gluten in beers that intended to achieve a gluten-free label. Thirty-seven samples of beers in which gluten was detected (range 10–80 mg/kg gluten) were analyzed under intermediate precision conditions (e.g., different days and different analysts). Each sample was analyzed 3–20 times. A total of 185 tests were performed and statistically analyzed. The mean calculation of the relative standard deviation (RSD) has a median of 13.6% (range 2.1–23.4%). The samples were pooled according to their gluten content (expressed as mg/kg or ppm gluten) and the median for each interval as follows: beers containing 10–20 ppm (n = 9): RSD 16.1% (range 2.7–19.9%); 21–40 ppm (n = 20): RSD 12.7% (range 2.8–21.5%); and 41–140 ppm (n = 8): RSD 13.7% (range 2.1–23.4%). The main variability in precision was found in the samples with a low gluten content, close to the limit of quantification. This could be due to the fact that small differences in the measured absorbances in this range make a significant difference in quantification. Our results suggest that the precision of the assayed method in our laboratory was satisfactory, in line with the expectable results of other ELISA methods. An internal reproducibility of 20% could be a reliable limit for any testing laboratory. Without evaluating other factors such as accuracy, the data findings point to an elevated uncertainty value for this analytical method.

Keywords: gluten analysis; beers; reproducibility



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