Analysis of Pork Adulteration in Commercial Burgers Targeting Porcine-Specific Mitochondrial Cytochrome B Gene by TaqMan Probe Real-Time Polymerase Chain Reaction

Abstract

A TaqMan probe real-time polymerase chain reaction assay was developed for the determination of pork adulteration in commercial burgers. The assay combined porcine-specific primers and TaqMan probe for the selective amplification and detection of a 109-bp fragment of swine cytochrome b (cytb) gene. Specificity test with 10 ng DNA of 11 different meat-providing animal and fish species yielded a quantification cycle (Cq) of 15. 5 ± 0. 20 for the pork and negative results for the others in a 40-cycle reaction with a change of analysts and sources. Analysis of beef burger formulations with spiked pork showed the assay can determine 100-0. 01% contaminated pork with a PCR efficiency (E) of 93. 8% and a correlation coefficient (R^2) of 0. 991. A plot of actual value against real-time PCR-predicted value also yielded a good linear regression, R^2 0. 998, and small root mean square error of calibration, RMSEC 0. 42. A strong correlation was found between the partial least square (PLS)-predicted values and real-time PCR-determined values. The accuracy of the method was ≥90% in all determinations of the standard set. Residual analysis also revealed a high precision in all determinations. Finally, a random analysis of 10 ng DNA of commercial burgers from pork, beef, chicken, mutton, and chevon yielded a Cq of 15. 56 ± 0. 22 to 16. 24 ± 0. 35 from pork burgers, and negative results from the others, showing the suitability of the assay to determine pork in commercial burgers with a high accuracy and precision.