609/E2

PCR analysis of pork contamination in processed chicken and beef products available in the Sri Lankan market

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Processed meat such as chicken sausages, meatballs, and corned beef are highly susceptible to contamination due to improper handling and processing. Polymerase chain reaction (PCR) was used to detect whether there was pork contamination. Four brands each of chicken sausages S1, S2, S3, S4, chicken meatballs M1, M2, M3, M4, and corned beef B1, B2, B3, B4were randomly selected from the local market, and DNA was extracted using CTAB method. PCR amplification was carried out to confirm the presence of chicken in processed chicken products using the primers, 5'-GGGACACCCTCCCCCTTAATGACA-3' (forward) and 5'-GGAGGGCTGGAAGAAGGAGTG-3' (reverse) which gave an amplicon of 266 bp. The presence of beef was confirmed in corned beef using primers, 5'-GCCATATACTCTCCTTGGTGACA-3' (forward) and 5'-GTAGGCTTGGGAATAGTAGTACGA-3' (reverse) which gave a band of 271 bp. Porcine primers 5'-ATGAAACATTGG-AGTAGTCCTACTATTTACC-3' (forward) and 5'-CTACGAGGTCTGTTCCGATATAAGG-3 (reverse) gives an amplicon at 149 bp. The amplifications were carried out with 2.5 µL 10X PCR buffer, 0.5 µL 10mM dNTP (Promega), 0.8 µL 10µM PCR primers, 0.2 µL Taq DNA polymerase (DreamTaq), 5 µL of genomic DNA, made up to 25 µl with DNAse free water, in an Applied Biosystems thermal cycler, for 30 cycles for chicken samples, and 35 cycles for beef samples. Analysis was carried out in duplicate. The positive control was porcine DNA and the negative control was sterilized water. Electrophoresis of 10 µL portions of the PCR products were carried out for 45 min at 100 V in a 2% agarose gel containing ethidium bromide in TBE buffer. The DNA fragments were visualized under UV light (E-Gel iBase™ Power System) with GelCapture Acquisition Software. The products of PCR amplification were visualized using agarose gel electrophoresis.The PCR analysis was conclusive for the presence of chicken in processed chicken products and presence of beef in the corned beef products that were analyzed. There was no evidence for pork contamination in any of the analyzed processed meat products as seen in Fig 1.

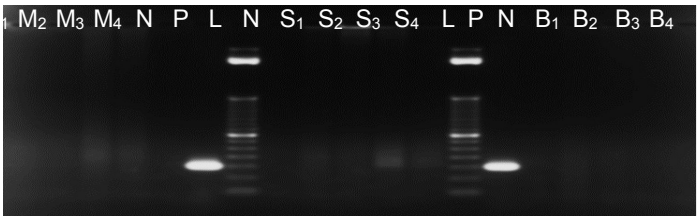

Fig.1 Agarose Gel Electrphoresis of PCR products to detect pork contamination. Lanes M_{1-4} – chicken meatballs brands. M_{1-4}, Lanes S_{1-4} – chicken sausages brands S_{1-4}, Lanes B_{1-4} – corned beef brands B_{1-4}. L – 50 bp ladder. N and P- negative and positive control for pork

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