Tetraplex PCR assay involving double gene-sites discriminates beef and buffalo in Malaysian meat curry and burger products

Abstract

Replacement of beef by buffalo and vice versa is frequent in global markets, but their authentication is challenging in processed foods due to the fragmentation of most biomarkers including DNA. The shortening of target sequences through use of two target sites might ameliorate assay reliability because it is highly unlikely that both targets will be lost during food processing. For the first time, we report a tetraplex polymerase chain reaction (PCR) assay targeting two different DNA regions in beef (106 and 120-bp) and buffalo (90 and 138-bp) mitochondrial genes to discriminate beef and buffalo in processed foods. All targets were stable under boiling, autoclaving and microwave cooking conditions. A survey in Malaysian markets revealed 71% beef curries contained buffalo but there was no buffalo in beef burgers. The assay detected down to 0.01 ng DNA and 1% meat in admixed and burger products.