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## Meat species identification and Halal authentication analysis using mitochondrial DNA

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### ABSTRACT

A method utilizing PCR-restriction fragment length polymorphism (RFLP) in the mitochondrial genes was developed for beef (*Bos taurus*), pork (*Sus scrofa*), buffalo (*Bubalus bubali*), quail (*Coturnix coturnix*), chicken (*Gallus gallus*), goat (*Capra hircus*), rabbit (*Oryctolagus cuniculus*) species identification and Halal authentication. PCR products of 359-bp were successfully obtained from the *cyt b* gene of these six meats. *AluI*, *BsaI*, *RsaI*, *MseI*, and *BstUI* enzymes were identified as potential restriction endonucleases to differentiate the meats. The genetic differences within the *cyt b* gene among the meat were successfully confirmed by PCR-RFLP. A reliable typing scheme of species which revealed the genetic differences among the species was developed.

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### 1. Introduction

Meat species identification and Halal authentication are a major concern in Asia, France, Russia, Sweden, Germany, Switzerland, Greece, Spain, Italy, United Kingdom, South and North America and most other countries. There is a call for the availability of reliable and rapid methods to identify animal species in food.

An accurate method for the differentiation of meat species is of great importance in forensic science cases. Law in many countries requires that products should be labeled with official names. Regulation by the EC legislation (178/2002) on food traceability (European Commission, 2002) requires all stakeholders within the food supply chain must be able to identify the source of all raw materials.

Meat which is Halal is the major concern of Muslim consumers; permitted animal and bird meats according to Islamic law are considered Halal. To differentiate meat species, many approaches

have been presented, and typing schemes for precise species identification of pork, which is a non-Halal meat, and other Halal meat by PCR technique are generally available (Kesmen, Yetim, & Sahin, 2007; Rastogi et al., 2007). The mitochondrial 12S ribosomal RNA (mt12S rRNA) gene was used by Rodríguez et al. (2004) for a PCR assay to detect pork in raw and heat-treated meat mixtures, also, for beef, sheep and goat detection. Trace amounts of pork DNA in processed foods were successfully detected by Tanabe et al. (2007), whereas, based on D-loop mtDNA, Montiel-Sosa et al. (2000) detected pork meat and its fat in meat mixtures.

PCR-restriction fragment length polymorphism (RFLP) analysis is widely developed for the typing of species, with several genes especially within the mitochondrion having been targets for this method (Bellagamba, Moretti, Comincini, & Valfrè, 2001; Cespedes et al., 1998; Fajardo et al., 2008; Hold et al., 2001; Russell et al., 2000; Sanjuan & Comesana, 2002; Sotelo et al., 2001; Zhang, Huang, Cai, & Huang, 2006). Girish et al. (2004) has applied PCR-RFLP to a segment of the mt12S rRNA gene for distinguishing between meats with a single pair of universal primers yielding a 456-bp amplicon. The banding patterns resulting from restriction of the amplicons

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