A PCR-RFLP assay for discrimination of Echinococcus granulosus sensu stricto and Taenia spp. in dogs stool

Hidalgo, Alejandro

Melo, Angélica

Romero, Fernando

Villanueva, José

Carrasco, Carolina

Jara, Paulina

Venegas, Juan

Fonseca-Salamanca, Flery

In order to develop a method of identification and discrimination of Echinococcus granulosus sensu stricto from faecal samples of dogs infected with taeniid eggs (Echinococcus spp., Taenia spp.), a combined strategy of Polymerase Chain Reaction (PCR) and Restriction Fragments of Lenght Polymorphisms (RFLP) was proposed. Initially, a pair of primers was designed to amplify a fragment of the 12 Subunit of ribosomal RNA gene (12SrRNA) from mitochondrial DNA. The amplified product was digested by SspI restriction enzyme, which in E. granulosus kept the intact fragment of 160 basis pairs (bp), while in Taenia spp. produced two fragments (62 bp and another of 98 bp). The method was tested using positive controls of DNA, in faecal samples experimentally contaminated with eggs of E. granulosus and Taenia spp. and in dogs naturally infected. In all of them, reproducible results were obtained and the primers were specific to amplify only Taeniidae DNA. The sensitivity of the technique was tested, achieving amplification of DNA extractions with a single egg. In conclusion, the technique developed was optimal and easy to identify patent infections by E. granulosus s.s., constituting a possible alternative for epidemiological studies in dogs, especially in endemic areas where this infection occurs.