

Development of an Optimized Polymerase Chain Reaction Protocol for the Successful Amplification of Nuclear *C-mos* Gene in Endemic Skink Genus *Lankascincus*

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Lankascincus is a skink genus endemic to Sri Lanka. The genus is represented by ten species and are commonly referred to as Lanka skinks. Studies on the phylogenetics of these species have used different mitochondrial genes and nuclear genes such as *c-mos*. This study was carried out to develop an optimized protocol for the polymerase chain reaction (PCR) amplification of nuclear *c-mos* gene in endemic skink species to aid in their phylogenetic relationship estimation. Genomic DNA extraction from tail tissue was performed using commercial DNeasy® Blood and Tissue kit. Potential primers were designed using NCBI primer BLAST tool considering their lengths, GC content, melting temperatures and amplicon size. Amplifications were done on a final volume of 20µL containing 1X FIREPol® Master Mix, PCR grade water and varied PCR conditions; primer concentration, template DNA concentration, annealing temperature, and cycle parameters. The amplicons mixed with loading dye were electrophoresed in agarose gel at 60V for 2 hours along with a 100bp DNA ladder. The PCR conditions; 0.1µM primer concentration (forward: 5'-AGAACCGTTTGGCATCACGA-3', reverse: 5'-GTGAATGGAGAAAGACCAAGCC-3'), 10ng/µl DNA concentration together with cycle conditions; initial denaturation at 94°C for 3 minutes, denaturation at 94°C for 30 seconds, annealing at 54°C for 40 seconds, extension at 72°C for 45 seconds, final extension at 72°C for 5 minutes, and final hold at 4°C, yielded the optimal amplification of 300bp *c-mos* gene fragment.

Keywords: Skinks, *Lankascincus*, *c-mos*, PCR