APPLICATION OF POLYMERASE CHAIN REACTION FOR DIAGNOSIS OF *Dirofilaria repens* IN DOGS

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ABSTRACT

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The aim of this study was to establish a Polymerase Chain Reaction (PCR) assay to diagnose *Dirofilaria repens* in dogs in Sri Lanka.

Dirofilaria repens causes subcutaneous infections in dogs, cats, wolves, foxes and sea lions. Dogs show symptoms such as dermal swelling, subcutaneous nodules prurities and hyper pigmentation.

Dirofilaria repens infection has been reported in humans in Sri Lanka. It causes asymptomatic infections, prurities or eczematous eruptions, sub mucosal and subcutaneous nodules.

Current studies show the prevalence of *Dirofilaria repens* infection is increasing in world wide.

The most common method for diagnosis of *Dirofilaria repens* infection in dogs is wet smear diagnosis under microscope. The reliability of this method depends on a number of parasites present in the sample and the experience of the technician.

Several scientists have developed molecular diagnosis methods to detect *Dirofilaria repens* infection in dogs.

In this study ten blood samples from dogs were taken and 10 serial dilutions were prepared from each sample and microfilaria were counted.

PCR was performed each diluted sample using Co1 Int forward and reverse primers. PCR products (10 µl) were loaded to 0.8% agarose gel for the electrophoresis and visualized by a UV transillumination. In agarose gel it showed bands. The primer CO1 Int was precisely amplify the CO1 gene of *Dirofilaria repens.*

This study shows the detection level of PCR is higher than the wet microscopy method. Highest detection level of direct blood drop test is 33 microfilaria per milliliter and PCR method was 3 microfilaria per milliliter. Therefore PCR is not depend on the number of parasites present in the sample and gives results when the number of parasites are extremely low. Large number of samples can be diagnosed by PCR method. Therefore it is a best method for the microfilaria screening in highly endemic areas as Sri Lanka.