# CLONING AND PARTIAL CHARACTERIZATION OF REPETITIVE SEQUENCES FROM WUCHERERIA BANCROFTI

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The gene cloning techniques have enabled the characterization of repetitive sequences and the development of species-specific oligonucleotide probes, for the identification of human filarial parasite, Brugia malayi.[1,2] In contrast no such probes have been developed for Wuchereria bancrofti, the filarial parasite causing filariasis in Sri Lanka. The present studies were undertaken to develop a DNA probe for W. bancrofti.

Purified microfilariae ($2x10^6$) of W.bancrofti were treated with Proteinase K and Sarcosine at 37°C over-night and DNA was extracted and further purified by Cesium Chloride density gradient centrifugation with Hoechst dye 33258. The purified DNA was randomly sheared, size fractionated and fragments of 3.0-8.0 kilo bases in size were recovered from low melting agarose. These fragments were then cloned in the EcoR1 site of λgt11 as described elsewhere.[3]

The amplified genomic library was subjected to differential screening using $\alpha$-$^{32}$P-dCTP nick translated W.bancrofti and human DNA. After 3-4 cycles of screening several putative clones giving specific and strong signals with $^{32}$P-labelled W.bancrofti were isolated. Southern blotted EcoR1 and Hind III cleaved W.bancrofti genomic DNA, when probed with $^{32}$P-labelled recombinant DNA, gave strong signals indicating the presence of parasite specific genomic fragments in the recombinants. The restriction mapping and sub-cloning experiments of these clones are currently in progress.

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References:

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