A HIGHLY REPEATED DNA SEQUENCE FROM SETARIA DIGITATA

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Setaria digitata is a filarial nematode, the microfilariae of which causes cerebrospinal nematodiasis in goats and sheep in Sri Lanka[2]. In an attempt to develop a species-specific DNA probe for the diagnosis and epidemiological surveys of cerebrospinal nematodiasis in Sri Lanka, a genomic library of S. digitata was established in EMBL 3. The number of recombinants were approximately 75,000.

The library was screened by in situ plaque hybridization technique[1] using nick-translated ^{32}p-labelled total genomic DNA as the probe. Initial screening of approximately 20,000 recombinants resulted in the isolation of 190 putative clones of which 25 gave very strong signals on autoradiography. These were further purified by several cycles of plaque purification and one clone designated EMBL3Sd41, was selected for further characterization.

Restriction endonuclease cleaved genomic DNA of S.digitata when resolved by agarose gel electrophoresis and Southern blotted and probed with nick translated ^{32}p-labelled clone (EMBL3Sd41) gave strong signals of ladder like pattern indicating the repetitive nature of the cloned fragment. Furthermore, the clone did not hybridize to denatured and dot-blotted genomic DNA of cattle, W. bancrofti, sheep, goat and Armigeres subalbatus which indicated the clone is specific for S.digitata.

^{32}p-labelled cloned fragment detected 25 ng of genomic DNA of S.Digitata while the biotin labelled cloned fragment detected 100 pg of genomic DNA. The cloned fragment which is highly repeated to an extent of 4% in the entire genome of S.digitata may prove to be useful as a DNA probe in the diagnosis and epidemiological surveys of cerebrospinal nematodiasis caused by S.digitata.

References:

1) Benton, W.D. & Davis R.W. (1977) Screening λgt recombinant clones by hybridization to single plaques in situ. Science 196, 180-182.

2) Shoho, C. & Nair, V.K. (1960) Studies of cerebrospinal nematodiasis in Ceylon, Experimental production of cerebrospinal nematodiasis by the innoculation of infective larvae of S.digitata into susceptible goats. Ceylon Vet. J., March 2 - 10.