

Genetic Characterization of Acanthamoeba Isolates from Amoebic Keratitis Patients

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Abstract : Background: Amoebic keratitis is a painful vision threatening infection caused by a free living pathogenic amoeba Acanthamoeba. It can be misdiagnosed and very difficult to treat if not suspected early. The epidemiology of Acanthamoeba genotypes causing infection in our geographical area is not yet known to the best of our knowledge. Objective: To characterize Acanthamoeba isolates from amoebic keratitis patients. Methods: A total of 19 isolates obtained from patients with amoebic keratitis presenting to the Advanced Eye Centre at Postgraduate Institute of Medical Education and Research, a tertiary care centre of North India over a period of last 10 years were included. Their corneal scrapings, lens solution and lens case (in case of lens wearer) were collected for microscopic examination, culture and molecular diagnosis. All the isolates were maintained in the Non Nutrient agar culture medium overlaid with E.coli and 13 strains were axenised and maintained in modified Peptone Yeast Dextrose Agar. Identification of Acanthamoeba genotypes was based on amplification of diagnostic fragment 3 (DF3) region of the 18srRNA gene followed by sequencing. Nucleotide similarity search was performed by BLAST search of sequenced amplicons in GenBank database (<http://www.ncbi.nlm.nih.gov/blast>). Multiple Sequence alignments were determined by using CLUSTAL X. Results: Nine out of 19 Acanthamoeba isolates were found to belong to Genotype T4 followed by 6 isolates of genotype T11, 3 T5 and 1 T3 genotype. Conclusion: T4 is the predominant Acanthamoeba genotype in our geographical area. Further studies should focus on differences in pathogenicity of these genotypes and their clinical significance.

Keywords : Acanthamoeba, free living amoeba, keratitis, genotype, ocular

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