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Genetics and Molecular Biology Research

Molecular cloning of borrelia

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Abstract

Gene cloning or molecular cloning is an important step in recombinant DNA technology wherein a sequence of one or more DNA fragment is introduced into another DNA sequenced for the purpose of amplification or functional studies. It is also used for downstream applications such as sequencing, mutagenesis, genotyping or heterologous expression of a protein. The traditional techniques of gene cloning involve the transfer of DNA fragment of interest from one organism to a self-replicating genetic element such as a bacterial plasmid. The process involves amplification of the desired gene fragment, splicing of the gene with restriction enzyme, if required and ligating into a plasmid. The final assembly includes promoter element fused to a reporter gene under the control of a ubiquitous promote. The assembly is then inserted into a host, which may be an eukaryotic or a prokaryotic cell. Gene cloning is important to many areas of modern biology and medicine.

Borrelia is a spirochete bacterium which cause Lyme Disease in humans. Transmission to human is caused by ticks of Ixodes spp. Several species of Borrelia cause Lyme disease some of which are B. burgdorferi, B. afzelii and B. garinii. Of these B. burdorferi is the most important and the most studied cause of Lyme Disease. The disease is worldwide in distribution including India (Vasudevan and Chatterjee, 2013; Praharaj et al., 2008). B. burgdorferi enters the skin at the site of the tick bite, the organism then migrate locally in the skin around the bite, spread via the lymphatics to cause regional adenopathy or disseminate in blood to organs or other skin sites. Clinical symptom may include fever, chills, headache, fatigue, muscle, joint aches and erythema migrans rash. For effective treatment, early diagnosis is of paramount importance. Among the many techniques employed for diagnosis of Lyme Disease in the past, molecular detection using PCR remains the most rapid and sensitive test (Marconi and Garon, 1992)

The present work involved cloning of a segment of a 16S rRNA gene of Borrelia burgdorferri for further characterization and downstream applications. The process involves PCR amplification of a fragment of the 16S rRNA gene of Borrelia burgdorferi, ligating into a plasmid vector and transformation into an E. coli host.

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Biography

Kaushiki Bhattacharya is pursuing her PhD at the age of 25 years from Birla Institute of Technology – Mesra. Her research area is Molecular Cloning of Borrelias. She has done

her training in molecular cloning and knows different techniques of extraction and is emerging to become an expertise in the field.