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Trapping enhancers by transgenic expression of BACs in zebrafish

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Bacterial Artificial Chromosomes (BACs) are large extra-chromosomal plasmids in bacteria that faithfully propagate large pieces of DNA from the chromosomes of organisms. Because they represent tiny contiguous pieces of the chromosome, BACs are ideally suited for expression of genes in their chromosomal contexts. Using loxP-Tn10 mini-transposons to introduce enhancer-traps into BACs, regulation of the Amyloid Precursor Protein (*appb*) gene expression in zebra fish were analyzed. Clusters of two known transcription factor protein-binding sites, E4BP4/ NFIL3 and Fork head, residing in two discontinuous DNA domains distal to the gene were found to be required for expression of *appb* in neurons. These sites are statistically over-represented in both the zebra fish and human APP genes, although their locations are different. The results suggest that the clock-regulated and immune system modulator transcription factor E4BP4/ NFIL3 likely regulates the expression of both *appb* in zebra fish and APP in humans. Intriguingly, a cluster of four E4BP4 sites in intron 4 of human APP exists in actively transcribing chromatin in a human neuroblastoma cell-line, SHSY5Y, expressing APP as shown using chromatin immunoprecipitation (ChIP) experiments. It suggests potential human APP gene regulatory pathways, not on the basis of comparing DNA primary sequences with zebrafish *appb* but on the model of conservation of transcription factors. Functional identification of long-range regulatory sequences of *appb* in zebrafish has provided important clues for regulation of the APP gene in humans.

Biography

Pradeep K Chatterjee is an Associate Professor in Chemistry at North Carolina Central University, and JLC Biomedical/Biotechnology Research Institute. His research interests include developing novel technology to map genetic markers and gene-regulatory elements that influence expression of genes from large distances along the DNA and application of such genomic/molecular genetic technologies to understand eukaryotic gene regulation using the zebra fish model system. He completed his undergraduate studies in Chemistry from IIT-Kharagpur and IIT-Kanpur, India. He received his PhD in Chemistry from Columbia University, New York. His Postdoctoral training was at MIT-Biology, Cambridge, Massachusetts, and Princeton University, Princeton, NJ.

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