**ABSTRACT**

Multiple sequence alignment is an alignment of three or more biological sequences, generally protein, DNA, or RNA. The input set of query sequences are assumed to have an evolutionary relationship i.e., they are descended from a common ancestor. The resulting MSA, sequence homology can be inferred and phylogenetic analysis can be conducted to assess the sequences' shared evolutionary origins. In this paper, six different mammalian species gene sequence were compared with human gene sequences. Metabolically important genes such as Amylase, ATPase, Cytochrome-B, Haemoglobin, and Insulin where chosen for comparison. The DNA sequences of FASTA format was retrieved from NCBI databank and used as input sequences for Multiple sequence analysis using ClustalW, MUSCLE, and T-Coffee. Multiple sequence alignment score and phylogenetic trees where obtained from all the three tools and discussed with the snapshots and findings.