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RESEARCH AREAS:

Gene Silencing

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RESEARCH INTERESTS:

Oligo-glycos:

Polysaccharides and oligosaccharides are known to have therapeutic value. Oligosaccharides were obtained from *Moringa oleifera* gum and oligos of size 3000 Da were purified and bioactivities of the oligos were studied. They could inhibit mitosis in plant cells and animal cell lines. The oligos had anticoagulant activity preventing blood clotting and were able to stabilize nucleic acids. Mitotic arrest has been found to be due to up-regulation of certain cell cycle proteins such as P21 and P27. Other bio-activities of the oligos have to be unveiled. It would be an important step if the suitable length of the oligos and their composition are well understood.

Therapeutic proteins:

Biologically active peptides (BAP) include both the peptides derived from food proteins and the peptides naturally present in the food. In our laboratory, we have cloned and expressed a bio-active peptide casoplatelin in *E. coli*. Protein recursor of the peptide is kappa-casein that is present in bovine and human milk. The peptide showed inhibition of ADP induced platelet aggregation.

In our laboratory we have cloned Staphylokinase encoding gene in an expression vector of *E. coli.* We are also interested in cloning therapeutic proteins such as Streptokinase, plasmin, enteropoietin etc.

Gene silencing using ribozyme:

β- amyloid protein significantly reduces the ability of nAChR to transmit messages relayed by the brain's signaling proteins leading to Alzheimer's disease. Therefore, in our laboratory, a hairpin ribozyme has been designed against the amyloid A4 precursor mRNA. This is to reduce amyloid formation thereby reducing the symptoms of the disease. Target was selected close to the 5' cap site. Catalytic activity of the hairpin ribozyme has been demonstrated *in vitro*. Once the catalytic efficiency of the ribozyme is known, *in vivo* experiments with cell lines etc., can be carried out. This is more advantageous compared to hammer head ribozyme since it requires Mg^{2+} ions at near physiological pH. Further, the secondary structure of the designed hairpin ribozyme with extended Helix-4 could confer better stability *in vivo*.

Gene silencing using DNAzyme:

Glutamic acid decarboxylase (GAD65) has been implicated as the major auto-antigen in Insulin Dependent Diabetes Mellitus (IDDM). The GAD65 protein in β-islet cells is proteolytically cleaved and the derived peptide is presented by the MHC class I

molecules. This complex is recognized by the anti-islet T-cells, leading to the destruction of the islet β cells. Therefore, a DNAzyme was designed that cleaves efficiently the GAD65 m RNA of humans. The catalytic motif used was 10-23. Cleavage of GAD65 m RNA would help the cells to eliminate the expression of the protein that acts as the auto-antigen. The anti-GAD DNAzyme has 9-nucleotide recognition arms specific to the GAD65 m RNA. Effective use of DNAzymes *in vivo* requires some modifications to render them resistant to nucleases.

PUBLICATIONS

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