

Scholar Name: SAIMA KHURSHEED

Supervisor Name: Prof. Tasneem Fatma

**Department of Biosciences , Faculty of Natural Sciences, Jamia Millia
Islamia, New Delhi-110025.**

**Title: “ISOLATION AND CHARACTERIZATION OF INSULIN LIKE
ANTIGENS FROM CYANOBACTERIA”**

Finding: The success in isolation of insulin like antigen from plant sources their less toxic and less cross reactive effect and low cost provides an alternate/substitute to human/animal based insulin. Not only this use of transgenic photosynthetic prokaryotic and eukaryotic systems, may appear to be revolutionary in future. My results as well as results of many investigators suggest that insulin is a molecule present in all organisms, from unicellular to multicellular organisms. Insulin’s wide phylogenetic distribution would be easily accepted if its function as a metabolic signalling pathway involved in glucose metabolism were confirmed.

ABSTRACT

Cyanobacteria are a rich source of potentially useful natural compounds (Moor, 1996). These natural compounds have found prominent applications in treatment of several infectious diseases such as tuberculosis, malignancies, viral infections and other metabolic syndromes. Among these disorders, diabetes is a chronic disease characterize by the body’s reduced capacity to use glucose. The world spent an estimated 215 – 375 billion Diabetics in 2007 to care for diabetes and the many complications that arise from the disease (IDF, 2008). Having being termed the silent epidemic, diabetes continues to ignite and sustain motivation in finding a cure.

A number of studies are in progress to find natural sources, which are effective in reducing the intensity of diabetes. There is also an increasing demand by patients to use natural products with antidiabetic activity due to the undesirable side effects accompanied with the use of insulin and other hypoglycaemic drugs (Kaleem et al, 2008).

The discovery of Insulin like antigen in tissues of the plants, yeast, as well as in cyanobacteria opens up a new field of work. Insulin obtained from animal pancreas has a very low yield, (one pound of pure insulin per 10,000 animals). Side effects of animal insulin are well known. As compared to animal insulin, Cyanobacterial insulin may offer the possible potential for safe, economical, high-capacity production for such important biopharmaceuticals. For finding out additional photosynthetic prokaryotic bioresource present study was performed with following objectives.

Screening (ELISA based) of Cyanobacteria for Insulin Like Protein

Screening of 31 species of cyanobacteria for insulin like antigen was done by ELISA technique utilizing anti-human insulin antibody. The insulin like antigen was present only in *Spirulina platensis* NCCU S5.

To purify Cyanobacterial Insulin Like Protein

During present study insulin like antigen was found in *Spirulina platensis* S5 with concentration of 31.414 µg/g that was purified and characterized by various techniques.

Characterization of Cyanobacterial Insulin Like Protein by---

SDS-PAGE

After identification of insulin like antigen by ELISA, SDS-PAGE was run and it was found that the protein band had similar molecular weight as that of bovine insulin (6 KDa).

Western Blotting

Further characterization was done by Western Blotting which showed similar antigenicity as was in ELISA.

RP-HPLC

After that same isolated protein was further characterized by RP-HPLC and standard bovine insulin showed similar retention time.

MALDI-TOF (Sequences)

Amino acid sequencing of *Spirulina*'s insulin like antigen was done and found only one blast hit on the query sequence. Silva et al, (2002) could not find any sequence similar to bovine insulin but they found same immunoactivity in ELISA and Western blotting in *Spirulina maxima*, *Gracilariopsis*, *Selaginella*, *Equisetum*, *Saccharomyces cerevisiae*.

Circular Dichroism

Circular dichroism (CD) spectroscopy has been a valuable method for the analysis of protein secondary structure. *Spirulina platensis* S5 insulin like antigen was also studied by Circular dichroism analysis and showed α -helical structure in far UV region.