Details of the Project sanctioned under the Human Resource Development scheme of Department of Health Research

1. Project Title: Role of Gfi-1 in Long Term–Hematopoietic Stem cells (LT-HSC) and leukaemia stem cells self -renewal and differentiation

2. Category of fellowship:

Programme to encourage research personnel [Non-resident Indian (NRI), Persons of Indian Origin (PIO), Overseas Citizen of India (OCI)] serving abroad, to come back to India for undertaking health research in identified areas.

3. PI (Name & Address):

Dr. Satyendra Kumar Singh, Assistant Professor, Stem cells/Cell Culture Lab, Centre for Advance Research, King George Medical University, Lucknow, U.P., 226003

4. Qualifications:

Degree	Institution	Field(s)	Year
B.Sc.	Purvanchal .University,Jaunpur,UP	Zoology,Botany,Chemistry	1996
M.Sc.	JNU, New Delhi	Life Sciences	2002
PG Diploma	JNU, New Delhi	Bioinformatics	2003
Ph.D.	Duisburg-Essen University, Essen, Germany	DNA Damage Response	2010
Post Doc	NCI-NIH, Maryland, USA	Hematopoietic Stem Cells	2016

5. Mentor or Co.PI (Name & Address): N/A

6. Duration of the project: 5 years

7. Broad area of Research: Stem cell Research

7.1 Sub Area: Hematopoietic Stem Cells and Leukaemia Stem Cells

8. Summary of the Project: (Give in about 300 words)

Gfi-1 is required for the proper development of hematopoietic cells. Mutation in the zinc finger domains of Gfi-1 results in hyperplasia and severe congenital neutropenia in humans. Gfi-1 expression is very low in early leukemic cells (Blast cells), like FAB subtype M0 and M1 associated with poor prognosis. The precise role of GFI-1 in human HSPC is not known; therefore, I would like to evaluate the role of GFI-1 in human HSPC and leukaemia stem cells growth and differentiation. The mechanism by which GFI-1 regulates HSC self-renewal, quiescence and maintenance remains an area of active research. AML is very heterogeneous in nature; I will try to find association of Gfi-1 expression in different AML subtype with self-renewal and differentiation. **Therefore, the precise mechanism by which GFI-1 regulates HSC self-renewal, quiescence and maintenance remains an area of active research**. **Thus, current studies are focused on defining the function of specific transcription factors in normal and leukemic HSPC development, and how these transcription factors activate and repress genes and gene programs/networks to regulate HSC self-renewal and cell fate and differentiation.**

Some of the GFI-1 target genes have been identified, and have been shown to mediate the hematopoietic defects observed in *Gfi-1*-/- mice, including the maturational defect in granulocyte development (CSF-1, RasGRP1, and PU.1), B cell development (PU.1 or Id2), and myeloid hyperplasia (Id2 or HoxA9). *Gfi-1-/-* HSPC show increased cell cycling, which suggests that GFI-1 functions to restrict HSC proliferation and prevent exhaustion.

I will use mouse model to find novel Gfi-1 targets and its role in HSC maintenance. Gfi-1 targets in murine HSC and leukemic cells will be validated. I will use them for human AML cell lines (M0-7e, MB02, KG1a, TF-1 etc.). I will improve conditions to grow AML cells in vitro by seeding them on human fetal liver stroma. This will help us to establish a great drug screening condition.

9. Objectives of the Proposal:

Aim 1. How does GFI-1 regulate HSC maintenance and self-renewal *in vivo*?

Aim 2. How does GFI-1 promote growth arrest and differentiation of normal human HSPC, AML cell lines and in human Leukemic cells?

Aim 3. Gfi-1 transcription level manipulation by investigating mechanism(s) of upstream transcription factors regulation.

10. Innovations in the project: (Give in about 100 words)

I will get a set of transcription factors those are required for HSC self-renewal and differentiation in normal and leukemic stem cells. Analysis of this study will give us knowledge and tool to regulate hematopoietic stem cells. I will use umbilical cord blood (CB) cells to translate the observation from mouse to human Stem cells. AMLs are not easy to grow and I will try to grow on human fetal liver stroma to grow them.

11. Significance of the outcome of the project: (Give in about 150 words)

This project will highlight the transcriptional network that regulates leukaemia stem cell self-renewal and differentiation along with normal hematopoietic stem cell. I will use small molecule inhibitor to modulate expression or regulation of different up stream Gfi-1 regulator to modulate its level in different leukaemia to differentiate them. *In vitro* study will give us a way to understand regulation of leukaemia stem cells self-renewal and differentiation.

12. Relevance in Public Health:

Outcome of this project will focus on regulation of leukaemia stem cells (LSCs). The inhibitor of transcription factors responsible for self-renewal and differentiation of LSCs can be used for treatment. Inhibitor in combination with radiotherapy or chemotherapy will kill LSCs more efficiently especially in case of FAB M0 and M1.

Date 04/04/2016

Signature of the Fellow /Faculty

Assistant Professor
Stem cells/ Cell Culture Lab
Centre for Advance Research
KGMU, Lucknow,
226003