DATE: Day 10 Month 05 Year 2018

SUMMARY of
2017 RESEARCH RESULTS REPORT
For International Collaborative Research with IPR, Osaka University

Research Title		
Applicant	**Name**	**K. Sekar**
	Affiliation	**Indian Institute of Science**
	Present Title	**Professor**
Research Collaborator (Host PI)		**Professor Atsushi Nakagawa**

Summary

Structural and functional studies on enzymes involved in Essential Amino-acid biosynthetic pathways.

Pyruvate kinase is an enzyme that catalyzes the conversion phosphoenolpyruvate (PEP) to pyruvate by transfering phosphate group, yielding one molecule of ATP and it is an important rate limiting steps in energy production. This enzyme is involved in the final step of glycolysis. Thus, any inhibitor designed against that could bind Pyruvate kinase will lead to cell death by preventing vital energy production metabolic pathway in the cells. It has been demonstrated that the production of ATP by cancer cells is much higher than the normal cells that is enhancing the cell growth and changing the physiology of the surrounding tissues. Thus, pyruvate kinase can be used as a potential drug target in cancer cells. Detailed analysis of the complex structures may reveal the reaction mechanism involved in phosphate transfer. Further, co-crystallization with other potential ligands will also be carried out. The protein crystals were mounted and checked in the X-ray facility present in Indian Institute of Science, Bangalore.

Structural and functional characterization of enzymes that are involving carbon-di-oxide fixation.

In the past 150 years, human activities have pumped enough carbon dioxide into the atmosphere to raise its levels higher than they have been for thousands of years. Reducing the atmospheric CO_2 level has received a great deal of attention recently as an approach to combat global warming and fossil-fuel shortages, but this process remains challenging. Biological CO_2 fixation is one of the most important approaches to solve these problems. Enzymatic CO_2 reduction has been examined extensively as a promising approach to greenhouse gas fixation and the production of renewable fuels and chemicals. Pyrococcus horikoshii FDH (FDH-Py) produces formate in *E.coli* after overexpressing in anaerobic condition. The reaction mechanism is not well understood. Identification of proteins interacting with FDH-Py is important to understand the reaction mechanism. In order to identify the interaction partners of Pyrococcus horikoshii FDH in vivo, both in aerobic and anaerobic conditions, protein-protein interactions assay will be done to find out interaction partners. Myc-tag needs to be introduced into the C-terminus of pER-22b-FDH. Myc tagged recombinant FDH-py will be immunoprecipitated using anti-Myc antibody. Mass spectrometry will be used to identify the nature of the interactions and the same will be optimized using formaldehyde. Identification of FDH-Py interacting partners will shed light on the reaction mechanism of formate production from CO_2.

***Deadline: May 18, 2018**
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