DATE: Day <u>25</u> Month <u>07</u> Year 2016 SUMMARY of 2015 RESEARCH RESULTS REPORT For International Collaborative Research with IPR, Osaka University

Research Title		Structure of a deubiquitylating enzyme, AMSH
Applicant	Name	Hyun Kyu Song
	Affiliation	Division of Life Sciences, Korea University, Korea
	Present Title	Structural study of full-length deubiquitylating enzyme, AMSH
		and an Atg protein
Research Collaborator (Host PI)		Atsushi Nakagawa

Summary

The research proposal was study on the full-length AMSH, an associated molecule with Src homology 3 domain of STAM. We and other researchers solved the domain structures and they are published and we only focused on the full-length AMSH, but so far unsuccessful and instead, we worked on other project, an autopahgic molecule AtgX ("X" is used instead of number because it is not a completed project yet). The AtgX is a membrane protein required for autophagy and forms a complex with AtgX as well as AtgX. We are planning to solve the first AtgX structure and to do further biochemical and cell biologic study with interacting partners.

Most of the AtgX protein crystals were diffracted about 4.2 angstrom and they were exhibited severe anisotropism (less than 7.0 angstrom) according to a specific axis. It may be derived from intrinsic helical property of the protein. The best crystal was diffracted up to 3.2 angstrom and it was collected as the highest resolution data set. Because of radiation damage during data collection, the data was cut off by 3.6 angstrom. Three-wavelength MAD method was tried with Seleno-methionine incorporated crystals (peak=0.974917, edge=0.979296, and remoteH=0.8) but the crystals were vulnerable to radiation damages and poorly diffracted to solve the phase problem.

To obtain better quality of AtgX protein crystals, we are currently trying seeding technique for different crystal form, generating different length constructs, mutating several residues for decreasing surface entropy and introducing metal binding properties.

^{*}Deadline: July 31, 2016

^{*}Please submit it to E-mail: tanpakuken-kyoten@office.osaka-u.ac.jp.

^{*}We accept only PDF file. Please file it after converting WORD to PDF.

^{*}Please describe this summary within 1 sheet. Please DON'T add some sheets.

^{*}This summary will be published on the web.