DATE: Day 13 Month 05 Year 2020

SUMMARY of

2019 RESEARCH RESULTS REPORT

For International Collaborative Research with IPR, Osaka University

Research Title		**Development of a new method to conjugate the defensin peptide to the carrier protein P64K using the thioester chemistry.**
Applicant	**Name**	**Hilda Elisa Garay Pérez [(1)] and David Diago Abreu [(2)]**
	Affiliation	**Center for Genetic Engineering and Biotechnology (CIGB)**
	Present Title	**(1)Senior Researcher. Head of Synthetic Peptides Group.** **(2) Head of Analytical and Purification Laboratory on Peptides Group**
Research Collaborator (Host PI)		**Hironobu Hojo (Professor)**

Summary

Actually, is a big problem in a worldwide and in Cuba the plant diseases caused by different microorganism, focused the impact in crop production. As an effort to get a solution for disease control, the researchers have identified the plant defensins as a good possibility. Plant defensins are small, cysteine-rich peptides, which inhibit the growth of a wide range of microorganisms and are part of the defense mechanisms of plants against diseases. This defensin was isolated from a plant and develops a strong anti- microbe activity in vitro and ex vitro against important bacteria, fungus and oomycetes pathogens. Herewith, the constitutive expression of this defensin in transgenic plants increased the resistance to many plant diseases with high economic impact. An important issue is the production of mono- and polyclonal antiserum that would specifically detect the recombinant defensin from transgenic soybean lines. The low immunogenicity of simple peptide is a main problem to generate immune response against these molecules. The problem presented can be solved with the use of adequate systems of presentation to the immune system, which allows an increase in the levels of induced antibodies and their functionality. Conjugation to carrier proteins, presentation of the antigenic sequence by multimerization by linear polymerization, strong adjuvants like recombinant cytokines, conjugation to polysaccharides and the use of multiple antigen peptides are some of successfully strategies.

Currently at CIGB; we generate antibodies against defensin using the conventional protocol for conjugation using Succinic Anhydride- Carbodiimide Method to P64K. Could be an attractive methodology conjugate the defensing to a carrier protein using the thioester chemistry to generate effective immune response and compare it with the response obtained by previously protocol

-Different methods for P64K activation and conjugation studies using a model peptide were performed. The results obtained were not successful.

-Different methods for Dendrimer (as a carrier protein model) activation and reaction with 2-aminoethanol as a conjugation model were performed. The results obtained were not successful.

-Different methods for Dendrimer (as a carrier protein model) activation and reaction with Metoxiglicin as a conjugation strategy were performed. The results obtained were not successful.

The activation and conjugation methods studied were not successful, therefore is necessary to perform additional studies in order to achieve the desired results. The MeOGly methodology could be an alternative to obtain the conjugation of defensin peptide to P64K.

***Deadline: May 15, 2020**

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

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

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