Idaho Incubation Fund Program

Progress Report Form

Proposal No. IF13-006

Name: Juliette Tinker

Name of Institution: Boise State University

Project Title: Characterization of Antigens for a Staphylococcal Bovine

Mastitis Vaccine

Information to be reported in your progress report is as follows:

1. Provide a summary of project goals/milestones for the period just completed, accomplishments for the period just completed, and plans and goals for the coming quarter:

We have previously constructed and purified chimeric *S. aureus* proteins as potential vaccines against bovine mastitis in dairy cattle. Preliminary data indicates that the vaccine containing the *S. aureus* IsdA antigen is immunogenic when delivered intranasally. The proposed project will permit a better understanding of the role of IsdA and additional vaccine candidates in bovine mastitis. The goals of the proposed project were two-fold: 1) to characterize the immune response to the IsdA antigen in healthy dairy cows, and cows with subclinical and clinical *S. aureus* mastitis and 2) to characterize the immune response to additional *S. aureus* antigens, including ClfA, IsdH and FnBP.

For the project period just completed (first three months of work) the proposed timeline includes the identification and sampling of S. aureus positive infected cows, and S. aureus negative healthy cows as well as the cloning and purification of *S. aureus* antigens for subsequent ELISA. Work began immediately at the University of Idaho (Dr. Mark McGuire) to prepare and submit Institutional Animal Care and Use Committee protocols for the sampling of blood and milk from commercial dairies. Dr. McGuire also contacted and arranged for the involvement of two potential commercial dairies, as well as the University dairy, during this period. Three University of Idaho students and one Post-doctoral fellow (Katherine Hunt) are, and will continue to be, involved in culturing, sampling and shipping samples to Boise State. U of I students include an undergraduate (Katelyn Steinkamp), a MS (Brittany Casperson) and a Ph.D. student (Janet Williams). Efforts have been somewhat hampered by difficulties with IACUC approvals and arranging a funding contract between universities. Work at Boise State has included the employment of an undergraduate student (Shandra Jeffries) and a MA student (Tyler Wines) to develop milk and blood ELISA assays, quantitative PCR and cloning and purification of S. aureus antigens. The IsdA antigen has been previously cloned and

purified (Arlian and Tinker, 2011). During the project period the ClfA antigen was cloned into the pBAD41 vector for purification using nickel chromatography and also cloned into the pARLDR19 vector for construction of a chimeric protein as a potential vaccine. These clones have been confirmed by PCR and the ClfA chimera was successfully purified. Current efforts are focused on purification of ClfA using nickel chromatography and optimization of ELISA assays.

Plans for the upcoming quarter include the completion of screening, sampling and shipping milk and blood from the University of Idaho, and initiation of ELISA analysis on the ClfA and IsdA antigens. In addition, standard curves for quantitative RT-PCR will be developed. All milk obtained from University of Idaho will be plated and *S. aureus* isolates stocked for later DNA sequencing analysis of select antigens.

REQUESTED USED

2. Provide a summary of budget expenditures for the period just completed:

Budget Expenditures (The University of Idaho):

	NEGOLOTED	USLD
Total salaries and fringe benefits	\$10,265	\$0
Total supplies	\$7,360	\$0
Total other expenses	\$ 1,500	\$0
Total travel	\$ 875	<u>\$0</u>
Total direct costs	\$20,000	\$0
Budget Expenditures (Boise State):		

Total salaries and fringe benefits	\$16,663	\$1100
Total supplies	\$13,337	\$2654.43
Total direct costs	\$30,000	\$3754.43

3. List patents, copyrights, plant variety protection certificates received or pending:

Cholera toxin chimera and its use as a staph vaccine. U.S. Patent pending. Submitted 12/16/11. Application # 13328686

4. List invention disclosures, patent, copyright and PVP applications filed, technology licenses/options signed, start-up businesses created, and industry involvement:

Inclusion of new collaborator/consultant during project period: Dr. Brian Mitchell, DairyTeam Nutrition and Veterinary Consulting, Eagle, ID. Dr. Mitchell has expertise in veterinary vaccines and numerous ties to the dairy industry in Idaho.

5. Include funding burn rate:

\$1251.5/mth

6. Any other pertinent information: