



## REQUEST FOR PRE-PROPOSALS

*Please Copy and Distribute to All Interested Parties*

The USDA-NIFA Southern Regional Aquaculture Center solicits response from qualified multi-state teams interested in participating in the regional project:

### **Development of Lateral Flow Immunoassays for use in Aquaculture**

SRAC's Board of Directors has authorized up to \$250,000 for a 2-year project on *Development of Lateral Flow Immunoassays*. This project will be developed using the "comprehensive method" where a team of multi-state scientists having demonstrated records of expertise in the subject complete a single pre-proposal that addresses all project objectives. One proposal will be selected for funding based on review by a committee of scientists not involved in any of the proposals that are submitted.

#### **Background**

Lateral flow immunoassays (LFIA) are an emerging technology that have been used as effective Point-of-Care diagnostic tools for many disease conditions in human and veterinary medicine. Most recently, at-home COVID-19 test kits were widely distributed by the US government, which demonstrates the utility and portability of LFIA as a simple, yet effective POC diagnostic tool. In aquaculture, LFIA have been developed experimentally to detect some viral, bacterial, and parasitic pathogens, but these tests are not readily available for use or applicable to US aquaculture. Compared to other available diagnostic tools (i.e., virus isolation, PCR, qPCR), LFIA can be easily deployed in the field by fish farmers and fish health professionals to screen and monitor select infections in under an hour. This is especially useful for viral pathogens or fastidious microbes, which can often take days to yield positive results in culture. LFIA results are easily visualized without technical expertise or sophisticated equipment. Moreover, extended shelf life without refrigeration makes LFIA suitable for use in fish farm conditions and make LFIA an attractive option as a cost-effective, rapid, and simple test that can be used as a primary pondside screening tool at farms and resource limited diagnostic laboratories.

#### **Objectives**

1. Develop LFIA for important aquaculture pathogens affecting catfish and ornamental aquaculture, particularly those that require prolonged culture conditions for accurate confirmatory diagnosis (Channel catfish virus, Megalocytivirus, *Streptococcus iniae*, *Edwardsiella ictaluri*, *Erysipelothrix piscisicarius*, *Vibrio* spp., *Mycobacterium* spp., etc.).
2. Provide proof of concept using laboratory induced disease/experimental infectivity trials for target species.

## Experimental Approach

The research team will identify and express the aquatic pathogen's target antigens using a recombinant expression vector. Monoclonal antibodies against the expressed target antigens should be produced in a suitable species (e.g., mouse or rabbit), and the specificity of the monoclonal antibodies to the pathogen's target antigen confirmed using enzyme-linked immunosorbent assay (ELISA) prior to assembling the lateral flow immunoassay (LFIA). The purified anti-pathogen antigen IgG will be conjugated to gold particles. Glass fiber samples and conjugate pads will be treated and processed using the appropriate buffer and colloidal gold anti-pathogen antigen IgG conjugate. The LFIA should consist of test (T) and control (C) lines on a nitrocellulose membrane coated with anti-pathogen antigen IgG and species-specific anti-Ig antibodies, respectively.

The second phase of the research should provide proof of concept using laboratory-induced disease/experimental infectivity trials for target species (catfish/ornamental). The diagnostic sensitivity and specificity of the assay should be determined by evaluating performance on both blood samples and pooled internal organs (liver, spleen, kidney) samples collected from reference populations of fish defined by their exposure status from experimental trials. In addition, the assay's analytical sensitivity/ limit of detection and specificity will be evaluated. After proof of concept, the final processing should be simplified for user accessibility and ease, based on results from specific fish/pathogens.

## How to Respond

Pre-proposals must address all objectives. Preference will be given to pre-proposals that show a high degree of collaboration and coordination among participants. To meet the criterion for a regional project, the pre-proposal must include collaboration from scientists in two or more states or territories in the Southern Region (Alabama, Arkansas, Florida, Georgia, Kentucky, Louisiana, Mississippi, North Carolina, Oklahoma, Puerto Rico, South Carolina, Tennessee, Texas, U.S. Virgin Islands, and Virginia).

The pre-proposal must include a one-page vita for each participant and a proposed budget for each participating institution or organization. Pre-proposals, vitae, and budgets that are not in the proper format will not be considered. (See "Guidelines for Writing a SRAC Pre-Proposal (Comprehensive)" file attached or contact Kristen Thompson with the SRAC office at 662-686-3269.)

Send an electronic copy of the pre-proposal in Word format to Jimmy Avery, SRAC Director as an email attachment (jimmy.avery@msstate.edu) by **July 20, 2024**. Proposals received after that date will not be considered.