LAKE SUPERIOR
STATE UNIVERSITY

January 13, 2015

Dr. Jun Li
School of Biological Science

Dear Dr. Li,

Congratulations! I am pleased to inform you that the Sabbatical Committee has recommended approval of one semester of sabbatical leave for either Fall 2015 or Spring 2016 to the Provost and Board of Trustees. The committee, which met on January 13, found your proposal, entitled *"Study of fish infectious diseases and immunity in the Great Lakes: Enhancing the research ability of LSSU's Fish Disease Laboratory"*, well designed and having high potential of professional growth and enhancing the capabilities of Lake Superior State University. We look forward to reading the final report detailing your research and accomplishments from this sabbatical.

Sincerely,

Ron Hutchins, for the Sabbatical Committee
(Allan, Crandall, Childs, Kabke, Finley, McDonald, Swedene)

cc Morrie Walworth, Provost ✓
Barb Keller, Dean

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LAKE SUPERIOR STATE UNIVERSITY

OFFICE OF THE PROVOST

APPLICATION FOR SABBATICAL LEAVE

(Refer to Section 15.4 of the Faculty Association Agreement)

I. Name Jun Li Date 11/13/14

Department Biological Sciences Ext. No. 2094

Home Address [redacted] Home Phone [redacted]

II. Application for leave during the following (*indicate semester and/or year*):

☐ Fall ☐ Spring X Full Year

III. Number of years of faculty service (*minimum of 5 years required*) 5

IV. Year your tenure at LSSU was earned (*tenure required*) 2013

V. Semester or year of last sabbatical (*if applicable*) NA
(*minimum of 5 years since last sabbatical required*)

VI. I agree to return to the University and to provide a complete written report (electronic) to the Provost upon the completion of my sabbatical semester(s) as denoted in section 15.4 of the Faculty Associate Contract.

11-13-14

Signature of Faculty Applicant Date

Provost Office

VII. Signature of your Dean indicating his/her awareness of the application: NOV 14 2014

Lake Superior State University

11-14-14

Signature of Dean Date

VIII. Attachments:

a. Title and Description of Sabbatical Project (Required and described on the next page)
b. Support Documents (Optional but strongly suggested)
c. Curriculum Vitae (Required)

Title: Study of Fish Infectious Diseases and Immunity in the Great Lakes: Enhancing the Research Ability of LSSU's Fish Disease Laboratory

I. Executive Summary

Over the past 5 years, I have been collaborating on several research projects focusing on aquatic animal health with my Chinese collaborators. One project was completed in early 2014 and one will end in 2015. There are 2 newly funded projects will last from 2014/2015 through the end of 2018. In 2013/2014, I also applied and was funded for 3 research projects at LSSU from different agencies. These three new research projects will start soon and span 1 or two years (2015-2016). I would like to dedicate more time to these projects to ensure all research goes well in the Fish Disease Laboratory at LSSU. Additionally, I would like to spend more time collaborating with external scientists on research activities, data analysis and writing manuscripts. Thus, I am requesting a full year of sabbatical leave, which will allow me to focus on the scientific research activities in the field of aquatic animal health, develop new skills and research abilities for the Fish Disease Lab, and expand my professional network and research program. My sabbatical activities will benefit LSSU by: 1) strengthening the reputation of Biology faculty as scientists and teachers, 2) incorporating current research approaches into my curricula, especially for the unique fish health program, 3) developing new skills and expanding research abilities in the Fish Disease Lab, 4) providing more research training opportunities for current biology students and enhancing their research experiences, and 5) acquiring additional funding to create new student research experiences for LSSU students.

II. Project Description

Introduction

There are more than 170 fish species in the Great Lakes, Michigan, surrounded by Lake Michigan, Lake Huron, Lake Superior and Lake Erie, has the most diverse fishing opportunities in the USA even over the world. Healthy fisheries in Michigan waters are economically and culturally important to the state. In recent years, however, more and more emerging and reemerging infectious diseases have posed a large threat to the sustainability of Great Lakes fisheries. Habitat changes due to climate warming and anthropogenic influences have not only impacted the behaviors of most fish, but are considered as the key factors altering their physiological conditions leading to increased susceptibility to a variety of infectious diseases. Recent emerging fish health concerns include viral hemorrhagic septicemia (VHS), epizootic epitheliotropic disease (EED), bacterial kidney disease (BKD), bacterial cold water disease (BCWD) and furunculosis. A sustainable fishery cannot be maintained with such deadly viral and/or bacterial diseases, however our knowledge of their prevalence in the Great Lakes is too limited to provide managers with the key information necessary to create informed management strategies. In order to maintain more sustainable fishery resources in the Great Lakes, it is very

important to better understand: 1) effects of the aquatic ecological changes on the Great Lakes fisheries (e.g. large-scale movements, feeding, spawning and nursery habitats), 2) prevalence and distribution of potential pathogens in their natural habitats. Knowledge gained from these ecological studies will allow the development and implementation of effective diagnostic tests and control strategies for these pathogens.

To address these questions, I submitted several research proposals for funding in the past several years. Most recently, three proposed projects have been granted funds from Great Lakes Fishery Trust (GLFT), Bureau of Indian Affairs/Great Lakes Restoration Initiative Funds (BIA/GLRIF), and Algal Scientific Inc.

Project 1 : Bureau of Indian Affairs-Great Lakes Restoration Initiative Funds (BIA/GLRIF) *(2015, 1 -2015, 12; total fund: $142,964, LSSU part: $80,033, Dr. Li is the Co-PI)*

Project Title: Monitoring fish movement and fish condition in tributaries of Whitefish Bay.

Project summary: Whitefish Bay historically is the most important site to the Ojibwa tribes, especially to the Bay Mills Indian Community (BMIC) for their recreational and fishery activities. BMIC has actively managed the Lake Superior fishery since the 1970s, and recognizes the importance of protecting the habitat and forage fish that support this fishery to ensure its sustainability. However, management of Great Lakes fish populations can be complex due to variation in fish movement and condition. Habitat degradation is a leading cause of declines in fish populations in Lake Superior and its tributaries. Therefore understanding river habitat use and timing of use by Great Lakes fishes is important for conservation and rehabilitation efforts. Our objectives for this project includes: 1) characterize movement patterns (timing and size) of Great Lakes fishes in and out of Whitefish Bay tributaries, and 2) assess the physiological health status of Great Lakes fish species using Whitefish Bay tributaries.

I will collaborate with Dr. Ashley Moerke (LSSU professor) and Dr. Paul Ripple (Bay Mills Biological Services) in this project. We will start to collect fish samples and their related bacterial/viral samples from early April through the whole Summer/Fall of 2015. Then I will lead a research team including several LSSU students for bacteriological, virological, and immunological assays of the collected samples. I would like to use part of my sabbatical time to train LSSU students and perform related research and data analysis.

Project 2: Great Lakes Fishery Trust *(GLFT, 2014, 9-2016, 9; Total Fund: $334,591; LSSU part: $64,054, Dr. Li is the Co-PI)*

Project Title: Re-emergence of Epizootic Epitheliotropic Disease Virus: Potential

Effects and Development of Improved Diagnostics & Control Measures

Project summary: The Epizootic Epitheliotopic Disease Virus (EEDV) is a serious infection that can impede the Lake Trout Rehabilitation Program in the Great Lakes. Since information on EEDV is vastly lacking, the proposed study aims to unravel important aspects of EEDV pathology such as the disease course, latency, and host immune responses. The newly generated information will lead to the development of improved diagnostic tools and the development of novel control strategies.

I am the co-PI for the GLFT grant (PI is Dr. Faisal from MSU) and will work together with a group of scientists from Michigan State University, University of Florida, Michigan Department of Natural Resources (MDNR) and United States Fish and Wildlife Services (USFWS). This is a great opportunity for me, and for LSSU, to be able to collaborate with so many highly respected leading scientists and federal/governmental researchers in the field of fish health. It will allow me to expend my scientific networking and collaborations in the fish health area. More importantly, such collaborative research will generate more opportunities for LSSU students to gain research experience, internships and future postgraduate studies in those institutions.

In the GLFT projects, I intend to use my sabbatical time to generate a series of antibodies against the EEDV virus and other related pathogens, as well as antibodies against fish immunoglobulins (IgM/IgT), which are very important and essential reagents for the future diagnostic and host-pathogen interaction studies. Such information will enhance the reputation and research ability of LSSU Fish Disease Laboratory for future collaborations and funding opportunities.

Project 3: LSSU & Algal Scientific Inc. Beta-glucan Study *(2015,1-2015,8, Total fund: $20,576, Dr. Li is the PI)*

Project Title: Effect of Dietary Beta-glucan Derived from Algae on Growth Performance, Disease Resistance and Immune Response in Atlantic Salmon Fry

Project summary: Dietary beta-glucans, a nutritional supplement derived from the cell wall of various plants, fungi, bacteria, yeasts and seaweeds, have been well documented in the promising enhancement of growth, survival and protection against infectious disease in various animals including fish and shellfish. In the present project, we will assess the growth and disease resistance of Atlantic salmon as well as the involved immune-stimulating effects of dietary supplementation of a new algae derived beta-glucan (**Algamune AM**) compared with traditional yeast derived beta-glucan (**Macrogard**). Atlantic salmon fry (6-8 inches) from the Lake Superior State University Aquatic Research Laboratory (LSSU-ARL) will be used as test subjects. All tests will be performed at 10-12^{o}C in a controlled environmental chamber at LSSU. The project will

last 6 months and assess changes in condition factor, immune system response and disease resistance to Furunculosis (caused by a bacterium of *Aeromonas salmonicida*), as a result of incorporating algal derived beta-glucan in the diet of the fish.

The beta-glucan study is a contract project from a Michigan company (Algal Scientific Inc.). I generated this contract with the company to understand the immune-stimulating effects of one of their products (algal derived beta-glucan, AM) on fish disease resistance and growth performance. They also intend to support future studies for other products and in different fish species and on developmental stages pending the effectiveness of the current contract study. This project involves 2 LSSU senior thesis students and several junior students. It allows the LSSU's Fish Disease Lab to tie contracts with Michigan industry and provides opportunities for our students, not only for research experiences but also for their future carrier development. I would like to use part of my sabbatical time for data analysis and preparation of the final reports. More importantly I intend to secure further study contracts with them and other companies.

Outcomes

1. Finish the research projects and gain new knowledge for better understanding the aquatic ecological aspects of the Great Lakes fish habitats, and the prevalence of related fish pathogens in the Great Lakes. (*Outcomes: finish writing a review paper on the Great Lakes fish diseases; 1-2 peer-reviewed publications with 1-2 LSSU students as co-authors; new knowledge acquired to incorporate into Biology curricula).*

2. Generate novel reagents and techniques/skills for effective diagnosis of fish pathogens and disease control strategies to expand and strengthen the research abilities of LSSU's Fish Disease Lab (FDL) to successfully compete for more funding. (*Outcomes: skills/facilities improvement in FDL; new skills acquired to incorporate into Biology curriculum and apply to current research projects).*

3. Expand collaborations and build additional avenues for research in aquatic animal health, while creating more LSSU student research opportunities and enhancing the LSSU-FDL's reputation as a productive research facility *(Outcomes: publications; new research proposals)*

Timeline

Prior to Sabbatical:

January – April, 2015

1. Fish sample collection for GLFT project, preparation and purification of recombinant proteins/fish immunoglobulins.
2. Preparation of fish feeds with/without beta-glucan supplementation.

April –August, 2015

1. Start feeding trials for beta-glucan study and complete challenge test, and hematological/immunological analysis.
2. Start fish sample collections from Whitefish Bay (BIA-GLRIF projects), finish field analysis.

Sabbatical period:

September – October 2015

1. Complete data analysis and write final report for the contract study "Effect of Dietary beta-glucan Derived from Algae on Growth Performance, Disease Resistance and Immune Response in Atlantic Salmon Fry".
2. Finish writing the review paper on infectious diseases related to the Great Lakes Fishes.
3. Begin preliminary data analysis on Whitefish Bay project; start bacteriological analysis of collected samples.
4. Immunize animals to generate antibodies (collaborate with Dr. Faisal at MSU and my postdoctoral supervisor Dr. Sunyer at University of Pennsylvania, (UPenn)

November, 2015– January 2016

1. Complete Whitefish Bay analyses and submit publication(s).
2. Visit MSU and UPenn to discuss research progress and submit 1-2 collaborative proposals on aquatic animal health to GLFT or NSF.
3. Finish MDNR contract for BKD diagnostic assay.
4. EEDV diagnostic assays in fish samples from MDNR state fish hatcheries.

February – May 2016

1. EEDV diagnostic assays in fish samples from MDNR state fish hatcheries (continue)
2. Complete MDNR contract for thiamine assay.
3. Present research findings at 2016 Annual Fish Health Workshop.
4. Submit 1-2 manuscripts to peer-reviewed journals (*Journal of Aquatic Animal Health, Journal of Fish Diseases or Fish & Shellfish Immunology*).

UNIVERSITY of PENNSYLVANIA

The School of Veterinary Medicine
Department of Pathobiology
3800 Spruce Street
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11/10/2104

To Whom It May Concern,

I am very pleased to write this letter in support of Dr. Jun Li's application for his sabbatical research project. As his post-doc supervisor, I am very happy to know he is doing well at Lake Superior State University.

Dr. Li had been working in my lab for 8 years and he did an awesome research and made great contributions to the field of developmental and comparative immunology. As a previous lab member, he is always welcome back home. We would certainly be happy to have him here again for a couple weeks or months to work on his sabbatical research project to produce recombinant proteins for generating salmonid fish antibodies in my laboratory. We will do whatever is necessary to help him out and provide our supports for more success on his sabbatical research project.

Oriol Sunyer, Ph.D

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Bureau of Indian Affairs
Great Lakes Restoration Initiative Funds

Bay Mills Indian Community Request for Funding
2014-2015

Monitoring fish movement and fish condition in tributaries of Whitefish Bay

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Introduction

Over 85 fish species inhabit Lake Superior, many of which historically and currently are viewed as vital commercial, recreational, subsistence, and cultural resources to the Ojibwa tribes. As a result, the Bay Mills Indian Community (BMIC)hasactively managed the Lake Superior fishery, with an emphasis on Whitefish Bay, since the 1970s and recognizes the importance of protecting the habitat and forage fish that support this fishery to ensure its sustainability. However, management of Great Lakes fish populations can be complex due to variable life history characteristics, including variation in fish movement and condition.

Fish may exhibit large-scale movements to feeding, spawning, and nursery habitats. However, the location and timing of historical and current fish movements between lake and river ecosystems are poorly documented, especially in Lake Superior waters (see Herbert et al. 2011 for Lake Huron information). Studies that have been done to date generally are species-specific, and focus on a handful of recreationally important species (i.e., walleye, lake trout, lake sturgeon) in specific geographic regions (Landsman et al. 2011). Movement patterns of forage species receives little attention and remains a research gap across all lake systems (Landsman et al. 2011). An understanding of the timing and location of fish movements, of both forage and game species, can help understand the reproductive biology of species, identify critical habitats (Cooke 2008) and potential barriers to that habitat, improve or regulate recreational fishing opportunities (Huckins and Baker 2008), and assess population size and fish condition for species that may otherwise be dispersed (Landsman et al. 2011).

The majority of all Great Lakes fish species spawn in nearshore or river environments, with nearly half known to migrate from lake environments to spawn in rivers (23 species spawn in lakes and streams, 28 species primarily spawn in streams, 19 exclusively spawn in streams) (Lane et al. 1996), in some cases followed by larval or juvenile out-migration. Timing and size of spawning runs may vary depending on species, natural environmental factors, and anthropogenic influences. For example, flooding events have been shown to alter movement of juvenile coho salmon (Lonzarich et al. 2009), while changes in thermal regimes related to thermal discharge activity altered smallmouth bass movement (Cooke et al. 2004).

The ecological importance of streams is likely magnified by the time species spend in spawning aggregations or early life stages in or near streams. In past years we have studied the extent to which nutrient and energy inputs from tributaries and nearshore areas are available to these stocks. We know little about the movement patterns of fishes between these ecosystems as well as the condition/health status of fishusing these streams.

Healthy fisheries in the Great Lakes Waters are economically and culturally important to the State and Bay Mills Indian Community. However,in recent years, more and more emerging and reemerging infectious diseases have posed a large threat to the sustainable Great Lakes fisheries. Habitat changes due to the climate warming and anthropogenic influences have not only impacted the behaviors of most fish, but also have been considered as the key factors to alter theirphysiological conditions as a result of increasedsusceptibility to various infectious diseases. Recent emerging fish health concerns include Viral HemorragicSepticimia (VHS), Epizootic Epitheliotropic Disease (EED),Bacterial Kidney Disease (BKD), Bacterial Cold Water Disease

(BCWD) and Furunculosis.A sustainable fishery cannot be established with such deadly viral and bacterial diseases, however our knowledge on their prevalence in the wild is too limited to provide managers with the key information necessary to make informed management strategies. Spawning migrations often result in individual fishes aggregating in small areas, which creates a fitting time to assess fish physiological condition.

Needs

Habitat degradation is a leading cause of declines in fish populations (Warren and Burr 1994) across the United States, and this trend is no different in Lake Superior and its tributaries. Therefore understanding river habitat use and timing of use by Great Lakes fishes is important for conservation and rehabilitation efforts. The Bay Mills Indian Community recognizes the importance of having functioning systems to support natural flow of water, native fish and wildlife, and a healthy tribal community.

We are requesting funding from the Bureau of Indian Affairs under their Great Lakes Restoration Initiative to support research on the use of Whitefish Bay tributaries by Great Lakes fishes and the health of those fish. Project costs will cover the technical expertise from the Bay Mills Indian Community and Lake Superior State University, will support four technicians (Bay Mills members or LSSU students), and will pay for supplies and travel for the proposed research.

Project Objectives

1) Characterize movement patterns (timing and size) of Great Lakes fishes in and out of Whitefish Bay tributaries.

2) Assess the physiological health status of Great Lakes fish species using Whitefish Bay tributaries.

Project Approach

Objective 1: Characterize movement patterns (timing and size) of Great Lakes fishes in and out of Whitefish Bay tributaries.

- Three tributaries will be selected from Naomikong Creek, Halfaday Creek, Roxbury Creek, and Angkodosh Creek

- Fish movement will be assessed in the three tributaries using paired fyke nets (anchored back-to-back) longitudinally in each stream (approximately 100 m, 500 m, and 2 km upstream from the rivermouths)

- Fish captured will be identified and measured for length and weight to characterize species use and size; reproductive state will be recorded; scale samples and genetic material will be collected for future use

- Fish captured will be tagged using external tags (e.g., FLOY tags) and released in the direction which they were captured (i.e., fish caught in the downstream net are assumed to be moving upstream and will be released upstream)

- Sampling will be collected on a biweekly basis from ice out to ice on (approximately mid-April to early December) to identify spring, summer, and fall migrations

Objective 2: Assess the physiological health status of Great Lakes fish species using Whitefish Bay tributaries.

- The health of 5-10 individuals of each Great Lakes fish species captured in each Whitefish Bay tributaries will be determined

- External deformities of all fish will be recorded and blood will be drawn in the field prior to releasing fish

- Bacterial or viral pathogens will be isolated from infected fish (e.g. fish with rotted fin, skin ulceration, hemorrhagic syndromes)

- Stress hormones (cortisol, prolactin etc.) and serum glucose levels, total white blood cell count, total red blood cell count, hematocrit, hemoglobin will be assessed

- Fish health status (have been exposed or not to the above mentioned viral and bacterial diseases) will be diagnosed by ELISA or qPCR

Table 1.Tribal Members of the Bay Mills Indian Community.

Total Tribal enrollment	1,820 people
Total on Reservation (Bay Mills & Sugar Island)	799 people
Total in Service area	1,201 people
Total in Chippewa County	1,179 people
Total in Michigan	1,452 people

Table 2.Resources of the Bay Mills Indian Community.

Reservation/Trust land	3,225 acres
Whitefish Bay Reserve	77,170 acres
Waishkey Bay	1, 500 acres
Inland Lakes	72.5 acres
Streams/Rivers	5 miles
Wetlands	1,085 acres
Wetland Preserve	460 acres
Shoreline (Great Lakes & inland)	5.16 miles
Forestland	1,653 acres
Ash Preserve	10 acres
Low Density Residential	250 acres
Governmental (Tribal Center, etc.)	10 acres
Commercial (Casinos, etc.)	37 acres
Golf Course	220 acres

References

Cooke, S.J., C.M. Bunt, and J.F. Schreer. 2004. Understanding fish behavior, distribution and survival in thermal effluents using fixed telemetry arrays: a case study of smallmouth bass in a discharge canal during winter. Environmental Management 33:140-150.

Cooke, S.J., 2008. Biotelemetry and biologging in endangered species research and animal conservation: relevance to regional, national, and IUCN Red List threat assessments. Endangered Species Research 4:165-185.

Herbert, M., M. Khoury, T. Bowe, and L. Cole. 2012. Development of conservation priorities for migratory, river-spawning fishes in the Michigan waters of Lake Huron. Final Report to Michigan Coastal Management Program, MDEQ, Project #10D-0.10.

Huckins, C.J. and E.A. Baker. 2008. Migrations and biological characteristics of adfluvial coaster brook trout in a south shore Lake Superior tributary. Transactions of the American Fisheries Society 137:1229-1243.

Landsman, S.J., V.M. Nguyen, L.F.G. Gutowsky, J. Gobin, K.V. Cook, T.R. Binder, N. Lower, R.L. McLaughlin, and S.J. Cooke. 2012. Fish movement and migration studies in the Laurentian Great Lakes: research trends and knowledge gaps. Journal of Great Lakes Research 37:365-379.

Lane, J.A., C.B. Portt, and C.K. Minns. 1996. Spawning habitat characteristics of Great Lakes fishes. Can. MS Rep. Fish.Aquat. Sci. 2368: v+48p.

Lonzarich, D.G., R.P. Franckowiak, and M.D. Allen. 2009. Summer movements of juvenile coho salmon under variable stream flow conditions. Transactions of the American Fisheries Society 138:397-406.

Warren, M.L. and B.M. Burr. 1994. Status of freshwater fishes of the United States: overview of an imperiled fauna. Fisheries 19:6-18.

Budget

Bay Mills Indian Community		
Personnel		
Salary		
Fishery/Administration Biologist (0.05 FTE)	2,668	
Supervisory technician (1.0 FTE, $12.88/hr.)	27,228	
Fall Research technician (13 weeks, 40 hrs. per week, 10.50/hr.)	5,460	
Spring Intern (8 weeks, 40 hrs per week, $10.50/hr.)	3,360	
Total Salary		**38,716**
Fringe Benefits		
Fishery/Administration Biologist (30% of salary)	800	
Supervisory technician (30% of salary)	8,168	
Fall Research technician (12% of salary)	655	
Spring intern (12% of salary)	403.2	
Total Fringe		**10,027**
Supplies		
Field Gear (boots, raingear, waders, ect.)	520	
3 Fyke nets and blocker nets	2400	
Purchase of fish for analysis (50 fish @ $5.00 per fish)	250	
Total Supplies		**3,170**
Travel		
Travel (20 trips, 90 mile average @ $.57 per mile)	1026	
Boat operations and maintenance (10 sampling trips @ 60)		
Total Travel		**1,026**
BMIC Total Direct Costs		**52,939**
BMIC Indirect Costs		
20.5% of salary and fringe		**9,992**
Contract		
Lake Superior State University		**80,033**
Total Budget		**142,964**

Lake Superior State University Contract expense breakdown

SALARY	
AHM Summer Salary (1 month)	6000
JL Summer Salary (0.5 month)	2800
Fisheries Co-PISummer Salary (0.5 month)	2800
Student field technicians (1 full time for 34 wks, 1 full-time 12 wks @ $10/hr)	18400
Total Salary/Wages	**30000**
FRINGE BENEFITS	
AHM Summer Salary	1186
JLSummer Salary	553
Fisheries Co-PI Summer Salary	553
Two student field technicians	4352
Total Fringe Benefits	**6644**
Total Personnel Costs	**36644**
SUPPLIES	
Fish disease analysis - hormones and blood (300 samples @$10/sample)	3000
Fish disease analysis - ELISA (300 samples x 3 tests @$12/sample)	10800
Fish disease analysis - qPCR/VHS exposure (100 samples @ $25/sample)	2500
Nets (fyke nets, block nets, anchors)	2400
Field supplies & equipment (waders, fish totes, electronic scale)	1500
Equipment maintenance & repair	1000
Field expendables (tags, collection jars, ethanol, data sheets)	1500
Total Operating Costs	**22700**
TRAVEL	
Routine travel to field sites (150 60-mi round trips @ $0.57 per mile)	10260
Extra travel (sample collection and processing, meetings)	2000
Total Travel	**12260**
TOTAL DIRECT COSTS	**71604**
Indirect cost base (ARL 23% on Salary and Fringe)	0.23
INDIRECT COSTS	**8428**
TOTAL COSTS by source	**$80,033**

PROJECT TITLE: RE-EMERGENCE OF EPIZOOTIC EPITHELIOTROPIC DISEASE VIRUS: POTENTIAL EFFECTS AND DEVELOPMENT OF IMPROVED DIAGNOSTICS& CONTROL MEASURES

Applicant eligibility: yes

Applicant Information and History

Principal investigator (PI) contact information.
Dr. Mohamed Faisal, Department of Pathobiology and Diagnostic Investigation, College of Veterinary Medicine, Michigan State University, 1129 Farm Lane, Room 174, Food Safety & Toxicology Building, East Lansing, MI, 48824, (517) 884-2019, faisal@cvm.msu.edu, lochthom@cvm.msu.edu.

Co-investigator(s): Dr. Thomas Loch, Michigan State University, College of Veterinary Medicine, Department of Pathobiology and Diagnostic Investigation.
Dr. Gavin Glenney, United States Fish and Wildlife Service, Northeast Fishery Center
Dr. Jun Li, Lake Superior State University, School of Biological Sciences
Dr. Tom Waltzek, University of Florida, Department of Infectious Diseases and Pathology
Mr.John Coll, United States Fish and Wildlife Service, Northeast Fishery Center
Ms.Patricia Barbash, United States Fish and Wildlife Service, Northeast Fishery Center
Mr.Gary E. Whelan, Michigan Department of Natural Resources, Fisheries Division

Has the PI or co-PI applied to the GLFT before?
Dr. Faisal: yes; Dr. Loch: no; Dr. Glenney: no; Dr. Li: yes; Dr. Waltzek: no; Mr. Coll: no; Ms. Barbash: no; Mr. Whelan: yes (as CoPI).

Has the PI or co-PI previously received a grant award from the GLFT? (Y/N)
Dr. Faisal: yes; Dr. Loch: no; Dr. Glenney: no; Dr. Li: no; Dr. Waltzek: no; Mr. Coll: no; Ms. Barbash: no; Mr. Whelan: yes (as CoPI).

Prior GLFT funded projects summary (200 words)
Dr. Faisal has received multiple grants dealing with viruses and bacteria threatening Great Lakes fish stocks, as PI on #2012.1257, "Elucidating the role of herd immunity in protecting Lake Michigan fish against VHSV" (publications in preparation); CoPI on #2007.883 "VHSV in the Great Lakes," with publications including two PhD dissertations and 8 published papers; PI on #2010.1147, "Emerging *Flavobacterium* spp. in the Great Lakes basin: Identification and assessment of their impacts on fish health," with publications including a PhD dissertation, 2 submitted, 5 published papers; CoPI on #2004.581, "Ecological and genetic approaches to develop sustainable and disease free fisheries in the Great Lakes," with publications from Faisal's group including one published, one accepted, and two submitted manuscripts; and Co-PI on #2009.1058, "Mechanistic approach to identify the role of pathogens in causing *Diporeia* spp. decline in the Laurentian Great Lakes," with publications including a PhD dissertation, one published, 3 in press, and one accepted manuscript. Mr. Whelan was named on the Great Lakes Watershed Interpretive Center Project in which watershed interpretive centers were installed at all six Michigan state fish hatcheries that he manages.

Active research commitments.
PI Dr. Faisal is committed to the following research: "Development and Implementation of a Fish Health Initiative for Michigan Inland and Great Lakes Fisheries," Michigan-DNR, 0%; "Formulating a Vision for Fish Health Management in Fishery Conservation" U.S Fish

&Wildlife Service, 10%; "The Use of Sensitive Serological Assays for Determining the Distribution of VHSV in the Great Lakes," U.S. Fish and Wildlife Service, 5%; "Identifying risks of emerging viral infections in the Great Lakes, U.S. Fish and Wildlife Service, 5%; "Elucidating the role of herd immunity in protecting Lake Michigan fish against VHSV," Great Lakes Fishery Trust, 10%.

Grant Request

Requested amount: $349,812
Match amount: $96,680
Project start date: September 1, 2014
Project end date: August 31, 2016
Has your organization proposed this project to the GLFT in the past: No

Project Summary.The Epizootic Epitheliotopic Disease Virus (EEDV) is a serious infection that can impede the Lake Trout Rehabilitation Program in the Great Lakes. Since information on EEDV is vastly lacking, the proposed study aims to unravel important aspects of EEDV pathology such as the disease course, latency, and host immune responses. The newly generated information will lead to the development of improved diagnostic toolsand the development of novel control strategies.

Project Description

1. Problem Statement.
In the 1980's, Great Lakes fishery managers were confronted with the emergence of a deadly disease of lake trout (*Salvelinus namaycush*) in sevenstate and federal hatcheries in three states (lakes Michigan and Superior watersheds). These outbreaks resulted in the destruction of 15 million fish, and were of particular concern due to the severe depletion of hatchery lake trout populations. The causative agent wasidentified as the Epizootic Epitheliotropic Disease Virus (EEDV). Fearing from its spread, fishery managers adopted stringent disease control measures that included depopulation of infected stocks. EEDV outbreaks ceased. EEDV disappearance left scientists wondering about the nature of EEDV, where it came from, how it is transmitted, whether fish remain latently infected, and most importantly, if EEDV resurfaced, what would be the best control strategy? Unfortunately, none of these questions could be answered since the virus could not be grown on cell lines and little material remained in a few archived infected tissues.

In 2003, EEDV reappeared at the Les Voigt State Fish Hatchery (WI) with low mortalities. This allowed scientists to develop a PCR assaywhich was used in additional testing. EEDV was present in healthy wild and hatchery lake trout in the Great Lakes and Wyoming but no epizootics were seen. In 2012, an EEDVepizootic occurred at Marquette State Fish Hatchery (MSFH) in Michigankilling 20% of the production lake trout. Given the reemergence of this pathogen, it is critical to fully understand EEDVproperties and host interactions. Hence, we propose to study in detail EEDV biology, pathogenicity, and host immune responses. The recent epizootic at MSFH has provided a windfall of freshly collected tissues laden with EEDV that present a unique opportunity to unravel the virus properties that until now have hampered the development of control strategies against this deadly virus.

2. Project Goal. (**300 words**).
The goal of this proposal is to develop sufficient information on the ecology of EEDVto allow the development and implementation of effective diagnostic tests and control strategies.

This proposal spans two years with the following objectives:

Objective 1:To fully elucidate the biological and pathological properties of EEDV using genomic characterization.
From the heavily infected tissues collected during the recurrent mortality episodes in MSFH lake trout, EEDV will be purified in large quantities and used in a number of experiments.

Objective 2: To determine EEDV host range, disease course, and modes of transmission. The PIs' unique collection of infected tissues will allow experiments aiming at determining the susceptibility of selected salmonid and non-salmonid species and their potential role as EEDV reservoir.

Objective 3:To elucidate humoral immune mechanisms involved in defense against EEDV.

Objective 4:To develop specific and sensitive diagnostic assays for EEDV detection in fish including carrier and previously exposed fish.

Objective 5:To test the efficacy of current biosecurity practices in hatcheries for the inactivation of EEDV including egg disinfection.

3. Methods and/or Activities.
Objective 1:To fully elucidate the biological and pathological properties of EEDV using genomic characterization. Full genome sequencing using next generation sequencing will be performed. The virulence mechanisms will be deciphered using a bioinformatics approach. Potential targets for novel control strategies will be identified. The phylogeny of EEDV will be determined using multiple phylogenetic methodologies (e.g. Maximum Likelihoodand Bayesian).

Objective 2: To determine EEDV host range, disease course, and modes of transmission. Experimental exposure of lake trout and other fish species (e.g., *Oncorhynchus* spp., *Salmo* spp., coregonids, & mottled sculpins) will be used to investigate disease course, shedding, and likely modes of horizontal and vertical transmission. The role of stressors in initiation of EEDV epizootics and EEDV shedding will be ascertained.

Objective 3: To elucidate humoral immune mechanisms involved in defense against EEDV. Secretory and circulating lake trout immunoglobulins will be purified and used to develop antibodies. These antibodies will be used to develop ELISA assays to assess antibody levels against EEDV throughout both experimental exposure and in survivors of naturally occurring outbreaks.

Objective 4: To develop specific and sensitive diagnostic assays for EEDV detection in lake trout including carrier and previously exposed fish. To identify the most specific and sensitive molecular diagnostic assayto detect EEDV even in carrier fish. Monoclonal antibodies will be used to create diagnostic ELISA assays. Susceptible cell cultures to propagate EEDV and serological assays will be developed.

Objective 5: To test the efficacy of current biosecurity practices in hatcheries for the inactivation of EEDV including egg disinfection. The efficacy of currently employed hatchery disinfectants, egg disinfection procedures, and ultra-violet water sterilization will be

evaluated. The ability of the stress test to facilitate the detection of EEDV will also be assessed.

4. Geographic Focus Area.
Enter the lake basin(s) in which the proposed project would have management implications: Lake Michigan.
The deliverables from the proposed study will provide immediate benefits to the lake trout rehabilitation program in the entire Great Lakes basin. Great Lakes state and federal fish management agencies are implementing a landscape scale rehabilitation program to re-establish self-sustaining lake trout populations in Great Lakes waters using millions of hatchery fish annually that must be high quality, disease-free fish for the program to be effective. The information and tools generated in the course of this study will be indispensable for the re-habilitation efforts.

5. Potential Management Benefits and Outcomes of Proposed Project. (**200 words**) This project will provide critical information for the management of lake trout rehabilitation programs and hatcheries, as it unravelsEEDV ecology, pathogenesis and transmission. Bridging thecurrent knowledge gaps is required to develop effective control strategies. Specifically, the production of purified EEDV will revolutionize EEDV research that has been hampered by insufficient viral concentrations. Assuming the proposed efforts to develop a permanent cell line that supports EEDV replication is successful, copious quantities of pure virus could be obtained and used to develop vaccines. Moreover, findingsof the proposed study will provide managers with vital information on species at risk, reservoirs of infection, and efficacy of disinfection. Furthermore, the development of enhanced diagnostic reagents of a serological and molecular nature will allow, for the first time, the ability to follow the EEDV epidemiological cycle in lake trout populations with precision. Finally,the transmission routes of this deadly virus will be ascertained, a key piece of data that will broadly influence the management of very valuable and irreplaceable captive lake trout broodstock. The deliverables from the proposed study will ensure the viability of current and future lake trout rehabilitation programs.

6. Relationship of the Project to Ongoing Activities.
Currently, research and control approaches are suffering from the knowledge gap on the ecology and biology of EEDV that is in part the result of the lack of viable virus. Other than the work being done by the PI and two CO-PIs, no other research on EEDV is being conducted; therefore, it is certain that no duplication of efforts would occur. On the contrary, once the proposed research is concluded, further research can be performed as entirely new sources of this herpesvirus will be available to other researchers. Drs. Waltzek and Glenney are working on the phylogeny of alloherpesvirus including the emerging salmonid herpesviruses 4 and 5. This work compliments and enriches the proposed study as it allows for comparative phylogenetic analysis.

7. Prior Experience.
The study's team consistsof Dr. Faisal, an endowed professorwho has successfully managed grants in excess of $50 million over his academic career;Dr. Loch, Research Associate; Dr. Glenney, fish health biologist; Dr. Waltzek, Assistant Professor; Dr. Li,Associate Professor; Mr. Coll,Project Leader; Ms. Barbash, senior fish health biologist; and Mr. Whelan,the Fish Production Program Manager for the State of Michigan and one of the competent fish health authorities for the state.

Communications

8. **Target Audience.**
Our main target audiences are all fisheries management entities involved in lake trout rehabilitation in the Great Lakes includingfisheries managers, fish health specialists, and diagnostic laboratories. Project reports will be given at: the Lakes Michigan, Superior, and Huron Technical Committee meetings;the Council of Lake Committees; and the Fish Health Committee. Presentations will be given on study results at appropriate venues that will likely include AFS-Fish Health Section and Annual Meetings. Project deliverables will be published as both fact sheets and in scientific articles in appropriate journals. Finally, a webpage will be designed to facilitate dissemination of the data.

9. **Usefulness of Results:** This research is critical to achieving Great Lakes fish community objectives forself-sustaining native fish populations. The re-emergence of EEDV has created urgent challenges for fisheries managers. New diagnostic assays developed by this projectwill be critical in understanding the disease course, the effectiveness of disinfection techniques, and development ofpathogen control plans.Furthermore, determination of susceptibility and carrier status of species is critical towards understanding how this virus is spread. Optimization of diagnostic assays and development of pathogen control strategies are key priorities for the GLFC- Fish Health Committee and the GL Fish and Wildlife Restoration Act of 2006.

10. **Distribution of Findings**. (**100 words**)
Semiannual progress reports and one final report of results will be delivered to GLFT. We will present our findings to the Great Lakes Fish Health Committee along with other fisheries managers and fish health professionals to ensure it is fully circulated to other state, provincial, tribal and federal hatcheries. We will present results of this study at scientific conferences, such as the annual meetings of the American Fishery Society and the Eastern Fish Health Workshop. Ultimately, our results will be published in peer-reviewed journals such as *J. Fish Diseases*, *Diseases of Aquatic Organisms*, and *J. Wildlife Diseases*.

Project Budget

Budget Narrative (150 words)
Salaries and Fringe Benefits: To support this research, we request 50% salary ($51,426) and fringe ($11,296) for CoPI Dr. Loch; funds for salary ($38,577), tuition/fees ($23,658), and health insurance ($5,787) for a PhD level student; and wages ($16,064) and FICA ($1,229) for student research assistants.

Materials/Supplies: funds are requested ($52,500) to cover chemicals and laboratory supplies needed for completion of this project.

Other Direct Expenses: None requested.

Contracted Services: Co-PIs from several institutions will collaborate on this project. Dr. Waltzek of the University of Florida requests $30,000 for supplies/materials. Researchers from USFWS request $40,000 for supplies/materials. Dr. Li of LSSU requests $54,054 for three months/year of salary/wage/fringe for himself and for student assistants, and $10,000 for supplies/materials and travel.

Total Direct Costs: $334,591.

Indirect Costs: IDC ($15,221) are 10% of salaries/wages for MSU and collaborators.

Amount Requested from GLFT: $349,812.

Matching Funds:MSU's unrecovered IDC ($96,654) is match for the difference between the negotiated rate of 53.5% of TDC and the allowable 10% of salaries/wages only.

Total Project Cost: $446,492.

LAKE SUPERIOR STATE UNIVERSITY
my.LSSU

Jun Li <jli@lssu.edu>

Fwd: GLFT - EEDV award

19 messages

Mohamed Faisal <faisal@cvm.msu.edu> Tue, Sep 2, 2014 at 4:04 PM
To: Gavin_Glenney@fws.gov, john_coll@fws.gov, Patricia_Barbash@fws.gov, jli@lssu.edu, WHELANG@michigan.gov, twaltzek@ufl.edu

Hello all,
Thanks to your great knowledge, brainstorming, and support we got the GLFT award in full, Congratulations to all of us. I am honored to be collaborating with you.

That was the good news, the bad news is we have to do the work.

Apparently, the grant went to four reviewers, actually outstanding reviewers who took the time to make an excellent critique. We have to address these points before we actually get the money. I have attached the comments and I am now working on addressing these comments to the best we can. Then I will send it back to you asking for input.

Tom W, can you tell us more about the survival of herpesviruses after one cycle of freezing and thawing?
Many thanks
Mohamed

>>> healthyfish 09/02/14 8:25 AM >>>
Docs,

Today we received written notification of the award for the GLFT-EEDV project - congratulations! Before the award can be processed, we are required to address the peer review comments listed in the attached "Peer Review Summary." The attached Scope of Work needs to be modified (as necessary) to address each of these comments. I have also attached the full peer review responses for reference. We'll need to make any revisions to the Scope of Work in blue and I will send this to CGA and GLFT so that the account can be set up and the award can be disbursed. Then let the fun begin!

Michelle

3 attachments

Scope-of-Work.docx
117K

Peer Review Summary.pdf
71K

Peer Review Responses.pdf
1718K

Mohamed Faisal <faisal@cvm.msu.edu> Wed, Sep 3, 2014 at 2:46 AM

PEER REVIEW SUMMARY: PROJECT NO. 2014.1455

Re-emergence of Epizootic Epitheliotropic Disease Virus: Potential Effects & Development of Improved Diagnostics and Control Measures

Principal Investigator: Mohamed Faisal
Organization: Michigan State University
Grant Request: $346,450

PI's Project Description

"The Epizootic Epitheliotopic Disease Virus (EEDV) is a serious infection that can impede the Lake Trout Rehabilitation Program in the Great Lakes. Since information on EEDV is vastly lacking, the proposed study aims to unravel important aspects of EEDV pathology such as the disease course, latency, and host immune responses. The newly generated information will lead to the development of improved diagnostic tools and the development of novel control strategies."

Peer Review

Four reviewers submitted a peer review of this proposal. Comments submitted by peer reviewers are provided following the summary of the panel discussion.

Panel Discussion

The panel noted the PI introduced the problem well and included a strong team of partners to address the project objectives. The panel noted that the peer reviewers identified a number of matters that should be considered by the research team before moving forward with the project, namely:

- Clarify the role of subcontractors that will be included in the project.
- Since the source of the test fish is the Michigan hatchery system, there is a chance that the virus could have inadvertently been transmitted among the hatcheries. The panel recommended obtaining test fish from outside Michigan, where the virus is less likely to be present.
- The PI should consider adding a species of coregonid to the susceptibility trials because of the single detection in cisco from a hatchery in New York.
- Panel members expressed concern regarding the effectiveness of the injection method. The PI should develop a repeatable challenge method, perhaps through emersion, to achieve repeatable results.
- The PI should clarify what size fish will be infected and may need to increase the number of fish collected to generate a sufficient amount of tissue for analysis.
- The PI should consider including additional diagnostic models such as immune-histochemistry (IHC) and in situ hybridization (ISH) to help trace infected areas. These diagnostic models may be able to determine better treatment methods (e.g., Is the infection occurring within an egg or on the egg shell?).
- The PI should consider if there is a way to measure and replicate fish stress levels (such as cortisol levels) to determine how stress affects the disease response.
- The PI should clarify how he will better understand transmission of the disease. Is this specific to hatcheries or in the field?
- The panel noted that injection of brood stock has not been a proven method of infection for EED. Would it be better to test wild brood stock that is known to have some level of infection using a PCR method, then select those females for the studies?

This proposal was recommended for funding, contingent upon a satisfactory response to panel comments.

Effect of Dietary β-glucan Derived from Algae on Growth Performance, Disease Resistance and Immune Response in Atlantic Salmon Fry

Principal Investigator:

Jun Li, Ph.D.
Associate Professor for Fish Health
School of Biological Sciences
Lake Superior State University
Tel: 906-635 2094(office); 610-931 4979 (cell)
Email: jli@lssu.edu

Co-PI: Barbara I. Evans, Ph.D.
Professor of Biology
School of Biological Sciences
Lake Superior State University

Duration: Jan, 2015 –Aug, 2015

Overview: Dietary β-glucans,a nutritional supplement derived from the cell wall of variousplants, fungi, bacteria, mushrooms, yeasts and seaweeds,hasbeen well documented in thepromising enhancement of growth, vival and protection against infectious disease in various animals including fish and shellfish. In the present project, we will assess the growth and disease resistance of Atlantic salmon as well as the associated immunostimulating effects of dietary supplementation of algae derived β-glucan (**Algamune AM**) compared with yeast derived β-glucan(**Macrogard**). Atlantic salmon fry (6-8 inches) from the Lake Superior State University Aquatic Research Laboratory(LSSU-ARL) will be used as test subjects. All tests will be performed at 10-12°C in a controlled environmental chamber at LSSU. The project will last 6 months and assess changes in condition factor, immune system response and disease resistance to Furunculosis (caused by the bacterium *Aeromonassalmonicida*), as a result of incorporating algal derived β-glucan in the diet of the fish.

I: Acclimation:Day 0-14

Twelve 30-gallon tanks will be set up with filtration and aeration. Thirty fish will be added to each tank, and fed normally for 2 weeks (2 times/day, 3% body weight/fish). Water will be changed (half volume) every other day.

Test: At day 0 and day 14, length and weight of 10 randomly selected fish from each tankwill be measuredfor calculation of condition factor.

II: β-glucan feeding trials: Day 15-45

a) Control Tanks: 4 tanks will continue to receive normal food withoutβ-glucan

Yeast derivedβ-glucan(**Macrogard**): 2 tanks of fish will be fed pellets supplemented with yeast derivedβ-glucan (dosage based on Manufacture suggestion).

c) Algal derived β-glucan(**Algamune AM**): 6 tanks of fish will be fed pellets supplemented with yeast derivedβ-glucan. 2 tanks at high level (AM-1000; 1000 mg/kg);2 tanks at intermediate level(AM-500; 500mg/kg); 2 tanks at low level (AM-250; 250 mg/kg).

Test: At day 30 and 45, condition factor, immunology and hematology testing (n=5 fish removed from each tank at each sampling event). So in total, 10 fish will be removed from each tank by the end of this 30-day feeding trial.

III: *Aeromonas salmonicida* challenge(Days 46-75)

15 fish from each tank of the feeding trials will be placed in new tanks and then challenged with *A. salmonicida.*

a) Control Tanks: For the first 2 control tanks, 15 fish from each tank will be challenged by intraperitoneal injection of *A. salmonicida* at $1x10^5$cfu/fish (LD50)as positive controls. In the other 2 control tanks, 15 fish from each tank will be injected intraperitoneallywith PBS as the negative controls.

b) Yeast derived β-glucan (**Macrogard**): 2 tanks, 15fish will be challenged with *A. salmonicida* injected into peritoneal cavity as above.

c) Algal derived β-glucan(**AM-1000; AM-500 and AM-250**): (6 tanks, 2 tanks/dose of β-glucan)15 fish from each tank will be challenged with *A. salmonicida*injected into the peritoneal cavity as above.

Test: Mortality will be recorded each day for 30 days. At day 60 and 75, 3 fish from each tank will be removed for immunology and hematology testing.

Testing Parameters

Condition Factor: growth or weight gain/loss will be monitored by length weight ratio.

Hematology: total blood counts; RBC and WBC as well as hematocrits and hemoglobin will be measured..
Immunology: head kidney, spleen and liver will be weighed after dissection. Immune cells will be extracted from head kidney using mesh cell strainer and isolated through percoll gradient, then phagocytic activity and respiratory burst activity of leukocytes will be tested.

Serum complement activity, lysozyme activity and antimicrobial activity: Serum will be collected and keptat -80°C for further analysis (to be analyzed in Dec -Apr).Serum complement activity, lysozyme activity and antimicrobial activity,as well as serum glucose will be measured

Feed: Food base components and Macrogard will be purchased by LSSU and Algamune products (AM-1000, AM-500 and AM-250) will be supplied by Algal Scientific Corporation. Feed will be manufactured using a food mill to create pellets with and without beta-glucan.

Potential follow up study: given positive results of this study, we propose to investigate the resistance against viral diseases (VHS, or IHNV, IPNV) and the best developmental time of Atlantic salmon and/or other specie for algal beta-glucan supplementation in the future.

2014-2015 β-glucan Project Proposal for Algal Scientific

Budget

	Items	Cost
12	Tanks (30 gallon)	200
12	Filters	360
12	Aeration	200
1	Meat grinder	150
	Cell strainer and tests for phagocytic activity and respiratory burst activity.	300
	Miscsupplies (heparin, cell culture media, PBS, syringes, disposable plastic tubes and culture plates, chemicals/reagents from Sigma et al.)	800
	Dissection supplies	100
	Food base components	150
2	Student Salary (total 12 weeks @$10/hr + FICA)	5,160
	Indirect Cost: Student Salary (0.23)	1,186. 8
1	Faculty Salary (6 weeks @$5500/mo + fringe(0.19625) (Plus 6weeks time as free contribution)	9,869
	Indirect Cost: Faculty Salary (0.23)	2,269. 9
400	Atlantic salmon Fry (6-8 inches)	-
		$ 20,745. 7

Tank Plan

ık #	Acclimation (2 weeks)	Feeding Trial (4 weeks)	Challenge (4 weeks)
ı	No BG	No BG	No BG; no challenge
2	No BG	No BG	No BG; no challenge
3	No BG	No BG	No BG; challenge
4	No BG	No BG	No BG; challenge
5	No BG	Macrogard	Macrogard, challenge
6	No BG	Macrogard	Macrogard, challenge
7	No BG	AM 250 g/MT	AM 250 g/MT, challenge
8	No BG	AM 250 g/MT	AM 250 g/MT, challenge
9	No BG	AM 500 g/MT	AM 500 g/MT, challenge
10	No BG	AM 500 g/MT	AM 500 g/MT, challenge
11	No BG	AM 1000 g/MT	AM 1000 g/MT, challenge
12	No BG	AM 1000 g/MT	AM 1000g/MT, challenge

awarded 780,000 Chinese Yuan.

申请代码	C190604
受理部门	
收件日期	
受理编号	

解除保护

国家自然科学基金

申 请 书

（2014 版）

资助类别：面上项目

亚类说明：

附注说明：

项目名称：产生免疫保护性的灭活迟缓爱德华氏菌的作用机制

申 请 人：谢国驷　　电话：053285823062

依托单位：中国水产科学研究院黄海水产研究所

通讯地址：山东省青岛市南京路 106 号

邮政编码：266071　　单位电话：0532-85836340

电子邮箱：xgsi@hotmail.com

申报日期：2014年3月6日

国家自然科学基金委员会

基本信息

申请人信息	姓名	谢国驷	性别	男	出生年月	1976 年 12 月	民族	汉族
	学位	博士	职称	博士后		每年工作时间（月）		10
	电话	053285823062			电子邮箱	xgsi@hotmail.com		
	传真				国别或地区	中国		
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	主要研究领域	鱼类疫苗研究						
依托单位信息	名称	中国水产科学研究院黄海水产研究所						
	联系人	刘志鸿			电子邮箱	kyc@ysfri.ac.cn		
	电话	0532-85836340			网站地址	www.ysfri.ac.cn		

合作研究单位信息
单位名称
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项目基本信息				
	项目名称	产生免疫保护性的灭活迟缓爱德华氏菌的作用机制		
	英文名称	The immune mechanism to generate protective immunity for Edwardsiella tarda inactivated vaccine		
	资助类别	面上项目	亚类说明	
	附注说明			
	申请代码	C190604:水产生物疫苗学		
	基地类别			
	研究期限	2015 年 1 月 — 2018 年 12 月		
	申请经费	78.0000 万元		

中文关键词	迟缓爱德华氏菌;灭活疫苗;抗原特征;免疫机制;牙鲆
英文关键词	Edwardsiella tarda;Inactivated vaccine;Antigenic characteristic;Immune mechanism;Japanese flounder

中文摘要	**(限 400 字)：**迟缓爱德华氏菌（Edwardsiella tarda）是当前水产养殖业中的重要致病菌，对其实用性疫苗的开发具有重要意义，但采用常规技术制备的 E. tarda 灭活疫苗对鱼体不能产生有效的免疫保护。本项目拟以本实验室前期发明的几种可有效维持 E. tarda 抗原免疫保护力的灭活方法制备的灭活疫苗为基础，以牙鲆（Paralichthys olivaceus）为实验动物，通过灭活 E. tarda 的抗原特征、免疫保护性及其对鱼体的体液和细胞免疫激发效应的差异比较，分析灭活条件对 E. tarda 灭活疫苗得以保持或丢失其免疫保护性的原因，阐释可产生免疫保护性的 E. tarda 灭活疫苗的作用机制，为 E. tarda 灭活疫苗的研发提供基础依据。
英文摘要	**(限 3000 Characters):**Edwardsiella tarda has become one of the most important emerging pathogens in the current aquaculture, therefore, the design of an effective vaccine to prevent this pathogen is very important. But we find that the inactivated vaccine by the route methods can not produce the immune protection for fish. Based on our pre-invention which can effectively maintain E.tard antigen immune protection in the inactivated vaccine preparation, the new inactivated E.tarda vaccine prepared under different inactivation conditions were investigated using Japanese flounder (Paralichthys olivaceus) as a model. Comparing the results of the antigenic characteristics, the differences in immunogenicity, and its impact of humoral and cellular immune responses of the inactivated vaccine were evaluated. Based on the obtained results we try to explain the reasion to maintain and lose protective immunity for this vaccine. This work will help us to understand the immune protection mechanisms induced by inactivated E.tarda vaccine. This studies also provide a scientific basis for the developed an effective E.tarda inactivated vaccine in future.

项目组主要参与者（注：项目组主要参与者不包括项目申请人）

编号	姓 名	出生年月	性别	职 称	学 位	单位名称	电话	电子邮箱	项目分工	每年工作时间（月）
1	李军	1968-12-21	男	副教授	博士	美国 Lake Superior State University	1-906-635 2094	jli@lssu.edu	项目指导	3
2	史成银	1971-10-29	男	研究员	博士	中国水产科学研究院黄海水产研究所	05328582306 2	shicy@ysfri.ac.cn	抗原特征分析	3
3	王秀华	1969-9-25	男	研究员	硕士	中国水产科学研究院黄海水产研究所	05328582306 2	wangxh@ysfri.ac.cn	免疫机制分析	3
4	李杰	1982-10-20	男	博士后	博士	中国水产科学研究院黄海水产研究所	05328582306 2	blowapc@163.com	免疫因子应答分析	3
5	马璀艳	1982-8-18	女	博士后	博士	中国水产科学研究院黄海水产研究所	05328582306 2	mcy1982@163.com	鱼类细胞免疫机制	3
6	张晓静	1987-8-3	女	硕士生	学士	中国水产科学研究院黄海水产研究所	05328582306 2	374463656@qq.com	免疫实施及样品采集	8
7	王娜	1989-6-14	女	硕士生	学士	中国水产科学研究院黄海水产研究所	05328582306 2	924773671@qq.com	分子免疫指标测定	8
8	[在此录入修改]									
9	[在此录入修改]									

总人数	高级	中级	初级	博士后	博士生	硕士生
8	3	0	0	3	0	2

说明： 高级、中级、初级、博士后、博士生、硕士生人员数由申请人负责填报（含申请人），总人数由各分项自动加和产生。

Awarded 3,000,000 Chinese Yuan for 2014.1 – 2018.12)
(= 480,000 USD. 1 USD = 6.2 Chinese Yuan).

申请代码	C190602
受理部门	
收件日期	
受理编号	313300299

13000000020000028

Proposal for Funding From NSFC

国家自然科学基金

申 请 书

(2013版)

资助类别：重点项目

亚类说明：

附注说明：领域名称：水产动物重要病原流行病学与致病机理（C190602）

项目名称：迟缓爱德华氏菌关键免疫逃逸因子及其作用机制和应用潜能

申 请 人：孙黎　　电 话：0532-82898829

依托单位：中国科学院海洋研究所

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申报日期：2013年3月15日

国家自然科学基金委员会

基本信息

申请人信息	姓名	[redacted]	性别	女	出生年月	1965年12月	民族	汉族
	学位	博士	职称	研究员		每年工作时间（月）	6	
	电话	0532-82898829		电子邮箱		lsun@qdio.ac.cn		
	传真	0532-82898829		国别或地区		中国		
	个人通讯地址	山东省青岛市南海路七号						
	工作单位	中国科学院海洋研究所/海洋生物学重点实验室						
	主要研究领域	鱼类免疫学，鱼类细菌性病原，鱼类病毒，鱼类疫苗						
依托单位信息	名称	中国科学院海洋研究所						
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合作研究单位信息	单位名称							
项目基本信息	项目名称	迟缓爱德华氏菌关键免疫逃逸因子及其作用机制和应用潜能						
	英文名称	Immune evasion factors of Edwardsiella tarda: working mechanism and application in disease control						
	资助类别	重点项目		亚类说明				
	附注说明	领域名称：水产动物重要病原流行病学与致病机理（C190602）						
	申请代码	C190602						
	基地类别							
	研究期限	2014年01月 — 2018年12月						
	申请经费	370.0000万元						
中文关键词	迟缓爱德华氏菌；免疫逃逸；血清杀菌；细胞免疫；疫苗							
英文关键词	Edwardsiella tarda ; immune evasion ; serum bactericidal effect ; cellular immunity ; vaccine							

中文摘要

迟缓爱德华氏菌（Edwardsiella tarda）是一种鱼类致病菌，可以感染多种海水和淡水鱼类，给养殖业造成了巨大的经济损失。研究表明，迟缓爱德华氏菌是典型的胞内寄生菌，能够逃避宿主的各种免疫防御，在宿主血清和吞噬细胞内存活并繁殖。因此，免疫逃逸被认为是迟缓爱德华氏菌至关重要的致病机制，也是该菌相关疾病免疫防控的一个重大障碍。然而，尽管迟缓爱德华氏菌免疫逃逸现象在科学界是一种共识，但是其中蕴含的分子机制却完全不清楚。基于此，本申请项目拟系统筛选迟缓爱德华氏菌的抗血清杀伤因子和抗免疫细胞杀伤因子，对关键因子进行功能解析，阐明这些因子的作用方式和表达调控，从而揭示迟缓爱德华氏菌的免疫逃逸机制。同时，在理论研究的基础上，项目还将探索免疫逃逸因子在迟缓爱德华氏菌病防御中的应用潜能。本项目研究结果将不但促进我们对鱼类胞内寄生菌致病机理的深入了解，而且也为相关疫病的免疫防控提供靶点和理论指导。

英文摘要

Edwardsiella tarda is an important fish pathogen and can infect many species of marine and freshwater fish, thus causing heavy economic losses to aquaculture industries. Studies have indicated that E. tarda is a typical intracellular pathogen that evades the immune defense of the host and survives in host serum and phagocytes. Hence, immune evasion is an important virulence mechanism of E. tarda and a barrier to prevention of edwardsiellosis. However, the molecular basis of this phenomenon is unknown. Based on these observations, we in this study plan to identify E. tarda factors that are involved in anti-serum and anti-phagocyte immunity and, through analyzing the function, expression, and regulation patterns of these factors, explore the mechanism of immune evasion. In addition, we also plan to investigate the applicability of immune evasion-related factors in the control of edwardsiellosis. Taken together, our study will add insights to the infection mechanism of E. tarda and provide targets and theoretical guidance for disease control.

项目组主要参与者（注：项目组主要参与者不包括项目申请人）

编号	姓名	出生年月	性别	职 称	学 位	单位名称	电话	电子邮箱	项目分工	每年工作时间（月）
1	张敏	1980-02-06	女	副研究员	博士	中国科学院海洋研究所	13791987318	zhangmin@qdio.ac.cn	抗血清杀菌因子筛选与功能研究	6
2	胡永华	1978-04-23	男	副研究员	博士	中国科学院海洋研究所	13780680059	huyonghua@ms.qdio.ac.cn	抗免疫细胞杀伤因子筛选与作用	6
3	孙铂光	1980-03-29	男	助理研究员	博士	中国科学院海洋研究所	13853270995	sunboguang@qdio.ac.cn	细菌胞内感染	10
4	迟恒	1980-09-02	男	助理研究员	博士	中国科学院海洋研究所	13969862133	chiheng@qdio.ac.cn	抗细胞免疫因子功能作用	10
5	李军	1968-12-21	男	助教	博士	Lake Superior State University，中国科学院海洋研究所	13341231999	jli@lssu.edu	抗血清杀菌因子功能作用	3
6	周志侠	1982-02-02	女	博士后	博士	中国科学院海洋研究所	15275230797	zhou_zhixia@126.com	鱼类抗感染免疫	10
7	李墨非	1986-11-15	男	博士生	硕士	中国科学院海洋研究所	15969820960	murphy210@163.com	抗血清杀菌因子功能分析	10
8	龙昊	1980-07-16	男	博士生	硕士	中国科学院海洋研究所	18661882764	longhao01@foxmail.com	免疫逃逸因子的应用研究	10
9	张宝存	1985-11-12	男	博士生	硕士	中国科学院海洋研究所	13685424022	virusouc@hotmail.com	免疫逃逸因子的应用研究	10

总人数	高级	中级	初级	博士后	博士生	硕士生
10	3	3		1	3	0

Granted 500,000 Chinese Yuan. for 2012 - 2015.
(about 80,000 USD).

申请代码	C190601
受理部门	
收件日期	
受理编号	

解除保护

Proposal for funding from Chinese National Natural Foundation (NSFC).

国家自然科学基金

申 请 书

（2011 版）

资助类别：面上项目

亚类说明：

附注说明：

项目名称：Immunological Mechanism of Marine Fish by Immersing with Saponins as an immune adjuvant.
皂角苷增强海水鱼类细菌疫苗浸泡免疫效果的作用机理

申 请 人：王秀华 电话：0532-85823062

依托单位：中国水产科学研究院黄海水产研究所

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申报日期：2011年3月10日

国家自然科学基金委员会

基本信息

申请人信息	姓　　名	王秀华	性别	男	出生年月	1969 年 9 月	民 族	汉族
	学　　位	硕士	职称	副研究员		每年工作时间（月）	7	
	电　　话	0532-85823062		电子邮箱		wangxh@ysfri.ac.cn		
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	工 作 单 位	中国水产科学研究院黄海水产研究所						
	主要研究领域	海水养殖动物疾病控制与免疫防治						
依托单位信息	名　　称	中国水产科学研究院黄海水产研究所						
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合作研究单位信息	单 位 名 称
	[在此录入修改]
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项目基本信息				
	项目名称	皂角苷增强海水鱼类细菌疫苗浸泡免疫效果的作用机理		
	资助类别	面上项目	亚 类 说 明	
	附注说明			
	申请代码	C190601:水产免疫生物学		
	基地类别			
	研究期限	2012 年 1 月 — 2015 年 12 月	研究属性	应用基础研究
	申请经费	50.0000 万元		

摘　要

(限 400 字)：

鱼类疫苗浸泡免疫为简捷的接种方式，但免疫保护率低阻碍浸泡疫苗的产业化进程。前期研究结果表明，皂角苷可显著提高大菱鲆细菌浸泡疫苗免疫保护率，具有佐剂活性，且在使用的有效浓度范围内对鱼体不表现毒性。为探讨皂角苷的佐剂效能机制，利用免疫组化、免疫电镜、ELISPOT 免疫分析技术，研究皂角苷促进大菱鲆体表黏膜组织对抗原吸收的效果及对黏膜 B 细胞与抗体分泌细胞数量增殖的作用机制；利用 RT-PCR 技术，研究黏膜中免疫细胞内 HMC II α、IL-1 β、TNF-α、TGF-β 免疫调控因子的基因表达水平；采用免疫印迹法分析皂角苷对疫苗抗原活性的影响；应用激光共聚焦技术，分析皂角苷对血细胞及黏膜免疫细胞的损害程度，评价皂角苷对大菱鲆的毒性。阐明皂角苷提高浸泡疫苗免疫效果的机制，为高效鱼类浸泡疫苗的研制提供技术支撑。

关 键 词(用分号分开，最多 5 个)	皂角苷，海水鱼类，浸泡疫苗，佐剂机理

项目组主要参与者（注：项目组主要参与者不包括项目申请人）

编号	姓 名	出生年月	性别	职 称	学 位	单位名称	电话	电子邮箱	项目分工	每年工作时间（月）
1	李军	1968-12-21	男	助教	博士	Lake Superior State Univ. 美国苏必略湖州立大学	001906-6352094	jli@lssu.edu	细胞标记与细胞分型	1
2	史成银	1970-10-29	男	研究员	博士	中国水产科学研究院黄海水产研究所	0532-85823062	shicy@ysfri.ac.cn	疫苗制备与效果评价	6
3	梁艳	1976-2-24	女	助理研究员	博士	中国水产科学研究院黄海水产研究所	0532-85823062	liangyan@ysfri.ac.cn	体表黏液蛋白分析	7
4	边慧慧	1983-6-6	男	博士生	硕士	中国水产科学研究院黄海水产研究所	0532-85823062	bhui-83@163.com	抗体制备与抗原检测	7
5	王玉娟	1986-11-13	女	硕士生	学士	中国水产科学研究院黄海水产研究所	0532-85823062	yj729506@126.com	黏膜抗原抗体定量	8
6	朱岩松	1987-9-10	男	硕士生	学士	中国水产科学研究院黄海水产研究所	0532-85823062	zhuyansong3@126.com	佐剂安全评价	8
7	[在此录入修改]									
8	[在此录入修改]									
9	[在此录入修改]									

总人数	高级	中级	初级	博士后	博士生	硕士生
7	2	2	0	0	1	2

说明： 高级、中级、初级、博士后、博士生、硕士生人员数由申请人负责填报（含申请人），总人数由各分项自动加和产生。

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EDUCATION

1998-2002 Ph.D. Fish Pathobiology
Department of Biology, The Chinese University of Hong Kong.

Dissertation Title: *Vibrio alginolyticus: pathogenicity and its immunological control via vaccination in silver sea bream (Sparus sarba)*

1992-1995 M.S. Genetics (Molecular Virology concentration)
Institute of Hydrobiology, Chinese Academy of Sciences, China.

Thesis Title: *Development of a rapid diagnosis technique for hemorrhagic virus of grass carp (GCHV) based on reverse transcription-polymerase chain reaction (RT-PCR).*

1988-1992 B.S. Biology (Microbiology and Immunology Concentration)
Department of Microbiology, Wuhan University, China.

Thesis Title: *Isolation and identification of photosynthetic bacteria from Donghu Lake.*

PROFESSIONAL EXPERIENCE

5/2014- present **Associate Professor (Fish Health)** (Tenured)
Director of Fish Diseases Laboratory
School of Biological Sciences; Lake Superior State University
Sault Ste. Marie, MI, 49783

11/2009- 4/2014 **Assistant Professor (Fish Health)** *(Granted Tenure in 11/2013)*
Director of Fish Diseases Laboratory
School of Biological Sciences; Lake Superior State University
Sault Ste. Marie, MI, 49783

4/2002-10/2009 **Research Associate (Fish Immunology)**
Department of Pathobiology, School of Veterinary Medicine
University of Pennsylvania, Philadelphia, PA 19014

1/2001- 2/2002 **Postdoctoral Researcher (Cell Biology)**
Lab Biologie Cellulaire, Faculte de Medecine
Universite Paul Sabatier, Toulouse, France

7/1995-1/1998 **Assistant professor (Marine Microbiology)**
Department of Marine Biology,

Ocean University of China, China.
and

Faculty member
Biotechnology Education and Training Center (BETCEN),
(UNESCO Chinese Center of Marine Biotechnology)
UNESCO

TEACHING

11/2009-present, Teach the following courses for undergraduate students in the School of Biological Sciences at the Lake Superior State University.
Biol. 499: Senior Research Seminars
Biol. 495: Senior Project
Biol. 434: Histopathology (lab)
Biol. 433: Histology (lab)
Biol. 426: Ecology of Infectious Diseases (lecture)
Biol. 425: Virology (lecture & lab)
Biol. 423: Immunology (lecture & lab)
Biol. 400: Microbial Ecology
Biol. 372: Freshwater Fish Culture (lecture & lab)
Biol. 389: Fish Aquaculture Internship
Biol. 389: Fish Health Internship
Biol. 389: Marine Biology Internship
Biol. 240: Natural History of Vertebrate
Biol. 204: Microbiology (lecture & lab)
Biol. 131: General Biology-Cell (Lab)

4/2002- 10/2009, **Fish Anatomy Lab** for VMD candidates at School of Veterinary Medicine, University of Pennsylvania, USA.

1/1998-12/2000, **General Microbiology Lab** and **Cell Biology Lab** for undergraduates at The Chinese University of Hong Kong, Hong Kong.

9/1995-12/1997 **General Microbiology** and **Marine Microbiology** (lecture & lab) for undergraduates at Ocean University of China, Qingdao, China .

HONORS

2014-2017, Chair of the Consulting Committee for Aquatic Animal Health/Aquatic Animal Medicine Programs, Shanghai Ocean University, Shanghai, China.

2014-2018, **Adjunct Professor** in College of Fisheries, Huazhong Agriculture University, Wuhan, China.

2013 Nominee for the Distinguished Teacher Award at Lake Superior State University.

2011 Nominee for the membership to the North Central Regional Aquaculture Center's (NCRAC) Technical Committee/Research Subcommittee.

2011-present **Guest professor**, School of Biotechnology, East China University of Science and Technology, Shanghai, China.

2011- present Selected as a member of External Peer-reviewer Panel for National Natural Science Foundation of China (NSFC).

2010-present **Visiting professor**, Institute of Oceanology, The Chinese Academy of Sciences, Qingdao, China

2009- 2013 Dr. Li was awarded a **"Taishan Scholarship"** (1,000, 000Chinese Yuan) for recognized outstanding oversea scholar from the Government of Shandong Province, China for supporting the collaborative research in the area of **"Infectious Diseases and Immunological Control in Marine Fish"**).

2001 National Innovation Award in Marine Sciences and Technology, State Oceanic Administration of China. (Second Prize)

2001 National Award for Advancement of Science and Technology, National Commission of Science and Technology of China. (Second Prize)

2000 International Outstanding Young Scientists Award (250,000 Franc), French Ministry of Research (I was one of 50 awardees worldwide of this year)

GRANTS & CONTRATS

2009- present Contracts with the Michigan Department of Natural Resources and Environment (MI-DNR): BKD and EMS diagnosis in Fish Gametes in MI State Fish Hatcheries. $5,000-7,500/year (PI)

2015 Bureau of Indian Affairs-Great Lakes Restoration Initiative Funds (BIA-GLRIF). Monitoring fish movement and fish condition in tributaries of Whitefish Bay. Co-PI ($ 80,033 for LSSU).

2014-2015 Contract from *Algal Scientific Company*, Effect of Dietary beta-glucan Derived from Algae on Growth Performance, Disease Resistance and Immune Response in Atlantic salmon. $20,746. (PI).

2014-2016 Great Lakes Fishery Trust. Re-Emergence of Epizootic Epitheliotropic Disease Virus: Potential Effects and Development of Improved Diagnostics &Control Measures. $446,492. Co-PI. (PI, Dr. Mohamed Faisal from Michigan State University, East Lansing, MI). ($64,054 for LSSU)

2014 The Great Lakes Council of the Federation of Fly fishers Research Grant, $400 (PI).

2014 LSSU Undergraduate Student Research Grant $500 (Awarded to support Dr Li's student Tyler Jackson for his senior research project).

2014-2019 Key Project from Chinese Natural Science Fund for the study Immuno-escape Mechanisms of *Edwarsiella tarda* in Turbot. Chinese Yuan (RMB)

3,000,000.00, Co-PI. (PI, Dr. Li Sun, from Institute of Oceanology, Chinese Academy of Sciences, Qingdao, China)

2011-2014 Chinese Natural Science Fund for Application of Immuno-stimulants against Fish Diseases. RMB 500,000. Co-PI, (PI, Dr. Xinhua Wang, Yollow Sea Fishery Research Institute, Qingdao, China).

2012 Superior Aqua-System Development for Michigan Clean Energy Venture Challenge. $39,000 (Co-PI, PI, Dr. Evans School of Biological Sciences, LSSU).

2012 Evaluation of the Relationship of Total Suspend Solids, Bacteria and Fish Health Status in Marquette Fish Hatchery. $ 8990 (PI)

2011 The Great Lakes Council of the Federation of Fly fishers Research Grant, $400 (To support Dr Li's student for his collecting water samples from Marquette State Fish Hatcheries).

2011 LSSU Strategic Planning Initiative funding, $19,500. (PI)

2010 LSSU Undergraduate Student Research Grant $500 (Awarded to Dr Li's student Heather R. Millard for supporting her senior research project).

2010 The Great Lakes Council of the Federation of Fly fishers Research Grant, $300 (To support Dr Li's student for her summer research in Platter River State Fish Hatcheries).

2009- present Contracts with the Michigan Department of Natural Resources and Environment (MI-DNR): BKD and EMS diagnosis in Fish Gametes in MI State Fish Hatcheries. $3,500-5,000/year (PI)

2009-2013 **"Taishan Scholarship"** from Shandong Province, China to support Dr. Li's International Collaboration Research on Fish Infectious Diseases and Immunological Control. RMB 1,000,000 (PI).

THESIS COMMITTEES

11/2009- present: Mentor for 15 senior students' projects.

12/2010- present: Co-supervisor for Ph.D. candidates (Fish Diseases and Immunology Program) in Institute of Oceanology, The Chinese Academy of Science, Qingdao, China

9/1996-7/1999: Co-supervisor for Hui Xiao, M. Sc. Candidate at Marine Biology Program of Ocean University of China, China. And also provide mentoring for over 10 undergraduates and master graduates on their research projects.

MEMBERSHIPS

2006- present: International Society of Developmental and Comparative Immunology

2007- present: American Association of Immunologists.

2009-present: American Fisheries Society-Mi Chapter

2013-present: International Society of Fish and Shellfish Immunology

EDITORIAL REFEREE

Invited to peer-review manuscripts for the following scientific journals:

Aquaculture
Central European Journal of Biology
Developmental and Comparative Immunology
Fish & Shellfish Immunology
Hydrobiologica
General and Comparative Endocrinology
Journal of Aquatic Animal Health
Journal of Immunology
Journal of Ocean University of China
Journal of Virological Methods
Journal of Visualized Experiments
Marine Genomics
Molecular Biology Reports
North America Fish Aquaculture
Oceanologica et Liminologica Sinica
PNAS
Veterinary Microbiology
Virology Journal

PUBLICATIONS

1. Wu N., LaPatra S.C., **Li J.**, Sunyer J. O. and Zhang Y.A. 2014. Complement C5a acts as molecular adjuvant in fish by enhancing antibody response to soluble antigen. ***Fish & Shellfish Immunology.*** 40(2) 616-623.

2. Li M.F., Zhang B.C., **Li J.**, and Sun L. 2014. Sil: A Streptococcus iniae Bacteriocin with dual role as an antimicrobial and an immunomodulator that inhibits innate immune response and promotes S. iniae infection. ***Plos One.*** 9(4) e96222.

3. Wang C, Hu Y, Sun B, **Li J**, Sun L. 2013. *Edwardsiella tarda* Ivy: a lysozyme inhibitor that blocks the lytic effect of lysozyme and facilitates host infection in a manner that depends on the conserved cysteine residue. ***Infection & Immunity.*** 81:3527-3533.

4. Yu L, Sun B, **Li J**, Sun L. 2013. Characterization of a c-type lysozyme of

Scophthalmus maximus: expression, activity, and antibacterial effect. ***Fish & Shellfish Immunology***. 34: 46-54.

5. Li M, Chen C, **Li J**, Sun L. 2013. The C-reactive protein of tongue sole *Cynoglossus semilaevis* is an acute phase protein that interacts with bacterial pathogens and stimulates the antibacterial activity of peripheral blood leukocytes. ***Fish & Shellfish Immunology***. 34: 623-631.

6. Qiu R, Sun B, **Li J**, Liu X, Sun L. 2013. Identification and characterization of a cell surface scavenger receptor cysteine-rich protein of *Sciaenops ocellatus*: bacterial interaction and its dependence on the conserved structural features of the SRCR domain. ***Fish & Shellfish Immunology***. 34:810-818.

7. Wang C, Hu Y, Sun B, Chi H, **Li J**, Sun L. 2013. Environmental isolates P1SW and V3SW as a bivalent vaccine induce effective cross-protection against *Edwardsiella tarda* and *Vibrio anguillarum. Diseases of Aquatic Organisms.* 103:45-53.

8. Wang Y., Wang X.H., Huang J. and **Li J**. 2013. Adjuvant effects of *Quillaja saponaria* saponins (QSS) on humoral immune responses in turbot. ***Fish & Shellfish Immunology***. 34: 1745.

9. Chen L., Wang C., Sun L. and **Li J.** 2013. Survival of *Edwarsiella tarda* in fish serum relates to bacteria surface LPS. ***Fish & Shellfish Immunology***. 34: 1646.

10. David Parra, Rieger A*, **Li J***, Zhang YA, Randall L, Hunter C, Barreda D and Sunyer JO. 2012. Peritoneal cavity B-1 B cells have phagocytic and microbicidal capacities, and present phagocytosed antigen to CD4+ T cells, ***Journal of Leukocyte Biology***, 91(4) :525-536. (* equal contribution).

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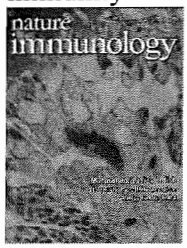
11. Zhang YA, Salinas I., **Li J**., Parra D., Bjork S., LaPatra S.E., Bartholomew J., and Sunyer J.O. 2010. IgT, a primitive immunoglobulin class specialized in mucosal immunity. ***Nature Immunology***, 11:827-835.



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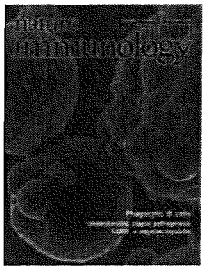
See also: News and Views by Flajnik in the same issue of ***Nature***

12. **Li J**, Barreda D R, Zhang Y A, Boshra H, Gelman A E, LaPatra S, Tort L and Sunyer J O. 2006. B lymphocytes from early vertebrates display potent phagocytic and micribicidal abilities. ***Nature Immunology***, 7, 1116-1124.



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See also "Research Highlights" in ***Nature Medicine***, 2006, Vol:12,

13. Zhang YA, Hikima J., **Li J.** Lapatra S.E., Luo Y.P., Sunyer J.O. 2009. Conservation of Structural and Functional Features in a Primordial CD80/86 Molecule from Rainbow Trout (*Oncorhynchus mykiss*), a Primitive Teleost Fish. ***Journal of Immunology,*** 183:83-96.

14. Sunyer J.O., Zhang Y.A., Li J., Parra D. and LaPatra S. 2009. Is IgT the evolutionary equivalent of IgA? Insights into its structure and function. ***Journal of Immunology***. 182. 81.21.

15. **Li J.**, Zhang Y.A. and J.O. Sunyer. 2007. Identification of a phagocytic complement C3d receptor in rainbow trout. ***Journal of Immunology***, 178: 53.15.

16. **Li J**, Barreda DR, Zhang YA, Boshra H, Gelman A.E, LaPatra S, Tort L and J.O. Sunyer. Complement and B cell cooperation in teleost fish: Role in phagocytosis and inflammation. *Molecular Immunology*, 44: 131

17. Boshra H*, Wang T*, Hove-Madsen L*, Hansen J*, **Li J***, Matlapudi A, Secombes C, Tort L and Sunyer JO. 2005. Cloning and characterization of a C3a receptor in rainbow trout and *Xenopus*: The first identification of C3a receptors in nonmammalian species. *Journal of Immunology*, 175, 2427-2437. (*equal contribution).

18. Boshra H*, Peters R*, **Li J*** and Sunyer JO. 2004. Production of recombinant C5a from rainbow trout (*Oncorhynchus mykiss*): role in leucocyte chemotaxis and respiratory burst. *Fish & Shellfish Immunology*, 17, 293-303. (*equal contribution).

19. **Li J**, Peters R, Lapatra S, Vazzana M, Sunyer JO. 2004. Anaphylatoxin-like molecules generated during complement activation induce a dramatic enhancement of particle uptake in rainbow trout phagocytes. *Developmental and Comparative Immunology*, 28,1005-1021.

20. Boshra H*, **Li J***, Peters R*, Hansen J, Matlapudi A, Sunyer JO. 2004. Cloning, expression, cellular distribution, and role in chemotaxis of a C5a receptor in rainbow trout: The first identification of a C5a receptor in a nonmammalian species. *Journal of Immunology*, 172, 4381-4390. (*equal contribution)

21. Deane EE, **Li J,** and Woo NYS. 2004. Modulated heat shock protein expression during pathogenic *Vibrio alginolyticus* stress of sea bream. *Diseases of Aquatic Organisms*, 62, 205-215.

22. **Li J,** Zhou LR and Woo NYS. 2003. Invasion route and pathogenic mechanisms of *Vibrio alginolyticus* to silver sea bream (*Sparus sarba*). *Journal of Aquatic Animal Health*, 15, 302-313.

23. Xiao H, **Li J**, Wang XH, Li Y, Ji WS and Xu HS. 2003. Preparation of vaccine against vibriosis of sea perch (*Lateolabrax japonicus*) and evaluation of its protective efficiency. *Journal of Ocean University of China.* 2:37-41.

24. Fulladosa E, Delmas F, **Li J,** Villaescusa I, and Murat JC. 2002. Cellular stress induced in cultured human cells by exposure to sludge extracts from water treatment plants. *Ecotoxicology and Environmental Safety*, 53, 134-140.

25. Deane EE, **Li J** and Woo NYS. 2001. Hormonal status and phagocytic activity in sea bream infected with vibriosis. *Comparative Biochemistry and Physiology*, 129 B, 687-693.

26. Xiao H, **Li J**, Wang XH, Ji WS, Xu HS. 1999. Studies on pathogens of rotted gill and rotted caudal fins of sea perch (*Lateolabrax japonicus*). *Journal of Ocean University of Qingdao*, 29 (1): 87 - 93.

27. Xu HS, **Li J,** Brayton P, Woo NYS, Swartz D, Zhang ST and Rita R. Colwell. 1999. Occurrence and distribution of halophytic vibrios in subtropical coastal water and seafood of Hong Kong. *Acta Oceanographica Sinica.* 17: 545 - 553.

28. **Li J**, Yie J, Foo RWT, Ling JML, Xu HS, and Woo NYS. 1999. Antibiotic resistance and plasmid profiles of vibrio isolates from cultured silver sea bream, *Sparus sarba. Marine Pollution Bulletin*, 39, 245 -249.

29. Vandenberghe J, Li Y, Verdonek L, **Li J**, Xu HS and Swings J. 1999. Vibrios associated with *Penaeus chinensis* larvae in Chinese shrimp hatcheries. *Aquaculture,* 169, 121- 132.

30. **Li J**, Wang TH, Zhou LR and Xu HS. 1998. Comparison studies of two strains of grass carp haemorrhagic virus, GCHV-861 and GCHV-873. *Journal of Fishery Sciences of China,* 5: 115-118.

31. **Li J,** Feng J, Liu X, Li Q F, Woo NYS and Xu HS 1998. *Vibrio alginolyticus*, a pathogen of silver sea bream, *Sparus sarba*, cultured in Hong Kong. *Journal of Fisheries of China*, 22: 275-278

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33. **Li J**, Wang TH, Yi YL, Liu HQ, Lu RH, and Chen HX. 1997. A detection method of haemorrhagic virus of grass carp (GCHV) based on reverse transcription-polymerase chain reaction (RT-PCR). *Diseases of Aquatic Organisms*, 29, 7-12.

INVITED REVIEWS

1. Boshra H*, **Li J*** and Sunyer JO*. 2006. Recent advances on the complement system of teleost fish. *Fish & Shellfish Immunology*, 20, 239-262. (*equal contribution).
2. Sunyer JO, Boshra H and **Li J**. 2005. Evolution of anaphylatoxins, their diversity and novel roles in innate immunity: Insights from the study of fish complement. *Veterinary Immunology and Immunopathology*, 108, 77-89.
3. **Li J** and Woo N YS. 2003. Pathogenicity of vibriosis in fish: an overview. *Journal of Ocean University of Qingdao*, 2, 117-128.
4. **Li J**, Wang T, Lu RH, Chen HX. 1999. Advances of the grass carp heamorrhagic virus research in China (A review). *Oceanologia et Limnologia Sinica*, 30:, 445 - 453.
5. Wang XH, **Li J,** Ji WS and Xu HS. 1998. Application of probiotics to aquaculture. *Transactions of Oceanology and Limnology*. 1, 33~39.
6. **Li J,** Wang TH, Lu RH and Xu HS. 1996. Techniques for assaying fish viruses, *Transactions of Oceanology and Limnology*. 2, 58~65.

PRESENTATIONS (*LSSU undergraduate, 2010-present)

Regional/National Conference or Workshop:

1. **Li J.** 2010. Thiamine Deficiencies Workshop, Ann Arbor, MI.
2. **Li J.** 2011. Annual Meeting of the Michigan Chapter of the American Fisheries Society. Petoskey, Michigan.
3. Williams R.* **Li J.**, 2011. Annual Meeting of the Michigan Chapter of the American Fisheries Society. Petoskey, Michigan.
4. Millard H.R.*, **Li J.**, 2011. Annual Meeting of the Michigan Chapter of the American Fisheries Society. Petoskey, Michigan.
5. **Li J.**, Williams R*. 2012. The joint Annual Meeting of the Wisconsin Chapter and Michigan Chapter of the American Fisheries Society. Marinette, WI.
6. Evans B., **Li J**. 2012. Immune Cells in the Brian of Larval Lake Sturgeon (*Acipenser fulvescens*). 2012 Great Lakes Lake Sturgeon Coordination Meeting. Sault Ste. Marie, MI.
7. **Li J.** 2014. Joint Annual Meeting for MI-OH Aquaculture Association. Toledo, OH.
8. **Li J.** 2014. Immune Evasion Mechanisms of *Edeardsiella tarda* and Application in Disease Control. The 39th Eastern Fish Health Workshop. April 28-May 2, 2014. Shepherdstown, WV.

International Conference or Workshop:

9. David Parra, **J. Li,** Aja Rieger, Yong-An Zhang, Louise Randall, Christopher Hunter, Daniel Barreda, J. Oriol Sunyer. 2012. Peritoneal cavity B-1 B cells have phagocytic and microbicidal capacities, and present phagocytosed antigen to CD4+ T cells. *The 12th Congress of International Society of Developmental and Comparative Immunology,* July 9th-13th, 2012, Fukuoka, Japan.

10. Ling Chen, Chong Wang, Li Sun and **Li J**. 2012. Survivability of Edwardsiella tarda in fish serum relates to bacterial surface LPS. *The 12th Congress of International Society of Developmental and Comparative Immunology,* July 9th-13th, 2012, Fukuoka, Japan.

11. Chen L., C. Wang, L. Sun and **Li J**. 2013. Survival of *Edwardsiella tarda* in fish serum relates to bacterial surface LPS. *The 1st International Conference of Fish and Shellfish Immunology.* June 25-28, 2013. Vigo, Spain.

12. Wang Y., Wang X.H., Huang J. and **Li J.** 2013. Adjuvant effects of *Quillaja saponaria* saponins (QSS) on humoral immune responses in turbot. *The 1st International Conference of Fish and Shellfish Immunology.* June 25-28, 2013. Vigo, Spain.

13. **Li J.** 2013. The Involvement of Fish Complement in Innate and Adaptive Immune Responses. *2013 Fish Immunology Workshop.* July 18-20, 2013. Wuhan, China.

14. **Li J.** 2014. Infection and Immunity: Involvement of Innate and Adaptive Immune Responses in Fish Defense. In: *The Symposium on Infection and Immunity in Marine Organisms. July 25-26, 2014. Guangzhou, China(Keynote Speaker)*

INVITED SEMINARS (2010-present)

2010:

1. **Jun Li**. 2010, Phagocytosis and phagocytic B cells in teolest fish. Ocean University of China, Qingdao, China
2. **Jun Li**, 2010. Evolution of phagocytic B cells. Yellowsea Fishery Research Institute, Chinese Fishery Academy of Sciences, Qingdao, China.
3. **Jun Li**, 2010. A New Discovery of Phagocytic B cells in Teolest Fish. The Hong Kong University of Sciences and Technology, Hong Kong, China.
4. **Jun Li**, 2010. Evolution of phagocytic B cells. School of Fishery and Life Sciences, Shanghai Ocean Univeristy, Shanghai, China.

2011:

5. **Jun Li**, 2011. Evolution of phagocytic B cell and macrophage. Shandong University, Jinan, China.
6. **Jun Li**, 2011. Evolution of phagocytic B cells. Invited seminar. East China University of Science and Technology, Shanghai, China.
7. **Jun Li**. 2011. IgT, IgT+ B cells and mucosal Immunity in Teolest Fish. Yellow Sea Fishery Research Institute, The Chinese Fishery Academy of Science, Qingdao, China.

2012:

8. **Jun Li**, 2012. Evolution of phagocytic B cells. Institute of Hydrobiology, The Chinese Academy of Sciences, Wuhan, China.
9. **Jun Li**. 2012. IgT, IgT+ B cells and mucosal Immunity in Teolest Fish. East China University of Science and Technology, Shanghai, China.
10. **Jun Li.** 2012. Influence of complement and antibody opsonization in the uptake of particles by Rinbow trout phagocytes. Yellow Sea Fishery Research Institute, The Chinese Fishery Academy of Science, Qingdao, China.
11. **Jun Li**. 2012. New discoveries of fish B cells and immunoglobulins. Institute of Oceanology, The Chinese Academy of Sciences, Qingdao, China.
12. **Jun Li.** 2012. Influence of complement and antibody opsonization in the uptake of particles by Rinbow trout phagocytes. Ocean University of China, Qingdao, China.

2013:

13. **Jun Li**, 2013. Evolution of phagocytic B cells. The Third Institute of Oceanography, SOA, Xiamen, China.
14. **Jun Li**. 2013. Survival of *Edwardsiella tarda* in fish serum relates to bacterial surface LPS. East China University of Science and Technology, Shanghai, China.
15. **Jun Li**. 2013. The Involvement of Fish Complement in Innate and Adaptive Immune Responses. Institute of Oceanology, The Chinese Academy of Sciences, Qingdao, China.

2014:

16. **Jun Li**, 2014. The Mucosal Immunity in Teolest Fish. The Third Institute of Oceanography, SOA, Xiamen, China.
17. **Jun Li**, 2104. Phagocytic B cells: Innate or Adaptive? Zhejiang University, Hangzhou, China.
18. **Jun Li**, 2104. Phagocytic B cells: Innate or Adaptive? Huazhong Agriculture University, Wuhan, China.
19. **Jun Li**, 2104. Phagocytic B cells: Innate or Adaptive? Agriculture University of China, Beijing, China.
20. **Jun Li,** 2014. Innate and Adaptive Immunity in Aquatic Animals. Shanghai Ocean University. Shanghai, China.
21. **Jun Li,** 2014. Recent Progress on the Study of Fish Immunity. Qingdao Agriculture University. Qingdao, China.