THE UNIVERSITY OF ARIZONA
TUCSON ARIZONA

AQUACULTURE PATHOLOGY LABORATORY
School of Animal & Comparative Biomedical Sciences
BioSciences West; Building 88, room 226
1041 E. Lowell Street, Tucson, Arizona 85721-0090
Phone: 520-621-4438; Email: dhuie@email.arizona.edu

November 2, 2018

Armangul Zhdanova
Arsal, LLC (OOO "Arsal")
1 C Gagarin Str.
Yarovoye, Altay region, 658839
Russian Federation

E-mail: Arsal.export1@gmail.com

Case: 18-943

Dear Armangul Zhdanova:

The PCR and RT-PCR tests you requested for the detection of WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci have been completed. One bag of Brine-shrimp eggs/Artemia Salina cysts (Identification No. Great Yarovoye Lake (Russia); Lot No. non) arrived on October 24, 2018 in good condition. The samples were collected from Arsal LLC Fishing Area No. 148 in the year 2017. Representative samples (approx. 30mg) were collected for DNA and RNA extractions. WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci were not detected in the sample tested. The testing was completed on November 2, 2018. A summary of the tests and results is provided on the following pages.

We hope that this information will be helpful to you. A hard copy of this report will be mailed to you. If there are any questions regarding this case, please feel free to contact us.

UAZ Policy on certification: This report provides our findings on the samples submitted to our laboratory for examination, health status evaluation, disease diagnosis, or pathogen detection. It is our policy and intent to perform the most appropriate assay(s) for the determination of the health/pathogen status of all specimens submitted to our laboratory. However, this report in no way constitutes a stock or facility "certification" or a "certificate" of health/pathogen status for the sample(s) tested or for the stocks, or facility, from which the sample(s) were derived.
PCR: disclaimer: This report provides our findings on the samples submitted to our laboratory for pathogen detection. The PCR assay used by this laboratory for the detection of shrimp pathogens is a research tool. The results should be considered as experimental and tentative. Whenever possible, PCR results should be confirmed by alternative assay. This report in no way constitutes a stock or facility "certification" or a "certificate" of health/pathogen status for the sample(s) tested or for the stocks, or facility, from which the sample(s) were derived.
The **UAZ Aquaculture Pathology Lab** is a **OIE** (Office International des Épizooties or the Organization or World Animal Health Organization) Reference Laboratory for Taura Syndrome and a **U.S.D.A. A.P.H.I.S.** Approved Laboratory for export testing for White Spot Disease, Taura Syndrome, Infectious Hypodermal and Hematopoietic Necrosis, Infectious Myonecrosis, Yellowhead Disease, Acute hepatopancreatic necrosis disease, Crayfish plaque (*Aphanomyces astaci*), White tail disease (*Macrobrachium rosenbergii* nodavirus), and Necrotizing hepatopancreatitis *(Hepatobacter penaeid*).

Sincerely yours,

Michelle Garfias
Research Specialist

Arun K. Dhar, Ph.D.
Associate Professor
Aquaculture Pathology Laboratory Director

Case: 18-943

Tests Requested:	**Sample Type:**	**Species:**
WSSV, IHHNV	N/A	Brine-shrimp eggs/Artemia Salina cysts
TSV, YHV, IMNV, *Mr*NV	N/A	Brine-shrimp eggs/Artemia Salina cysts
AHPND/EMS, NHP-B, HPV, CP/*A. astaci*	N/A	Brine-shrimp eggs/Artemia Salina cysts

Protocols:
Real time WSSV: qPCR protocol by Durand and Lightner (J. Fish Dis. 2002, 25:381-389)
IHHNV: PCR described by Tang et al. (Virus Research, 2006, 118: 185-191) Real time
Real time TSV: RT-PCR protocol described by Tang, et al. (J Vir Methods, 2004, 109-114)
Real time YHV: qPCR protocol described by Aranguren, et al. (DAO. 2012, 98:185-192)
Real time IMNV RT-PCR: modified from Andrade, et al. (Aquaculture. 2007, 264: 9-15)
*Mr*NV: RT-PCR protocol described by OIE Manual of Diagnostic tests for Aquatic Animals 2014 Ch. 2.27
HPV: PCR protocol described by Tang, et al. (DAO 2008, 80:105-112)
AHPND/EMS: PCR protocol described by Han, et al. (DAO 2015, 113:33-40)
NHP-B: PCR method described by Aranguren et al. (Aquaculture 2010, 307:187-192)
CP/A. astaci: PCR protocol developed in this laboratory based on the OIE Manual of Diagnostic Tests for Aquatic Animals 2012

Table: PCR and RT-PCR result

UAZ #	Sample Ref	WSSV	IHHNV	TSV	YHV	IMNV	*Mr*NV	HPV	AHPND/ EMS	NHP-B	CP/*A. astaci*
18-943	Identification No. Great Yarovoye Lake (Russia) Lot No. non	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected

Conclusion: WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci were not detected in the sample tested.

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Armangul Zhdanova
Arsal, LLC (OOO "Arsal")
1 C Gagarin Str.
Yarovoye, Altay region, 658839
Russian Federation

E-mail: Arsal.export1@gmail.com

Case: 18-943-1

Dear Armangul Zhdanova:

The PCR and RT-PCR tests you requested for the detection of WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci have been completed. One bag of Brine-shrimp eggs/Artemia Salina cysts (Identification No. Great Yarovoye Lake (Russia); Lot No. non) arrived on October 24, 2018 in good condition. The samples were collected from Arsal LLC Fishing Area No. 148 in the year 2017. Representative samples (approx. 30mg) were collected for DNA and RNA extractions. WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci were not detected in the sample tested. The testing was completed on November 2, 2018. A summary of the tests and results is provided on the following pages.

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PCR: disclaimer: This report provides our findings on the samples submitted to our laboratory for pathogen detection. The PCR assay used by this laboratory for the detection of shrimp pathogens is a research tool. The results should be considered as experimental and tentative. Whenever possible, PCR results should be confirmed by alternative assay. This report in no way constitutes a stock or facility "certification" or a "certificate" of health/pathogen status for the sample(s) tested or for the stocks, or facility, from which the sample(s) were derived.
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Sincerely yours,

Michelle Garfias
Research Specialist

Arun K. Dhar, Ph.D.
Associate Professor
Aquaculture Pathology Laboratory Director

Case: 18-943-1

Tests Requested:	**Sample Type:**	**Species:**
WSSV, IHHNV	N/A	Brine-shrimp eggs/Artemia Salina cysts
TSV, YHV, IMNV, *Mr*NV	N/A	Brine-shrimp eggs/Artemia Salina cysts
AHPND/EMS, NHP-B, HPV, CP/*A. astaci*	N/A	Brine-shrimp eggs/Artemia Salina cysts

Protocols:

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Table: PCR and RT-PCR result

UAZ #	Sample Ref	WSSV	IHHNV	TSV	YHV	IMNV	*Mr*NV	HPV	AHPND/ EMS	NHP-B	CP/*A. astaci*
18-943-1	Identification No. Great Yarovoye Lake (Russia) Lot No. non	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected	Not Detected

Conclusion: WSSV, IHHNV, TSV, YHV, IMNV, *Mr*NV, HPV, AHPND/EMS, NHP-B, and CP/A. astaci were not detected in the sample tested.