



Procurement Hub – Fredericton  
301 Bishop Drive  
Fredericton, NB E3C 2M6

13 December 2019

**F5211-190589**

**ADVANCED CONTRACT AWARD NOTICE**

**TITLE: Genomic characterization of *Piscirickettsia salmonis* and *Aeromonas* species**

**ACAN:**

The purpose of this Advance Contract Award Notice (ACAN) is to signal the government's intention to award a contract for these services to BC Centre for Aquatic Health Sciences, 871A Island Highway, Campbell River, BC V9W 1C2. Before awarding a contract, however, the government would like to provide other suppliers with the opportunity to demonstrate that they are capable of satisfying the requirements set out in this Notice, by submitting a statement of capabilities during the ACAN posting period.

If other potential suppliers submit a statement of capabilities during this ACAN posting period that meets the requirements set out in the ACAN, the government will proceed to a full tendering process on either GETS or through traditional means, in order to award the contract.

If no other supplier submits, on or before the closing date, a statement of capabilities meeting the requirements set out in the ACAN, a contract will be awarded to the pre-selected supplier.

**INTRODUCTION:**

Fisheries and Oceans Canada (DFO) requires the services of specialized analysis of isolated from British Columbia waters to inform fish health management and to support improved diagnostics.

**BACKGROUND:**

The contract is to support the ACRDP Project "Genomic characterization of *Piscirickettsia salmonis* and *Aeromonas* species isolated from British Columbia waters to inform fish health management and to support improved diagnostics".

**OBJECTIVES:**

The objective of this requirement is to obtain high-quality draft genome sequences for Psal and Asal isolates obtained from disease outbreaks in BC. The sequence data from this project will be used to develop new molecular diagnostic tests that are species specific and/or suitable for identification of isolates which are highly pathogenic and/or have reduced antibiotic sensitivity. Information on antimicrobial sensitivity and virulent factors will help DFO and company veterinarians make decisions with respect to the treatment and management of these diseases as well as support vaccine development.

**WORK REQUIREMENT:**

The work we require includes:

1. obtaining high-quality draft genome sequences for 12 isolates of Psal selected based on the locations, clinical signs and antibiotic resistance.
2. obtaining high-quality draft genome sequences for select isolates of Asal, and the other 3 *Aeromonas* species we found in BC. Asal isolates will be selected for sequencing based on the MIC profiles, knowledge of disease severity/treatment outcome, and will include isolates from seawater and freshwater sites.

The Project Coordinator will be responsible for the identification of which isolates to sequence.

Qualified Fish Health Technicians will recover isolates on blood agar with 2% salt (BAS) and/or blood agar with haemoglobin.

Genomic DNA will be extracted using a phenol-chloroform protocol to isolate long DNA fragments (up to 200 kb). Briefly, Gram negative bacteria will be lysed using Proteinase K, DNA will be purified using phenol-chloroform steps and precipitated using ethanol. DNA quality control will be performed by assessing fragment size on an agarose gel, DNA purity will be evaluated by a spectrophotometric method (absorbance ratios 260/280 and 260/230 equal to 1.8 and 2-2.2 are indicative of pure DNA respectively), and DNA concentrations will be measured using dsDNA HS Assay.

#### Library Construction and Nanopore Sequencing

Extracted DNA will be sheared using g-Tube to obtain approximately 8 kb fragments. Shared DNA samples will be further cleaned and sized using solid-phase reversible immobilization beads. Library preparation will be performed using ID Native Barcoding genomic DNA and Ligation Sequencing ID kits (Oxford Nanopore Technologies). Briefly, DNA ends will be repaired and dA-tailed using the NEBNext End Repair/dA-tailing module. Then the dT-tailed barcode adapters are ligated on the dA-tailed template. Barcoded samples will be pooled and used for barcoding adaptor ligation step using Ultra II ligation module. A final library cleanup will be performed using the solid-phase reversible immobilization beads. Libraries will be loaded to the flow cells (Oxford Nanopore Technologies) according to the manufacture's recommendation.

#### Sequence Analysis

Fast5 generated from the Minion sequencer will be analysed using Albacore or MinKNOW basecallers. The generated Fastq files will be analysed further for sequence quality and control using Minion QC ([https://github.com/roblanf/minion\\_qc](https://github.com/roblanf/minion_qc)) in R using the summary data file as input. Adaptors will be trimmed using the PoreChop command line. As a confirmatory step, taxonomy classification of the reads will be performed using Kraken program. Reads will be mapped to the appropriate reference genome using the BWA and SAMTOOLS command lines. Statistical data of the read mapping will be generated using samtools stats and coverage will be visualized using ggplot2 library in R. Sequence annotation will be performed using the Prokka program. The phylogenetic analysis of the genome sequences will be analysed using the pan genome pipeline Roary. Antimicrobial and virulent factors will be assessed using RAST Annotation Server and the Pathosystems Resource Integration Center (PATRIC platform <https://www.patricbrc.org/>). Sequence similarities and differences will be characterized and selected for further analysis. Staff of the contractor have received formal training on the assembly and analysis of bacterial genomes obtained using MinION sequencing.

All data generated from sequencing and analysis will be provided to the Project Coordinator.

#### **TASKS AND DELIVERABLES:**

The Contractor will work with the Project Coordinator to deposit all sequence data into public databases.

A short progress report detailing the status of sequencing and data analysis will be submitted to the Project Coordinator or Technical Authority at the mid-point of the project.

A final report (in English) will include detailed descriptions of the laboratory and sequencing methods used, accession numbers of sequences, and results of the analysis of virulence and antibiotic resistance.

The contractor will participate in monthly progress meetings with the Project Coordinator or Technical Authority that will be arranged at a mutually agreed upon times. These meetings will be held in person or via conference call.

#### **MANDATORY CRITERIA:**

- The contractor must be able to provide isolates of *Piscirickettsia salmonis* (*Psal.*) (minimum of 12 isolates) and *Aeromonas* species (*As.*) (minimum of 6 isolates) from British Columbia salmon farming industry. Each isolate needs to be phenotypically characterized with respect to antibiotic sensitivity. For each isolate associated epidemiological data, such as site of location, treatment outcome etc. must be available.

- The contractor must have a minimum of 12 months of experience in library construction and Nanopore “MinIon” sequencing of bacterial genomes.
- The contractor must have minimum of 48 months of experience of bioinformatics experience including experience in the analysis of Nanopore “MinIon” sequence data which will includes base calling and polishing and mapping of sequences to reference genomes.
- The contractor must have a minimum of 48 months of experience in phylogenetic analysis of nucleotide and amino acid sequences using Bayesian methods (BEAST).

**ESTIMATED VALUE**

The total value of the contract shall not exceed \$49,500 excluding all applicable taxes for the firm period from contract award to 15 March 2021.

**SECURITY REQUIREMENT**

No Security Requirement

**TRADE AGREEMENTS APPLICABILITY OR OTHER OBLIGATIONS:**

No Trade Agreements apply.

**GOVERNMENT CONTRACTS REGULATIONS EXCEPTION AND LIMITED TENDERING REASONS:**

The following policy requirements are applicable to this ACAN process:

**Applicable Exceptions to Soliciting Bids under the Government Contracting Regulations (GCRs)**  
(Section 6):

Section 10.2.1 Section 6 (d) only one person or firm is capable of performing the work applies to this ACAN for the following reasons:

There are no known alternative sources of supply. It is feasible and affordable to compete the requirement.

**CONTRACT PERIOD:**

The period of the contract is from contract award to 15 March 2021 inclusive.

**SUPPLIER’S RIGHT TO SUBMIT A STATEMENT OF CAPABILITIES:**

Suppliers who consider themselves fully qualified and available to provide the services described herein, **must submit a Statement of Capabilities in writing to the Contracting Officer identified in this Notice on or before closing date. The Statement of Capabilities must clearly demonstrate how the supplier meets the advertised requirements.**

**CLOSING DATE FOR SUBMITTING STATEMENT OF CAPABILITIES:**

**30 December 2019 at 2:00 p.m. (AST)**

Inquiries and statements of capabilities are to be directed to:

**Michael Peters**

Contracting Specialist – Contracting Services

Procurement Hub – Fredericton

Telephone: (506) 429-2359

Email: DFOtenders-soumissionsMPO@dfo-mpo.gc.ca