GenomeQuébec CES | CENTRE D'EXPERTISE ET DE SERVICES

Centre d'expertise et de services Génome Québec

SEPTEMBER 8, 2020

SEQUENCING SERVICES

Centre d'expertise et de services Génome Québec

User Guide
SNP discovery and *de novo* sequencing by primer walking services Sanger-type technology

Version 4.0

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Introduction

This document describes the procedure for submitting samples for SNP discovery and *de novo* sequencing by primer walking. In order to use these services, an account must first be opened in our web application Nanuq. Samples must then be sent to the Centre d'expertise et de services Génome Québec (CES) in accordance with the submission requirements described in this guide.

Service Request

Quote Request

Contact the client management office for more information regarding prices or quote requests:

Phone: 514-398-7211
E-mail: infoservices@genomequebec.com

Note: A contract is required for all services that total or exceed $25,000.00 (prior to taxes).

Service Request

To complete a service request:

1. Download the sequencing service's request form.

2. Carefully complete the form following the given instructions.

3. Send the completed form by e-mail to the client management office and include also:

   a. A copy of the ethics review committee's approval form(s) for all submitted samples obtained from human subjects.

   b. A Purchase Order number, if it is the chosen method of payment.

Note:

Within 24 hours a username and password to access Nanuq will be forwarded, after which online sample submission is possible.

Billing Policy

Projects that last no longer than 3 months will be invoiced at the end of the project. However, those that last longer will be invoiced at regular intervals. In either case, the billing information must be provided on the sequencing services request form.

Method of Payment

Payments can be made by cheque (purchase order mandatory) or credit card. The cheque must be addressed and sent to:

Génome Québec
630, boulevard René Lévesque, suite 2660,
Montréal (Québec) Canada
H3B 1S6

Important:

A purchase order number is required for services that total or exceed $2,500.00 (prior to taxes).

For security purposes do not enter the credit card information in the sequencing services request form. A separate form will be sent at time of billing.

Sample Preparation and Submission – General Guidelines

It is crucial that the guidelines mentioned in the User Guide be carefully followed so that unnecessary delays can be avoided.

Sample Submission Requirements

Sending genomic DNA samples for a SNP discovery project:

It is important to provide a sufficient amount of good quality genomic DNA in order to complete the project in its entirety, that is 2 µl of DNA per fragment to be analyzed. The concentration of genomic DNA samples should be about 20 ng/µl.

A 96-well plate must be used for submissions of more than 48 samples. A list including the name, volume and concentration of each sample must be provided by email.

Sending a sample for a de novo sequencing project:

The concentration of plasmid DNA required is between 100 and 500 ng/µl. The DNA must be of good quality in order to ensure that the sequencing reactions work adequately.

The required quantity of plasmid DNA for a project is approximately 2 µl or 4 µl per 600 bases. For example, if the region to be sequenced has a length of 5000 bases, 8 to 9 sequencing reactions will be necessary. Such a project requires a minimum of 18 µl of plasmid DNA. If the sequencing of both DNA strands is requested, the minimum amount of plasmid DNA is twice as much.

It is imperative to remove all traces of phenol/chloroform or ethanol from plasmid DNA samples. It is recommended to resuspend DNA in 10 mM Tris-HCl pH 8 or water and to avoid EDTA. The ratio of optical density (OD) 260/280 must be between 1.7 and 1.9.

Note

When PCR or sequencing primers are provided, the following rules should be followed:

- **Primer length should be between 18 and 24 bases**
- **PCR products should be between 250 and 750 bases**
- **The annealing temperature (Tm) of the primers should be greater than 50°C.**
- **The primers should be located at least 50 bases from the region to be sequenced**
- **The concentration of primers should be provided**
- **The Tm as well as the % GC of the primers should be provided**

Sending Samples

Samples can be sent by courier or brought to the CES in person from Monday to Friday between 9h00 am and 5h00 pm.

IMPORTANT!

Samples are stored at -20°C throughout the duration of the project. Once the project is completed, the client can pick up the left-over samples.

Address for sending samples

Please refer to the waybill for instructions on how to ship your samples.

Transmission of Results

When a SNP discovery analysis is required, a SNP report will be sent to the client. This report comprises the following information:

-the position of the SNPs found in the genome;

-the accession number (rs) if the SNP is already known;

-the genotype of each analyzed sample;

-location of the SNP in an intron or an exon and, in the latter case, if it is a synonymous or non-synonymous SNP;

-the change in amino acid if applicable.

A tutorial is sent along with the report for better understanding of the data. The client can contact the scientific director, Alexandre Montpetit, at all times for any question regarding the results.

A partial report can be requested during the project in order to see its progress.

The sequencing results are directly accessible via **Nanuq**. The chromatograms or the texts can be viewed (FASTA or GenBank format) and downloaded locally.

The downloaded sequences can be viewed by using a free version of Chromas for PC available on the internet: **http://www.technelysium.com.au/chromas.html**

All sequences are stored and accessible on **Nanuq** for a minimum of one year. The sequences are then archived but remain accessible upon request. Customers may request the removal of their data from Nanuq at any given time.

For More Information

Client Management Office

Phone : 514-398-7211
Email : infoservices@genomequebec.com

Sequencing Services

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