

COFACTOR

THE LEADERS IN RNA

Cofactor Prism Software

Version 0.7

Introduction

Cofactor Genomics' ImmunoPrism enables researchers to profile the immune response at the site of a solid tumor. The protocol accepts total RNA extracted from solid tumor tissue and delivers a fully-analyzed immune report.



This document explains how to use Prism, a web portal that facilitates cloud-based analyses of samples prepared using the ImmunoPrism Kit. Prism allows users to share samples via Illumina's BaseSpace, generate Individual Sample Reports and Biomarker Discovery Report, and view the status of submitted projects and results from completed projects.

IMPORTANT: Analyses in Prism were specifically developed for libraries using the optimized reagents and protocol of the ImmunoPrism Kit. As such, immune profiling results in the Individual and Biomarker Discovery reports may be dramatically aberrant for samples prepared without the ImmunoPrism Kit.

0. BaseSpace Preparation

The ImmunoPrism Kit generates libraries to sequence on an Illumina sequencing machine. To run analyses on Prism, sample FASTQs must be accessible to BaseSpace. Depending on the sequencer model and the IT infrastructure of users, sequenced reads via FASTQ files will be stored on BaseSpace via two methods.

Note: Free BaseSpace accounts can store up to 1 Terabyte of sequencing data and can be created at <https://basespace.illumina.com/>

Many users will have a locally run pipeline to demultiplex sequencing runs and generate a single FASTQ file per sample. In this situation, we recommend users upload FASTQ files to BaseSpace using the web uploader. Instructions for this method can be found in this Illumina article:

<https://help.basespace.illumina.com/articles/tutorials/upload-data-using-web-uploader>.

Other users may have sequencing runs upload in real-time to BaseSpace from select sequencing machines (e.g MiSeq) and demultiplex runs using BaseSpace tools. This setup and procedure is beyond this guide. Please refer to your machine manual to understand how to configure machines and how to setup runs to upload directly to BaseSpace. For additional assistance, we recommend the “New to BaseSpace” article for configuration and

demultiplexing in BaseSpace: <https://help.basespace.illumina.com/articles/new-to-basespace>. When employing this method, each sample should have only 1 FASTQ file.

Note: Other methods may exist for creating or uploading FASTQ files to BaseSpace. Any method that creates individual FASTQ files per sample can be used to run analysis on Prism.

1. Creating and Logging into a Prism Account

The Prism web portal is found at <http://prism.cofactorgenomics.com/>

The first step to run the immune profiling analysis is to create a Prism account. Visit the web portal at the above link and click “Login to Start”.

From this login page, click “create user”. Follow the form and input the relevant information to create your account. Note the username and password; this will be used in later sessions to log into your account and access current and past analyses. Upon account creation, you will be directly logged in. From here, you can proceed to instructions for submitting a project for analysis (**Step 2**).

To log into the Prism web portal visit the above web page and click “Login to Start”. Then input your account credentials and click “Login”. From here, you can proceed to instructions for submitting a project for analysis (**Step 2**) or looking at previous results (**Step 3**).

2. Submitting a Project for Analysis

To submit a project, you will share the files stored on BaseSpace with your Prism account.

After logging in, click “Submit New Project” in the top toolbar from any page in Prism. This action will navigate you to the submission portion of the user home page. This portion has instructions on the sample upload process using BaseSpace. Click “Upload via BaseSpace” to start the process.

Clicking the “Upload via BaseSpace” button will momentarily take you away from Prism and to BaseSpace. First, you will log into BaseSpace using your BaseSpace credentials (these are different than your Prism credentials). Second, you will fill out a form for the new project. Inputs include the project name, the tissue disease, and samples by group. The grouping of samples, and the corresponding grouping names, are necessary to generate

the Biomarker Discovery Report. Note that a minimum of 3 samples per group are required to generate the Biomarker Discovery Report. When you have input all of the necessary fields, click the “Launch Application” button to submit the form. Lastly, a dialogue will ask you to permit Prism to download the sample files that you specified. Click “accept” to finish the BaseSpace process. You will be redirected back to Prism and will see a confirmation message.

Congratulations, you have successfully created a Prism analysis project!

From this confirmation page, click “Go Home” to return to the user home page. To see the status of this and other submitted projects, proceed to instructions for looking at project status and analysis results (Step 3).

Note: You will receive an email notification when your project has completed analysis. A link in the email will take you directly to Prism to view the project’s Individual Sample Reports and Biomarker Discovery Report.

3. View Project Status and Analysis Results

Prism enables a user to see the status of submitted projects and to view sample and biomarker reports per project. While logged in, click “See Results” in the top toolbar or any page of Prism. Here you will see a table of projects you have created on Prism. The table has four columns for the status, name, date of submission, and Biomarker Discovery Report link for projects.

The status of each project can be:

- “Waiting for Approval”, where Cofactor Genomics could potentially hold a project and review it before starting analysis;
- “Queued”, where the project is in line for computational resources to run analysis;
- “Running”, where the project analysis is currently running;
- “Success”, where the project analysis is complete and reports are available; and

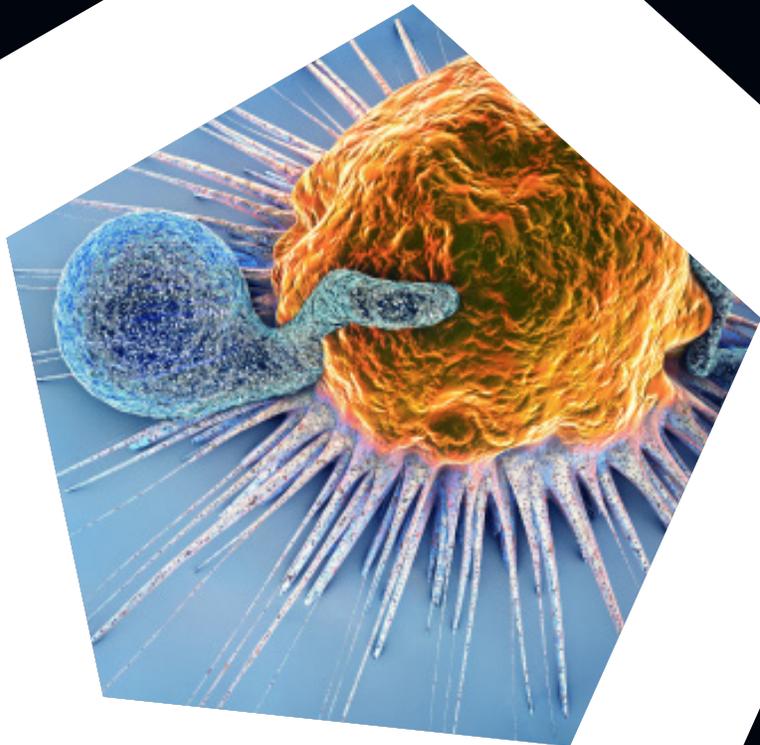
If a project has finished analysis (indicated by a “Success” status), Individual Sample Reports and a Biomarker Discovery Report will be available. To access these reports, return to the table of projects and click on the name of the project. On this project page, you will see a table with a row for each sample in the project. Click the link

in each row, under the “Report” column, to access the Individual Report of each sample. Immediately below the table, click the link for the Biomarker Discovery Report.

If no links are in this page, your project has not completed analysis.

4. Getting Help

If you have any challenges with using Prism, or have questions about project status or reports, please contact Cofactor Genomics at support@cofactorgenomics.com.



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