


|  | Work instruction         | Code       | Revision | Date       | Owner         |
|---|--------------------------|------------|----------|------------|---------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit |
|   |                          |            |          |            | Page: 1 of 19 |



## Agendo User Guide


### Genomics Unit

|  |                                  |              |
|--|----------------------------------|--------------|
| Created by: <ul style="list-style-type: none"> <li>• Clemente Borges</li> <li>• Berta Fusté</li> </ul> | Last modification:<br>12.02.2025 | Version: 1.0 |
|--|----------------------------------|--------------|

|  | Work instruction         | Code              | Revision | Date       | Owner         |
|---|--------------------------|-------------------|----------|------------|---------------|
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit |
|   |                          |                   |          |            | Page: 2 of 19 |

## Contents

|   |    |
|---|----|
| 1. Agendo Registration.....                               | 3  |
| 2. List of Genomics Unit Agendo Requests .....            | 3  |
| 3. Agendo workflow .....                                  | 4  |
| 4. Submitting Agendo Requests for the Genomics Unit ..... | 6  |
| 4.1 General submission.....                               | 6  |
| 4.2 Importing samples in plates.....                      | 11 |
| 4.3 Importing libraries.....                              | 13 |
| a) Importing single/individual libraries .....            | 13 |
| b) Importing pooled libraries .....                       | 13 |
| 5. Quick Agendo Sample Submission Guide .....             | 14 |

|  | Work instruction         | Code       | Revision | Date       | Owner                          |
|---|--------------------------|------------|----------|------------|--------------------------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit<br>Page: 3 of 19 |

## 1. Agendo Registration

To be able to work with the Genomics Unit all users must be registered at Agendo:

- URL: <https://crg.agendoscience.com/>
- You must register yourself at “Register” link at the bottom of the login form. Fill all the required information under the “Create a new account” form.
- On the registration form, you will have to fill in your credentials and select the list and the group (laboratory). If your institute or group is not in the lists, select “Other” and complete the information to register a new institute and group.
- Then, request access to the facilities that you plan to use by clicking on the empty text box. Check the facility “Genomics Unit” to gain access to all the Genomics Unit resources.
- Upon registration, your account is reviewed by an institutional administrator. Once your account is accepted, you will receive a temporary password that must be changed upon the first login. After that, you are ready to go.

if you need:

- help with your registration
- to add a financial account/project number
- to delete an old member of your group
- if you have changed from one group to another


Please, write to [isabel.jurado@crg.eu](mailto:isabel.jurado@crg.eu)

For more information you may also follow the official Agendo [Quick user guide](#).

## 2. List of Genomics Unit Agendo Requests

The Genomic Unit offers multiple services distributed in different request classes:

- NGS Library Prep and Sequencing: Whole and Targeted Genome
- NGS Library Prep and Sequencing: Transcriptome
- NGS Library Prep and Sequencing: Epigenome
- NGS Library Prep and Sequencing: Single Cell genomics and transcriptomics
- NGS Library Prep and Sequencing: RNA plates
- NGS Library Prep and Sequencing: DNA plates
- NGS Libraries Ready for Sequencing
- NGS complementary services: Automated electrophoresis
- NGS complementary services: Covaris

|   |                          |                   |          |            |               |
|---|--------------------------|-------------------|----------|------------|---------------|
|  | Work instruction         | Code              | Revision | Date       | Owner         |
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit |
|   |                          |                   |          |            | Page: 4 of 19 |

| Whole and Targeted Genome | Transcriptome                  | Epigenome  | Single Cell genomics and transcriptomics |
|---------------------------|--------------------------------|------------|--|
| WG-Seq                    | mRNA-seq                       | ChIP-seq   | Single cell RNA-seq                      |
| ExomeCapture-seq          | Ribodepleted RNA-seq           | ATAC-seq   | Single cell ATAC-seq                     |
| CustomCapture-seq         | Low input mRNA-seq             | HiC-seq    | Single cell Multiome_ATAC                |
| PanelCapture-seq          | Low input ribodepleted RNA-seq | Methyl-seq | Single cell Multiome_GE                  |
| Amplicon-seq              | smallRNA-seq                   | Other      | SMARTseq2                                |
| Genome editing CRISPR     | Ribo-seq                       |            | Other                                    |
| WG SNP genotyping         | Other                          |            |  |
| Other                     |                                |            |  |


How to access to Genomics Unit requests:

- The fastest way to find a Genomics Unit request or resource is by typing its name in the search tool on the top navigation bar.
- If you do not know the name of the resource, you can find the complete list in Requests, accessible through the menu on the left, and sort by class or Facility.

### 3. Agendo workflow

All requests follow a simple workflow:

- **Draft:** complete the request and save it as draft allowing you to leave and return later to continue editing it until it's ready for submission.
- **Submission:** when a request is submitted it cannot be changed again by the user unless the Genomics Unit suggests changes after the reviewing of it.
- **Locked:** once a submitted request is opened by Genomics Unit, its status is automatically set to locked and the requesting user will no longer be able to alter it. Managers can edit the request while it is not completed.
- **Approved/rejected:** once reviewed by the Genomics Unit the request can be Approved or Rejected:
  - The unit may return the request with comments or suggestions for revisions, at which point the status will change to "Rejected", and allowing the user to edit again, and resubmit the request.
  - If the request is satisfactory and "Approved," the Genomics Unit may issue a quote for it. Once the quote is issued, it requires the PI's approval before processing can continue. After the PI approves the quote "approval for funds", the Genomics Unit will begin the experimental process.
- **Completed:** the request will be considered completed when NGS data is already sent. For sample quality results or any small document (pdf or doc) you can access them by clicking on the folder icon located in the top right corner of the request details, as shown in the following figure. You will be redirected to a new page, "NextCloud", where you have to login with your Agendo credentials (user and password). **IMPORTANT:** the capacity of the NextCloud is limited, download the results as soon as possible.

|  | Work instruction         | Code       | Revision | Date       | Owner         |
|---|--------------------------|------------|----------|------------|---------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit |
|   |                          |            |          |            | Page: 5 of 19 |



Note that:


During the whole process you may receive comments from the Genomics Unit. The comments may be seen by clicking on the “Comments” tab at the request upper left menu.


General

- Comments 8
- Notes
- History
- Files 2
- Participants
- Milestones

**REQUEST DETAILS**

|                 |                                  |                  |           |   |
|-----------------|----------------------------------|------------------|-----------|---|
| Request #       | 6938                             | Custom reference | 2024BD303 | <span style="color: green;">👍 Approved</span> |
| Class           | Data analysis (Bioinformatics) ⓘ |                  |           |   |
| User            | Irene González ⓘ                 |                  |           |   |
| Group           | Genomics Unit (Berta Fuste) ⓘ    |                  |           |   |
| Institute       | CENTRE DE REGULACIÓ GENÒMICA     |                  |           |   |
| Requesting date | 2024-12-20 13:47:07              |                  |           |   |
| Account         | 006341 - Genomics Unit ↻         |                  |           |   |



|  | Work instruction         | Code              | Revision | Date       | Owner         |
|---|--------------------------|-------------------|----------|------------|---------------|
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit |
|   |                          |                   |          |            | Page: 6 of 19 |

## 4. Submitting Agendo Requests for the Genomics Unit

### 4.1 General submission

Each Genomics Unit Agendo Request has 4 blocs to be completed:

- General information
- Request form
- Sample list
- Comments

Some fields are common for all the Requests and others are Request specific fields. Moreover, some fields are mandatory (they show an asterisk \*) and others are optional fields.

First add the administrative information into the **GENERAL INFORMATION** area

**GENERAL INFORMATION** FOR MORE INFORMATION, CLICK HERE

Submit request on behalf of another user

Select user


Account

Research line


Use this field to reference a previous request

Title

- Account: only for CRG users, it is associated to a project number already introduced in the system (Ex. 00XXXX – Genomics Unit). If the project it is not introduced, please send an email to Isabel Jurado, [isabel.jurado@crg.eu](mailto:isabel.jurado@crg.eu)
- Research line: corresponds to a previous Agendo request in case of generating multiple Agendo request that must be connected
- Title: free text, add the information related to your project

|  | Work instruction         | Code       | Revision | Date       | Owner         |
|---|--------------------------|------------|----------|------------|---------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit |
|   |                          |            |          |            | Page: 7 of 19 |

Second complete the experimental design that you would need into the **REQUEST FORM** area. Most fields will be self-explanatory as you may have to select an option from a list, or they may show some text below the field to help you understand what is needed.

**REQUEST FORM** 

Order number   
Specify here the official order number for this request provided by your institution (only to be filled by non-CRG users)

Subproject name   
To be filled by Genomics Unit

Application \*

Input \*

Sample type \*


Number of samples \*

Sample preparation only? \*

Sequencing depth \*   
M reads / sample

Read length \*

Full run required   
Specify in comments sequencer type and run format (read1,index1,index2,read2) (e.g. 50,8,8,50 or 51,10,0,0)

|   |                          |                   |          |            |               |
|---|--------------------------|-------------------|----------|------------|---------------|
|  | Work instruction         | Code              | Revision | Date       | Owner         |
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit |
|   |                          |                   |          |            | Page: 8 of 19 |

Third you need to add the information related to the samples to be processed at the **SAMPLE LIST** area.

Sample information can be completed manually into the request form by filling in the table shown and adding extra rows as needed by sample using the “+ Add sample” icon as shown in the figure.

### SAMPLE LISTS


Before filling the CSV or the table below, check mandatory fields (\*)

Download 
Upload 
+ Add sample
Remove all 

Validate




| SAMPLE NO. | CODE | SAMPLE NAME<br>*  | SAMPLE TYPE *<br> | SAMPLE SPECIES NAME<br>(USE NCBI TAXONOMY) *<br> | VOLUME (UL) *<br> | CELL OR NUCLEI<br>CONCENTRATION<br>(CELLS/UL OR NUCLEI/UL) *  | TARGET<br>RECOVERY *  |
|------------|------|--|--|---|--|--|--|
|            |      |  |  |   |  |  |  |

Sample number and custom IDs will be automatically created upon submission | \* Required fields







In the case of many samples, the information can be also uploaded through a CSV file. Be aware that each type of request requires different information, so each request type has its own and specific CSV template. To download the CSV template provided by Agendo, simply click on the icon (**Download** ) as shown in the next Figure.

### SAMPLE LISTS

Before filling the CSV or the table below, check mandatory fields (\*)

Download 
Upload 
+ Add sample
Remove all 


Validate

| SAMPLE NO. | CODE | SAMPLE NAME<br>*  | SAMPLE TYPE *<br> | SAMPLE SPECIES NAME<br>(USE NCBI TAXONOMY) *<br> | VOLUME (UL) *<br> | CELL OR NUCLEI<br>CONCENTRATION<br>(CELLS/UL OR NUCLEI/UL) *  | TARGET<br>RECOVERY *  |
|------------|------|--|--|---|--|--|--|
|            |      |  |  |   |  |  |  |

Sample number and custom IDs will be automatically created upon submission | \* Required fields

When working with CSV files all information related to one sample will appear in one cell separated by comas. If you prefer to work by columns, please modify the document using the feature “Text to columns” found in the Excel menu and save it as CSV (Comma delimited) (\*.csv).



|   |                          |                   |          |            |               |
|---|--------------------------|-------------------|----------|------------|---------------|
|  | Work instruction         | Code              | Revision | Date       | Owner         |
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit |
|   |                          |                   |          |            | Page: 9 of 19 |

Entering the information using the correct format or specific text is essential for uploading the data from the CSV file. If this is not filled out correctly, an error will occur, and the data upload will not be allowed. To facilitate this, some fields have extra information and guidelines to be completed. Please check them out by mousing over the information icon (i) for each of the sample field names. You will get a help tooltip as shown in the next Figure or you can find all the information at the end of this document “Quick Agendo Sample Submission Guide”.

**SAMPLE LISTS**

Before filling the CSV or the table below, check mandatory fields (\*)

| SAMPLE NO.   | CODE | SAMPLE NAME * | SAMPLE TYPE * | SAMPLE SPECIES NAME (USE NCBI TAXONOMY) * i | VOLUME (UL) * | CONCENTRATION (NG/UL) * i | NANODROP 260/280 * i |
|--|------|---------------|---------------|---|---------------|---------------------------|----------------------|
| Sample number and custom IDs will be automatically created upon submission   * Required fields |      |               |               |   |               |                           |                      |

Between 2 and 15 alphanumeric characters, underscore included (e.g. TE\_123)

Once the CSV file is ready with all your samples, go back to the “Sample list” section under your Agendo request, and click on “**Upload**”. It is mandatory to upload a CSV file (not Excel files).


**SAMPLE LISTS**

Before filling the CSV or the table below, check mandatory fields (\*)

| SAMPLE NO.   | CODE | SAMPLE NAME * | SAMPLE TYPE * | SAMPLE SPECIES NAME (USE NCBI TAXONOMY) * i | VOLUME (UL) * | CONCENTRATION (NG/UL) * i | NANODROP 260/280 * i |
|--|------|---------------|---------------|---|---------------|---------------------------|----------------------|
| Sample number and custom IDs will be automatically created upon submission   * Required fields |      |               |               |   |               |                           |                      |


Finally, a message will appear to confirm if you want to either append or replace the CSV data to the Agendo sample list. If the Agendo sample list is empty, it does not really matter which option you select. But if you have already samples in the Agendo list, and you want to recreate them, the best option is to choose the “Replace” option. The append option would add new samples at the end of already existing samples. You should see all your samples on the list.

Do you want to replace (and loose the IDs) or to append more information?


|  | Work instruction         | Code       | Revision | Date       | Owner          |
|---|--------------------------|------------|----------|------------|----------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|   |                          |            |          |            | Page: 10 of 19 |

Before finalizing the process and in case you need to clarify something, please add free text in the **COMMENTS** area.

To finish the submission, click at the end of the page “submit” and confirm the submission.

 Save draft

Reset


 Submit

●

Request is about to be submitted. Do you want to proceed?

Confirm

Cancel

|   |                   |            |          |            |                                 |
|---|-------------------|------------|----------|------------|---------------------------------|
|  | Work instruction  | Code       | Revision | Date       | Owner                           |
|   | Agendo User Guide | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit<br>Page: 11 of 19 |

## 4.2 Importing samples in plates

There are only three available Agendo requests involving the submission of samples in plate:

- NGS Library Prep and Sequencing: Plates – DNA: for large scale projects such as WGS or Amplicon-seq experiments.
- NGS Library Prep and Sequencing: Plates – RNA: for large scale RNA projects for standard input mRNA or SMARTseq2 experiments.
- NGS complementary services: Automated electrophoresis

For these requests, you must download a *pre-filled Excel template* that includes a plate number and a well position for placing a sample (see Figure below with red IMPORTANT message with links).

**SAMPLE LISTS**

**IMPORTANT** Download the [Excel pre-filled plates template](#). Read the [guidelines](#).

| SAMPLE NO. | CODE | PLATE NUMBER<br>* ⓘ | PLATE NAME *<br>ⓘ | WELL POSITION<br>* ⓘ | SAMPLE NAME<br>* ⓘ | SAMPLE TYPE ⓘ | SAMPLE SPECIES NAME<br>(USE NCBI TAXONOMY) *<br>ⓘ |
|------------|------|---------------------|-------------------|----------------------|--------------------|---------------|---|
|------------|------|---------------------|-------------------|----------------------|--------------------|---------------|---|

Sample number and custom IDs will be automatically created upon submission | \* Required fields

By default, the template contains up to three plates with the “Plate Number” and the “Well Position” are already pre-filled, you just need to fill the sample data in the right positions (See figure below). It is **mandatory to remove the rows that you are not using**. In the end, you will need to convert the Excel file into a CSV file, which is the only format supported by Agendo to upload samples (CSV (Comma delimited) (\*.csv)).

| A          | B    | C            | D          | E             | F           | G           | H            | I           | J             | K            | L            | M            | N            |
|------------|------|--------------|------------|---------------|-------------|-------------|--------------|-------------|---------------|--------------|--------------|--------------|--------------|
| Sample No. | Code | Plate Number | Plate Name | Well Position | Sample Name | Sample Type | Sample Speci | Volume (ul) | Concentration | Nanodrop 260 | Nanodrop 260 | Sample Repla | Observations |
|            |      | 1            | plate_1A01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1B01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1C01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1D01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1E01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1F01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1G01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1H01 |               |             |             |              |             |               |              |              | no           |              |
|            |      | 1            | plate_1A02 |               |             |             |              |             |               |              |              | no           |              |


Plates – DNA: specific plate distribution by columns, and **well G12 and H12** should always be **empty**. Do not leave empty wells between samples.

Plates – RNA: specific plate distribution by columns, and **well H12** should always be **empty**. Do not leave empty wells between samples.

Plates – Automated Electrophoresis: specific plate distribution by columns, and **well D12 and H12** should always be **empty**. Do not leave empty wells between samples. Please, use the “AE\_pre-filled\_plate\_template” to submit samples information.

|  | Work instruction         | Code       | Revision | Date       | Owner          |
|---|--------------------------|------------|----------|------------|----------------|
|   | <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|   |                          |            |          |            | Page: 12 of 19 |

Accepted plates: Eppendorf twin-tec PCR 96 semi-skirted (ref. 30128575) and NZYTECH 96 well PCR plates half skirt (ref. LW00801). If sending plates with dry ice, we recommend using the Bio-Rad Microseal "B" seals (ref.MSB1001) to prevent detachment of the foil.

|  | Work instruction         | Code              | Revision | Date       | Owner          |
|---|--------------------------|-------------------|----------|------------|----------------|
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit  |
|   |                          |                   |          |            | Page: 13 of 19 |

### 4.3 Importing libraries

If you are bringing already prepared libraries to the Genomics Unit, there is a specific Agendo request for it *NGS Libraries Ready for Sequencing*. Users can submit single/individual libraries, pooled libraries or a mix of single and pooled libraries by adding information within the Sample list section.

#### a) Importing single/individual libraries

To import single libraries, you just need to ignore the sample list field called *“Pool name (mandatory for each library belonging to a pool)”*. In this way, the system will consider the libraries as single/individual.


| Library Name * | Species Name (Use NCBI taxonomy) * | Sequence index I7 * | Sequence index I5 * | Pool name (mandatory for each library belonging to a pool) * | Library or Pool Volume (ul) * |
|----------------|------------------------------------|---------------------|---------------------|--|-------------------------------|
| lib_01         | Homo sapiens                       | AGGTCGGA            | AACCTAGA            |  | 1.4                           |

#### b) Importing pooled libraries

In order to import pooled libraries, you must fill the *“Pool name (mandatory for each library belonging to a pool)”* field as it is clearly stated in the field name.

| Library Name * | Species Name (Use NCBI taxonomy) * | Sequence index I7 * | Sequence index I5 * | Pool name (mandatory for each library belonging to a pool) * | Library or Pool Volume (ul) * | Library or Pool Molarity (nM) * |
|----------------|------------------------------------|---------------------|---------------------|--|-------------------------------|---------------------------------|
| R13R           | Homo sapiens                       | CCTTGATC            | ACTCCATC            | pool6  | 29                            |                                 |
| R18R           | Homo sapiens                       | GTGAAGTG            | GATTGCTC            | pool6  | 29                            |                                 |
| R36R           | Homo sapiens                       | ATGCCTGT            | CGCAATCT            | pool6  | 29                            |                                 |
| R36C           | Homo sapiens                       | CAACACCT            | TGGTAGCT            | pool6  | 29                            |                                 |

As illustrated in the Figure above, the field *“Pool name (mandatory for each library belonging to a pool)”* has the value “pool6” (which would be the name in the tube), indicating that all these libraries will be grouped into this one pool. If you add multiple libraries with various pool names, the system will generate a separate pool for each unique pool name. Lastly, if your sample list includes libraries with specified pool names and others where the “Pool name (mandatory for each library belonging to a pool)” field is left empty, the system will create pools for those with pool names while treating the others as single/individual libraries.

|   |                          |                   |                 |             |                                 |
|---|--------------------------|-------------------|-----------------|-------------|---------------------------------|
|  | <b>Work instruction</b>  | <b>Code</b>       | <b>Revision</b> | <b>Date</b> | <b>Owner</b>                    |
|   | <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1               | 12/02/2025  | Genomics Unit<br>Page: 14 of 19 |

## 5. Quick Agendo Sample Submission Guide

| NGS complementary services: Automated electrophoresis |  |
|---|--|
| Sample field  | Description  |
| Sample No.  | Automatically generated by Agendo  |
| Code  | Automatically generated by Agendo  |
| Plate Number  | It allows only numbers. Starting from 1 to indicate plate 1 up to 3 plates   |
| Well Position   | A01 to H12 by columns. Positions D12 and H12 must remain empty.  |
| Sample Name (same as in tube)                         | Between 1 and 4 alphanumeric characters, underscore included (e.g. TE_1)   |
| Sample Type   | Choose 1 of the following (spelling must match exactly): gDNA, fragmented DNA, cDNA, PCR amplicons, Total RNA, polyA RNA, rRNA-depleted RNA, smallRNA, WG Library, Exome-Capture Library, Amplicon Library, mRNA Library, rRNA-Ribodepleted Library, smallRNA Library, ChIP Library, ATAC Library, Other |
| Sample Species Name (Use NCBI taxonomy)               | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota)  |
| Concentration (ng/ul)                                 | Integer or decimal with . as separator   |
| Volume (ul)   | Integer or decimal with . as separator   |
| Expected size (bp) for libraries                      | Integer  |
| Observations  | Any desired comment about the sample   |

| NGS complementary services: Covaris |  |
|-------------------------------------|--|
| Sample field                        | Description  |
| Sample No.                          | Automatically generated by Agendo  |
| Code                                | Automatically generated by Agendo  |
| Sample Name (same as in tube)       | Between 1 and 4 alphanumeric characters, underscore included (e.g. TE_1) |
| Desired fragment size (bp)          | Base Pairs (bp)  |
| Volume (ul)                         | Integer or decimal with . as separator                                   |
| Duty Factor (%)                     | Completed by the Genomics Unit   |
| Peak Power (W)                      | Completed by the Genomics Unit   |
| Cycles / burst                      | Completed by the Genomics Unit   |
| Time (s)                            | Completed by the Genomics Unit   |
| Observations                        | Any desired comment about the sample                                     |

| Work instruction         | Code              | Revision | Date       | Owner          |
|--------------------------|-------------------|----------|------------|----------------|
| <b>Agendo User Guide</b> | <b>GEN-MG-002</b> | 1        | 12/02/2025 | Genomics Unit  |
|                          |                   |          |            | Page: 15 of 19 |

### NGS Library Prep and Sequencing: Whole and Targeted Genome

| Sample field                            | Description   |
|---|---|
| Sample No.                              | Automatically generated by Agendo   |
| Code                                    | Automatically generated by Agendo   |
| Sample Name                             | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type                             | Choose 1 of the following (spelling must match exactly): cDNA,cfDNA,Cloned DNA,ctDNA,gDNA,WGA-DNA,PCR amplicons,Other                                     |
| Sample Species Name (Use NCBI taxonomy) | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)                             | Integer or decimal with . as separator  |
| Concentration (ng/ul)                   | Integer or decimal with . as separator  |
| Nanodrop 260/280                        | Integer or decimal with . as separator  |
| Nanodrop 260/230                        | Integer or decimal with . as separator  |
| Sample Replacement                      | Is it a replacement of a previous sample? yes or no   |
| Observations                            | Any desired comment about the sample  |

### NGS Library Prep and Sequencing: Transcriptome

| Sample field                            | Description   |
|---|---|
| Sample No.                              | Automatically generated by Agendo   |
| Code                                    | Automatically generated by Agendo   |
| Sample Name                             | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type                             | Choose 1 of the following (spelling must match exactly): cfRNA,nuclearRNA,polyA RNA,rRNA-depleted RNA,smallRNA,Total RNA,Other                            |
| Sample Species Name (Use NCBI taxonomy) | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)                             | Integer or decimal with . as separator  |
| Concentration (ng/ul)                   | Integer or decimal with . as separator  |
| Nanodrop 260/280                        | Integer or decimal with . as separator  |
| Nanodrop 260/230                        | Integer or decimal with . as separator  |
| Sample Replacement                      | Is it a replacement of a previous sample? yes or no   |
| Observations                            | Any desired comment about the sample  |

| Work instruction         | Code       | Revision | Date       | Owner          |
|--------------------------|------------|----------|------------|----------------|
| <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|                          |            |          |            | Page: 16 of 19 |

| NGS Library Prep and Sequencing: Epigenome |   |
|--|---|
| Sample field                               | Description   |
| Sample No.                                 | Automatically generated by Agendo   |
| Code                                       | Automatically generated by Agendo   |
| Sample Name                                | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type                                | Choose 1 of the following (spelling must match exactly): CHIP DNA, Cloned DNA, ctDNA, CUT&RUN DNA, gDNA, HiC DNA, WGA-DNA, PCR amplicons, Other           |
| Sample Species Name (Use NCBI taxonomy)    | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)                                | Integer or decimal with . as separator  |
| Concentration (ng/ul)                      | Integer or decimal with . as separator  |
| Nanodrop 260/280                           | Integer or decimal with . as separator  |
| Nanodrop 260/230                           | Integer or decimal with . as separator  |
| Sample Replacement                         | Is it a replacement of a previous sample? yes or no   |
| Observations                               | Any desired comment about the sample  |

| NGS Libraries Ready for Sequencing                         |   |
|--|---|
| Sample field   | Description   |
| Sample No.   | Automatically generated by Agendo   |
| Code   | Automatically generated by Agendo   |
| Library Name   | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Species Name (Use NCBI taxonomy)                           | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Sequence index i7  | i7 index sequence, at least 6bp long, must only contain A,C,G,T; in case the library only has an i5 index the i7 sequence must be XXXXXX                  |
| Sequence index i5  | i5 index sequence, at least 6bp long, must only contain A,C,G,T   |
| Pool name (mandatory for each library belonging to a pool) | Between 2 and 6 alphanumeric characters (i.e. letters, numbers and underscore)  |
| Library or Pool Volume (ul)                                | Integer or decimal with . as separator  |
| Library or Pool Molarity (nM)                              | Integer or decimal with . as separator  |
| Library or Pool Concentration (ng/ul)                      | Integer or decimal with . as separator  |
| Library or Pool Average Size (bp)                          | Integer   |
| Library Replacement  | Is it a replacement of a previous library? yes or no  |
| Observations   | Any desired comment about the library   |



| Work instruction         | Code       | Revision | Date       | Owner          |
|--------------------------|------------|----------|------------|----------------|
| <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|                          |            |          |            | Page: 17 of 19 |

| NGS Library Prep and Sequencing: Single Cell genomics and transcriptomics |   |
|---|---|
| Sample field  | Description   |
| Sample No.  | Automatically generated by Agendo   |
| Code  | Automatically generated by Agendo   |
| Sample Name   | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type   | Choose 1 of the following (spelling must match exactly): Cell pellet, Cell suspension, Cells in lysis buffer, Frozen tissue, Isolated nuclei, cDNA, Other |
| Sample Species Name (Use NCBI taxonomy)                                   | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)   | Integer or decimal with . as separator  |
| Cell or Nuclei Concentration (cells/ul or nuclei/ul)                      | Integer or decimal with . as separator  |
| Target Recovery   | Integer or decimal with . as separator. For SMART-seq put 1.  |
| Low RNA Content (only for scGE)   | Yes or No   |
| Sample Replacement  | Is it a replacement of a previous sample? yes or no   |
| Observations  | Any desired comment about the sample  |

| Work instruction         | Code       | Revision | Date       | Owner          |
|--------------------------|------------|----------|------------|----------------|
| <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|                          |            |          |            | Page: 18 of 19 |

| NGS Library Prep and Sequencing: Plates - DNA |   |
|---|---|
| Sample field                                  | Description   |
| Sample No.                                    | Automatically generated by Agendo   |
| Code  | Automatically generated by Agendo   |
| Plate Number                                  | It allows only numbers. Starting from 1 to indicate plate 1 up to 3 plates  |
| Plate Name                                    | Between 2 and 50 alphanumeric characters, underscore included (e.g. PL_123)   |
| Well Position                                 | A01 to H12 by columns. Positions D12 and H12 must remain empty.   |
| Sample Name                                   | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type                                   | Choose 1 of the following (spelling must match exactly): cDNA,cfDNA,ChIP DNA,Cloned DNA,ctDNA,CUT&RUN DNA,gDNA,HiC DNA,WGA-DNA,PCR amplicons,Other        |
| Sample Species Name (Use NCBI taxonomy)       | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)                                   | Integer or decimal with . as separator  |
| Concentration (ng/ul)                         | Integer or decimal with . as separator  |
| Nanodrop 260/280                              | Integer or decimal with . as separator  |
| Nanodrop 260/230                              | Integer or decimal with . as separator  |
| Sample Replacement                            | Is it a replacement of a previous sample? yes or no   |
| Observations                                  | Any desired comment about the sample  |

| Work instruction         | Code       | Revision | Date       | Owner          |
|--------------------------|------------|----------|------------|----------------|
| <b>Agendo User Guide</b> | GEN-MG-002 | 1        | 12/02/2025 | Genomics Unit  |
|                          |            |          |            | Page: 19 of 19 |

| NGS Library Prep and Sequencing: Plates - RNA |   |
|---|---|
| Sample field                                  | Description   |
| Sample No.                                    | Automatically generated by Agendo   |
| Code  | Automatically generated by Agendo   |
| Plate Number                                  | It allows only numbers. Starting from 1 to indicate plate 1 up to 3 plates  |
| Plate Name                                    | Between 2 and 50 alphanumeric characters, underscore included (e.g. PL_123)   |
| Well Position                                 | A01 to H12 by columns. Positions D12 and H12 must remain empty.   |
| Sample Name                                   | Between 2 and 15 alphanumeric characters, underscore included (e.g. TE_123)   |
| Sample Type                                   | Choose 1 of the following (spelling must match exactly): Total RNA,Other  |
| Sample Species Name (Use NCBI taxonomy)       | Must start with a capital letter word, might be followed by more words, separated by spaces from each other (e.g. Homo sapiens, Mus musculus, Microbiota) |
| Volume (ul)                                   | Integer or decimal with . as separator  |
| Concentration (ng/ul)                         | Integer or decimal with . as separator  |
| Nanodrop 260/280                              | Integer or decimal with . as separator  |
| Nanodrop 260/230                              | Integer or decimal with . as separator  |
| Sample Replacement                            | Is it a replacement of a previous sample? yes or no   |
| Observations                                  | Any desired comment about the sample  |