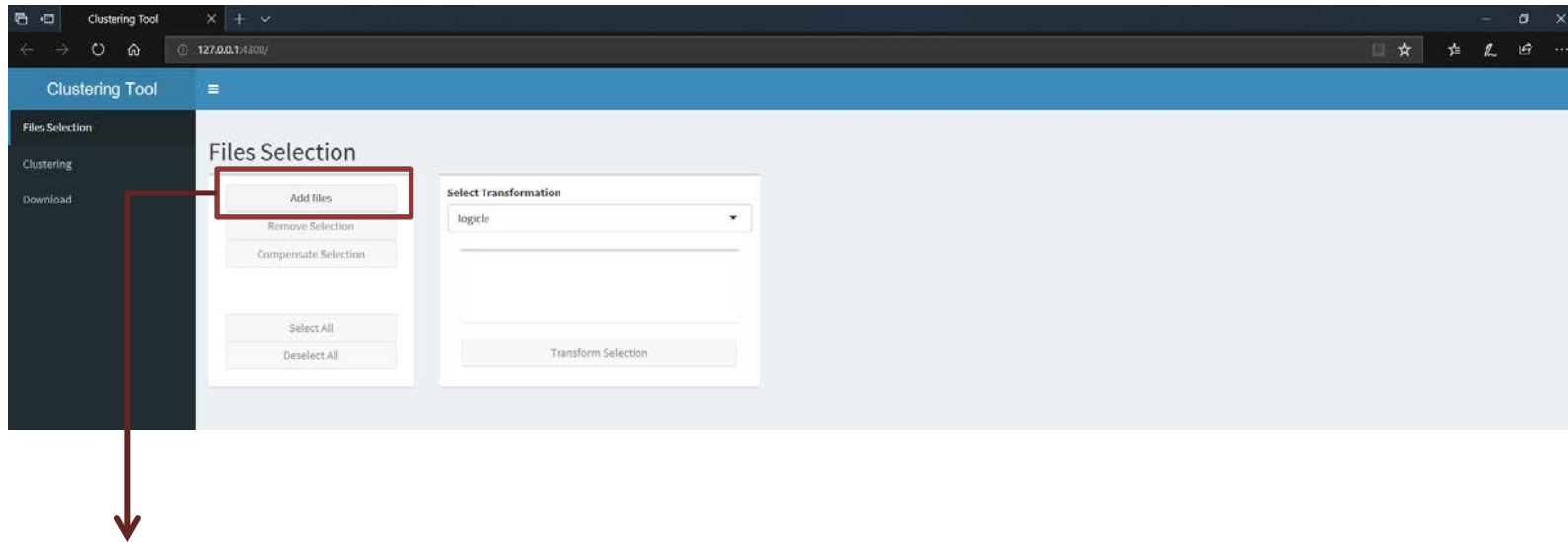


Clustering Tool

Manual

Files Selection Tab

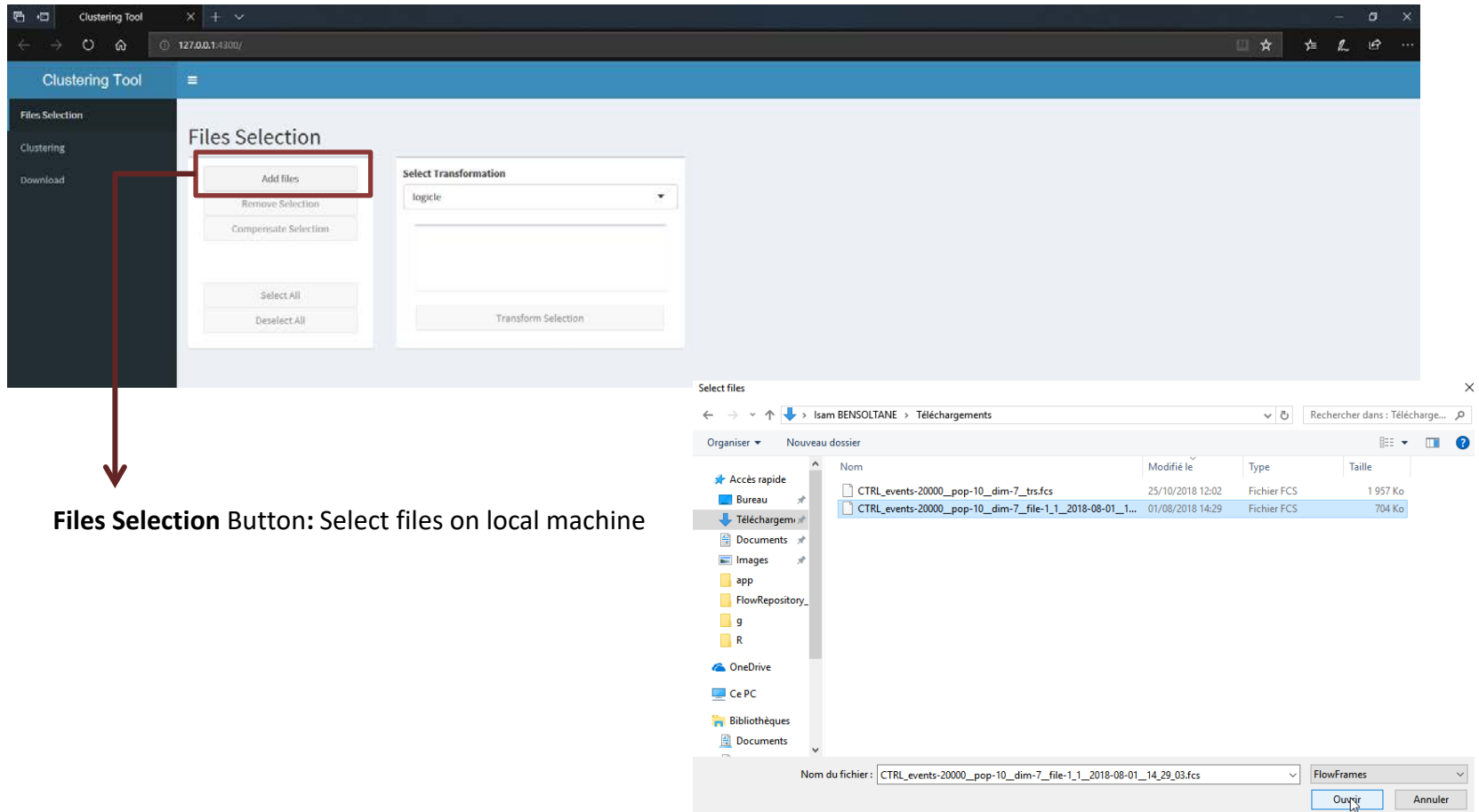
Files Selection, Markers Selection, Downsampling, Compensation, Transformation



Files Selection Button: Select files on local machine

Files Selection Tab

Files Selection, Markers Selection, Downsampling, Compensation, Transformation

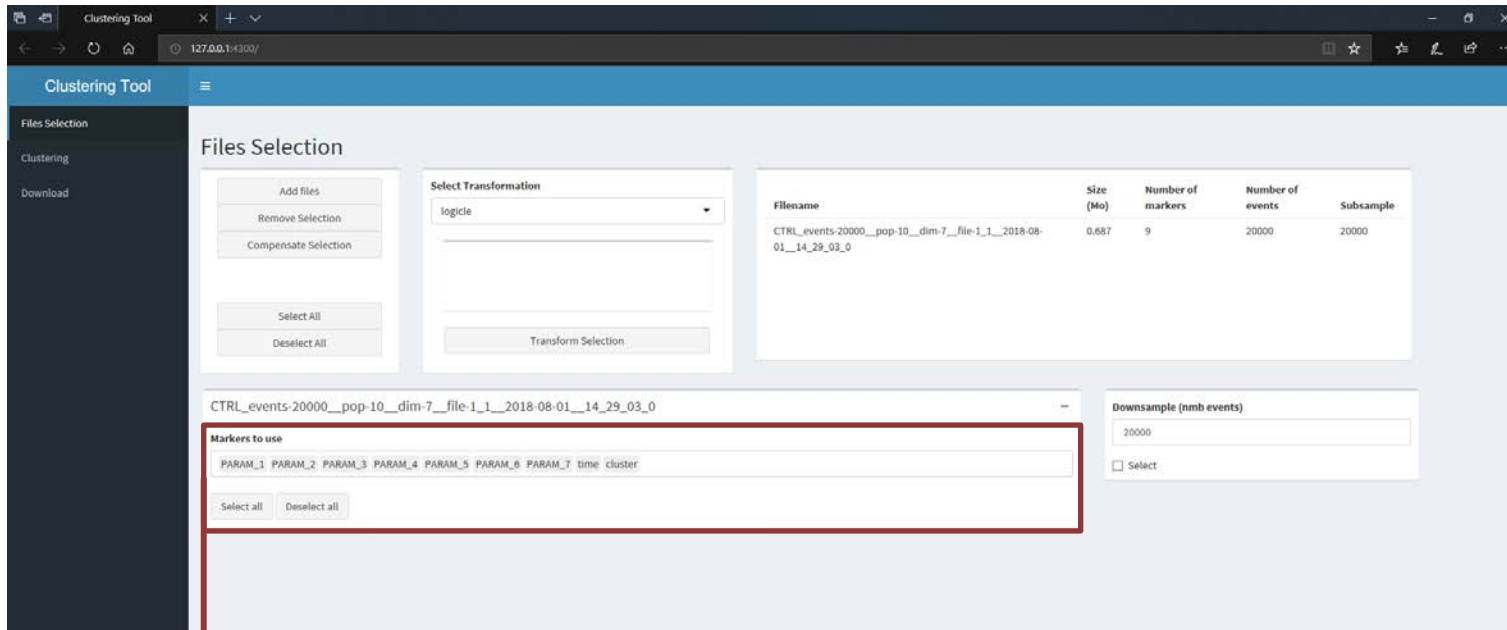


The screenshot displays the Clustering Tool interface. On the left, a sidebar contains the 'Files Selection' tab, which is highlighted. A red arrow points from the 'Add files' button in the 'Files Selection' panel to a file selection dialog. The dialog shows a list of files in the 'Téléchargements' folder. The file 'CTRL_events-20000_pop-10_dim-7_file-1_1_2018-08-01_1...' is selected. The dialog also shows the file's name, type (Fichier FCS), and size (704 Ko).

Files Selection Button: Select files on local machine

Files Selection Tab

Files Selection, **Markers Selection**, Downsampling, Compensation, Transformation



Clustering Tool

Files Selection

Add files
Remove Selection
Compensate Selection

Select All
Deselect All

Select Transformation
logicle

Transform Selection

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0	0.687	9	20000	20000

CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0

Markers to use

PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster

Select all Deselect all

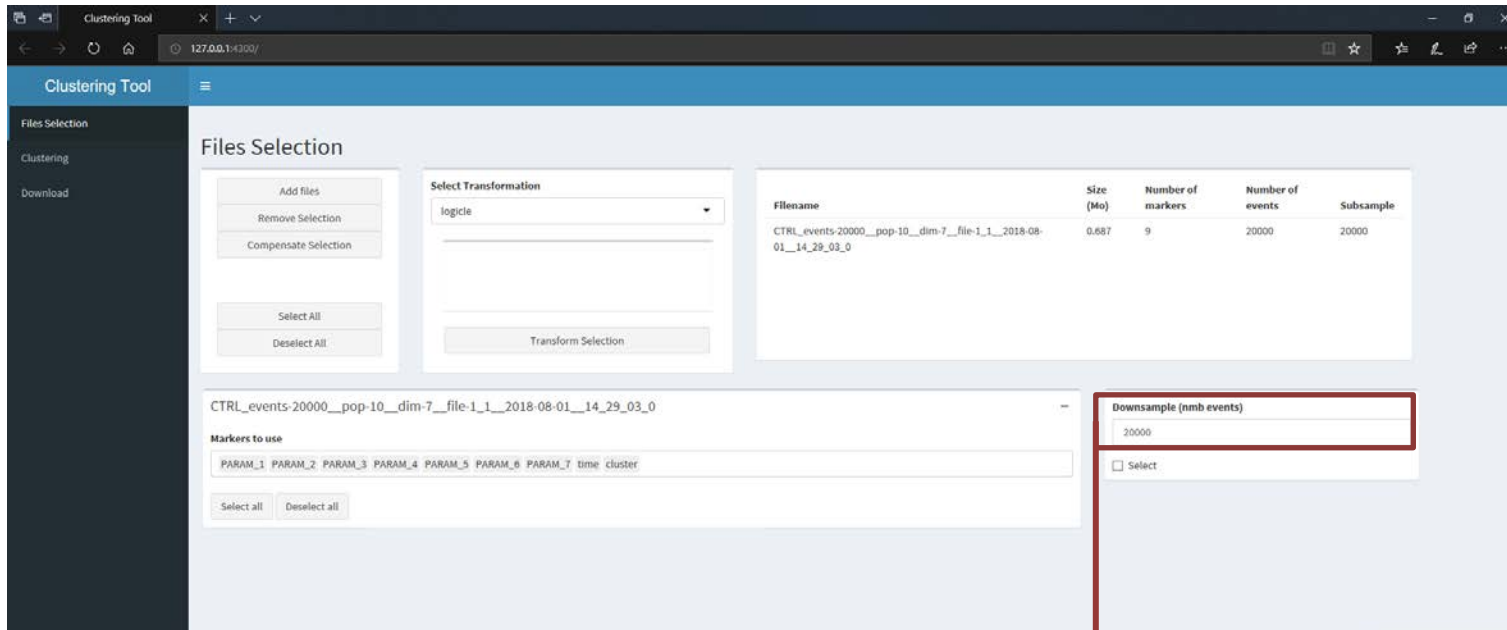
Downsample (nmb events)
20000
☐ Select

Select the markers to use for Compensation, Transformation & Clustering:

- Individually (list)
- **Select/Deselect all** buttons

Files Selection Tab

Files Selection, **Markers Selection**, **Downsampling**, Compensation, Transformation



The screenshot shows the 'Files Selection' tab in the 'Clustering Tool' interface. The interface includes a sidebar with 'Files Selection', 'Clustering', and 'Download' options. The main area is titled 'Files Selection' and contains several controls:

- File Management:** Buttons for 'Add files', 'Remove Selection', 'Compensate Selection', 'Select All', and 'Deselect All'.
- Transformation:** A 'Select Transformation' dropdown menu currently set to 'logicle', with a 'Transform Selection' button below it.
- File List:** A table displaying file details:

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0	0.687	9	20000	20000
- Markers Selection:** A section titled 'Markers to use' with a text input field containing 'PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster' and 'Select all'/'Deselect all' buttons.
- Downsampling:** A 'Downsample (nmb events)' input field with the value '20000' and a 'Select' checkbox below it.

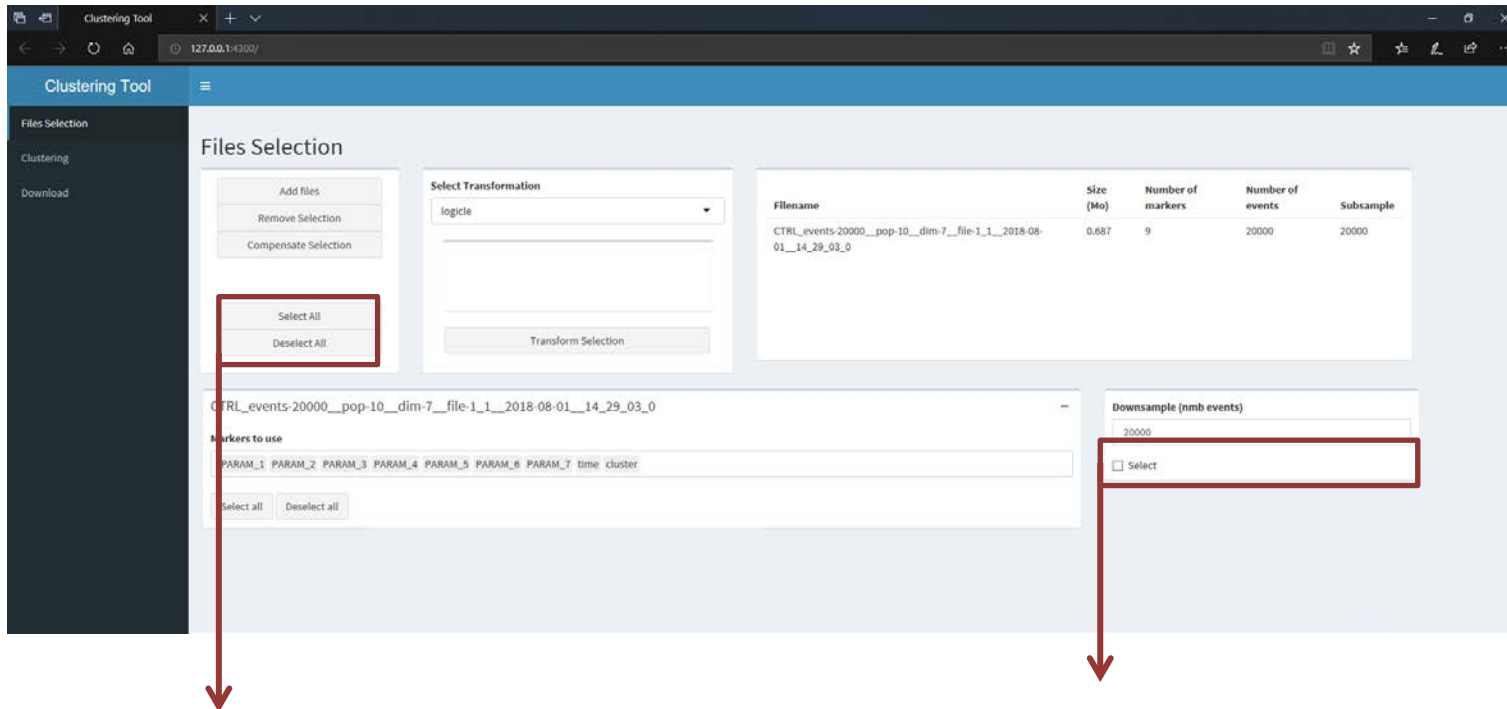
A red arrow points from the 'Downsample (nmb events)' input field to the text below.

Choose the number of events to use when clustering:

- By default: all the events within the file
- If downsampled, the output fcs contains only the selected events

Files Selection Tab

Files Selection, **Markers Selection**, **Downsampling**, Compensation, Transformation



Clustering Tool

Files Selection

Add files
Remove Selection
Compensate Selection

Select All
Deselect All

Select Transformation
logicle

Transform Selection

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0	0.687	9	20000	20000

CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0

Markers to use

PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster

Select all Deselect all

Downsample (nmb events)

20000

Select

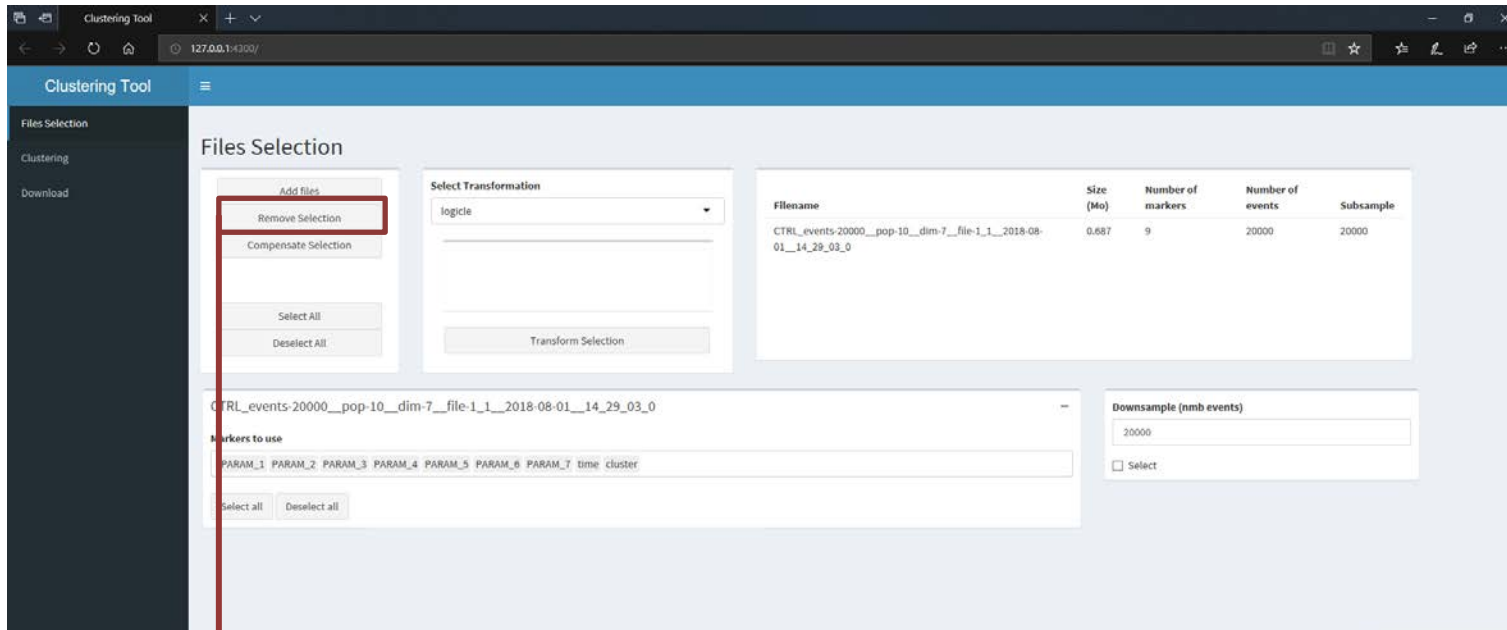
Select the file for Compensation or Transformation:

- Individually (checkbox)
- **Select/Deselect all** buttons

ALL THE FILES ARE USED IN THE CLUSTERING TAB

Files Selection Tab

Files Selection, **Markers Selection**, Downsampling, Compensation, Transformation



The screenshot shows the 'Clustering Tool' interface with the 'Files Selection' tab active. The interface includes a sidebar with 'Files Selection', 'Clustering', and 'Download' options. The main content area has a 'Files Selection' section with buttons for 'Add Files', 'Remove Selection', 'Compensate Selection', 'Select All', and 'Deselect All'. A 'Select Transformation' dropdown is set to 'logicle', with a 'Transform Selection' button below it. A table displays file information:

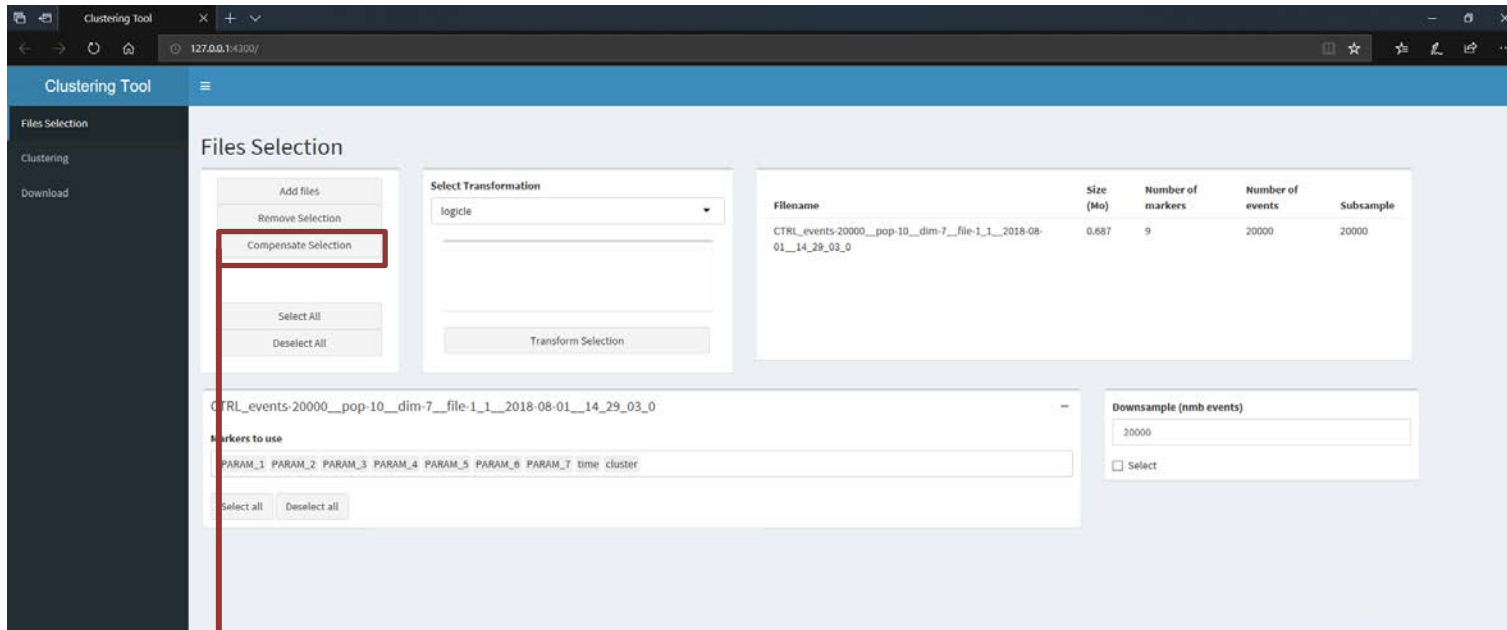
Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0	0.687	9	20000	20000

Below the table, the filename 'CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0' is listed. A 'Markers to use' section contains a text input with 'PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster' and 'Select all'/'Deselect all' buttons. A 'Downsample (nmb events)' section has a text input with '20000' and a 'Select' checkbox.

Remove Selection Button: Remove the selected files

Files Selection Tab

Files Selection, Markers Selection, Downsampling, **Compensation**, Transformation



The screenshot shows the Clustering Tool interface with the Files Selection tab active. The interface includes a sidebar with 'Files Selection', 'Clustering', and 'Download' options. The main area contains several sections: 'Add files', 'Remove Selection', 'Compensate Selection' (highlighted with a red box), 'Select All', 'Deselect All', 'Select Transformation' (set to 'logicle'), 'Transform Selection', a table of selected files, 'Markers to use', and 'Downsample (nmb events)'.

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

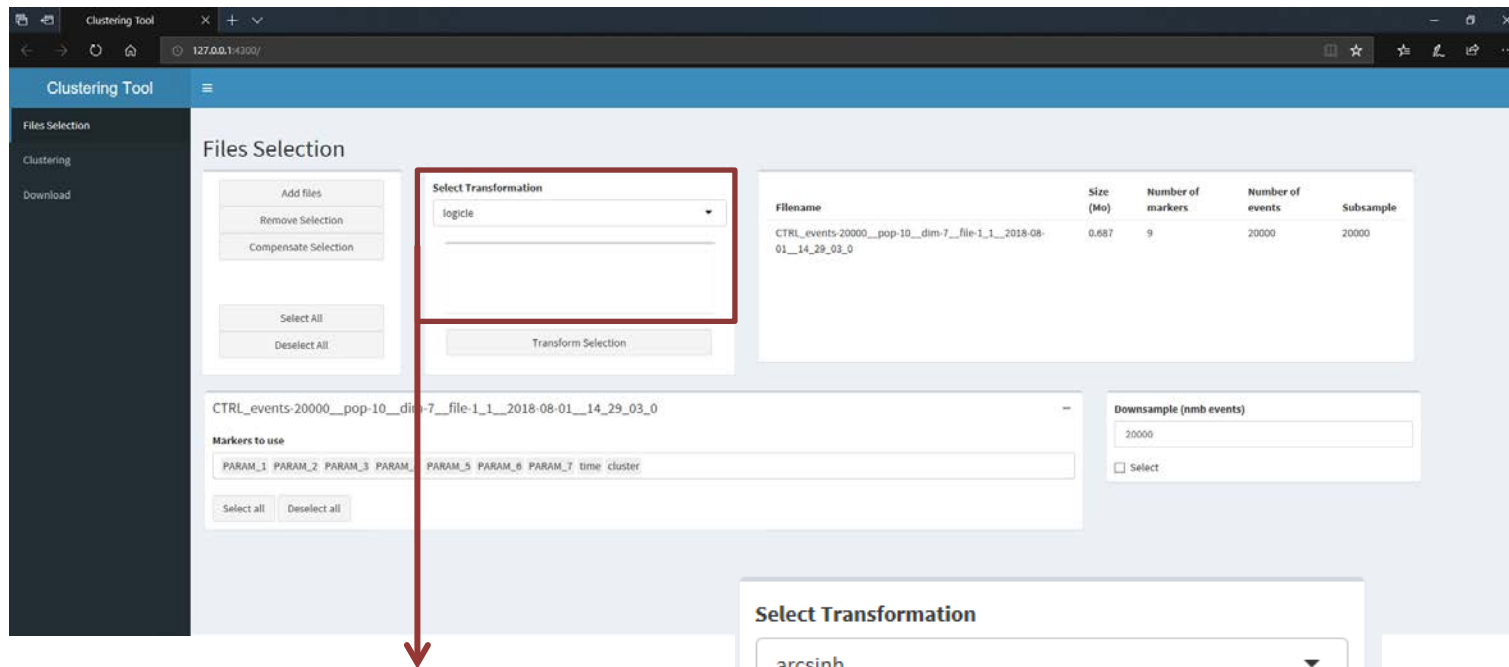
Markers to use: PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster

Downsample (nmb events): 20000

Compensate Selection Button: Compensate the selected files (if possible)

Files Selection Tab

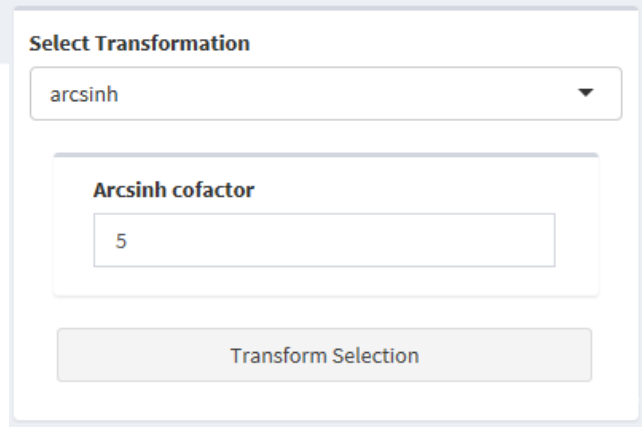
Files Selection, Markers Selection, Downsampling, Compensation, **Transformation**



The screenshot shows the 'Clustering Tool' interface. On the left is a sidebar with 'Files Selection', 'Clustering', and 'Download'. The main area is titled 'Files Selection'. It contains several buttons: 'Add files', 'Remove Selection', 'Compensate Selection', 'Select All', and 'Deselect All'. A 'Select Transformation' dropdown menu is highlighted with a red box and labeled 'logicle'. Below it is a 'Transform Selection' button. To the right is a table with columns: 'Filename', 'Size (Mo)', 'Number of markers', 'Number of events', and 'Subsample'. The table contains one row: 'CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0', '0.687', '9', '20000', and '20000'. Below the table is a 'Markers to use' section with a list of parameters: 'PARAM_1', 'PARAM_2', 'PARAM_3', 'PARAM_4', 'PARAM_5', 'PARAM_6', 'PARAM_7', 'time', and 'cluster'. There are 'Select all' and 'Deselect all' buttons. To the right of the markers section is a 'Downsample (nmb events)' input field with '20000' and a 'Select' checkbox.

Select Transformation:

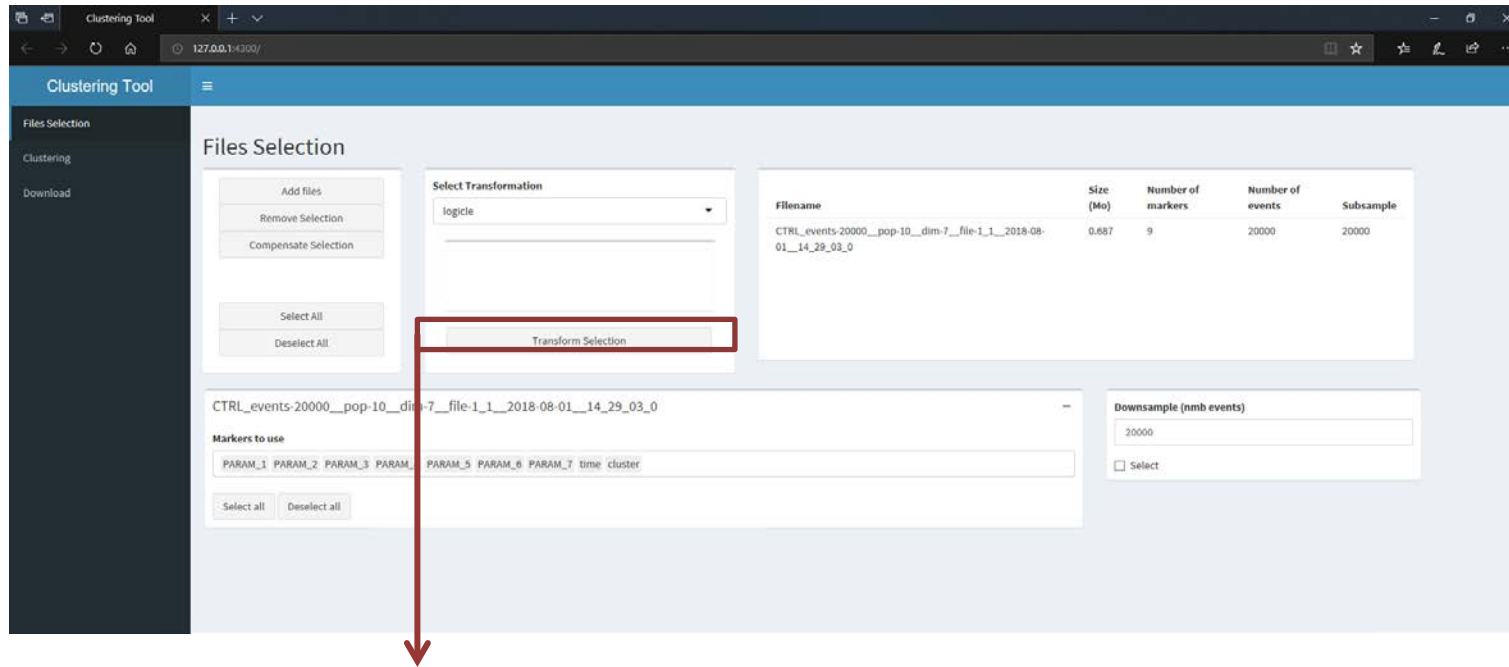
- Logicle (no parameters)
- Arcsinh (cofactor)



The 'Select Transformation' dialog for 'Arcsinh' shows a dropdown menu with 'arsinh' selected. Below it is a section titled 'Arcsinh cofactor' with an input field containing the value '5'. At the bottom is a 'Transform Selection' button.

Files Selection Tab

Files Selection, Markers Selection, Downsampling, Compensation, **Transformation**

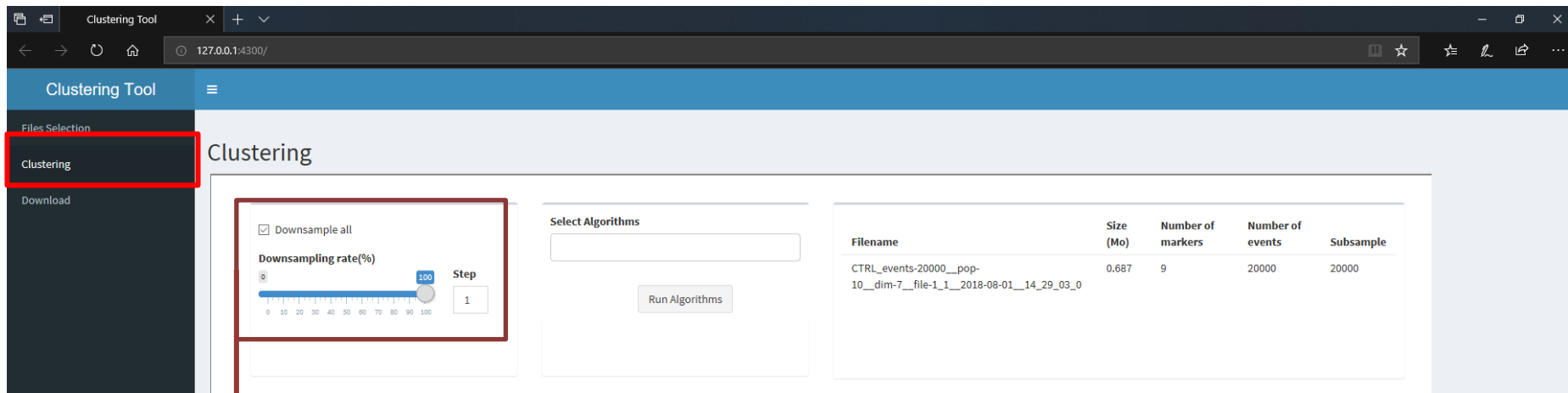


The screenshot shows the Clustering Tool interface with the Files Selection tab active. The interface includes a sidebar with 'Files Selection', 'Clustering', and 'Download' options. The main content area displays the 'Files Selection' section with buttons for 'Add files', 'Remove Selection', 'Compensate Selection', 'Select All', and 'Deselect All'. A 'Select Transformation' dropdown menu is set to 'logicle'. A table lists files with columns for 'Filename', 'Size (Mo)', 'Number of markers', 'Number of events', and 'Subsample'. The 'Transform Selection' button is highlighted with a red box, and a red arrow points from it to the text 'Transform Selection Button' below the screenshot.

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01_14_29_03_0	0.687	9	20000	20000

Transform Selection Button

Clustering Tab



Clustering Tool

Files Selection

Clustering

Download

Clustering

☒ Downsample all

Downsampling rate(%)

0 100 Step 1

Select Algorithms

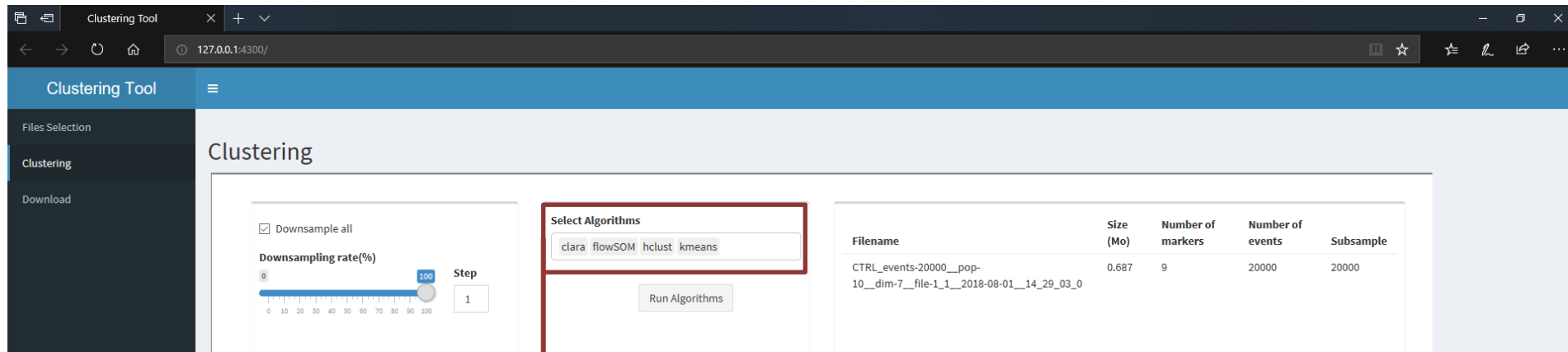
Run Algorithms

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

Downsample all checkbox :

- Checked: Use the selected downsampling rate on all files (% of the events of each file)
- Unchecked: Use the individual downsampling rate chosen in the *Files Selection Tab*

Clustering Tab



The screenshot shows the Clustering Tool interface. On the left is a sidebar with 'Files Selection', 'Clustering', and 'Download' tabs. The main area is titled 'Clustering'. It contains a 'Downsample all' checkbox, a 'Downsampling rate(%)' slider (set to 100), and a 'Step' dropdown (set to 1). A red box highlights the 'Select Algorithms' section, which contains a text input field with 'clara flowSOM hclust kmeans' and a 'Run Algorithms' button. A red arrow points from this box down to the 'Select Algorithms:' text below. To the right of the 'Select Algorithms' section is a table with the following data:

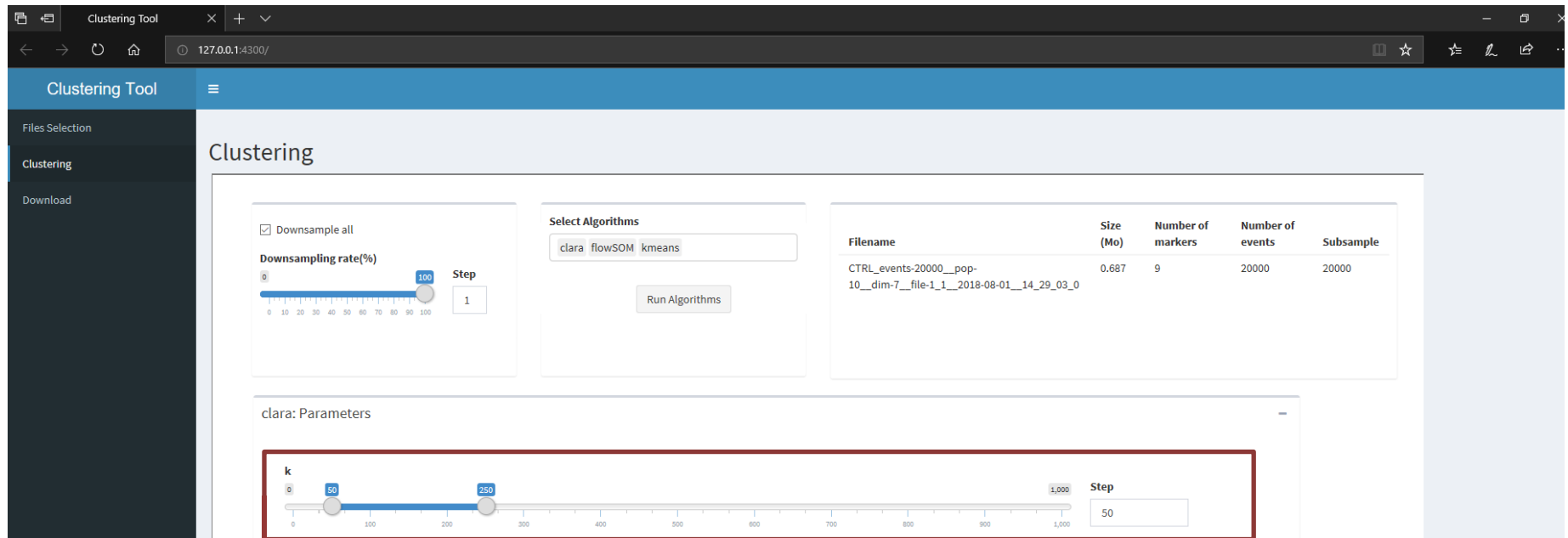
Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

Select Algorithms:

Currently:

- Clara
- flowSOM
- Hclust **LESS THAN 40 000 EVENTS ONLY**
- Kmeans

Clustering Tab



The screenshot shows the Clustering Tool interface. On the left is a sidebar with 'Files Selection', 'Clustering', and 'Download'. The main area is titled 'Clustering'. It contains three panels:

- Downsampling all:** A checkbox is checked. Below it is a slider for 'Downsampling rate(%)' ranging from 0 to 100, with a 'Step' input set to 1.
- Select Algorithms:** A text box contains 'clara flowSOM kmeans' and a 'Run Algorithms' button.
- Filename Table:**

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10_dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

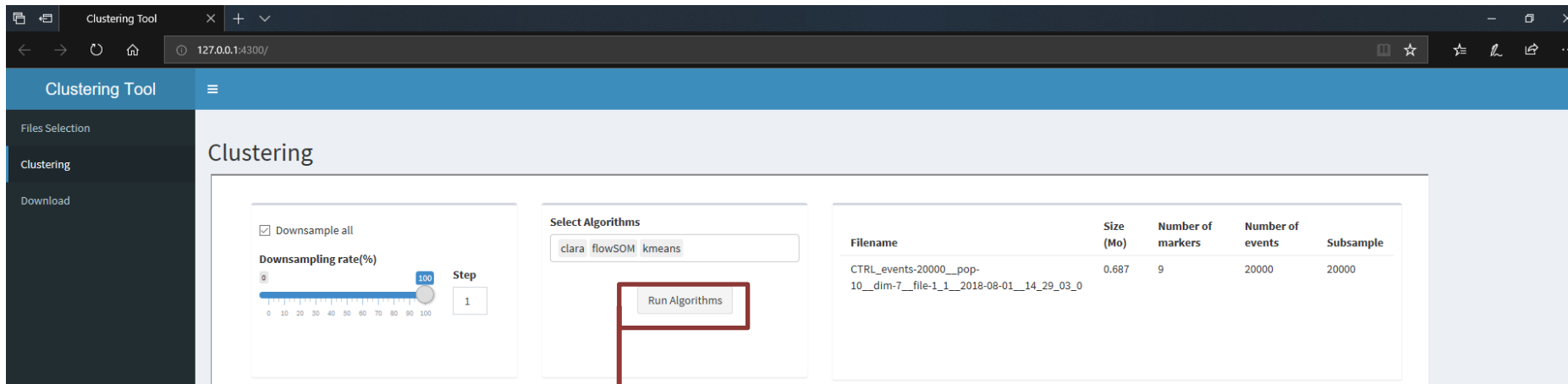
Below these panels is a section for 'clara: Parameters' which includes a slider for 'k' ranging from 0 to 1,000, with markers at 50 and 250, and a 'Step' input set to 50. A red box highlights the 'k' slider and its step value.

For each algorithm, select the range of values for all parameters:

- Sliders: Min and Max values for the parameters
- Step: Values step between 2 runs

eg: Min=50, Max=150, STEP=50
 ➔ K = 50,100,150

Clustering Tab



Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

Run the algorithms with the combinations of all the chosen values

Eg: K-means centers=(50,100,150), Iterations=(1000,1100,1200)

→ Runs:

k	Iterations
50	1000
100	1000
150	1000
50	1100
100	1100
...	...



Download Tab

Clustering Tool

Download Files

Select Transformation

logicle

Decompensate Selection

Select All

Deselect All

Download Enriched Files

Filename	Size (Mo)	Number of markers	Number of events	Subsample
CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0	0.687	9	20000	20000

CTRL_events-20000__pop-10__dim-7__file-1_1__2018-08-01__14_29_03_0

Markers to use

PARAM_1 PARAM_2 PARAM_3 PARAM_4 PARAM_5 PARAM_6 PARAM_7 time cluster cluster_K-Means.10 cluster_K-Means.11 cluster_K-Means.12

Select all Deselect all

Select

Download all the generated fcs: original fcs + clustering columns added

!! Reverse Transformation, Decompensate FIRST!!