

## PiVAT 2020.1 CNV Analysis User Guide:

1. Set SA (Secondary Analysis) Parameters. (This step is optional.)

**Secondary Analysis Parameters**

SA Parameters Name

Panel Selection

Save SA Parameters

Saves a new SA Parameters or update the placeholder you can ignore the

1 Secondary Analysis Parameters 2 Post Filter Parameters 3 QC Parameters

**CORE\_MODULE** Local Realignment Paired End Assembly Bam Export

Read To Position Variant Call Reduce Annotation/Filtering

Parameter	Value
UID_OPTION	NONE
VCF_FORMAT_FIELD_VER	True
LD_XAMPLICON_CROSSING	False
WRITE_DEBUG_EXCEL_FILES	False

a. Click on the SA Parameters button

b. Define SA Parameters Name

c. Choose the panel name from the dropdown menu

d. Change the analysis parameters if needed.

e. Save SA parameters for future runs with the same panel and parameters

2. After clicking on the “Save SA Parameters” button, click on the “Go To Start An Analysis” button in the pop out window.

The screenshot shows the Pillar Biosciences web interface for configuring Secondary Analysis Parameters. The left sidebar contains navigation options: Home, Secondary Analysis (expanded), SA Parameters (selected), Start Analysis, Output & Visualizations, Data Management, Tools, and Help. The main content area is titled "Secondary Analysis Parameters" and includes a "Save SA Parameters" button. Below this, the "SA Parameters Name" field contains "CNV\_test" with a warning message: "Custom preset name already exists. Please choose another one." The "Panel Selection" dropdown is set to "BRCA FFPE with CNV (BR283)". A modal dialog box is displayed in the center, indicating a success message: "SA parameters file CNV\_test has been successfully created." with "Cancel" and "Go To Start An Analysis" buttons. Below the dialog, a table lists parameters and their values:

Parameter	Value
UID_OPTION	NONE
VCF_FORMAT_FIELD_VER	True
LD_XAMPLICON_CROSSING	False
WRITE_DEBUG_EXCEL_FILES	False

### 3. Start analysis from fastq files.

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4. Pick the fastq files.

Start Analysis Step2: Select Analysis Fastq Files

Sample Type: Sample Panel: - SA Parameters File:

Home / input / user

a. Pick the folder that contains the fastq files

You haven't selected any files yet.

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Start Analysis Step2: Select Analysis Fastq Files

Sample Type: Sample Panel: - SA Parameters File:

Home / input / pbs / 20200605\_M03294\_0425\_000000000-J4M6K\_BRCA\_FFPE\_CORRECTED /

b. Pick the panel name from the dropdown menu.

d. Click on "Next" button.

c. SA Parameters File as defined in step 1.

a. Pick the fastq files.

Definition Name	Size	Modified Date
<input type="checkbox"/> BD4_S3_L001_R1_001.fastq.gz		
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-BD4_S3_L001_R2_001.fastq.gz	164.59 MB	Jun 8, 2020 3:48 PM
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-BD6_S4_L001_R1_001.fastq.gz		
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-BD6_S4_L001_R2_001.fastq.gz	100.56 MB	Jun 8, 2020 3:48 PM
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-CPMB1_S5_L001_R1_001.fastq.gz	120.73 MB	Jun 8, 2020 3:48 PM
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-CPMB1_S5_L001_R2_001.fastq.gz	124.1 MB	Jun 8, 2020 3:48 PM
<input checked="" type="checkbox"/> RDvSMpBRCAFFPEgCNVd200605IM1-CPMB45_S6_L001_R1_001.fastq.gz	98.88 MB	Jun 8, 2020 3:48 PM

## 5. Define Sample Type.

For CNV analysis, it is recommended to provide 3-5 in-run negative control samples. Follow the steps below to define the sample types.

Panel: SA Parameters File

Panel :: SA Parameters File	Sample...	Sample Type	Number
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2
<input type="checkbox"/>	RDvSMpBR	Sample	2

\*Tips: To make batch operations on panel, SA parameter file, or sample files, make your selections on the sample items above.

Panel: SA Parameters File

Panel :: SA Parameters File	Sample...	Sample Type	Number
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2
<input checked="" type="checkbox"/>	RDvSMpBR	Sample	2

\*Tips: To make batch operations on panel, SA parameter file, or sample files, make your selections on the sample items above.

SA Parameters File

BRCA FFPE with CNV (BR283) CNV\_test

Edit Files Ignore

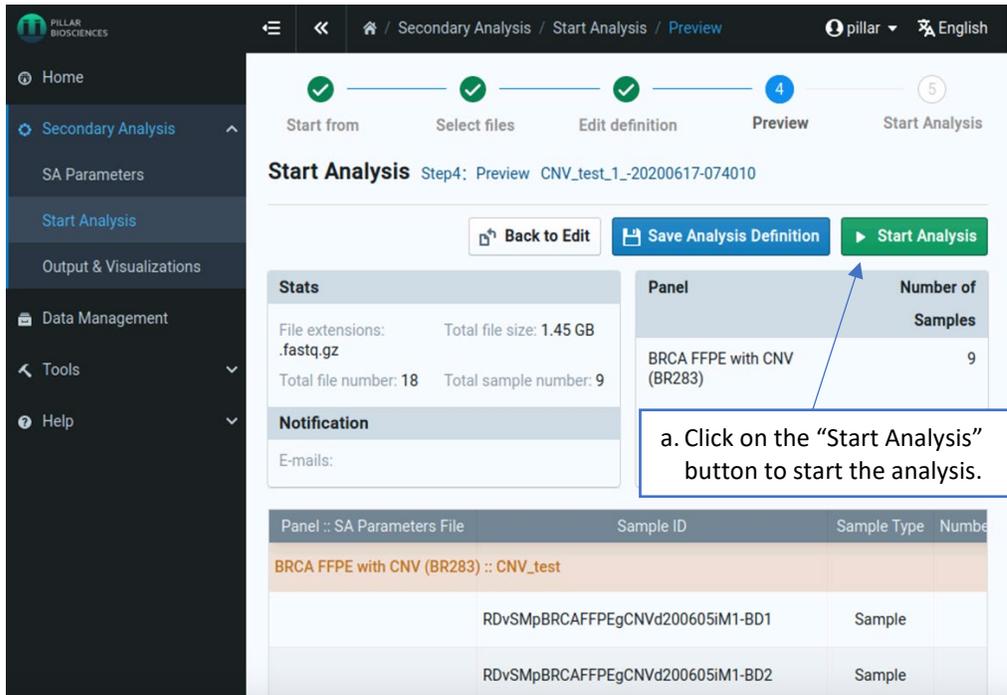
d. After the defined the sample types, click on the “Next” button to go to “Preview” step.

The screenshot displays the PILLAR BIOSCIENCES web application interface. The browser address bar shows `pivat.pillar.bio/secondary-analysis/launch/edit`. The navigation menu on the left includes Home, Secondary Analysis, SA Parameters, Start Analysis, Output & Visualizations, Data Management, Tools, and Help. The main content area shows a progress bar with five steps: Start from, Select files, Edit definition (Step 3), Preview, and Start Analysis. A green 'Next' button is visible, with a callout box pointing to it containing the text: "d. Click on the 'Next' button to go to the next step." Below the progress bar, the 'Start Analysis' section shows 'Step3: Edit Analysis Definition' with a text input field containing 'CNV\_test\_1\_' and a date '20200617-071516'. There is also a notification field with the text 'Please input E-mail addresses for notification'. Below this, there is a 'Show:' dropdown menu with options 'All', 'Active', and 'Insert Sample item'. The main table displays sample definitions for 'BRCA FFPE with CNV (BR283) :: CNV\_test'.

Sample ID	Sample Type	Number
<input checked="" type="checkbox"/> RDvSmpBRCAFFPEgCNVd200605iM1-PB09	NegCtrl	2
<input checked="" type="checkbox"/> RDvSmpBRCAFFPEgCNVd200605iM1-PB06	NegCtrl	2
<input checked="" type="checkbox"/> RDvSmpBRCAFFPEgCNVd200605iM1-BD6	NegCtrl	2
<input checked="" type="checkbox"/> RDvSmpBRCAFFPEgCNVd200605iM1-TV2	NegCtrl	2

6. Final review of the setting and start analysis.

- Review the details of the analysis definition. If everything looks fine, click on the “Start Analysis” button to start the analysis.
- You can also click on the “Save Analysis Definition” button to save the current analysis definition for rerun similar analysis in the future.



Start from Select files Edit definition **Preview** Start Analysis

**Start Analysis** Step4: Preview CNV\_test\_1\_-20200617-074010

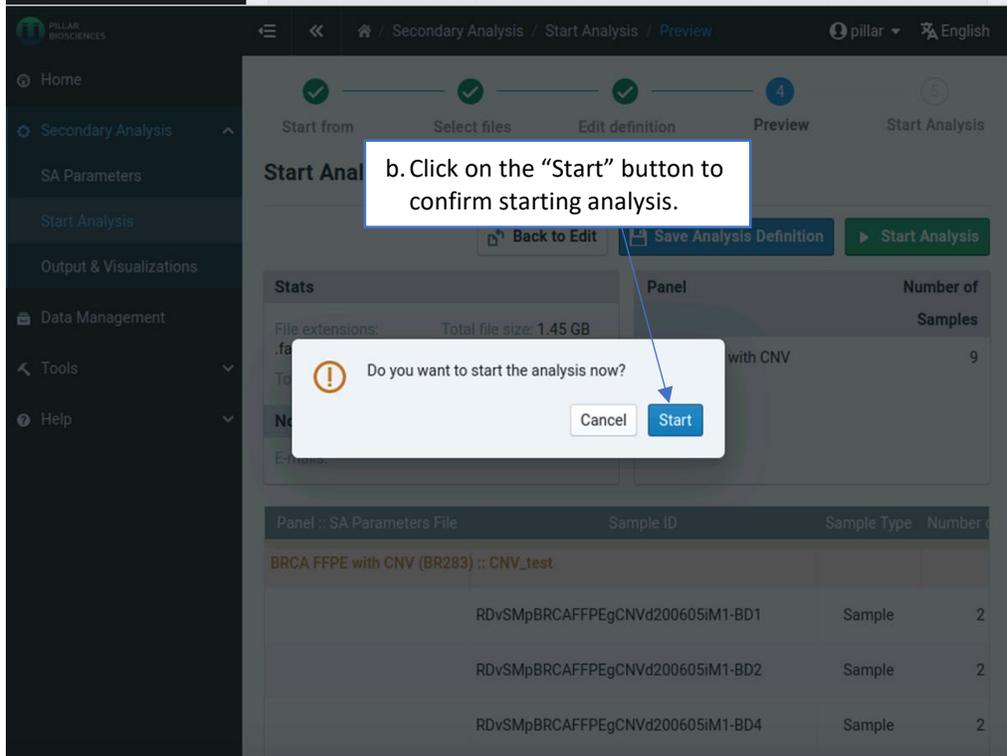
Back to Edit Save Analysis Definition **Start Analysis**

Stats	Panel	Number of Samples
File extensions: .fastq.gz	BRCA FFPE with CNV (BR283)	9
Total file size: 1.45 GB		
Total file number: 18		
Total sample number: 9		

Notification

E-mails:

Panel :: SA Parameters File	Sample ID	Sample Type	Number
<b>BRCA FFPE with CNV (BR283) :: CNV_test</b>			
	RDvSMpBRCAFFPEgCNVd200605iM1-BD1	Sample	
	RDvSMpBRCAFFPEgCNVd200605iM1-BD2	Sample	



Start from Select files Edit definition **Preview** Start Analysis

**Start Anal** b. Click on the “Start” button to confirm starting analysis.

Back to Edit Save Analysis Definition **Start Analysis**

Do you want to start the analysis now?

Cancel **Start**

Panel :: SA Parameters File	Sample ID	Sample Type	Number
<b>BRCA FFPE with CNV (BR283) :: CNV_test</b>			
	RDvSMpBRCAFFPEgCNVd200605iM1-BD1	Sample	2
	RDvSMpBRCAFFPEgCNVd200605iM1-BD2	Sample	2
	RDvSMpBRCAFFPEgCNVd200605iM1-BD4	Sample	2

- Right after the start, the task will be queued for a few seconds. Then you will see the progress bar in the task "Status" column, which indicate the task is running on PiVAT. Once the task is done, the "Status" will become "completed".

**Output and Visualizations** [View Task History and Result Status](#)

Search...

ID	Task	Panel	Date	Status	Operation
855	CNV_test_1_...	BRCA FFPE witl	Jun 17, 2020 7:44 AM	queued	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
854	CFTNA_linea...	cFTNA Fusion /	Jun 16, 2020 7:31 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
853	QC_BRCAV2_s...	BRCA-FFPE (BR'	Jun 14, 2020 9:46 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
852	QC_CF131spi...	ONCO/Reveal M	Jun 13, 2020 2:59 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
849	CFTNA_QCpoo...	cFTNA UID Ana:	Jun 11, 2020 1:44 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
848	BR283_CNV_T...	BRCA FFPE witl	Jun 10, 2020 7:14 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
847	20200608_BR...	BRCA FFPE witl	Jun 8, 2020 3:51 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
846	L81-Demo-20...	ONCOReveal Lur	Jun 4, 2020 3:21 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>

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**a. This indicates the current task is in the queue of PiVAT analysis.**

**Output and Visualizations** [View Task History and Result Status](#)

Search...

ID	Task	Panel	Date	Status	Operation
855	CNV_test_1_...	BRCA FFPE witl	Jun 17, 2020 7:44 AM	running	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
854	CFTNA_linea...	cFTNA Fusion /	Jun 16, 2020 7:31 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
853	QC_BRCAV2_s...	BRCA-FFPE (BR'	Jun 14, 2020 9:46 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
852	QC_CF131spi...	ONCO/Reveal M	Jun 13, 2020 2:59 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
848	BR283_CNV_T...	BRCA FFPE witl	Jun 10, 2020 7:14 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
847	20200608_BR...	BRCA FFPE witl	Jun 8, 2020 3:51 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>
846	L81-Demo-20...	ONCOReveal Lur	Jun 4, 2020 3:21 PM	completed	<input type="button" value="Abort"/> <input type="button" value="Rerun"/>

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**b. This indicates the current task is running on PiVAT. Once the task is done, the status will become "completed".**

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