



PathAI PD-L1 Multi-Clone NSCLC

Version 1.0

REF 20025001

User Manual





NOTE: PathAI PD-L1 Multi-Clone NSCLC and **navify**[®] Digital Pathology software are intended for Research Use Only. Not for use in diagnostic procedures. Access is intended for authorized users only, contact your institution for access approval.

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Chapter 1: Introduction

This chapter includes:

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Product Description

The PathAI PD-L1 Multi-Clone NSCLC is a software tool meant to identify and enumerate cells within tissue stained with the following PD-L1 staining assays 22C3, 28-8, SP142 and SP263 and is intended for Research Use Only. Not for use in diagnostic procedures:

Algorithm	Manufacturer
PathAI PD-L1 Multi-Clone NSCLC RUO	<p>The PathAI PD-L1 Multi-Clone NSCLC algorithm is a software tool intended for identification and quantification of tumor cells, immune cells, and tissue regions within whole slide images (WSI) of formalin-fixed, paraffin-embedded (FFPE) Non-Small Cell Lung Cancer (NSCLC) tissue stained with PD-L1 (22C3, 28-8, SP142 or SP263 clone) and scanned on a VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner.</p> <p>The PathAI PD-L1 Multi-Clone NSCLC algorithm is intended for Research Use Only. Not for use in diagnostic procedures.</p>

Supported Software





The PathAI PD-L1 Multi-Clone NSCLC algorithm runs on Roche **navify**® Digital Pathology, Version 2.5. This PathAI user manual applies to the PathAI PD-L1 Multi-Clone NSCLC algorithm.

About This User Manual

The user manual provides details about the slide and scanner requirements, image analysis workflow, and the image analysis results generated by the PathAI PD-L1 Multi-Clone NSCLC algorithm. For instructions on running the IHC using the Dako Autostainer Link 48 or VENTANA Benchmark Ultra / UltraPlus stainers, scanning slides using the VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner, or using the features and supported workflows in the **navify** Digital Pathology, see their respective user manuals.

Symbol Definitions

The following table defines symbols that appear in this user manual:

Symbol	Definition
	Indicates the product is "For Research Use Only. Not for use in diagnostic procedures."
	Indicates the manufacturer's catalog number so the product can be identified.
	Indicates the need for the user to consult the Instructions for Use.
	Indicates the manufacturer of the product.

Product Names

The following product names are referenced throughout this user manual:

Product Names	Manufacturer
PathAI PD-L1 Multi-Clone NSCLC	PathAI, Inc.
navify Digital Pathology	Roche Diagnostics
VENTANA DP 200 Slide Scanner	Roche Diagnostics
VENTANA DP 600 Slide Scanner	Roche Diagnostics
VENTANA Benchmark Ultra	Roche Diagnostics
VENTANA Benchmark UltraPlus	Roche Diagnostics
PD-L1 IHC 22C3 pharmDx assay kit	Agilent
PD-L1 IHC 28-8 pharmDx assay kit	Agilent
PD-L1 IHC SP142 RxDx assay kit	Roche Diagnostics
PD-L1 IHC SP263 RxDx assay kit	Roche Diagnostics
EnVision FLEX visualization system	Agilent

About Algorithm Limitations

The PathAI PD-L1 Multi-Clone NSCLC analyzes tissue in whole slide images stained with H&E.

The PathAI PD-L1 Multi-Clone NSCLC algorithm analyzes tissue in whole slide images stained with one of four major PD-L1 IHC assays - 22C3, 28-8, SP142 and SP263.

Guidelines for Ensuring Image Quality

To improve image quality, use the following guidelines:

- For each staining run, follow the manufacturer's recommendations for each applicable assay using all the positive and negative quality control materials.
- Validate the applicable assay staining run by reviewing the control slides manually using conventional light microscopy to ensure the stain quality of the slide before generating a whole slide image with the slide scanner.
- If you determine the quality of the control slides are not acceptable during your manual examination, re-stain the slide and review the quality again using conventional light microscopy. For recommendations, see the manufacturer's assay documentation.
- For each PD-L1 assay, follow the manufacturer's interpretation guidelines to help you evaluate stain quality.

Guidelines for Understanding Algorithm Limitations

When reviewing algorithm results, note the following algorithm limitations:

- The algorithm results are only as good as the quality of the immunohistochemistry (IHC) and the quality of the subsequent whole slide image that is analyzed.
- The algorithm may generate incorrect scores if the slide was improperly scanned or has:
 - Significant artifacts obscuring cancer cells; or
 - Abnormal staining.

Contacting Roche Support

If you have questions pertaining to the appropriate scanning of images (VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner), or effective utilization of **navify** Digital Pathology, contact your local affiliate or Roche Service representative and reference the following product information:

- *PathAI PD-L1 Multi-Clone NSCLC*
- *Roche product number 10686138001*

Chapter 2: Product Overview

This chapter includes:

- About the Algorithm Test Principles 8
 - Steps in Image Analysis 9
 - Cell Identification and Scoring 9
 - How the Algorithm Reports Scores 9

About the Algorithm Test Principles

The PathAI PD-L1 Multi-Clone NSCLC uses derived overlays to visualize classification of tissue stained with major PD-L1 assays (Figure 1).

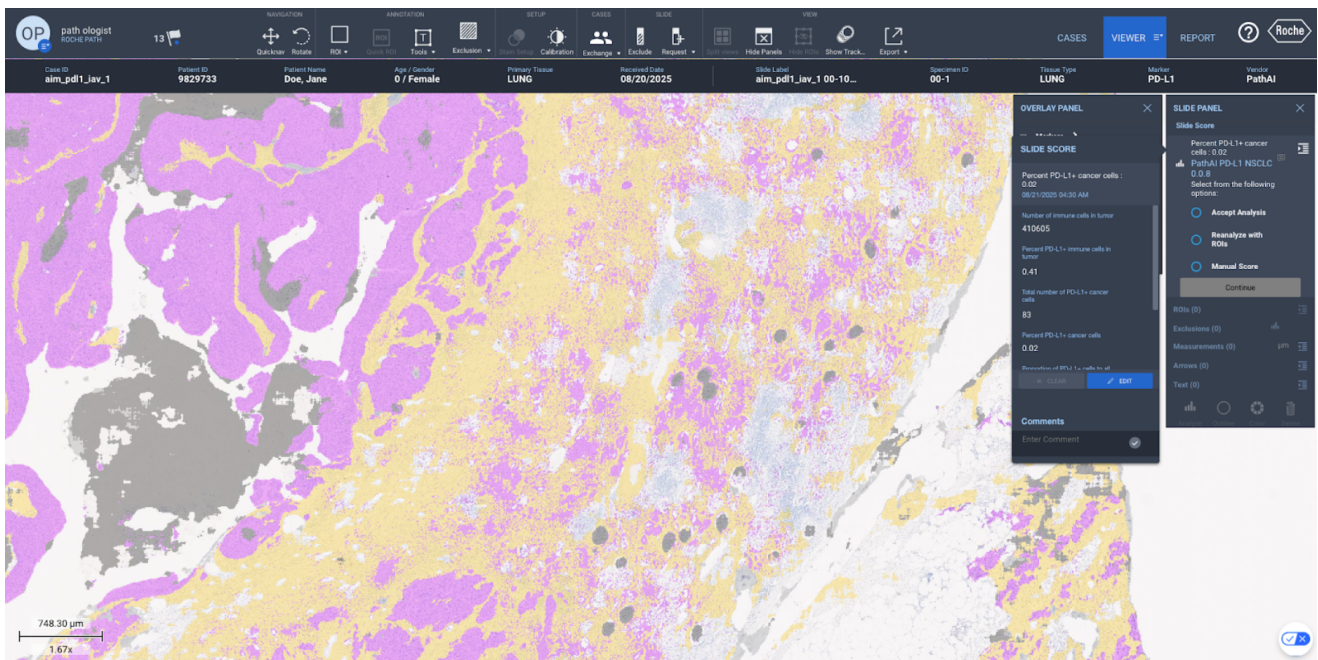


Figure 1 - navify Digital Pathology Showing PD-L1 Overlays

Steps in Image Analysis

This process happens in the background.

1. Split image into tiles for processing by the convolutional neural network.
2. Classify tissue regions as one of the following tissue types:
 - Cancer
 - Cancer Stroma
 - Necrosis
 - Artifact
3. Classify cells as one of the following cell types:
 - PD-L1+ Cancer Cell
 - PD-L1 Negative Cancer Cell
 - PD-L1+ Immune Cell
 - PD-L1 Negative Immune Cell
 - Other Cell
4. Calculate the slide-level results which summarize the number and proportions of different cell types within different tissue regions.

Cell Identification and Scoring

Cell locations and classifications are identified by parsing overlays generated by the deep learning network. The classification for any individual cell is based on the cell type with the highest probability as assessed by the deep learning network.

How the Algorithm Reports Scores

The PathAI PD-L1 Multi-Clone NSCLC reports each score to one digit in the tenths place (for example, 4.8%).

Chapter 3: Algorithm Workflow

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About the Algorithm Workflow

The workflow for the PathAI PD-L1 Multi-Clone NSCLC includes the following procedures:

- Staining the tissue on the slide using one of four major PD-L1 IHC assays - 22C3, 28-8, SP142, SP263, and applicable assay kit with the Dako Autostainer Link 48 (22C3, 28-8) or VENTANA Benchmark Ultra/Ultra Plus (SP263, SP142).
- Scanning the slide with the VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner.
- Analyzing and scoring the whole slide image with the PathAI PD-L1 Multi-Clone NSCLC.

Antibody Staining

The PD-L1 IHC antibody identifies PD-L1+ cells in tissue:

- Follow the instructions provided in the respective PD-L1 assay manual for tissue preparation, staining, and review of controls.
- Restain the slides if staining does not meet the guidelines outlined in the respective PD-L1 assay manual.

Slide and Scanning Criteria

The PathAI PD-L1 Multi-Clone NSCLC accepts the following whole slide image (WSI) and scanner types as input per accession:

Types	Description
Slides	<ul style="list-style-type: none"> • 1 PD-L1 stained slide with the 22C3, 28-8, SP142 or SP263 clone (required) • 1 Negative Reagent Control (NRC) stained slide (optional) • 1 H&E stained slide (optional)
Image	<ul style="list-style-type: none"> • BIF(F)
Scanner	<ul style="list-style-type: none"> • VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner* 20x, 40x magnification

**For scanning instructions, see the applicable scanner user manual.*

Creating and Analyzing Slide Images

To perform this task, complete the following steps:

1. Use the recommended staining and visualization system as mentioned in the user manuals for the assay to stain the tissue sample with one of the four applicable major PD-L1 IHC assays - 22C3, 28-8, SP142 and SP263.

Scan the slide with the VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner in BIF (F) file format at 20x or 40x magnification and at one z-plane.

2. Create a case in **navify** Digital Pathology either manually in **Data Management** or automatically through communication with the laboratory information system. For instructions, see the *Roche navify Digital Pathology User Guide*.

Chapter 4: Algorithm Results Overview

This chapter includes:

- Viewing Colored Overlays 12
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Viewing Colored Overlays

After an algorithm run, the software produces colored overlays on the analyzed slide image in the **VIEWER**. You can view the definitions of each color and toggle them on and off from the **OVERLAY PANEL** (Figure 2).

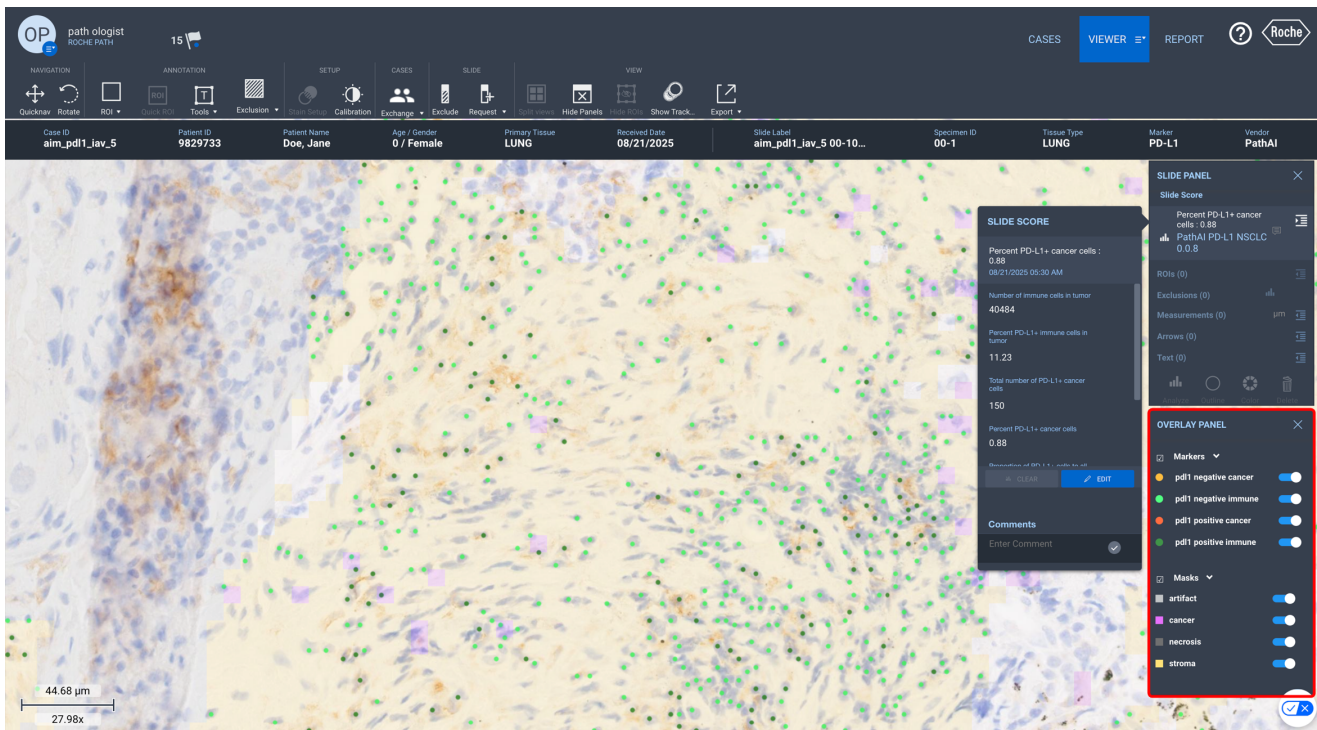










Figure 2 - navify Digital Pathology OVERLAY PANEL

Color Definitions in Overlays

Cells > Cell Overlays

Group	Color		Definition
Cancer Cells		Red	PD-L1 Positive Cancer Cell
		Orange	PD-L1 Negative Cancer Cell
Immune Cells		Dark green	PD-L1 Positive Immune Cell
		Light green	PD-L1 Negative Immune Cell

Tissue Regions > Tissue Overlays

Group	Color		Definition
Evaluable Tissue		Violet	Cancer
		Yellow	Stroma
		Black	Necrosis
Slide Quality		Grey	Artifact

Viewing Quantitative Scoring Results

After the algorithm run, you can view the quantitative scoring results in the **VIEWER's** slide panel. To open this panel, click **VIEWER** and select **Slide Panel** (Figure 3).

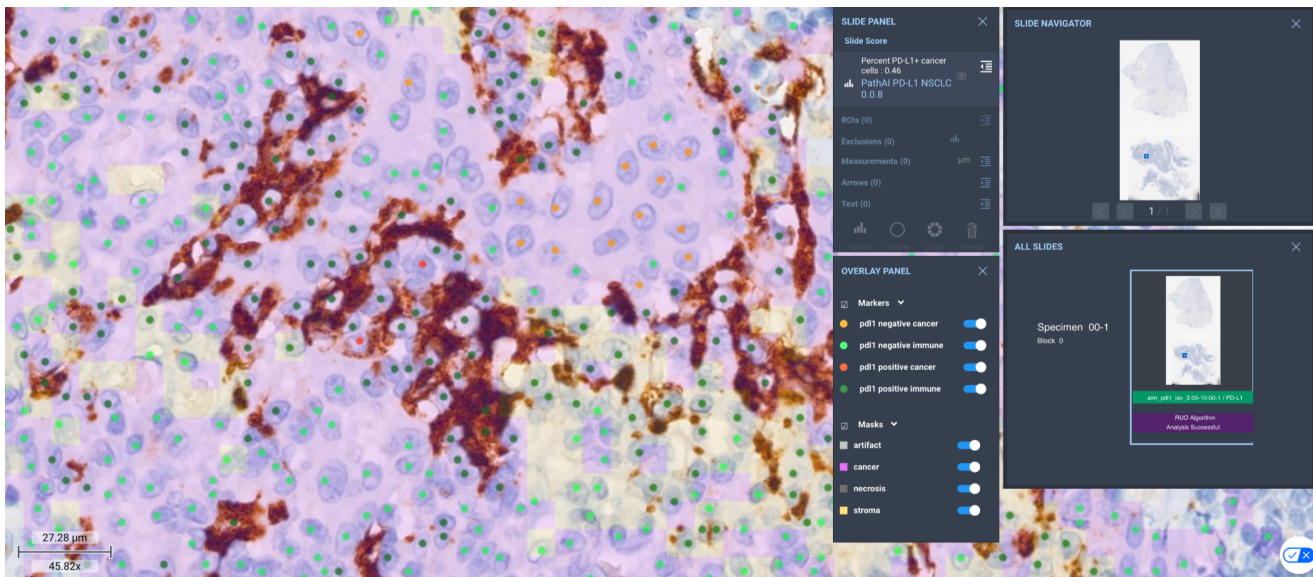


Figure 3 - navify Digital Pathology OVERLAY PANEL

Results	Description
Percent PD-L1+ cancer cells	Number of detected PD-L1+ cancer cells in tumor divided by number of detected cancer cells in tumor, expressed as a percentage (rounded to nearest hundredth).
Number of immune cells in tumor	Number of immune cells detected in evaluable tumor tissue.
Percent PD-L1+ immune cells in tumor	Number of detected PD-L1+ immune cells in tumor divided by number of immune cells in tumor, expressed as a percentage (rounded to nearest hundredth).
Total number of PD-L1+ cancer cells	Total number of PD-L1+ cancer cells detected in evaluable tissue.
Number of PD-L1+ cancer cells in cancer epithelium region.	Total number of PD-L1+ cancer cells detected in Cancer overlay region.
Proportion of PD-L1+ cells to all cancer cells	Number of detected PD-L1+ cells (both tumor and immune) in tumor divided by number of detected cells in tumor, multiplied by 100 and capped at 100 (rounded to nearest whole number).
Total number of cancer cells	Total number of cancer cells detected in evaluable tissue.

Results	Description
Number of PD-L1+ immune cells in tumor	Number of PD-L1+ immune cells detected in evaluable tumor tissue.
Total number of nucleated cells	Total number of nucleated cells (tumor and immune cells) in evaluable tissue.
Number of PD-L1+ immune cells in cancer epithelium region	Number of PD-L1+ immune cells detected in the Cancer overlay region.
Number of PD-L1+ immune cells in cancer stroma region	Number of PD-L1+ immune cells detected in the Cancer Stroma overlay region.
Evaluable tissue area (mm ²)	Area within the whole slide image of evaluable tissue.
Artifact detected (%)	Percent of evaluable tissue area with artifact detected.
Tumor area detected (%)	Percent of evaluable tissue area with tumor detected.

Chapter 5: Generating Reports

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About Generating Reports

Understanding the Report

The report includes a tabular summary of all the quantitative scoring results described in Chapter 4.

Chapter 6: Troubleshooting

This chapter includes:

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Re-triggering a Failed Analysis

You must have Administrator privileges to perform this troubleshooting task.

If an error occurs during the whole slide analysis, an error message appears under the slide. To address the failure, perform the following steps:

1. Login to **navify** Digital Pathology.
2. Navigate to **Administer Settings** and choose **Job Queue**.



NOTE: The administrator settings is only available if you have administrator privileges.

3. Choose the **Failed** tab.
All slides with failed analysis are displayed.
4. Choose the slide to be analyzed then choose the **Start Job** button.
The slide moves to **Canceled/Completed** tab when done.

Best Practices for Excluding Areas from Analysis

To ensure the algorithm is analyzing the tumor region appropriately, perform the following steps:

1. Turn off the algorithm overlays on the PD-L1 slide to identify the tumor area, consulting the matched H&E-stained slide as necessary.
2. Evaluate whether the algorithm is over-calling the tumor region. For example, determine if the algorithm overlay labels non-tumor tissue incorrectly as *cancer* or *cancer stroma*. Specifically:
 - a. Do the combined overlays for *cancer* and *cancer stroma* overlie non-tumor regions?
 - b. If so, draw exclusion ROI annotations to remove these overcalled tumor regions from the algorithm analysis.
3. If this mitigation does not address the problem, reject the finding and manually score the case.

Frequently Asked Questions

Overview

This topic provides answers to frequently asked questions (FAQ) about the PathAI PD-L1 Multi-Clone NSCLC algorithm for Non-Small Cell Lung Cancer which runs on **navify** Digital Pathology Version 2.5.

Frequently Asked Questions

Algorithm Output

Question	Answer
How many cell types do the PathAI PD-L1 Multi-Clone NSCLC algorithm detect?	<p>The PathAI PD-L1 Multi-Clone NSCLC algorithm will detect and show the following cell types:</p> <ul style="list-style-type: none"> • PD-L1+ Cancer Cells • PD-L1 Negative Cancer Cells • PD-L1+ Immune Cells • PD-L1 Negative Immune Cells
How many tissue types do the PathAI PD-L1 Multi-Clone NSCLC algorithm detect?	<p>The PathAI PD-L1 Multi-Clone NSCLC algorithm will detect and show the following tissue types:</p> <ul style="list-style-type: none"> • Cancer • Cancer Stroma • Necrosis • Artifact

Question	Answer
<p>What types of tissue should I exclude when using the algorithm?</p>	<p>The user should ensure that the algorithm is appropriately analyzing the tumor region. To do so, apply the following procedure:</p> <ol style="list-style-type: none"> A. Turn off the algorithm overlays on the PD-L1 slide to identify the tumor area, consulting the matched H&E-stained slide as necessary. B. Evaluate whether the algorithm is over-calling the tumor region. For example, determine if the algorithm overlay labels non-tumor tissue incorrectly as cancer or cancer stroma. Specifically: <ul style="list-style-type: none"> • Do the combined overlays for cancer and cancer stroma overlie non-tumor regions? • If so, draw exclusion ROI annotations to remove these overcalled tumor regions from the algorithm analysis. C. If this mitigation does not address the problem, reject the algorithm result and provide a manual score.
<p>What is the accuracy rate of the PathAI PD-L1 Multi-Clone NSCLC algorithm on this platform?</p>	<p>To find the reference paper for our PD-L1 algorithm, please follow this link to our publication in Modern Pathology.</p>
<p>How long should it take for image analysis to complete?</p>	<p>The time for whole slide analysis will vary based on details like the size of the image, the number of cases that are concurrently being analyzed, etc. That said, we expect total analysis time to take roughly 1 hour for primary analysis, and roughly 10 minutes for secondary analysis after ROI annotations have been saved.</p>
<p>Is there a color key on the platform for cell types or tissue regions that I can reference?</p>	<p>After an algorithm run, the software produces colored overlays on the analyzed slide image in the VIEWER. You can view the definitions of each color and toggle them on and off from the OVERLAY PANEL. For a color key, you can review the Instructions For Use, which can be found in the About section of the navify Digital Pathology platform.</p>
<p>Is there any customization built into the PathAI PD-L1 Multi-Clone NSCLC algorithm? Can I change it to only mark PD-L1+ Cancer Cells and ignore PD-L1 negative Cancer cells?</p>	<p>No, the algorithm has been trained to provide the whole set of outputs at the tissue and cell level. You can view the definitions of each color and toggle them on and off from the OVERLAY PANEL.</p>
<p>Can I use the PathAI PD-L1 Multi-Clone NSCLC algorithm to score NSCLC core biopsies or resections?</p>	<p>Yes, the PD-L1 algorithm may be used to score results on both core biopsies and resections.</p>

Question	Answer
I decided to manually score the slide - can I share my analysis results with others on the platform?	After an algorithm run, you can generate reports containing summary details about each case. The type of data included in the report is based on templates configured by the administrator. For details, see Producing and signing out a report in the Roche navify Digital Pathology User Guide.

Slides

Question	Answer
What image file formats do the PathAI PD-L1 Multi-Clone NSCLC algorithm support?	Users will only be able to run image analysis on BIF (F) and files scanned by the VENTANA DP 200 Slide Scanner or VENTANA DP 600 Slide Scanner.
Can I use PathAI PD-L1 Multi-Clone NSCLC algorithm stained with a different PD-L1 antibody other than 28-8, 22C3, SP142 or SP263?	No, the algorithm was developed specifically for the 28-8, 22C3, SP142 or SP263 antibody clones and is not intended for use with other clones.

Cloud / Infrastructure

Question	Answer
Which cloud provider does PathAI use?	PathAI uses Amazon Web Services as the cloud provider.
What PII/PHI is transferred to PathAI to process algorithm? How long does PathAI retain and store this data?	We do not maintain any PHI. PathAI receives no PHI to process algorithm.
What type of results are returned to navify Digital Pathology?	All slide-level results and overlays are returned to navify Digital Pathology.
Does the PathAI algorithm cloud environment have HITRUST or SOC-II certification?	PathAI is ISO 27001-certified.
What platform considerations should I recommend to my customers for PathAI PD-L1 Multi-Clone NSCLC algorithm?	PathAI algorithm only work with our navify Digital Pathology (cloud) solution and the server in the cloud is not customer configurable. Because the algorithm analysis runs in the PathAI cloud environment, there is no specific guidance necessary for the algorithm analysis, beyond high-speed internet.
How is data securely transferred between PathAI and Roche?	OAuth credentials are provided to navify Digital Pathology, in order to make requests via HTTPS. All API requests are expected to come only from navify Digital Pathology.

Product

Question	Answer
How often are the PathAI PD-L1 Multi-Clone NSCLC updated to reflect changes in technology and best practices?	PathAI is always looking to improve its algorithm to provide the best experience for customers. We will take customer feedback into account for updates to the algorithm and will inform the customers prior to any updates being released onto the platform.



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