510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
INSTRUMENT ONLY

I Background Information:

A 510(k) Number

K251511

B Applicant

Copan WASP S.r.l.

C Proprietary and Established Names

PhenoMATRIX

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
PPU	Class II	21 CFR 866.2190 - Automated Image Assessment System For Microbial Colonies On Solid Culture Media	MI - Microbiology
QQY	Class II	21 CFR 866.2190 - Automated image assessment system for microbial colonies on solid culture media	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain a substantial equivalence decision for PhenoMATRIX used with the WASPLab instrument.

B Type of Test:

The PhenoMATRIX is an *in vitro* diagnostic system which utilizes image analysis software for automated assessment and classification of images of solid culture media plates streaked with microbiological samples derived from the human body. The PhenoMATRIX requires the Copan WASPLab microbiology automation system in order to operate. WASPLab is an *in vitro* diagnostic device for handling, incubation, digital imaging and sorting of solid culture media plates.

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

PhenoMATRIX is a WASPLab optional module intended for the automatic sorting of images of blood-based agar, chocolate agar, MacConkey agar, and CHROMagar Orientation culture media plates according to classification parameters based on Image Analysis Software results and clinical and demographic data.

Image Analysis Software performs semi-quantitative and/or qualitative analysis of culture media plates by detecting microbial growth, estimating colony counts and differentiating isolates based on phenotypic colony characteristics.

The system determines how the images should be sorted based on image analysis results in addition to patient data according to expert rules defined by the laboratory.

All images shall be evaluated by trained personnel for final assessment and result definition.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

IV Device/System Characteristics:

A Device Description:

PhenoMATRIX is an *in vitro* diagnostic system which utilizes image analysis software for automated assessment and classification of images of solid culture media plates streaked with microbiological samples derived from the human body. PhenoMATRIX comprises software modules intended for analysis and automatic classification of high-resolution digital images of culture plates captured by the Copan WASPLab system. PhenoMATRIX analysis modules include semi-quantitative and qualitative assessment of microbial growth.

Solid media culture plates inoculated with microbiological samples are incubated within the WASPLab system. Digital images of the culture plate are acquired after appropriate incubation according to the media instructions for use. PhenoMATRIX interacts with WASPLab for analysis of the culture plate digital images. Within the PhenoMATRIX software, specific

imaging product set (IP) analysis modules perform analysis of microbial growth detection, colony count estimations and isolate differentiation based on phenotypic colony characteristics. IPs are independently developed for specific analysis functions. The image analysis result can be combined with laboratory information system (LIS) data (such as demographic, clinical, and/or sample data) according to customizable logic rules defined by the laboratory for image classification. The classification is used for image sorting into dedicated digital folders associated with specific, pre-defined expected results.

After PhenoMATRIX processing, the physical plates are left inside the WASPLab and images are available for digital inspection by the trained microbiologist through the WASPLab User Interface. The trained microbiologist reviews the plate images in each digital classification folder to confirm (or modify) the assigned folder and final result. After that, the plates follow the workflow that has been defined by the laboratory according to the assigned result.

PhenoMATRIX is intended to analyze the digital images of microbial growth on validated culture media from various clinical specimens including:

- Urine specimens (e.g., clean catch/mid-stream, catheter, suprapubic aspirate)
- Upper respiratory tract specimens (e.g., anterior nares swab, sputum, throat swabs)
- Lower respiratory tract specimens (e.g., bronchoalveolar lavage, bronchial wash, endotracheal aspirate)
- Genital swab specimens (e.g. vaginal swabs, urethral swabs)
- Vaginal-rectal swab specimens
- Body fluid specimens
- Skin specimens
- Stool specimens

PhenoMATRIX is intended to analyze the digital images of whole and bi-plates of different categories of culture media including:

- Blood-based agar, such as tryptic soy agar with blood, Columbia agar with blood, Columbia agar with blood and colistin-nalidixic acid, Schaedler agar, selective agar plates intended to detect or differentiate colonies of Group A and Group B *Streptococcus* spp. based on hemolytic reaction.
- Chocolate agar plates for the isolation of fastidious bacteria
- MacConkey agar plates for the selective isolation of gram-negative organisms
- CHROMagar Orientation plates for the isolation and differentiation of urinary pathogens

B Instrument Description Information:

1. Instrument Name:
   WASPLab

2. Specimen Identification:
   Users can identify specific culture media images for PhenoMATRIX analysis through the user interface. All culture plates processed by the WASPLab are identified by scanning a manually applied linear barcode on the side of each plate. The barcode is visible through the graphical user interface when using PhenoMATRIX.

3. Specimen Sampling and Handling:
PhenoMATRIX is intended to process culture media plate images acquired from plates managed by the WASPLab device. PhenoMATRIX classifies media plate images based on image analysis results and culture plate interpretation criteria defined by the user. Classification occurs based on analytical information provided from the PhenoMATRIX image analysis modules (e.g., colony count, growth, morphology, beta-hemolysis), as well as patient data provide by the laboratory information system (LIS).

Plates labeled with a barcode are loaded onto a conveyor and loaded into the WASPLab. The barcode is scanned, and the plate is incubated within the WASPLab. After appropriate incubation as determined by the culture media instructions for use, each plate is photographed, and the image is saved on the WASPLab server in a dedicated section of the WebApp User interface that interfaces with the PhenoMATRIX, where plate images are grouped into folders according to classification. When using PhenoMATRIX, the operator reviews the plate images and confirms or modifies the assigned folders and result(s). The plates then follow the user-defined follow-up workflow based on the classification.

4. Calibration:
No calibration is required for the PhenoMATRIX image analysis software that is installed on the WASPLab device. The WASPLab is calibrated by trained personnel during instrument installation and setup. Multiple calibration controls are also automatically executed during instrument use during image acquisition and interpretation to ensure proper function. If any calibration anomalies are detected, an alarm is shown in the graphical user interface and the plate involved in the error is identified by WASPLab software as "not processable" by PhenoMATRIX, and the plate is not processed.

5. Quality Control:
There is not specific Quality Control (QC) for the PhenoMATRIX. The device labeling indicates that QC checks for the WASPLab instrument should be conducted daily during use or at a frequency determined appropriate by the laboratory. QC checks are performed with cultures of *E. coli* ATCC 25922 and sterile TSB plated on blood agar to monitor for contamination and proper performance of the system processes from primary sample processing to culture plate incubation and imaging. To pass, results must meet the expected designation (i.e., plates inoculated with positive tubes exhibit growth, plates inoculated with negative tubes do not exhibit growth).

There was a total of 441 QC results from the three clinical study sites during PhenoMATRIX clinical validation. All QC results passed on each day of testing.

V Substantial Equivalence Information:

A Predicate Device Name(s):

APAS Independence with Urine Analysis Module, APAS Independence with IC Chromogenic MRSA BD Analysis Module; APAS Independence with IC Chromogenic MRSA TFS/S Analysis Module

B Predicate 510(k) Number(s):
K183648, K200839

C Comparison with Predicate(s):

Device & Predicate Device(s):	Candidate Device K251511	Predicate Device: K183648	Other Reference Device: K200839
Device Trade Name	PhenoMATRIX	APAS Indipendence with Urine Analysis Module	APAS Independence with IC Chromogenic MRSA BD Analysis Module; APAS Independence with IC Chromogenic MRSA TFS/S Analysis Module
General Device Characteristic Similarities			
Intended Use/Indications For Use	PhenoMATRIX is a WASPLab optional module intended for the automatic sorting of images of blood-based agar, chocolate agar, MacConkey agar, and CHROMagar Orientation culture media plates according to classification parameters based on Image Analysis Software results and clinical and demographic data. Image Analysis Software performs semi-quantitative and/or qualitative analysis of culture media plates by detecting microbial growth, estimating colony counts and differentiating isolates based on phenotypic colony characteristics. The system determines how the images should be sorted based on image analysis results in addition to patient data according to expert rules defined by the laboratory. All images shall be evaluated by trained personnel for final assessment and result definition.	The APAS Independence is an in vitro diagnostic system comprised of an instrument and software analysis module(s) for specific indications that are used to automate imaging and interpretation of microbial colonies on plates of solid culture media. The APAS Independence is an in vitro diagnostic system comprised of an instrument for automated imaging of agar culture plates and a software analysis module for the following use: The APAS Independence, when using its urine analysis module, automates urine culture plate imaging and interpretation to detect the presence or absence of microbial growth on sheep blood and MacConkey agar culture plates that are inoculated with a 1µL sample volume. The APAS Independence, when using its urine analysis module, provides a semi- quantitative assessment of colony counts that are used as an aid in the diagnosis of urinary tract infection. All urine culture plates that are identified as positive for growth by the APAS Independence, when using its urine analysis module, must be reviewed by a trained	The APAS Independence is an in vitro diagnostic system comprised of an instrument and software analysis module(s) for specific indications that are used to automate imaging and interpretation of microbial colonies on plates of solid culture media. The APAS Independence is an in vitro diagnostic system comprised of an instrument for automated imaging of agar culture plates and a software analysis module for the following use: 1. The APAS Independence, when using its IC MRSA Chromogenic BD analysis module, automates culture plate imaging and interpretation to detect the presence or absence of colonies with colors suggestive of methicillin-resistant Staphylococcus aureus (MRSA) growth on Beckton Dickson BBL CHROMagar MRSA II agar that has been inoculated with anterior nares swabs and incubated at 36°C ± 1°C for 24 hours. The APAS Independence, when using its IC MRSA Chromogenic BD analysis module, provides an aid in routine screening for colonization with MRSA. It provides one of two screening results: Presumptive MRSA or Negative. All culture plates that

		microbiologist.	are identified as Presumptive MRSA by the APAS Independence, when using the IC MRSA Chromogenic BD analysis module require review by a trained microbiologist. 2. The APAS Independence, when using its IC MRSA Chromogenic TFS/S analysis module, automates culture plate imaging and interpretation to detect the presence or absence of colonies with colors suggestive of methicillin- resistant Staphylococcus aureus (MRSA) growth on Thermo-Fisher Spectra MRSA agar that has been inoculated with anterior nares swabs and incubated at 36°C ± 1°C for 24 hours. The APAS Independence, when using its IC MRSA Chromogenic TFS/S analysis module, provides an aid in routine screening for colonization with MRSA. It provides one of three screening results: Presumptive MRSA, Presumptive nonMRSA, or Negative. All culture plates that are identified as Presumptive MRSA or Presumptive non-MRSA by the APAS Independence, when using the IC MRSA Chromogenic TFS/S analysis module, require review by a trained microbiologist.
Imaging Station	PhenoMATRIX analyzes culture plate digital images captured by the WASPLab. The WASPLab Imaging Station includes a LineScan camera that acquires the image of the culture, two Light Emitting Diode (LED) for the illumination of culture plates from the top and from the bottom of the plates, an imaging background that is a motorised white panel used to acquire plate pictures with white background and plate handling mechanisms to manage plates movements, lid opening and lid closings	A fixed Charge Coupled Device (CCD) camera, top and bottom Light Emitting Diode (LED) illumination for the culture plates, and image capture and a plate handling mechanism, all of which are housed in a light-sealed chassis.	A fixed Charge Coupled Device (CCD) camera, top and bottom Light Emitting Diode (LED) illumination for the culture plates, and image capture and a plate handling mechanism, all of which are housed in a light- sealed chassis.
Instrument control and management	The PhenoMATRIX software depends on the WASPLab instrument for the acquisition of culture plate digital images. The WASPLab system also manages the physical culture plates.	The APAS Controller PC manage image capture, storage and analysis. The Plate Controller PC controls movement of culture plates	The APAS Controller PC manage image capture, storage and analysis. The Plate Controller PC controls movement of culture plates

	WASPLab device is provided with: -A PANEL PC Touch Screen Monitor on the Imaging Station provided with the user interface to allow the user to manage the instrument and to check its status; -A Workstation to allow the user to visualize the acquired images; -A Control Unit where all the software managing the instrument are loaded and running to allow plates movements and handling. Moreover, in the Control Unit plates images are stored and can be evaluated by the PhenoMATRIX.	between the input carriers, imaging station and output carriers or stacks. The Instrument Controller PC provides the user interface for operation of the APAS Independence and coordinates the functions of the APAS and Plate Controller PCs.	between the input carriers, imaging station and output carriers or stacks. The Instrument Controller PC provides the user interface for operation of the APAS Independence and coordinates the functions of the APAS and Plate Controller PCs
Image Analysis Module Software	PhenoMATRIX is installed on the WASPLab Control Unit and configurable for plate images assessment.	Analyses modules are installed on the APAS Controller PC to provide the configuration and instructions for image capture and analysis.	Analyses modules are installed on the APAS Controller PC to provide the configuration and instructions for image capture and analysis.
LIS Interface	Analyses results can be sent to the LIS. Data related to the sample are retrieved by LIS.	Analyses results can be sent to the LIS. Data related to the sample are retrieved by LIS.	Analyses results can be sent to the LIS. Data related to the sample are retrieved by LIS.
QC Control	For the WASPLab, quality control is performed daily by the operator using known positive and negative samples to assess growth plates. No specific QC is included for the PhenoMATRIX.	Daily performed by the operator by using Positive and Negative for Growth Plates.	Daily performed by the operator by using Positive and Negative for Growth Plates.
General Device Characteristic Differences			
Sample type	Microbiological samples derived from the human body in liquid or semi-liquid phase.	Urine	Anterior nares specimens
Culture media plates	Blood-based, Chocolate, MacConkey and CHROMagar Orientation culture media plates	Sheep blood and MacConkey agar culture plates	Beckton Dickson BBL CHROMagar MRSA II agar
Result management	All digital images shall be reviewed by trained microbiologist for final assessment before result definition and sending to the LIS.	All plates assessed as positive for growth shall be reviewed by trained microbiologist. Negative for growth plates are automatically managed by the device.	All plates assessed as “Presumptive MRSA” or “Presumptive non-MRSA” shall be reviewed by trained microbiologist. Negative for MRSA plates are automatically managed by the device.
Instrumentation	WASPLab	APAS Independence	APAS Independence
Calibration	No calibration is required for PhenoMATRIX software that is installed on WASPLab device that is calibrated by Copan personnel during setup during manufacturing process.	Daily color calibration is required prior to use to be performed by the user.	Daily color calibration is required prior to use to be performed by the user.

VI Standards/Guidance Documents Referenced:

- ISO 14971 Medical devices - Application of Risk Management to Medical Devices, 2019.

- IEC 62304 Medical device software - software lifecycle processes, 2006.

- IEC 60601-1-2 Edition 4.0 2014-02, Medical electrical equipment - Part 1-2: General requirements for basic safety and essential performance

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

The reproducibility of the PhenoMATRIX was evaluated across three different WASPLab instruments over multiple test runs over multiple days. All IPs developed for the specific culture media and analysis capabilities were tested: blood-based agar (9 IPs for growth detection/quantification and 7 IPs for beta-hemolysis detection), chocolate agar (1 IP), CHROMagar Orientation (2 IPs), and MacConkey agar (6 IPs). Testing was performed using representative microbial species at three logarithmic concentrations (10^3, 10^4, and 10^5 CFU/mL) on all four claimed agar types: blood agar, MacConkey agar, CHROMagar Orientation, and chocolate agar. The study assessed the device capabilities, including growth detection, growth semi-quantification, isolate differentiation by morphology, growth purity classification, and beta-hemolysis detection (blood agar only). Each dilution was plated in triplicate and incubated under appropriate conditions, with images captured by three different WASPLab instruments and analyzed using the imaging product sets specific to each agar type over three consecutive days.

The results demonstrated acceptable reproducibility across all microbial growth characteristics (Tables 1-5). Growth detection achieved 100% agreement across all agar types, instruments and IPs for detecting presence or absence of microbial growth. Colony quantification showed consistent mean counts with acceptable coefficient of variation values (typically 15-60%) and acceptable reporting of semi-quantitative growth. Morphological detection and growth purity classification both achieved 100% agreement for all tested species and conditions, including detection of various colony morphologies (coliform, GBS-like, *Enterococcus*-like, etc.) and classification of growth patterns. Beta-hemolysis detection on blood agar also achieved 100% agreement. These findings support that PhenoMATRIX provides consistent and reliable automated assessment of microbial growth characteristics across different instruments and agar types.

Table 1. Reproducibility of growth detection obtained by the PhenoMATRIX

Media type	Species	Expected result	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
Blood agar	*Candida albicans*	Growth	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Media type	Species	Expected result	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
	Enterococcus faecalis	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	Growth	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Staphylococcus aureus	Growth	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Streptococcus pyogenes	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	No growth	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
MacConkey agar	*Escherichia coli* ATCC 25922	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli O157	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	No growth	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
Chocolate agar	*Streptococcus pneumoniae*	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Neisseria gonorrhoeae	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	No growth	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Media type	Species	Expected result	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
CHROMagar Orientation	*Enterococcus faecalis*	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Klebsiella pneumoniae	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Proteus mirabilis	Growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	No growth	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Table 2. Reproducibility of growth semi-quantification obtained by the PhenoMATRIX

Media type	Species	Load [CFU/mL]	WASPLab A		WASPLab B		WASPLab C		Overall	
			Mean*	CV%	Mean	CV%	Mean	CV%	Mean	CV%
Blood agar	*Candida albicans*	10^3	5.3	60.0%	5.3	60.0%	5.3	60.0%	5.3	57.7%
		10^4	56.0	43.5%	55.9	43.9%	56.1	43.7%	56.0	42.0%
		10^5	248.7	44.7%	250.4	45.7%	248.8	46.6%	249.3	43.9%
	Enterococcus faecalis	10^3	5.1	63.0%	5.1	63.0%	5.1	63.0%	5.1	60.5%
		10^4	46.0	62.1%	46.0	62.1%	46.2	62.3%	46.1	59.7%
		10^5	321.0	46.2%	320.8	46.4%	320.8	46.1%	320.9	44.4%
	Escherichia coli	10^3	5.4	55.2%	5.3	53.9%	5.3	53.9%	5.4	52.2%
		10^4	42.9	35.7%	42.9	35.7%	42.9	35.7%	42.9	34.3%
		10^5	224.2	26.3%	223.3	25.8%	224.9	26.3%	224.1	25.1%
	Escherichia coli / *Streptococcus agalactiae*	10^3	4.4	76.4%	4.4	76.4%	4.4	76.4%	4.4	73.4%
		10^4	54.0	54.4%	54.1	54.5%	54.1	54.3%	54.1	52.3%
		10^5	229.0	47.9%	229.9	48.1%	230.7	49.2%	229.9	46.5%
	Staphylococcus aureus	10^3	5.2	74.0%	5.2	74.0%	5.2	74.0%	5.2	71.1%
		10^4	54.1	54.9%	54.1	54.2%	54.6	55.2%	54.3	52.6%
		10^5	233.9	52.9%	234.0	52.6%	232.8	51.0%	233.6	50.1%
	Streptococcus pyogenes	10^3	4.7	52.5%	4.7	52.5%	4.7	52.5%	4.7	50.4%
		10^4	42.6	31.2%	42.4	31.2%	42.4	30.9%	42.5	29.9%
		10^5	181.0	17.6%	179.8	16.8%	178.7	16.8%	179.8	16.4%
	Saline	N/A	0.0	N/A	0.0	N/A	0.0	N/A	0.0	N/A
MacConkey agar	*Escherichia coli* ATCC 25922	10^3	9.4	57.5%	9.4	57.5%	9.4	57.5%	9.4	55.3%
		10^4	71.9	24.4%	71.1	24.7%	71.6	25.0%	71.5	23.7%
		10^5	291.9	22.3%	292.2	21.5%	293.8	21.7%	292.6	21.0%
	Escherichia coli O157	10^3	7.0	42.9%	7.0	42.9%	7.0	42.9%	7.0	41.2%
		10^4	74.1	34.7%	74.1	34.5%	74.2	35.0%	74.1	33.4%

Media type	Species	Load [CFU/mL]	WASPLab A		WASPLab B		WASPLab C		Overall	
			Mean*	CV%	Mean	CV%	Mean	CV%	Mean	CV%
		10^5	270.2	22.8%	269.8	23.3%	270.6	23.0%	270.2	22.1%
	Saline	N/A	0.0	N/A	0.0	N/A	0.0	N/A	0.0	N/A
Chocolate agar	*Streptococcus pneumoniae*	10^3	7.2	48.8%	7.2	48.8%	7.2	48.8%	7.2	46.9%
		10^4	76.1	31.3%	76.4	31.7%	76.8	31.8%	76.4	30.4%
		10^5	249.4	22.9%	251.9	23.7%	253.2	23.9%	251.5	22.6%
	Neisseria gonorrhoeae	10^3	5.3	50.5%	5.4	53.6%	5.4	53.6%	5.4	50.6%
		10^4	56.8	36.2%	56.2	34.3%	56.1	35.5%	56.4	34.0%
		10^5	173.6	17.3%	172.3	16.7%	171.0	16.4%	172.3	16.2%
	Saline	N/A	0.0	N/A	0.0	N/A	0.0	N/A	0.0	N/A
CHROMaga Orientation	*Enterococcus faecalis*	10^3	4.8	48.8%	4.7	45.5%	4.8	48.8%	4.7	45.9%
		10^4	47.0	17.9%	47.6	19.8%	47.2	18.7%	47.3	18.1%
		10^5	276.3	18.7%	275.9	19.1%	275.1	18.3%	275.8	18.0%
	Escherichia coli	10^3	6.9	59.2%	6.9	59.2%	6.9	59.2%	6.9	56.8%
		10^4	64.0	42.6%	63.2	40.7%	62.1	39.8%	63.1	39.5%
		10^5	275.4	39.6%	272.8	39.1%	275.2	40.3%	274.5	38.1%
	Escherichia coli / *Streptococcus agalactiae*	10^3	5.9	60.3%	5.9	60.3%	5.9	60.3%	5.9	57.9%
		10^4	54.2	42.9%	54.9	43.2%	54.4	44.1%	54.5	41.7%
		10^5	261.3	48.9%	262.3	48.4%	261.1	49.7%	261.6	47.1%
	Klebsiella pneumoniae	10^3	4.0	53.0%	4.0	53.0%	4.0	53.0%	4.0	51.0%
		10^4	37.4	30.2%	37.3	30.5%	37.4	30.2%	37.4	29.1%
		10^5	224.7	27.3%	221.0	26.9%	221.6	27.3%	222.4	26.1%
	Proteus mirabilis	10^3	5.4	52.0%	5.4	52.0%	5.4	52.0%	5.4	50.0%
		10^4	52.8	19.2%	52.7	18.6%	52.6	18.7%	52.7	18.1%
		10^5	259.9	27.0%	256.0	26.7%	258.9	27.8%	258.3	26.1%
	Saline	N/A	0.0	N/A	0.0	N/A	0.0	N/A	0.0	N/A

* Mean values represent the average colony count detected by the WASPLab automated system across replicate measurements at each inoculum load level (10^3, 10^4, or 10^5 CFU/mL).

Table 3. Reproducibility of morphology labels obtained by the PhenoMATRIX

Media type	Species	Expected morphology label	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
Blood agar	*Candida albicans*	*Candida*-like	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Enterococcus faecalis	*Enterococcus faecalis*-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Coliform	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	Coliform and GBS-like	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Staphylococcus aureus	*Staphylococcus aureus*-like	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Media type	Species	Expected morphology label	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Streptococcus pyogenes	GAS-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	N/A (no morphology reported)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
MacConkey agar	*Escherichia coli* ATCC 25922	Not defined	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli O157	Non-fermenter	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	N/A (no morphology reported)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
CHROMaga Orientation	*Enterococcus faecalis*	*Enterococcus*-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	*E. coli*-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	*E. coli*-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Klebsiella pneumoniae	*Klebsiella, Enterobacter, Serratia,* and *Citrobacter* group (KESC)-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Proteus mirabilis	*Proteus, Morganella, Providencia* group (PMP)-like	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	N/A (no morphology reported)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Table 4. Reproducibility of beta-hemolysis obtained by the PhenoMATRIX

Media type	Species	Expected result	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
Blood agar	*Candida albicans*	No beta-hemolysis detected	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Enterococcus faecalis	No beta-hemolysis detected	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Beta-hemolysis detected	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	Beta-hemolysis detected	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Staphylococcus aureus	Beta-hemolysis detected	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Streptococcus pyogenes	Beta-hemolysis detected	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	No beta-hemolysis detected	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27

Table 5. Reproducibility of growth purity obtained by the PhenoMATRIX

Media type	Species	Expected purity bin	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
Blood agar	*Candida albicans*	Predominant growth	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Enterococcus faecalis	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli /	Co-predominance	10^3	100%	100%	100%	100%

Media type	Species	Expected purity bin	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
	Streptococcus agalactiae	– ≥2 isolates		8/8	8/8	8/8	24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Staphylococcus aureus	Predominant growth	10^3	100% 8/8	100% 8/8	100% 8/8	100% 24/24
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Streptococcus pyogenes	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	N/A (No growth)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
MacConkey agar	*Escherichia coli* ATCC 25922	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli O157	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Saline	N/A (No growth)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27
CHROMagar Orientation	*Enterococcus faecalis*	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Escherichia coli / *Streptococcus agalactiae*	Not predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Klebsiella pneumoniae	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100% 9/9	100% 9/9	100% 9/9	100% 27/27
	Proteus mirabilis	Predominant growth	10^3	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^4	100% 9/9	100% 9/9	100% 9/9	100% 27/27
			10^5	100%	100%	100%	100%

Media type	Species	Expected purity bin	Load [CFU/mL]	WASPLab A	WASPLab B	WASPLab C	Combined
				9/9	9/9	9/9	27/27
	Saline	N/A (No growth)	N/A	100% 9/9	100% 9/9	100% 9/9	100% 27/27

2. Linearity:

To determine the system's ability to provide semi-quantitative assessment of growth, studies were conducted to assess the linearity and accuracy of microbial load quantification of the PhenoMATRIX with each claimed media type in four separate studies. Each study tested multiple IPs developed for the specific culture media and analysis capabilities: blood-based agar (9 IPs), chocolate agar (1 IP), CHROMagar Orientation (2 IPs), and MacConkey agar (6 IPs). The test datasets comprised stock images from various WASPLab instruments, selected by independent microbiologists and representing diverse microbial growth characteristics and with varied clinically relevant starting inoculum volumes. Images were categorized into logarithmic growth bins (0, 1-9, 10-99, 100-999, and ≥1000 CFU) representing different microbial concentrations. Reference results were provided by experienced technicians through visual inspection of the digital image of the culture plate, and software performance was evaluated using confusion matrices with a primary acceptance criterion of ≥90% accuracy for logarithmic colony counts, stratified by media type, IP, and starting inoculum volume. The results demonstrate consistent acceptable performance across all culture media types, with overall agreement rates varying by media, IP, and starting inoculum volume.

Blood-based agar plates demonstrated acceptable overall agreement ranging from 97.7% to 99.0% for all IPs across the starting inoculum volumes tested. When stratified by individual IP, most IPs exceeded 95% agreement, although two demonstrated agreements of 82.4% when used with starting inoculum volumes of 1 µL. Chocolate agar demonstrated 97.0 and 99.2% overall agreement for the one IP across the starting inoculum volumes tested. MacConkey agar plates acceptable demonstrated overall agreement ranging from 94.7% to 98.9% for all IPs across the starting inoculum volumes tested. When stratified by individual IP, all IPs exhibited >90% agreement.

CHROMagar Orientation plates demonstrated an overall agreement of 83.6% for the two IPs with the starting inoculum volume tested. CHROMagar Orientation was also evaluated with morphology-specific quantification testing with acceptance criteria of ≥80% accuracy, achieving results ranging from 83.8% to 95.3% across different colony morphologies including *Escherichia coli*-like, *Pseudomonas*-like, microcolonies, and various other bacterial morphological characteristics.

Discrepancies between software and technician results were systematically analyzed by senior microbiology laboratory experts. The primary sources of error included microcolonies that remained undetected by human technicians but were identified by the software (leading to apparent overestimation), and agar defects such as scratches that were misinterpreted as microbial growth by the software.

Agreements for individual IPs and specific inoculum volume combinations that fell below the acceptance criteria were considered acceptable on the basis of the lower bound of the 95% confidence interval and because all digital plate images are subject to follow-up review

by a trained microbiologist to confirm the final result; thus, the risk to patients from incorrect assignment of colony counts by the software is mitigated.

In several cases, PhenoMATRIX designated plates as positive for growth (i.e., $\geq 10^3$ CFU/mL) but for which no growth was reported by the trained microbiologist (reference method) and incorrectly designated some of the culture plates with growth as reported by the reference method as "No Growth". However, this occurred in <1% and <3% of cases, respectively, and is mitigated by the fact that a trained microbiologist reviews all culture plate images, including those with no growth, for final assessment and result definition.

3. Analytical Specificity/Interference:
To assess the system's analytical specificity and growth detection capabilities studies were conducted on PhenoMATRIX with each claimed media type in four separate studies. Each study tested multiple IPs developed for specific culture media and analysis capabilities: blood-based agar (9 IPs for growth detection, 7 IPs for beta-hemolysis detection, and 6 IPs for growth purity assessment), chocolate agar (1 IP), CHROMagar Orientation (2 IPs), and MacConkey agar (6 IPs). The test datasets comprised stock images from various WASPLab instruments, selected by independent microbiologists and representing diverse microbial growth characteristics. Images were categorized for growth detection (presence/absence), morphological differentiation (target colony morphologies), beta-hemolysis detection, and growth purity assessment. Reference results were provided by experienced technicians through visual inspection of the culture plate digital image, with primary acceptance criterion of ≥99% positive percent agreement (PPA) for growth detection, ≥80% agreement for morphological detection, ≥95% agreement for beta-hemolysis detection, and ≥80% overall agreement for growth purity assessment.

The results demonstrate consistently acceptable performance across all culture media types, with PPA for growth detection ranging from 99.3% to 100% across all studies. Blood-based agar plates showed excellent growth detection (99.3-100% PPA, 96.6-100% negative percent agreement [NPA]), morphological differentiation generally exceeding 80% agreement, beta-hemolysis detection achieving 87-99.6% PPA and 50.1-98.3% NPA, and growth purity assessment with 93.3-98.5% overall agreement. Chocolate agar achieved perfect growth detection with 100% PPA and 99.4% NPA. Morphological detection and growth purity were not assessed, as these are not claims for the PhenoMATRIX with chocolate agar.

CHROMagar Orientation plates demonstrated 99.9-100% PPA for growth detection though lower NPA (86.3-92.0%), with morphological detection generally exceeding 80% agreement except for *Staphylococcus saprophyticus*-like and *Pseudomonas*-like morphologies which exhibited gradient-like features challenging software performance, and growth purity assessment with 89.7-90.1% overall agreement. MacConkey agar plates performed exceptionally well with 99.9-100% PPA and 99.1-100% NPA for growth detection, high agreement rates for morphological detection of lactose fermenter and non-fermenter colonies, and 99.4% overall agreement for growth purity assessment.

Discrepancies between software generated and technician read results were systematically analyzed by senior microbiology laboratory experts. The primary sources of error included microcolonies and transparent colonies that remained undetected by the software (leading to false negatives), and artifacts such as bubbles, dust, scratches, or residual biological matrix that were misinterpreted as microbial growth by the software (leading to false positives). For

beta-hemolysis detection, some IPs showed lower NPA due to the software's high sensitivity in detecting beta-hemolysis. Performance was acceptable and additional risk is mitigated by the fact that a trained microbiologist reviews the culture plate digital image for final assessment and result definition as also defined in the procedure manual and package insert.

4. Detection Limit:

The ability of the PhenoMATRIX to detect colonies of varying sizes was assessed as part of the analytical and clinical studies. The rates of false positive results (i.e., growth detected by PhenoMATRIX but not detected by the reference method/trained microbiologist visual inspection) and false negative results (i.e., growth not detected by PhenoMATRIX but detected by the reference method) were assessed across all studies. In the analytical studies, growth detection and morphological interpretation false positive rates ranged from 0-2% and false negative rates ranged from 0-3.2%. In the clinical studies, growth detection and morphological interpretation false positive rates ranged from 1.0-1.3% and false negative rates ranged from 0.1-0.5%. These data demonstrate that the PhenoMATRIX can accurately identify colony growth and morphology across the range of colony sizes and morphologies expected during routine clinical use. Additionally, the risk of inaccurate determination of presence of growth by PhenoMATRIX is mitigated by the requirement in the procedure manual and package insert that all digital plate images will be evaluated by trained personnel for final assessment and result definition.

5. Accuracy (Instrument):

N/A

6. Carry-Over:

N/A

B Clinical Study:

A clinical study was conducted to evaluate the PhenoMATRIX with WASPLab for automated assessment of culture plate images across three U.S. clinical laboratories. In total, the study analyzed thousands of plate images from various specimen types and culture media. PhenoMATRIX results based on digital plate images were compared against reference interpretations by experienced technicians reviewing the digital plate images (Tables 6-10).

Clinical Site A analyzed specimens containing culturable aerobic bacteria using blood agar, Columbia CNA, chocolate agar, and MacConkey agar plates. In total, 3856 plate images were analyzed at Clinical Site A. Growth detection achieved acceptable performance with positive percent agreement (PPA) ranging from 99.5% to 100% across all media types. Growth semi-quantification demonstrated overall agreement between 96.6% and 99.3%, exceeding the 90% acceptance criterion. Morphological detection for *S. aureus*-like organisms achieved 100% accuracy, and growth purity assessment reached 91.8% overall agreement, exceeding the 80% acceptance criteria threshold.

Clinical Site B evaluated urine specimens with uropathogenic and normal flora using blood agar and MacConkey agar bi-plates. In total, 418 plate images were analyzed at Clinical Site B. Both culture media achieved 100% positive and negative percent agreement (NPA) for growth

detection. Growth semi-quantification showed acceptable performance with overall agreement ranging from 93.2% to 97.1% depending on inoculum volume, exceeding the acceptance criteria.

Clinical Site C assessed clean-catch and catheter-collected urines using CHROMagar Orientation and blood agar bi-plates. In total, 1571 plate images were analyzed at Clinical Site C. Growth detection demonstrated acceptable performance with 100% PPA for both media types. Growth semi-quantification achieved 97.8% agreement for blood agar and 90.1% for CHROMagar Orientation. The Clinical Site C demonstrated robust morphological detection capabilities across multiple organism types, with most morphologies achieving greater than 95% accuracy. Semi-quantification by specific morphologies varied from 88.8% to 99.6% depending on organism complexity. Growth purity assessment exceeded expectations with 98-99.6% overall agreement. Beta-hemolysis detection achieved 100% PPA and NPA.

Across all clinical sites combined, the PhenoMATRIX clinical performance met or exceeded the predefined acceptance criteria: ≥99% for growth detection, ≥90% for semi-quantification accuracy, ≥80% for morphological detection, ≥95% for beta-hemolysis detection, and ≥80% for growth purity assessment.

Table 6. Clinical performance of growth detection obtained by the PhenoMATRIX

Site	Culture Medium	PPA Results (n/N)	Positive Percent Agreement (95% CI)	NPA Results (n/N)	Negative Percent Agreement (95% CI)
A	Blood agar	774/774	100% (99.5%-100%)	186/190	97.9% (94.7%-100%)
	Columbia CNA	718/721	99.6% (98.8%-99.9%)	240/243	98.8% (96.4%-99.6%)
	Chocolate agar	763/763	100% (99.5%-100%)	198/201	98.5% (95.7%-100%)
	MacConkey agar	181/182	99.5% (97%-99.9%)	771/782	98.6% (97.5%-99.2%)
B	Blood agar	366/366	100% (99%-100%)	52/52	100% (93.1%-100%)
	MacConkey agar	198/198	100% (98.1%-100%)	220/220	100% (98.3%-100%)
C	Blood agar	1338/1338	100% (99.7%-100%)	233/233	100% (98.4%-100%)
	CHROMagar Orientation	1376/1376	100% (99.7%-100%)	188/195	96.4% (92.8%-100%)
Total (A+B+C)	Blood agar	2478/2478	100% (99.8%-100%)	471/475	98.3% (97.2%-99.1%)
Total (A only)	Columbia CNA	718/721	99.6% (98.8%-99.9%)	240/243	98.8% (96.4%-99.6%)
Total (A only)	Chocolate agar	763/763	100% (99.5%-100%)	198/201	98.5% (95.7%-100%)
Total (A+B)	MacConkey agar	379/380	99.7% (98.4%-99.9%)	991/1002	99.0% (98.2%-99.5%)
Total (C only)	CHROMagar Orientation	1376/1376	100% (99.7%-100%)	188/195	96.4% (92.8%-100%)

Table 7. Clinical performance of growth semi-quantification obtained by the PhenoMATRIX

Site	Culture Medium	Inoculum	Colony Count Bin [CFU/mL]	Software = Reference	Software < Reference	Software > Reference	Agreement Rate
A	Blood agar	30µL	No Growth	186/190 (97.9%)	N/A	4/190 (2.1%)	97.9%
			3.33×10^{1}-3.00×10^{2}	315/316 (99.7%)	0/316 (0%)	1/316 (0.3%)	99.7%
			3.33×10^{2}-3.30×10^{3}	84/85 (98.8%)	1/85 (1.2%)	0/85 (0%)	98.8%
			3.33×10^{3}-3.30×10^{4}	93/94 (98.9%)	0/94 (0%)	1/94 (1.1%)	98.9%
			$\geq3.33\times10^{4}$	279/279 (100%)	0/279 (0%)	N/A	100%
	Columbia CNA	30µL	No Growth	240/243 (98.8%)	N/A	3/243 (1.2%)	98.8%
			3.33×10^{1}-3.00×10^{2}	121/135 (89.6%)	3/135 (2.2%)	11/135 (8.1%)	89.6%

Site	Culture Medium	Inoculum	Colony Count Bin [CFU/mL]	Software = Reference	Software < Reference	Software > Reference	Agreement Rate
			3.33×10^2-3.30×10^3	94/110 (85.5%)	3/110 (2.7%)	13/110 (11.8%)	85.5%
			$\geq3.33\times10^3$	476/476 (100%)	0/476 (0%)	N/A	100%
	Chocolate agar	30µL	No Growth	198/201 (98.5%)	N/A	3/201 (1.5%)	98.5%
			3.33×10^1-3.00×10^2	106/119 (89.1%)	0/119 (0%)	13/119 (10.9%)	89.1%
			3.33×10^2-3.30×10^3	94/110 (85.5%)	1/110 (0.9%)	15/110 (13.6%)	85.5%
			$\geq3.33\times10^3$	533/534 (99.8%)	1/534 (0.2%)	N/A	99.8%
	MacConkey agar	30µL	No Growth	771/782 (98.6%)	N/A	11/782 (1.4%)	98.6%
			3.33×10^1-3.00×10^2	16/22 (72.7%)	1/22 (4.5%)	5/22 (22.7%)	72.7%
			3.33×10^2-3.30×10^3	21/29 (72.4%)	0/29 (0%)	8/29 (27.6%)	72.4%
			$\geq3.33\times10^3$	131/131 (100%)	0/131 (0%)	N/A	100%
B	Blood agar	1µL	No Growth	40/40 (100%)	N/A	0/40 (0%)	100%
			10^3	32/35 (91.4%)	0/35 (0%)	3/35 (8.6%)	91.4%
			10^4	43/51 (84.3%)	0/51 (0%)	8/51 (15.7%)	84.3%
			$\geq10^5$	258/258 (100%)	0/258 (0%)	N/A	100%
		10µL	No Growth	12/12 (100%)	N/A	0/12 (0%)	100%
			10^2	2/2 (100%)	0/2 (0%)	0/2 (0%)	100%
			10^3	1/2 (50%)	0/2 (0%)	1/2 (50%)	50%
			$\geq10^4$	18/18 (100%)	0/18 (0%)	N/A	100%
	MacConkey agar	1µL	No Growth	205/205 (100%)	N/A	0/205 (0%)	100%
			10^3	25/33 (75.8%)	0/33 (0%)	8/33 (24.2%)	75.8%
			10^4	11/29 (37.9%)	1/29 (3.4%)	17/29 (58.6%)	37.9%
			$\geq10^5$	117/117 (100%)	0/117 (0%)	N/A	100%
		10µL	No Growth	15/15 (100%)	N/A	0/15 (0%)	100%
			10^2	2/3 (66.7%)	0/3 (0%)	1/3 (33.3%)	66.7%
			$\geq10^4$	16/16 (100%)	0/16 (0%)	N/A	100%
C	Blood agar	1µL	No Growth	233/233 (100%)	N/A	0/233 (0%)	100%
			10^3	264/273 (96.7%)	0/273 (0%)	9/273 (3.3%)	96.7%
			10^4	309/318 (97.2%)	0/318 (0%)	9/318 (2.8%)	97.2%
			10^5	226/233 (97.0%)	0/233 (0%)	7/233 (3.0%)	97.0%
			$\geq10^6$	505/514 (98.2%)	9/514 (1.8%)	N/A	98.2%
	CHROMagar Orientation	1µL	No Growth	188/195 (96.4%)	N/A	7/195 (3.6%)	96.4%
			10^3	293/321 (91.3%)	0/321 (0%)	28/321 (8.7%)	91.3%
			10^4	308/357 (86.3%)	1/357 (0.3%)	48/357 (13.4%)	86.3%
			10^5	494/530 (93.2%)	18/530 (3.4%)	18/530 (3.4%)	93.2%
			$\geq10^6$	132/168 (78.6%)	36/168 (21.4%)	N/A	78.6%

Table 8. Clinical performance of morphology labels obtained by the PhenoMATRIX

Site	Morphology Type	PPA Results (n/N)	Positive Percent Agreement (95% CI)	NPA Results (n/N)	Negative Percent Agreement (95% CI)
A	*S. aureus*-like	217/217	100% (98.3%-100%)	747/747	100% (99.5%-100%)
C	Coliform	576/579	99.5% (98.5%-100%)	992/992	100% (99.6%-100%)
	Pseudomonas-like	52/53	98.1% (90.1%-100%)	1518/1518	100% (99.7%-100%)
	S. aureus-like	145/145	100% (97.4%-100%)	1426/1426	100% (99.7%-100%)
	E. coli-like	511/511	100% (99.3%-100%)	1045/1060	98.6% (97.7%-100%)
	Enterococcus-like	470/476	98.7% (97.3%-100%)	1072/1095	97.9% (96.9%-100%)
	GBS-like	249/249	100% (98.5%-100%)	1306/1322	98.8% (98%-100%)
Total (A+C)	*S. aureus*-like	362/362	100% (98.9%-100%)	2173/2173	100% (99.8%-100%)
Total (C only)	Coliform	576/579	99.5% (98.5%-100%)	992/992	100% (99.6%-100%)
Total (C only)	*Pseudomonas*-like	52/53	98.1% (90.1%-100%)	1518/1518	100% (99.7%-100%)
Total (C only)	*E. coli*-like	511/511	100% (99.3%-100%)	1045/1060	98.6% (97.7%-100%)
Total (C only)	*Enterococcus*-like	470/476	98.7% (97.3%-100%)	1072/1095	97.9% (96.9%-100%)
Total (C only)	GBS-like	249/249	100% (98.5%-100%)	1306/1322	98.8% (98%-100%)

Table 9. Clinical performance of beta-hemolysis detection obtained by the PhenoMATRIX

Site	Culture Medium	PPA Results (n/N)	Positive Percent Agreement (95% CI)	NPA Results (n/N)	Negative Percent Agreement (95% CI)
C	Blood agar	283/283	100% (98.7%-100%)	1288/1288	100% (99.7%-100%)

Table 10. Clinical performance of growth purity obtained by the PhenoMATRIX

Site	Culture Medium	Category Agreement	Agreement Results (n/N)	Overall Agreement (95% CI)
A	Blood agar	Predominant: 385/405 (95.1%); Not Predominant: 94/117 (80.3%)	479/522	91.8% (89.1%-100%)
C	Blood agar	Predominant: 837/837 (100%); Co-predominance 2: 264/269 (98.1%); Co-predominance ≥3: 231/232 (99.6%)	1332/1338	99.6% (99%-99.8%)
	CHROMagar Orientation	Predominant: 886/890 (99.6%); Not Predominant: 383/405 (94.6%)	1269/1295	98% (97.1%-98.6%)

C Other Supportive Instrument Performance Characteristics Data:

Digital Image Evaluation

A digital image equivalency study was conducted to evaluate the accuracy and reproducibility of PhenoMATRIX digital image interpretation compared to conventional visual inspection of physical culture plates. The study tested three WASPLab instruments using the claimed culture media (blood-based, chocolate, MacConkey, CHROMagar Orientation, and bi-plates) streaked with mixed bacterial cultures simulating respiratory and urine specimens. Three operators with different experience levels (junior, intermediate, senior) interpreted both physical plates and digital images across multiple working days and incubation times, with results compared using standardized questionnaires covering microbial growth detection, quantification, and phenotypical characteristics.

The results demonstrated agreement between digital image interpretation and physical plate inspection, with overall agreement consistently exceeding 95% across all tested parameters. Statistical analysis using χ^2 tests showed no significant differences in interpretation accuracy based on operator experience level, WASPLab instrument, working day, or incubation time,

confirming the system's reproducibility. The study demonstrates that WASPLab digital images allow accurate and consistent interpretation of culture plates by trained technicians after PhenoMATRIX classification, successfully meeting all predefined acceptance criteria for growth detection, semi-quantification, and isolate differentiation based on morphological characteristics including hemolysis patterns and colony appearance.

Validation of the customizable classification rules

A study was conducted to evaluate the performance of PhenoMATRIX when sorting digital images of culture plates according to user-defined classification rules. The software can use customizable classification rules based on laboratory information system (LIS) data and image analysis results to group plate images into digital folders for review by trained microbiologists. To assess its clinical performance, simulators were deployed at three U.S. clinical laboratories, each configured with specific classification rules that replicated the laboratory's standard decision-making workflow for microbial workup.

The study analyzed thousands of culture plate images across different configurations, comparing PhenoMATRIX classifications against microbiologist digital image reviews. Results demonstrated strong classification agreement rates: 98.3% for Clinical Site A (aerobic bacteria cultures), 98.1% for Clinical Site B (urine specimens), and 92.6% for Clinical Site C (clean-catch and catheter-collected urines with demographic data integration). All Clinical Sites exceeded the predefined acceptance criterion of >92% overall agreement. Only one classification category showed agreement below 80%: the “2 isolates” (co-predominance of 2 isolates) category in Clinical Site C achieved 64.4% agreement, while three other categories in the same Clinical Site showed 80-90% agreement, all involving complex mixed culture interpretations. This risk is mitigated by the fact that a trained microbiologist reviews the culture plate digital image for final assessment and result definition. The study demonstrated that PhenoMATRIX successfully classifies plate images according to user defined clinical laboratory criteria. Performance varied based on classification rule complexity. Simple cases like no growth or pure cultures achieved highest concordance, while mixed cultures showed lower agreement, but this was acceptable given that all PhenoMATRIX results require final review by trained personnel.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.