

Protocols for Laboratory Verification of Performance of the BioFire® FilmArray® Pneumonia Panel *plus*

Laboratory Protocols for Use with a ZeptoMetrix NATtrol™ Verification Panel

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Purpose

The Clinical Laboratory Improvement Amendments (CLIA), passed in 1988, establishes quality standards for all laboratory testing to ensure the accuracy and reliability of patient test results, regardless of where the test is performed. The CLIA regulations include a requirement for verifying the performance specifications of unmodified, moderate complexity tests cleared or approved by the FDA.

This document provides examples of verification procedures to assist your laboratory in developing a protocol for the verification of the BioFire Pneumonia Panel *plus* performance on BioFire® FilmArray® Systems as required by CLIA. Two possible verification schemes, compatible with the BioFire Pneumonia Panel *plus*, have been designed using non-clinical specimens. Each verification scheme provides positive and negative tests for each organism detected by the BioFire Pneumonia Panel *plus*, as well as semi-quantitative bin results for bacterial interpretations to demonstrate the ability to distinguish between relative abundance levels in a single test. Middle East Respiratory Coronavirus (MERS-CoV) is provided as two inactive recombinants, each targeting a unique assay on the BioFire Pneumonia Panel *plus*. The pooling scheme provided will give equivocal detections for MERS-CoV and is designed to reduce false positive detections. This scheme may be easily modified or expanded to meet specific criteria.

Day-to-day variation is evaluated by testing each sample on two separate days. To evaluate user-to-user variation, multiple laboratory technicians may test the same sample. In addition, testing patient samples representative of the different types expected to be tested by the laboratory (ex. Sputum, ETA, BAL) for verification of the performance of the BioFire Pneumonia Panel *plus* should be done under the guidance of the Laboratory Director, but is not described here.

As per the CLIA regulation, the Laboratory Director is ultimately responsible for ensuring that verification procedures meet the appropriate standards for CLIA and applicable laboratory accrediting agencies.

BioFire Intended Use

The BioFire® FilmArray® Pneumonia Panel *plus* is a multiplexed nucleic acid test intended for use with BioFire® FilmArray® 1.5, BioFire® FilmArray® 2.0, or BioFire® FilmArray® Torch systems for the simultaneous detection and identification of nucleic acids from Middle East Respiratory Syndrome Coronavirus (MERS-CoV) and multiple respiratory viral and bacterial nucleic acids, as well as select antimicrobial resistance genes, in sputum-like specimens (induced or expectorated sputum, or endotracheal aspirates) or bronchoalveolar lavage (BAL)-like specimens (BAL or mini-BAL) obtained from individuals meeting MERS-CoV clinical and/or epidemiological criteria.

Testing with BioFire Pneumonia Panel *plus* should not be performed unless the patient meets clinical and/or epidemiologic criteria for testing suspected MERS-CoV specimens. This includes: clinical signs and symptoms associated with MERS-CoV infection, contact with a probable or confirmed MERS-CoV case, history of travel to geographic locations where MERS-CoV cases were detected, or other epidemiological links for which MERS-CoV testing may be indicated.

The following bacteria are reported semi-quantitatively with bins representing approximately 10⁴, 10⁵, 10⁶, or ≥10⁷ genomic copies of bacterial nucleic acid per milliliter (copies/mL) of specimen, to aid in estimating relative abundance of nucleic acid from these common bacteria within a specimen:

Bacteria reported with bins of 10 ⁴ , 10 ⁵ , 10 ⁶ , or ≥10 ⁷ copies/mL		
<i>Acinetobacter calcoaceticus-baumannii</i> complex	<i>Klebsiella oxytoca</i>	<i>Serratia marcescens</i>
<i>Enterobacter cloacae</i> complex	<i>Klebsiella pneumoniae</i> group	<i>Staphylococcus aureus</i>
<i>Escherichia coli</i>	<i>Moraxella catarrhalis</i>	<i>Streptococcus agalactiae</i>
<i>Haemophilus influenzae</i>	<i>Proteus</i> spp.	<i>Streptococcus pneumoniae</i>
<i>Klebsiella aerogenes</i>	<i>Pseudomonas aeruginosa</i>	<i>Streptococcus pyogenes</i>

The following atypical bacteria, viruses, and antimicrobial resistance genes are reported qualitatively:

Atypical Bacteria		
<i>Chlamydia pneumoniae</i>	<i>Legionella pneumophila</i>	<i>Mycoplasma pneumoniae</i>
Viruses		
Middle East Respiratory Syndrome Coronavirus		
Adenovirus	Human Rhinovirus/Enterovirus	Parainfluenza Virus
Coronavirus	Influenza A	Respiratory Syncytial Virus
Human Metapneumovirus	Influenza B	
Antimicrobial Resistance Genes		
CTX-M	NDM	<i>mecA/C</i> and MREJ
IMP	OXA-48-like	
KPC	VIM	

The complete intended use statement and additional information about the use of the BioFire® FilmArray® System can be found in the *FilmArray Pneumonia Panel plus Instructions for Use*.

Performance Verification: Overview

Two different examples of performance verification procedures are described. Both procedures evaluate the performance of each assay on the BioFire Pneumonia Panel *plus* using samples in the presence of synthetic or clinical matrix background. A Synthetic Matrix Protocol describes the verification of the BioFire Pneumonia Panel *plus* performance in either a synthetic background (Negative) provided with the ZeptoMetrix NATrol control organisms or in Artificial

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Brochoalveolar Lavage Matrix (aBAL). Preparation of aBAL is described in the Performance Verification: Protocols section below. A Clinical Matrix Protocol evaluates the performance of each assay on the BioFire® FilmArray® Pneumonia Panel *plus* in a clinical specimen matrix (induced or expectorated sputum, endotracheal aspirates or bronchoalveolar lavage (BAL)-like specimens (BAL or mini-BAL). These protocols are examples of procedures to assist your laboratory in developing a protocol for the verification of BioFire Pneumonia Panel *plus* performance on BioFire Systems.



Note: It is important to characterize aBAL or clinical matrix specimens for Pneumonia Panel *plus* targets by screening the specimen on the BioFire Pneumonia Panel *plus* prior to starting the verification procedure. The optimal clinical matrix specimen will be negative for all analytes tested on the BioFire Pneumonia Panel *plus*. If a negative specimen is not available, note the positives and take that into account when reviewing results.

A BioFire® FilmArray® System is defined as all BioFire® FilmArray® Instruments or modules that are connected to and controlled by a single computer system. If the laboratory director chooses not to perform the entire verification protocol on each individual instrument, it is advised that test replicates are evenly distributed among the instruments or modules.

The procedures have been designed to take advantage of the multiplex nature of the BioFire Pneumonia Panel *plus*. Verification testing efficiency is maximized by evaluating multiple target organisms in a single test run. Each procedure described below will generate multiple positive and negative detections for each of the BioFire Pneumonia Panel *plus* assays. In addition, semi-quantitative bacteria are reported with relative bin values of 10^4 , 10^5 , 10^6 , or $\geq 10^7$ copies/mL. The protocols described will generate detections in high bins (10^6 - $\geq 10^7$) and low bins (10^4 - 10^5) to demonstrate the ability to differentiate organism levels in a single test run. Middle East Respiratory Coronavirus (MERS-CoV) is provided as two inactive recombinants, each targeting a unique assay on the BioFire Pneumonia Panel *plus*. The pooling scheme provided will give equivocal detections for MERS-CoV and is designed to reduce false positive detections. The procedures were developed using a Pneumonia Verification Panel available from ZeptoMetrix™ Corporation, Buffalo, NY (NATPPA-BIO + NATPPQ-BIO + NATMR-BIO).



Note: Feedback from discussions with federal regulatory bodies was used in the development of the verification schemes described here and is designed to reduce possible false positive MERS-CoV detections as a result of control material. MERS-CoV is provided as 2 unique recombinants that target specific assays on the BioFire Pneumonia Panel *plus*. The pooling scheme described in Table 3 will result in Equivocal detections for MERS-CoV in pool 4 and 5. Documentation of MERS-CoV equivocal detections in both pool 4 and pool 5 infers a MERS-CoV positive detection.

Clinical/patient samples may be used in place of or in addition to the verification schemes described here in order to assess clinical sensitivity/specificity and sample matrix effects as part of the performance verification of the BioFire Pneumonia Panel *plus*.

Table 1. Overview of Verification Protocols

Verification Protocol	Organisms per Pool	Number of Sample Pools	Replicates per Sample Pool	Pouches Required	Expected Positive Results	Expected Negative Results	Targeted Low Bin Results ^a	Targeted High Bin Results ^b	Approximate Days of Testing ^c
Example 1: Synthetic Matrix protocol	5, 6, or 7	5	4	20	4 per organism	16 per organism	32	36	4
Example 2: Clinical Matrix protocol	5, 6, or 7	5	4	20	4 per organism	16 per organism	32	36	4

^a Targeted Low Bin Results represents bacterial detections in the semi-quantitative bins of 10⁴-10⁵ copies/mL.

^b Targeted High Bin results represent bacterial detections in the semi-quantitative bins of 10⁶-≥10⁷ copies/mL.

^cThe approximate number of days for testing assumes a BioFire® FilmArray® System configured with one instrument/module.

Performance Verification: Materials

The following materials may be used to perform verification procedures:

Table 2. Recommended materials for verification protocol

Material	Part Number
BioFire® FilmArray® Pneumonia Panel <i>plus</i> Kit (30 tests per kit)	BioFire Diagnostics, LLC (RFIT-ASY-0143)
BioFire® FilmArray® Pneumonia Panel <i>plus</i> Instruction for Use	BioFire Diagnostics, LLC (RFIT-PRT-0894)
BioFire® FilmArray® Pneumonia Panel <i>plus</i> Quick Guide	BioFire Diagnostics, LLC (RFIT-PRT-0896)
Control Organism ^a	ZeptoMetrix NATPPA-BIO, NATPPQ-BIO, and NATMR-BIO
5 mL sample tubes	Various manufacturers
Transfer pipettes	VWR Part # 13-711-43 (or similar)
Human genomic DNA (1mL of 0.2mg/mL) or equivalent ^b	Roche 11691112001
Sterile normal saline (0.85 - 0.9% sodium chloride) ^b	Medline PCS1650 or equivalent
10 mL sample tubes ^b	Various manufacturers

^aAny appropriate source of organism may be used for verification of any or all of the assays in the BioFire® FilmArray® Pneumonia Panel. However, when alternate organism sources are used (i.e. not the ZeptoMetrix material), the sample volumes or pooling schemes suggested in the examples below may need to be adjusted.

^bOptional; needed for preparation aBAL matrix

Performance Verification: Protocols

Synthetic Matrix Protocol

The Synthetic Matrix Protocol evaluates BioFire Pneumonia Panel *plus* performance in a synthetic matrix. Sample material (ZeptoMetrix NATPPA-BIO, NATPPQ-BIO, and NATMR-BIO) is pooled and added to an equal volume of synthetic matrix. The proposed organism pooling scheme (Table 3) should be

followed to obtain the expected results for each assay in a time and resource-efficient manner.



Note: Dilution of ZeptoMetrix Pneumonia *plus* Verification Panel organisms beyond levels proposed in these guidelines may lead to inconsistent results and is not recommended.

The Synthetic Matrix Protocol can be followed to test 20 pooled samples, providing 4 positive results and 16 negative results per assay, as well as 32 low bin and 36 high bin detections. Sample distribution in the targeted low and high bins are approximate and bin variation may occur due to sampling differences and run variability. The pooling scheme provides sufficient volume for testing more samples if desired. The number of samples tested per day should be determined by the individual laboratory. The testing scheme can be modified to run more samples per day based on the number of instruments configured on the BioFire® FilmArray® System.



Note: Target bin levels are relative and may vary between test runs.

Pooled samples may be stored overnight (or up to 3 days) at refrigeration temperature (2–8°C) for subsequent testing to evaluate day-to-day variation. To evaluate user-to-user variation, multiple laboratory technicians may perform testing.

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Table 3. Proposed Organism Pooling Scheme

Organism	Target Bin Level ^a	Approximate Organism Volume	Approximate Volume of Synthetic or Clinical Matrix	Approximate Final Volume of Pool
Pool 1				
<i>Acinetobacter baumannii</i>	Low	0.2 mL	1.2 mL	2.4 mL
<i>Enterobacter cloacae</i>	High	0.2 mL		
<i>Escherichia coli</i> (IMP)	Low	0.2 mL		
<i>Klebsiella pneumoniae</i> Z138 (OXA-48-like, CTX-M)	High	0.2 mL		
<i>Proteus mirabilis</i>	Low	0.2 mL		
<i>Serratia marcescens</i>	High	0.2 mL		
Pool 2				
<i>Klebsiella aerogenes</i> (aka <i>Enterobacter aerogenes</i>)	High	0.2 mL	1.0 mL	2.0 mL
<i>Klebsiella oxytoca</i>	Low	0.2 mL		
<i>Klebsiella pneumoniae</i> KPC-2 (KPC)	High	0.2 mL		
<i>Pseudomonas aeruginosa</i> (VIM)	Low	0.2 mL		
<i>Streptococcus pyogenes</i>	High	0.2 mL		
Pool 3				
<i>Haemophilus influenzae</i>	High	0.2 mL	1.2 mL	2.4 mL
<i>Klebsiella pneumoniae</i> Z460 (NDM/ CTX-M)	High	0.2 mL		
<i>Moraxella catarrhalis</i>	Low	0.2 mL		
<i>Staphylococcus aureus</i> (<i>mecA/C</i> and MREJ)	High	0.2 mL		
<i>Streptococcus agalactiae</i>	Low	0.2 mL		
<i>Streptococcus pneumoniae</i>	Low	0.2 mL		
Pool 4				
Adenovirus Type 31	N/A	0.2 mL	1.4 mL	2.8 mL
Coronavirus NL63	N/A	0.2 mL		
Influenza A H3	N/A	0.2 mL		
MERS1 Recombinant	N/A	0.2 mL		
Parainfluenza virus Type 1	N/A	0.2 mL		
Respiratory Syncytial Virus (RSV) A2	N/A	0.2 mL		
<i>Legionella pneumophila</i>	N/A	0.2 mL		
Pool 5				
Adenovirus Type 3	N/A	0.2 mL	1.4 mL	2.8 mL
Human Metapneumovirus 8	N/A	0.2 mL		
Human Rhinovirus 1 A	N/A	0.2 mL		
Influenza B	N/A	0.2 mL		
MERS2 Recombinant	N/A	0.2 mL		
<i>Chlamydia pneumoniae</i>	N/A	0.2 mL		
<i>Mycoplasma pneumoniae</i>	N/A	0.2 mL		

^aTarget bin levels are relative and may vary between test runs. Low Bin results refer to relative abundance of 10⁴-10⁵ copies/mL; High Bin results refer to relative abundance of 10⁶-≥10⁷ copies/mL.

Synthetic Matrix Protocol Example

The estimated total time for completion for this verification example is 4 days for a BioFire® FilmArray® System configured with 1 instrument/module. A proposed organism pooling scheme is presented above in Table 3. Refer to Figure 1 for the suggested workflow.



Note: It is important to prepare only the number of sample pools that will be tested within 3 days of preparation. The suggestion to prepare 3 sample pools is based on testing up to 6 pouches per day. The number of samples prepared may be increased or decreased based on the laboratory's work schedule and number of instruments connected within a BioFire® FilmArray® System.

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Day 1

1. Organize materials needed (Table 2). If using Artificial Brochoalveolar Lavage Matrix (aBAL) - follow the optional protocol below to prepare the synthetic matrix before starting Step 2.
2. Prepare one sample pool (i.e. pool #1) from ZeptoMetrix NATPPQ-BIO and NATPPA-BIO control material. The MERS1 and MERS2 recombinants are contained in Zeptomatrix NATMR-BIO and should be used to prepare Pools 4 and 5. Organism vials should be well mixed prior to preparing each pool. Refer to Table 3 for example organism pooling schemes and specific volumes for each pool.
 - a. Using Table 3 as a guide, transfer 1.0, 1.2, or 1.4 mL of Synthetic Matrix (either ZeptoMetrix Negative or aBAL) into a tube large enough to hold the entire organism pool volume (at least 3 mL).
 - b. Use a transfer pipette to remove the entire contents of the ZeptoMetrix organism vial (approximately 0.2 mL) and transfer to the larger tube containing Synthetic Matrix.
 - c. Repeat with the second (and subsequent) organisms to combine the appropriate organisms for each pool into a single tube (approximately 2.0 mL total volume for five organisms, 2.4 mL for six organisms, or 2.8 mL for seven organisms).
 - d. Ensure the pooled sample is well mixed prior to removing a sample for testing.
 - e. Refrigerate samples (2–8°C) for up to 3 days for the evaluation of day-to-day variation.
3. Prepare and test 2 samples from a single pool (i.e. pool 1). The duplicate samples should be tested in a single day by different users.



Note: For each sample, follow instructions in the *BioFire® FilmArray® Pneumonia Panel plus Instructions for Use* and the *BioFire® FilmArray® Pneumonia Panel plus Quick Guide* for pouch preparation, pouch hydration, sample loading, and sample testing.

4. Repeat Steps 2 and 3 for remaining sample pools to be tested that day.



Note: The proposed organism pooling scheme (Table 3) provides sufficient material for running samples as described in Figure. 1. The volume is sufficient for testing more samples if desired.

Day 2

To evaluate day-to-day variation, test the remaining volume of the sample pools prepared on Day 1 by repeating Step 3 above.

Day 3

Prepare 2 new sample pools (i.e. pools #4 and #5) as described in Step 2. Test samples according to Step 3 above.

Day 4

To evaluate day-to-day variation, test the samples prepared on Day 3 by repeating Step 2 and 3 above.

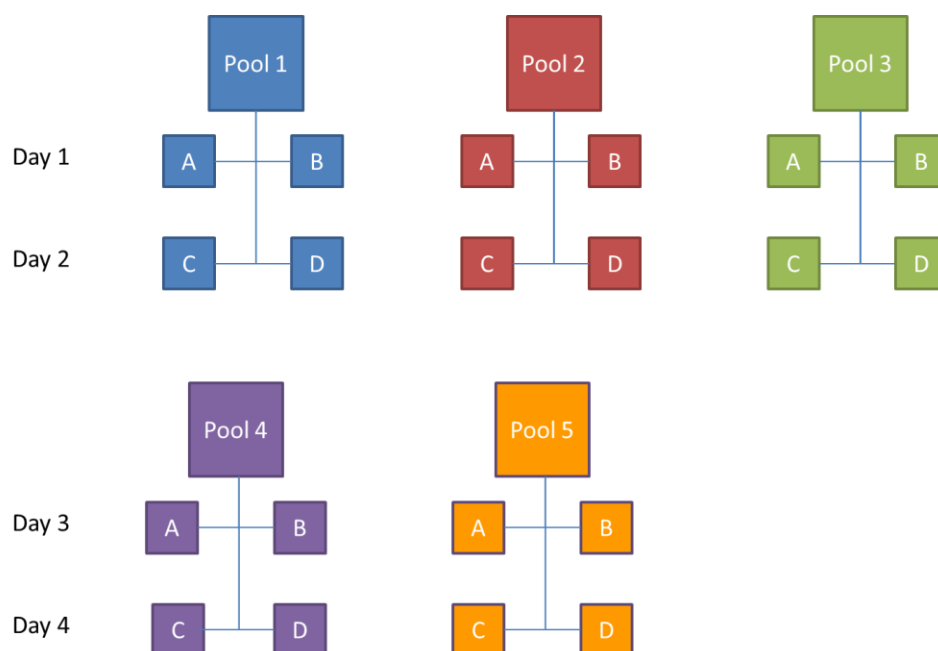
Optional Protocol: Preparation of Artificial Brochoalveolar Lavage Matrix (aBAL)

1. Thoroughly decontaminate the work area with 10% bleach followed by water wipes.
2. Prepare a 20 ng/uL solution of Human genomic DNA (hgDNA) by adding 1.0 mL hgDNA stock solution (0.2mg/mL) to 9 mL sterile saline in a 10 mL tube.
3. Mix by vortexing or inversion.
4. Characterize aBAL matrix by running a BioFire® FilmArray® Pneumonia Panel *plus* test prior to performing verification schemes. The BioFire Pneumonia Panel *plus* should test negative for all analytes on the panel.
5. Aliquots of aBAL matrix may be stored refrigerated at 4°C for up to 3 days or stored frozen at ≤ -70°C.



Note: It is important to characterize aBAL or clinical matrix specimens for Pneumonia Panel targets by screening the specimen on the BioFire Pneumonia Panel *plus* prior to starting the verification procedure. The optimal clinical matrix specimen will be negative for all analytes tested on the BioFire Pneumonia Panel *plus*. If a negative specimen is not available, note the positives and take that into account when reviewing results.

Figure 1. Workflow for Synthetic Matrix Protocol



Clinical Matrix Protocol

The Clinical Matrix Protocol evaluates BioFire® FilmArray® Pneumonia Panel *plus* performance in a clinical specimen matrix. The laboratory will require a clinical specimen (induced or expectorated sputum, endotracheal aspirates or bronchoalveolar lavage (BAL)-like specimens (BAL or mini-BAL) appropriate for the laboratory's verification needs and in a volume sufficient for testing as described in Table 3. The number of different sample types to be tested is up to the discretion of the laboratory director; multiple specimens may be used, if necessary. Clinical specimens should be screened in the BioFire Pneumonia Panel *plus* in order to characterize the sample.



Note: It is important to characterize aBAL or clinical matrix specimens for Pneumonia Panel *plus* targets by screening the specimen on the BioFire Pneumonia Panel *plus* prior to starting the verification procedure. The optimal clinical matrix specimen will be negative for all analytes tested on the BioFire Pneumonia Panel *plus*. If a negative specimen is not available, note the positives and take that into account when reviewing results.

Sample material (ZeptoMetrix NATPPA-BIO, NATPPQ-BIO, and NATMR-BIO) is pooled and added to an equal volume of clinical matrix. The proposed organism pooling scheme (Table 3) should be followed to obtain the expected results for each assay in a time and resource-efficient manner. Specimen consistency may make accurate measurement difficult, but care should be taken to try to add the volume indicated.



Note: Dilution of ZeptoMetrix Pneumonia *plus* Verification Panel organisms beyond levels proposed in these guidelines may lead to inconsistent results and is not recommended.

The Clinical Matrix Protocol can be followed to test 20 pooled samples, providing 4 positive results and 16 negative results per assay, as well as 32 low bin and 36 high bin detections. Sample distribution in the targeted low and high bins are approximate and bin variation may occur due to sampling differences and run variability. The pooling scheme provides sufficient volume for testing more samples if desired. The number of samples tested per day should be determined by the individual laboratory. The testing scheme can be modified to run more samples per day based on the number of instruments configured on the BioFire® FilmArray® System.



Note: Target bin levels are relative and may vary between test runs.

Pooled samples may be stored overnight at refrigeration temperature (2–8°C) for subsequent testing to evaluate day-to-day variation; but all tests should be completed within a day. To evaluate user-to-user variation, multiple laboratory technicians may perform testing.



Note: Clinical specimens may contain inhibitors and enzymes which may degrade the organism mixture and lead to unpredictable results. It is important to prepare the organism pools and test samples within a day to achieve expected results.

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Clinical Matrix Protocol Example

The estimated total time for completion for this verification example is 4 days for a BioFire® FilmArray® System configured with 1 instrument/module. A proposed organism pooling scheme is presented above in Table 3. Refer to Figure 2 for the suggested workflow.



Note: It is important to prepare only the number of sample pools that will be tested within a day of preparation. The suggestion to prepare 3 sample pools is based on testing up to 6 pouches per day. The number of samples prepared may be increased or decreased based on the laboratory's work schedule and number of instruments connected within a BioFire System.

Day 1

1. Organize materials needed (Table 2.) Clinical specimens should be screened in the BioFire® FilmArray® Pneumonia Panel *plus* in order to characterize the sample prior to preparing pools.
2. Prepare one sample pool (i.e. pool #1) from ZeptoMetrix NATPPQ-BIO and NATPPA-BIO control material. The MERS1 and MERS2 recombinants are contained in NATMR-BIO and should be used to prepare Pools 4 and 5. Organism vials should be well mixed prior to preparing each pool. Refer to Table 3 for example organism pooling schemes and specific volumes for each pool.
 - a. Using Table 3 as a guide, transfer 1.0, 1.2, or 1.4 mL of Clinical Matrix into a tube large enough to hold the entire organism pool volume (at least 3 mL). Care should be taken to transfer the correct volume. Specimen consistency may make accurate measurement difficult, but care should be taken to try to add the volume indicated. Organism to matrix ratios that differ from recommendations in Table 3 may affect detection results.
 - b. Use a transfer pipette to remove the entire contents of the ZeptoMetrix organism vial (approximately 0.2 mL) and transfer to the larger tube containing clinical specimen matrix.
 - c. Repeat with the second (and subsequent) organisms to combine the appropriate organisms for each pool into a single tube (approximately 2.0 mL total volume for five organisms, 2.4 mL for six organisms, and 2.8 mL for seven organisms).
 - d. Ensure the pooled sample is well mixed prior to removing a sample for testing.
 - e. Refrigerate samples (2–8°C) for up to a day for the evaluation of day-to-day variation.
3. Prepare and test 2 samples from a single pool (i.e. pool 1). The duplicate samples should be tested in a single day by different users.



Note: For each sample, follow instructions in the *BioFire® FilmArray® Pneumonia Panel plus Instructions for Use* and the *BioFire® FilmArray® Pneumonia Panel plus Quick Guide* for pouch preparation, pouch hydration, sample loading, and sample testing.

4. Repeat Steps 2 and 3 for remaining sample pools to be tested that day.

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Note: The proposed organism pooling scheme (Table 3) provides sufficient material for running samples as described in Figure. 2. The volume is sufficient for testing more samples if desired.

Day 2

To evaluate day-to-day variation, test the remaining volume of the sample pools prepared on Day 1 by repeating Step 3 above.

Day 3

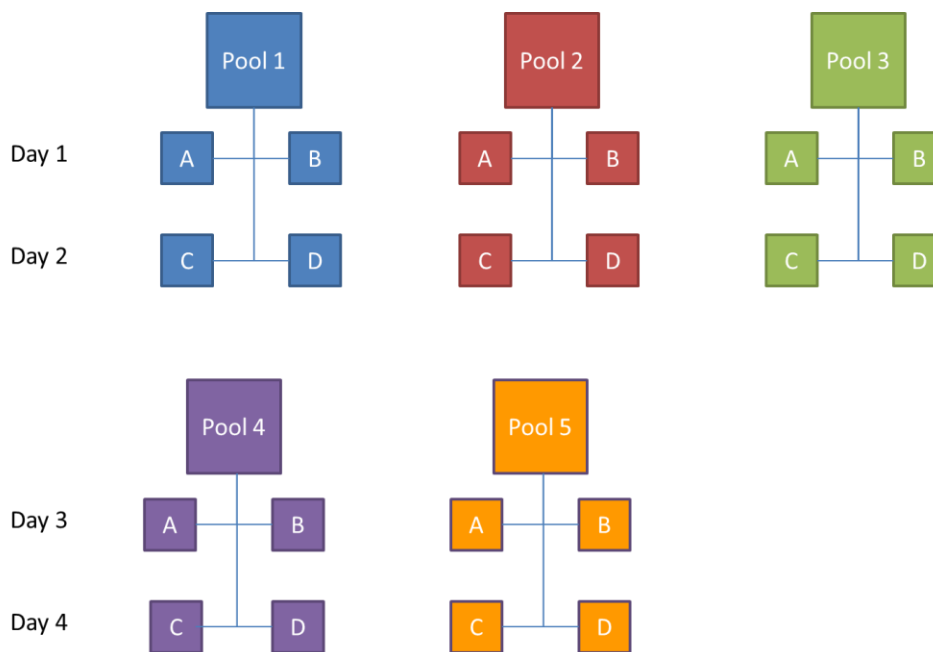
Prepare 2 new sample pools (i.e. pools #4 and #5) as described in Step 2. Test samples according to Step 3 above.

Day 4

To evaluate day-to-day variation, test the samples prepared on Day 3 by repeating Step 2 and 3 above.

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Figure 2. Workflow for Clinical Matrix Protocol



Expanding the protocols

The protocols described above can be expanded by increasing the number of tests from each of the organism pools. Each organism pool contains more than 2 mL, which is enough material to complete many tests for each pool.

Verification of Loaner, Repaired, and Permanent Replacement Instruments

If it becomes necessary to verify the performance of a loaner, repaired, or permanent replacement instrument, the following protocol may serve as a guideline but should be verified by the Laboratory Director.

1. Select a few specimens and/or proficiency samples (any combination of positives and negatives) previously tested on the BioFire® FilmArray® Pneumonia Panel *plus*. The Laboratory Director should determine the appropriate number of samples to test. Proficiency samples should not be pooled or diluted.
2. Select a set of controls that verify detection of all targets on the BioFire Pneumonia Panel *plus*.
3. Test the selected specimens/samples/controls on the loaner, repaired, or permanent replacement instrument and document the results.

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Technical Support Contact Information

BioFire is dedicated to providing the best customer support available. If you have any questions or concerns about this process, please contact the FilmArray Technical Support team for assistance.

BioFire Technical Support

Email: support@biofiredx.com

Phone: +1-801-736-6354, select Option 5

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BioFire® FilmArray® Pneumonia Panel *plus* Verification Record Page 1

System Serial # _____

BioFire® FilmArray® Pneumonia Panel *plus*, Kit Part #: _____

Lot #: _____

Organism/Sample Source and Lot #: _____

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Verification Record: Bacteria Reported in Bins

Organism	Representative Strains	System Serial #	Was the Organism Detected?	# of Positives	# of Negatives	Bin (copies/mL)				# Days Tested	# Users	Matrix Type Tested	Patient Samples Tested?
						Low Bin		High Bin					
						# 10 ⁴	# 10 ⁵	# 10 ⁶	# ≥10 ⁷				
Bacteria													
<i>Acinetobacter calcoaceticus-baumannii</i> complex			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Enterobacter cloacae</i> complex			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Escherichia coli</i> (with IMP call)			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Haemophilus influenzae</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Klebsiella aerogenes</i> (aka <i>Enterobacter aerogenes</i>)			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Klebsiella oxytoca</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Klebsiella pneumoniae</i> group	KPC-2 (with KPC call)		<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
	Z138 (with CTX-M and OXA-48 like calls)		<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
	Z460 (with CTX-M and NDM calls)		<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Moraxella catarrhalis</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Proteus</i> spp.			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Pseudomonas aeruginosa</i> (with VIM call)			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Serratia marcescens</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Staphylococcus aureus</i> (with <i>mecA/C</i> and MREJ calls)			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Streptococcus agalactiae</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Streptococcus pneumoniae</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
<i>Streptococcus pyogenes</i>			<input type="checkbox"/> Yes										
			<input type="checkbox"/> No										
Subtotal of Bin Detections at 10 ⁴ , 10 ⁵ , 10 ⁶ and ≥10 ⁷				N/A	N/A								
Total number of Positives, Negatives, and High and Low Bin Detections				#Positives	#Negatives	Low Bin Total		High Bin Total		N/A	N/A	N/A	N/A

BioFire® FilmArray® Pneumonia Panel *plus* Verification Record- Page 2

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System Serial # _____

BioFire® FilmArray® Pneumonia Panel *plus*, Kit Part #: _____

Lot #: _____

Organism/Sample Source and Lot #: _____

Verification Record: Atypical Bacteria and Viruses

Organism	Representative Strains	System Serial #	Was the Organism Detected?	Number of Positives	Number of Negatives	Bin (copies/mL)				# Days Tested	# Users	Matrix Type Tested	Patient Samples Tested?
						# 10 ⁴	# 10 ⁵	# 10 ⁶	# >10 ⁷				
Atypical Bacteria													
<i>Chlamydia pneumoniae</i>			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
<i>Legionella pneumophila</i>			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
<i>Mycoplasma pneumoniae</i>			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Viruses													
Adenovirus	Type 3		<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
	Type 31		<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Coronavirus			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Human Metapneumovirus			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Human Rhinovirus/Enterovirus			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Influenza A			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Influenza B			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Middle East Respiratory Coronavirus*	MERS1		<input type="checkbox"/> Yes			N/A	N/A	N/A	N/A				
	MERS2		<input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Parainfluenza virus			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Respiratory Syncytial Virus			<input type="checkbox"/> Yes <input type="checkbox"/> No			N/A	N/A	N/A	N/A				
Total number of Positives and Negatives						N/A	N/A	N/A	N/A	N/A	N/A		N/A

*MERS 1 and MERS2 tested in separate pools will each give an Equivocal detection. A MERS Equivocal detection in Pool 4 and Pool 5 infers a Positive detection.