Performance Assessment of an Independent Meningitis Control Panel Using the BIOFIRE® FILMARRAY® system

Introduction

Central nervous system (CNS) infections, including meningitis and encephalitis, represent acute medical emergencies with high mortality and long-term sequelae, especially in neonates, children, and immunocompromised patients. These infections may be caused by a wide range of pathogens, **bacteria**, **viruses**, and **fungi,** and early, accurate etiological diagnosis is critical for guiding appropriate treatment and improving prognosis.

Multiplex molecular assays such as the **BIOFIRE® FILMARRAY® 2.0 Meningitis / Encephalitis (ME) Panel** are widely adopted in emergency and point-of-care (POC) settings to provide **rapid, simultaneous detection of multiple CNS pathogens** directly from cerebrospinal fluid (CSF) in under two hours. However, maintaining diagnostic reliability requires routine use of **external quality controls** that are independent from the assay manufacturer and that **cover all potential targets** in the panel.

The **AMPLIRUN® TOTAL MENINGITIS CONTROL PANEL (Ref. MBTC025-R, Vircell)** is a third-party, external quality control specifically developed to challenge molecular assays for meningitis and encephalitis. It contains inactivated whole microorganisms (not just nucleic acids), suspended in a lyophilized matrix enriched with human cells to simulate the biological complexity of a true CSF sample. This enhances analytical realism and allows complete process monitoring, from nucleic acid extraction to amplification and detection, across a variety of platforms.

The purpose of this technical note is to demonstrate the compatibility and performance of the **AMPLIRUN® TOTAL MENINGITIS CONTROL PANEL** when used with the **BIOFIRE® FILMARRAY® 2.0 Meningitis / Encephalitis (ME) Panel**, the most widely used POC platform for rapid syndromic diagnosis of CNS infections.

Materials and methods

Control Description and Presentation

The **AMPLIRUN® TOTAL MENINGITIS CONTROL PANEL** consists of two vials:

- **Vial 1** (Bacterial/Fungal targets): Contains whole, inactivated cells of:
  - *Escherichia coli K1*
  - *Haemophilus influenzae*
  - *Listeria monocytogenes*
  - *Neisseria meningitidis serogroup C*
  - *Streptococcus agalactiae*
  - *Streptococcus pneumoniae*
  - *Cryptococcus neoformans*
- **Vial 2** (Viral targets): Contains inactivated viral particles of:
  - Cytomegalovirus
  - Enterovirus
  - Herpes Simplex virus 1 & 2
  - Human Herpesvirus 6
  - Human Parechovirus 1
  - Varicella Zoster virus

Each vial includes **lyophilized material** in **monodose format**, prepared at analytically challenging concentrations. Human cells are also incorporated into the lyophilized matrix to simulate real CSF.

- **Presentation:** 10 vials (5 of each type).
- **Reconstitution volume:** 400 µL per vial.
- **Shelf life:** 30 months at 2–8 °C.
- **Intended use:** External run control for validation, quality assurance, and accreditation compliance.
- **Compatibility:** Universal use for nucleic acid-based assays. Verified on BIOFIRE® FILMARRAY® 2.0 System.

Experimental Procedure

Each vial was reconstituted with 400 µL of nuclease-free water following manufacturer instructions. Reconstituted controls were tested using the **BIOFIRE® FILMARRAY® 2.0 System***, a fully automated, multiplex real-time PCR platform designed for syndromic testing directly using 200 µL in each case. The system integrates nucleic acid extraction, reverse transcription, amplification, and detection into a single, closed pouch workflow, delivering results in approximately one hour. See figure 1.

In the case of combined vials 1 + 2 of MBTC025, 400 µl of nuclease-free water was used to reconstitute vial 1 and then vial 2. A total of 200 µl of the resulting mixture was used for testing, as required by the assay.

Three test conditions were evaluated:

1. **Vial 1 only** – Sample ID: 25MBTC025101
2. **Vial 2 only** – Sample ID: 25MBTC025201
3. **Combined vials 1 + 2** – Sample ID: 25MBTC025101+25MBTC025201 (equal volumes mixed prior to loading)

Figure 1: BIOFIRE® FILMARRAY® 2.0 System

Each sample was processed using the **Meningitis / Encephalitis (ME) Panel – IVD v1.4** and **CSF protocol v3.1**, according to the manufacturer's standard operating procedure. Results were automatically generated and reviewed upon completion.

Results

In all three test conditions—**Vial 1**, **Vial 2**, and **Combined Vials 1 + 2**—the BIOFIRE® FILMARRAY® 2.0 System successfully detected **each and every pathogen present in the respective vials**, with **no false negatives or undetected targets**. Internal controls were valid in all runs, confirming correct system performance and sample processing.

These findings confirm that:

- All **bacterial and fungal targets** were detected in Vial 1.
- All **viral targets** were detected in Vial 2.
- When both vials were combined, the system simultaneously detected the **full panel of 14 pathogens**, spanning bacterial, viral, and fungal, without interference or signal loss.

BIOFIRE® FILMARRAY® reports are attached to this technical note, confirming correct target detection in both combined and individual vial tests.

Conclusion

The **AMPLIRUN® TOTAL MENINGITIS CONTROL PANEL** has demonstrated excellent compatibility and performance on the **BIOFIRE® FILMARRAY® 2.0 System**. It offers complete coverage of all relevant bacterial, viral, and fungal CNS pathogens included in the syndromic panel.

Its **clinically relevant weak-positive concentrations**, together with the use of a **matrix that mimics human cerebrospinal fluid**, including **human cells**, make it especially suitable for challenging the entire analytical process—**from nucleic acid extraction to amplification and detection**.

These characteristics position the panel as an essential, independent external control for **performance verification, routine quality assurance, assay validation, and troubleshooting** in molecular meningitis diagnostics.

** Filmarray® system and brand belongs to BioFire® Diagnostics LLC, a subsidiary of bioMérieux®*

FilmArray
Meningitis / Encephalitis (ME) Panel - IVD

BIOFIRE BY BIOMÉRIEUX

www.BioFireDx.com

Run Summary

Sample ID:	25MBTC025101+25MBTC025201	**Run Date:**	19 May 2025 11:21 AM
Detected:	*Escherichia coli* K1 *Haemophilus influenzae* *Listeria monocytogenes* *Neisseria meningitidis* *Streptococcus agalactiae* *Streptococcus pneumoniae* Cytomegalovirus Enterovirus Herpes simplex virus 1 Herpes simplex virus 2 Human herpesvirus 6 Human parechovirus Varicella zoster virus *Cryptococcus neoformans/gattii*	**Controls:**	Passed

⚠ **WARNING:** The FilmArray ME Panel does not distinguish between latent and active CMV and HHV-6 infections. Detection of these viruses may indicate primary infection, secondary reactivation, or the presence of latent virus. Results should always be interpreted in conjunction with other clinical, laboratory, and epidemiological information.

Result Summary

Bacteria	
✓ Detected	*Escherichia coli* K1
✓ Detected	*Haemophilus influenzae*
✓ Detected	*Listeria monocytogenes*
✓ Detected	*Neisseria meningitidis*
✓ Detected	*Streptococcus agalactiae*
✓ Detected	*Streptococcus pneumoniae*
Viruses	
✓ Detected	Cytomegalovirus
✓ Detected	Enterovirus
✓ Detected	Herpes simplex virus 1
✓ Detected	Herpes simplex virus 2
✓ Detected	Human herpesvirus 6
✓ Detected	Human parechovirus
✓ Detected	Varicella zoster virus
Yeast	
✓ Detected	*Cryptococcus neoformans/gattii*

Run Details

Pouch:	ME Panel v1.4	**Protocol:**	CSF v3.1
Run Status:	Completed	**Operator:**	Vircell Vircell (Vircell)
Serial No.:	106174938	**Instrument:**	2FA09157
Lot No.:	3HUE24		

FilmArray
Meningitis / Encephalitis (ME) Panel - IVD

BIOFIRE BY BIOMÉRIEUX

www.BioFireDx.com

Run Summary

Sample ID:	25MBTC025101	**Run Date:**	08 May 2025 3:07 PM
Detected:	*Escherichia coli* K1 *Haemophilus influenzae* *Listeria monocytogenes* *Neisseria meningitidis* *Streptococcus agalactiae* *Streptococcus pneumoniae* *Cryptococcus neoformans/gattii*	**Controls:**	Passed

Result Summary

Bacteria	
✓ Detected	*Escherichia coli* K1
✓ Detected	*Haemophilus influenzae*
✓ Detected	*Listeria monocytogenes*
✓ Detected	*Neisseria meningitidis*
✓ Detected	*Streptococcus agalactiae*
✓ Detected	*Streptococcus pneumoniae*
Viruses	
Not Detected	Cytomegalovirus
Not Detected	Enterovirus
Not Detected	Herpes simplex virus 1
Not Detected	Herpes simplex virus 2
Not Detected	Human herpesvirus 6
Not Detected	Human parechovirus
Not Detected	Varicella zoster virus
Yeast	
✓ Detected	*Cryptococcus neoformans/gattii*

Run Details

Pouch:	ME Panel v1.4	**Protocol:**	CSF v3.1
Run Status:	Completed	**Operator:**	Vircell Vircell (Vircell)
Serial No.:	106175021	**Instrument:**	2FA03108
Lot No.:	3HUE24		

FilmArray
Meningitis / Encephalitis (ME) Panel - IVD

BIOFIRE BY BIOMÉRIEUX

www.BioFireDx.com

Run Summary

Sample ID:	25MBTC025201	**Run Date:**	09 May 2025 10:49 AM
Detected:	Cytomegalovirus Enterovirus Herpes simplex virus 1 Herpes simplex virus 2 Human herpesvirus 6 Human parechovirus Varicella zoster virus	**Controls:**	Passed

⚠ **WARNING:** The FilmArray ME Panel does not distinguish between latent and active CMV and HHV-6 infections. Detection of these viruses may indicate primary infection, secondary reactivation, or the presence of latent virus. Results should always be interpreted in conjunction with other clinical, laboratory, and epidemiological information.

Result Summary

Bacteria	
Not Detected	*Escherichia coli* K1
Not Detected	*Haemophilus influenzae*
Not Detected	*Listeria monocytogenes*
Not Detected	*Neisseria meningitidis*
Not Detected	*Streptococcus agalactiae*
Not Detected	*Streptococcus pneumoniae*
Viruses	
✓ Detected	Cytomegalovirus
✓ Detected	Enterovirus
✓ Detected	Herpes simplex virus 1
✓ Detected	Herpes simplex virus 2
✓ Detected	Human herpesvirus 6
✓ Detected	Human parechovirus
✓ Detected	Varicella zoster virus
Yeast	
Not Detected	*Cryptococcus neoformans/gattii*

Run Details

Pouch:	ME Panel v1.4	**Protocol:**	CSF v3.1
Run Status:	Completed	**Operator:**	Vircell Vircell (Vircell)
Serial No.:	106174941	**Instrument:**	2FA03108
Lot No.:	3HUE24		