



# CERTIFICATION

## AOAC Research Institute *Performance Tested Methods*<sup>SM</sup>

Certificate No.  
**082102**

The AOAC Research Institute hereby certifies the method known as:

### **PathoSEEK<sup>®</sup> 5-Color Aspergillus Multiplex Assay with SenSATIVAx<sup>®</sup> Extraction**

manufactured by

**Medicinal Genomics Corp.**  
**100 Cummings Center, Suite 406L**  
**Beverly, MA 01915 USA**

This method has been evaluated and certified according to the policies and procedures of the AOAC *Performance Tested Methods*<sup>SM</sup> Program. This certificate indicates an AOAC Research Institute Certification Mark License Agreement has been executed which authorizes the manufacturer to display the AOAC Research Institute *Performance Tested Methods*<sup>SM</sup> certification mark on the above-mentioned method for the period below. Renewal may be granted by the Expiration Date under the rules stated in the licensing agreement.

A handwritten signature in black ink, appearing to read "Bradley A. Stawick".

Bradley A. Stawick, Senior Director  
Signature for AOAC Research Institute

Issue Date  
Expiration Date

October 27, 2024  
December 31, 2025

**METHOD NAME**

PathoSEEK® 5-Color Aspergillus Multiplex Assay with SenSATIVAx®

**CATALOG NUMBERS**

420147, 420148, 420001, 420004, 420201, 420330

**ORIGINAL CERTIFICATION DATE**

August 10, 2021

**PRINCIPLE OF THE METHOD**

The PathoSEEK 5-Color Aspergillus Multiplex Detection Assays use a multiplexing strategy with an internal plant DNA reaction control to ensure accurate detection of four species of *Aspergillus* as well as cannabis DNA in every reaction. Unlike other techniques, this multiplexing strategy verifies the performance of the assay when detecting pathogens, resulting in the minimization of false negative results due to reaction setup errors or failing experimental conditions.

Two multiplex assays are available for use, dependent on the thermocycler used for analysis. The PathoSEEK 5-Color Aspergillus Multiplex (AriaMx) Detection Assay for use on the Agilent AriaMx Real-Time PCR System uses the ATTO 425 Fluorophore for detection of the *A. terreus*, while the 5-Color Aspergillus Multiplex (CFX) Detection Assay, for use on the Bio-Rad CFX-96 Real-Time PCR System, uses the Cy5.5 Fluorophore for the detection of *A. terreus*.

SenSATIVAx is a proprietary DNA isolation process that uses magnetic particles to isolate and purify both plant and microbial DNA from a raw, homogenized plant or MIP sample. The use of magnetic particles affords 8 or 96 tip automation, enabling high throughput applications. DNA can be isolated from a single sample or a large batch in under 1 h. Hands-on time is less than 45 min.

**CERTIFIED CLAIM STATEMENT:** The PathoSEEK® 5-Color Aspergillus Multiplex Assay with SenSATIVAx® method is certified for the detection of *Aspergillus* species (*A. flavus*, *A. fumigatus*, *A. niger*, & *A. terreus*) within the scope of Tables 1 and 2.

**Certification includes:**

1. Agilent AriaMx G8830A Option 010, containing the following Optical Channels: FAM, ROX, HEX, Cy5, and ATTO 425 with AriaMx Version 2.1 software.
2. Bio-Rad CFX96 Touch™ (standard) with CFX Manager Version 3.1 software or CFX Maestro Version 2.2 software.

**Table 1. Method Performance Claims**

| Matrix                            | Test Portion | Enrichment Conditions |        |             |         | SMPR <sup>b</sup> | Claim <sup>c</sup> |
|-----------------------------------|--------------|-----------------------|--------|-------------|---------|-------------------|--------------------|
|                                   |              | Broth <sup>a</sup>    | Volume | Temperature | Time    |                   |                    |
| Dried cannabis flower (>0.3% THC) | 10 g         | TSB                   | 90 mL  | 37 ± 1°C    | 24–48 h | 2019.001          | NSDD <sup>d</sup>  |
| THC-Infused chocolate             | 25 g         | TSB                   | 60 mL  | 37 ± 1°C    | 24–48 h | 2019.001          | NSDD               |
| Cannabis concentrate              | 5 g          | TSB                   | 12 mL  | 37 ± 1°C    | 24–48 h | 2019.001          | NSDD               |

<sup>a</sup>TSB = Tryptic soy broth.

<sup>b</sup>AOAC Standard Method Performance Requirements (SMPR) 2021.009 for Detection of *Aspergillus* in Cannabis and Cannabis Products. Confirmation by streaking enrichment to a fungal specific agar (Potato Dextrose Agar or Dichloran Rose Bengal Agar (25 ± 2°C for 5-7 days). Confirm by microscopic examination of colony morphology.

<sup>c</sup>NSDD = No statistical difference detected using SLV study design from OMA Appendix J (2012). The SLV qualitative method comparison study design from OMA Appendix J (2012) is not intended to demonstrate statistical equivalence. Expert opinion is that the method is appropriate for its intended use. For cannabis matrixes, the comparison is between presumptive and confirmed results only.

**Table 2. Method Selectivity**

| Enrichment         |           | Inclusivity Strains     |              | Exclusivity Species     |                           |
|--------------------|-----------|-------------------------|--------------|-------------------------|---------------------------|
| Broth <sup>a</sup> | Temp., °C | No. Tested <sup>b</sup> | No. Positive | No. Tested <sup>c</sup> | No. Positive <sup>d</sup> |
| TSB                | 37 ± 1°C  | 50                      | 50           | 35                      | 3                         |

<sup>a</sup> TSB = tryptic soy broth.

<sup>b</sup> Comprising *Aspergillus flavus* (13), *A. fumigatus* (13), *A. niger* (11), and *A. terreus* (13).

<sup>c</sup> 35 Strains comprising 34 species. Exclusivity organisms were cultured under optimal conditions for growth.

<sup>d</sup> *Aspergillus oryzae*, and *Aspergillus parasiticus* tested positive for *A. flavus*, and *Aspergillus pseudoterreus* tested positive for *A. terreus*.

**Table 3. Method Summary**

| No. | Date          | Summary                                                                                                                           | Supporting Data       |
|-----|---------------|-----------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| 1   | August 2021   | Original Certification.                                                                                                           | Certification Report  |
| 2   | November 2021 | Level 2 Modification: Matrix extension to add cannabis concentrate (5 g) and a reduced incubation time for dried cannabis flower. | Modification Report 1 |
| 3   | May 2024      | Level 2 Modification: Evaluation of a modified elution volume.                                                                    | Modification Report 2 |