

soleris Direct Yeast and Mold

Product Number: DYM-109C



DYM-109C Vial uninoculated (left) and inoculated vial (right).

Introduction

The Direct Yeast and Mold Vial, 9 mL (DYM-109C) allows for rapid detection of yeast and mold in various matrices. The vial has broad inclusivity and an assay time of 48 hours for most applications. As yeast and molds grow in the broth medium, the carbon dioxide (CO_2) diffuses through a membrane layer into a soft agar plug containing a dye indicator. The Soleris[®] instrument reads the color change in the dye. The membrane layer also serves as a barrier, eliminating product interference within the reading frame.

In AOAC Research Institute *Performance Tested Method* Certificate #051301 studies, the Soleris DYM-109C vial was found to be an effective procedure for semi-quantitative determination of yeast and mold in the following sample types: nonfat dry milk, ice cream mix, salad dressing, yogurt, dried fruit, orange juice concentrate, tomato juice, saw palmetto powder, cornflour, cocoa powder, cocoa liquor, cocoa butter, dry pet food, black pepper, and dried cannabis flower. Test duration is 48 hours with the following exceptions: cocoa butter, which is 60 hours, and dried cannabis flower, which is 72 hours.

Materials Required

- 1. Direct Yeast and Mold Vial (DYM-109C).
- 2. Yeast and Mold Supplement (YI-110C).
- Neogen[®] rapid microbiology instrument (product no. BSX-32, BSX-128, BLX-INS32, SNG-INS32). Containing one or four temperature-controlled (18-50°C ± 0.5°C) incubator drawers with 32 test locations per drawer. Each test location contains a light-emitting diode (LED) based optical sensor for measurement of changes in absorbance over time.
- 4. Soleris computer (product no. SNG-COMPUTER or equivalent).
- 5. Soleris Vial Rack (product no. VR-300, VR-200, or equivalent): Holds 32 vials.

Dependent on Sample Tested

- 1. Sterile 1N to 5N sodium hydroxide (NaOH) and/or hydrochloric acid (HCl).
- 2. pH meter or pH paper.
- 3. Micropipettor and tips, 20–200 $\mu L.$
- 4. Micropipettor and tips, 100–1,000 $\mu L.$
- 5. Butterfield's Phosphate Buffer (BPB), 99 mL (BPB-99).
- 6. Buffered peptone water (BPW) (product no. NCM0015 or equivalent).



- 7. Tryptic Soy Broth (TSB), 90 mL (BLX-TSB90 or equivalent).
- 8. BPB, 90 mL (product no. 6654 or equivalent).
 - a. If required, use a designated neutralization broth, such as D/E Neutralizer, TAT Broth, Modified Letheen Broth, etc.
- 9. Stomacher or equivalent.
- 10. Stomacher-type bags with mesh filter (product no. 6827 or equivalent).
- 11. Balance: For weighing samples, minimum 100 g \pm 0.1 g capacity.
- 12. Dichloran Rose Bengal Chlortetracycline (DRBC) Agar (product no. NCM0029, NCM0082, or equivalent).

Vial Specifications

- 1. Vial pH is 5.6 ± 0.2 .
- 2. Vial sample capacity of 0.1–1.0 mL.

Yeast and Mold Supplement Preparation

Add 10 mL of sterile deionized water to the YI-110C Vial, mix well. Store in the refrigerator up to 28 days after rehydration. For more information, please see the YI-110C product insert.

Vial Preparation

- 1. Remove DYM-109C Vials from the refrigerator and allow them to equilibrate to room temperature.
- 2. Add supplement to the DYM-109C.
 - a. Sample without starter culture: Add 0.15 mL of YI-110C directly to the DYM-109C Vial, mix well. Add sample to vial within two hours after the addition of supplement. Refer to insert.
 - b. Sample with starter culture or unknown: Add 0.6 mL of YI-110C directly to the DYM-109C Vial, mix well. Add sample to vial within two hours after the addition of supplement. Refer to insert.

Sample Preparation

- 1. Add sample directly, or, if using dilute-to-specification, complete the dilution required.
 - a. For United States Pharmacopeia (USP) testing, perform 1:10 dilution by adding 10 g of sample in 90 mL of TSB or designated neutralization broth.
 - b. For cannabis testing, perform 1:10 dilution by adding 10 g of sample to 90 mL of TSB. Homogenize the sample thoroughly and decant the liquid. The liquid becomes the test sample.
 - c. For AOAC food testing, perform 1:10 dilution by adding 25 g of sample to 225 mL 0.1% peptone.
 - d. For all other testing, perform 1:10 dilution by adding 11 g of sample to 99 mL of BPB.
- 2. Check pH and adjust, if necessary, to 7.0 ± 1.0 .

Inoculation of Vial

- 1. Inoculate the vial with no more than 1.0 mL and no less than 0.1 mL of the sample to be tested. If using specification monitoring, add the volume of the appropriate dilution required.
 - a. For AOAC testing, add 1.0 mL to the Soleris vial.
- 2. Cap the vial and gently invert three times to mix the sample. Keep cap tight.
- 3. Insert the vial into the Soleris instrument utilizing the applicable algorithm below or as indicated by a trainer. The incubation temperature and test duration can be optimized if required. It is not recommended to adjust parameters without consulting Neogen technical service at 517.372.9200 or visiting our website at neogen.com.

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Algorithms Utilized (Yellow Test Type)

Threshold	Skip	Shuteye	Temperature	Test Duration	Validation Scope
8	2	50	28 ± 2°C	48 hours ^{1,2}	AOAC PTM # 051301: Nonfat dry milk, ice cream mix, salad dressing, yogurt, dried fruit, orange juice concentrate, tomato juice, saw palmetto powder, cornflour, cocoa powder, cocoa liquor, cocoa butter, dry pet food, and black pepper.
					Validated in accordance with AOAC International Methods Committee Guidelines for Validation of Microbiological Methods for Food and Environmental Surfaces ³ : Broad food and environmental surfaces.

Cannabis

Food

Threshold	Skip	Shuteye	Temperature	Test Duration	Validation Scope
8	2	50	28 ± 2°C	72 hours	AOAC PTM # 051301 : Dried cannabis flower [>0.3% delta 9-tetrahydrocannabinol (THC)].
				48 hours⁴	Validated in accordance with USP <1223> Validation of Alternative Microbiological Methods ⁵ : Broad cannabis and

Personal Care, Cosmetics, Nutraceuticals, and Dietary Supplements

Threshold	Skip	Shuteye	Temperature	Test Duration	Validation Scope
8	2	50	28 ± 2°C	48 hours ¹	Validated in accordance with USP <1223> Validation of Alternative Microbiological Methods ⁵ : Broad personal care, cosmetic, nutraceutical, and dietary supplement products.

¹ For some slow growing organisms, test duration may be extended from 48 to 72 hours.

² Cocoa butter requires a test duration of 60 hours.

³ U.S. Food and Drug Administration Bacteriological Analytical Manual (FDA-BAM), Chapter 18 (Yeasts, Molds, and Mycotoxins) dilution plating method referenced for food products and environmental samples.

⁴ Dried cannabis flower requires a test duration of 72 hours.

⁵ Compendial USP <2021> and <2022> methods referenced for cannabis, nutraceuticals, and dietary supplement products. USP <61> and <62> methods referenced for personal care and cosmetic products.

Confirmation Step (Optional)

1. From a DYM-109C Vial, invert to mix.

- 2. Using a 10 μ L inoculating loop, streak from the Soleris vial to a DRBC agar plate.
- 3. Incubate the plates at $25 \pm 1^{\circ}$ C for 5–7 days and examine for yeast and mold growth.

Caution

Products containing CO_2 releasing compounds (e.g., ascorbic acid, calcium carbonate, or calcium ascorbate) need to be carefully validated, as reactions with the vial chemistry may occur, causing false positive results.

Disclaimers

Information provided is based on validation procedures that Neogen performed in Neogen laboratories. Deviation from procedures is possible, but should be discussed with Neogen technical services.

Samples may need to be pH adjusted for all vials.

Appearance of the vials should be inspected prior to use.

If shuteye detections are observed, the threshold may need to be adjusted based on the product matrix. Certain product matrices may require parameter adjustments, including increased test duration. For more information contact Neogen technical services at 517.372.9200 or visit our website at neogen.com.

Some strains do not detect within the recommended test duration and will need an extended test duration. These organisms may have been strain-specific or described as being temperature sensitive.

Reference the Soleris Operating Manual for troubleshooting and instrument use information.

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Safety Precautions

Use of this test should be restricted to individuals with appropriate laboratory training in microbiology as some *Enterobacteriaceae* are potentially infectious. Reagents are for laboratory use only. Test samples and used Soleris vials may contain potentially infectious microorganism; follow appropriate laboratory procedures for the handling of microbial pathogens. (U.S. Department of Health and Human Services, Biosafety in Microbiological and Biomedical Laboratories (BMBL), 6th Edition, HHS Publication No. (CDC) 300859, Revised June 2020; found at: www.cdc.gov/labs/pdf/CDC-Biosafety Microbiological BiomedicalLaboratories-2020-P.pdf (or most current version, found at cdc.gov). All pipetting transfers must be made using either a disposable pipette and pipetting aid or a micro pipettor with disposable tips. Culture media contains antimicrobial selective agents and dyes: wear appropriate PPE and avoid contact with skin and mucous membranes. Refer to the Safety Data Sheet available from Neogen for more information. Used enrichment cultures and agar media should be handled and disposed of as potentially infectious material. The preferred method for decontamination of contaminated material is autoclaving. Items that cannot be autoclaved may be decontaminated using a disinfectant solution, e.g., 10% household bleach, followed by rinsing with water. Consult with your facility safety director for specific instructions.

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