

Path-Chek Hygiene Listeria Media

(Cat. nr. PC080)

Path-Chek Hygiene Listeria Media is intended for the detection of important food pathogens on food contact surfaces in food handling and manufacturing environments after cleaning and sanitizing has been performed. Path-Chek Hygiene Listeria Media should be considered as a fundamental component of Good Manufacturing Practice and an integral component of any HACCP (Hazard Analysis Critical Control Points) plan.

1. PRINCIPLE OF THE TEST

Path-Chek Hygiene Pathogen Detection consists of two components (available separately):

1. A pre-moistened **Path-Chek Hygiene Swab** which assists in the removal of bacteria on test surfaces, particularly from dry surfaces. The composition of the wetting agent also ensures that any residual sanitising agents on test surfaces are neutralised. The swabs have a small breakpoint (nick) approximately 45mm from the end of the swab. This is to assist in breaking the swab off at an appropriate length when it is placed into the tube of specific growth medium.
2. The **Path-Chek Pathogen Detection** Broth. Individual detection broths are available for Coliforms, Salmonella species and Listeria species. Each broth comprises a buffered growth medium containing growth promoters, selective agents and a specific indicator system.

2. KIT CONTENTS

- 100 x 3 mL Listeria media vial

3. MATERIALS REQUIRED BUT NOT PROVIDED

- Path-Chek Hygiene Swabs (100 piece) cat.nr. PCS100
- Incubator, set to 35 – 37 °C
- Incubator, set to 25 °C
- Rack to hold tubes during incubation
- Template 10 x 10cm to assist with sampling (See Warnings and Precautions)

4. PROCEDURE

- 1) Carefully remove the cap from the pre-moistened Path-Chek Hygiene swab (See Precautions).
- 2) Thoroughly swab a standard sample area (10 x 10 cm), rotating the swab as the sample is being collected.

If sample areas are irregular develop a standard sampling procedure which is documented and used consistently. Label the swab tube with the sample details.

- 3) If the swab cannot be transferred immediately into the Path-Chek Hygiene Pathogen Detection Broth, return it to its holding tube and store in a cool place. Swabs may be held at a maximum temperature of 20°C for up to 24 hours.
- 4) Place the swab into the Path-Chek Hygiene Pathogen Detection Broth at an angle with the tip of the swab against the side of the tube. Press down on the shaft of the swab. The shaft of the swab will break at breakpoint of the swab, 45mm from the swab tip. (See Precautions) Replace the cap and label the Pathogen Detection Broth tube with the sample details.
- 5) Place inoculated tubes into a suitable rack and incubate at the appropriate temperature for the appropriate time (See Below).
- 6) Observe for colour changes and record the results.

5. INCUBATION

PATHOGEN BROTH	INCUBATION CONDITIONS
Listeria*	25 ± 1°C for 24 – 48 hours

***NOTE: If Path-Chek Hygiene Listeria is incubated at 30 – 3 °C there will, in certain circumstances, be an increased risk of false positives**

6. INTERPRETATION

A **POSITIVE** result which indicates the presence of a particular pathogen is indicated by the development of a specific color change (See Below).

A **NEGATIVE** result which indicates the absence of a particular pathogen is indicated by no color change in the Path-Chek Pathogen Detection Broth (See Below).

PATHOGEN BROTH	INTERPRETATION	
	NEGATIVE	POSITIVE
Listeria	Yellow/Straw	Black

7. WARNINGS AND PRECAUTIONS

Path-Chek Pathogen Detection Broth may grow potentially dangerous pathogens. All positive samples should be handled using precautions appropriate for the handling of infectious materials.

When collecting samples aseptic procedures should be used. Do not touch the swab tip with hands or fingers.

If a template is used during sampling, it must be sanitized, rinsed and dried before using again.

Follow Good Microbiological Practices where appropriate.

8. STORAGE AND SHELF LIFE

Path-Chek Hygiene Pathogen Detection Broths should be stored at 2 – 8 °C when not in use. The pre-moistened sample swabs should be stored at 4 – 25 °C. Components should not be used after the expiry date printed on the carton label.

9. OPTIONAL CONFIRMATION PROCEDURE

Presumptive positive tests may be confirmed by sub-culturing the Path-Chek Detection Broth onto an appropriate selective agar plate medium for the pathogen being sought. After incubation at 35 – 37 °C for 24 – 48 hours, plates should be examined for colonies resembling the pathogen being sought. Any suspicious colonies should be further identified using more definitive tests.

See below for further suggestions that conform to recognised international standard methods such as AOAC, USDA etc.

PATHOGEN DETECTION BROTH	CONFIRMATION PROCEDURES	
	ISOLATION MEDIA	CONFIRMATION TESTS
Listeria	Oxford Palcam ALOA	Microgen® Listeria ID Microgen® Listeria Latex Rapid Catalase Test (See Below)

10. RAPID CATALASE TEST – PATH

Listeria spp. produce the enzyme catalase, while E. faecalis does not. The performance of a 2 minute catalase test will differentiate between positive Path-Chek tests growing Listeria spp and false positive tests growing E. faecalis.

- 1) Gently add 100 – 200 µL of 3 – 6% Hydrogen Peroxide to positive Path-Check Listeria test. **Do not mix!**
- 2) Allow to stand for up to 2 minutes.
- 3) Examine for the development of bubbles on top of the media in the Path-Check tube.

Positive – Development of bubbles on the top of the media in the Pat-Check tube.

Negative – No bubbles on the top of the media in the Pat-Check tube.

11. DISPOSAL

After reading, the tests may be:

1. autoclaved and disposed of in the normal manner, OR
2. Incinerated.

12. LIMITATIONS

1. Path-Chek Hygiene Pathogen is intended as a screening test. Positive results should, if required, be confirmed by standard bacteriological methods.
2. Path-Chek Hygiene Pathogen has not been validated for testing food or uncleaned food surfaces. The use of Path-Chek Hygiene Pathogen for these purposes may result in false positive results.
3. The absence of a colour change does not necessarily imply the absence of target bacteria. Reasons for this may include:
 - a. Inadequate neutralization of sanitizing agents.
 - b. Numbers of target organisms are below the detection level of the test.
 - c. The target organisms, although present may be severely damaged and unable to be resuscitated.
4. A colour change indicative of the target organism may arise due to the presence of excessively high numbers of other organism species. Non target organisms that when in sufficiently high numbers (>10⁶ per swab) may result in false positive test results.

The potential cause of false positive results is the presence of organisms of enteric origins. The occurrence of false positive results may be an important indicator of poor cleaning and sanitising practices.

13. PERFORMANCE CHARACTERISTICS

Comprehensive sensitivity and specificity data is available for all pathogen detection products. Please refer to: **Extended User Instructions for Path Chek Hygiene swabbing kits. As a general guide to product sensitivity refer to the following table.**

Target Organism	Direct Inoculation (1)	Wet Surface (2)	Dry Surface (3)
Listeria	<5 CFU	<5 CFU	<10 CFU

Notes:

- (1). Organisms inoculated directly onto swab.
- (2). Organisms recovered from a standard 100cm² wet surface.
- (3). Organisms recovered from a standard 100cm² surface, dried and then recovered.