

The OnSite logo, consisting of three teal circles of varying sizes arranged vertically to the left of the text.

**OnSite<sup>®</sup> III Allergen  
Mustard**

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## **Instructions For Use**

Rapid detection of mustard.  
Suitable for rinse water, environmental surfaces, food and  
beverage testing



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## 1. Introduction

Food allergies are potentially life-threatening immune-mediated reactions to consuming foods, most notably those containing milk, egg, fish, shellfish, tree nut, sesame, mustard, peanut, wheat, or soybean residues. Roughly 2% of adults and 4-8 % of children are affected by food allergies. Management of food allergies requires strict dietary avoidance. To minimize the risk of inadvertent exposures to food allergens, numerous regulatory bodies have adopted strict requirements. Compliance is achieved in part through the use of antibody-based assays that can detect allergen residues in foods and on surfaces. To assist the food industry in establishing effective food safety practices, Microbiologique has developed the OnSite® III Allergen Mustard test kit, which can detect mustard residues at 1.0 ppm in foods and on surfaces in roughly 20 min.

## 2. Intended Use

OnSite® III Allergen lateral flow devices (LFDs) are rapid, qualitative tests designed to detect specific allergen residues in raw and processed foods and on surfaces. The results obtained using these kits can be used as part of an effective Allergen Control Plan.

These tests are intended for laboratory and industry use, including within food production facilities, commercial kitchens, contract laboratories, and auditing programs. The tests should only be performed by trained personnel.

**Please read ALL instructions prior to use.**



## 3. Performance Characteristics

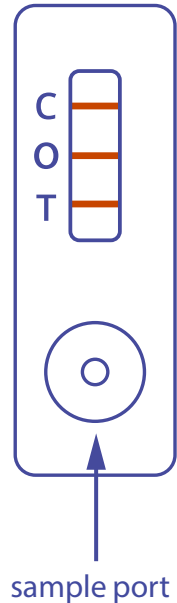
**Limit of Detection:** 1.0 mg/kg (ppm) food or rinse water, 1 µg/100 cm<sup>2</sup> from surfaces

**Operation Time:** 20 min

**Suitability:** Native and processed proteins, though detection of extensively hydrolyzed or thermally processed allergens may be reduced. See Section 10 for additional information including cross-reactivity.

## 4. Assay Principles

OnSite® III Allergen test kits are lateral flow devices (LFDs) that can detect specific allergens in foods and liquids, and on surfaces. The kits rely on the use of polyclonal antibodies (pAbs) directed against the target allergen. The test is configured with a test line (T), an overload line (O), and a procedural control line (C). The test line (T) will produce a red line at low to medium levels of contamination. The overload line (O) will produce a red line at blank or low analyte concentrations. Both lines will disappear in the presence of very high analyte concentrations. The procedural Control Line (C) should always produce a red line, regardless of the analyte concentration, and failure for this line to appear indicates a failure of the mixture to migrate across the membrane. To operate the kit, the sample is first extracted for 2 min with shaking followed by a 5 min settling time. Thereafter, 100  $\mu\text{L}$  of settled sample extract is applied to sample port, where it will begin to react with gold particles conjugated to the polyclonal antibodies. The mixture will then migrate across the membrane thus enabling visualization of the test line (T), overload line (O), and procedural control line (C). The test outcome is interpreted by visualizing the appearance of these lines at 15 min.



## 5. Kit Storage and Safe Handling

- ★ Store in original packaging at 2-8 °C (36-46 °F). DO NOT FREEZE.
- ★ Do not use kit after printed expiration date.
- ★ Avoid exposure of any components to direct sunlight or heat.
- ★ Dispose of kit components in regular trash, or recycle where appropriate.
- ★ Do not eat or drink any kit components. See SDS for additional information.

## 6. Kit Components

- A OnSite® III LFD Devices (10)
- B Single-use Droppers (10)
- C Single-use Sampling Spoons (10)
- D Bottle of OnSite® III LFD Buffer A, 120 mL (1)
- E Extraction Tubes, 14 mL (10)
- F Instructions for use

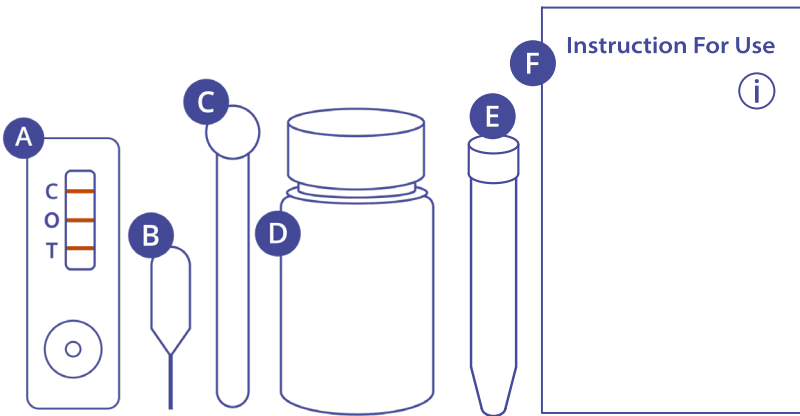


Figure 1. Kit Components

### 6.1. Optional Materials

- PA-46, OnSite® III Allergen Accessory Kit for surface sampling
- Blender, grinder, or similar device for homogenizing sample
- Laboratory timer
- Centrifuge
- Digital scale sensitive to 0.1 g for measuring solid samples
- Calibrated laboratory pipette for measuring liquid samples
- Fine-tipped marking-pen
- Lateral flow device reader
- Non-powdered disposable gloves
- pH test strips and NaOH solution
- 15 mL tube rack

## 7. Food and Liquid Sampling and Analysis

### 7.1. Sample Preparation and Analysis



**IMPORTANT: Prior to starting, ensure ALL test kit components have been brought to room temperature!**

**7.1.1.** Prior to beginning, thoroughly clean hands, the work area, and requisite utensils to reduce the risk of contamination. Disposable gloves may be used. To adequately perform this task, use detergent and water followed by thorough rinsing. A secondary cleaning with alcohol is strongly recommended.

**7.1.2.** A representative sample(s) must be taken from food products. To ensure an accurate analysis of the final product, ensure that all components of the final product are present in the sample.

**7.1.3.** To ensure consistent results, a suitable blender, food processor, or a similar mixing device should be used to homogenize **solid samples**. **Liquid samples** should be mixed or shaken vigorously before sampling. Samples of a doughy consistency, or high in viscosity can be mixed using clean metal utensils.

**7.1.4.** Pour LFD extraction buffer into a 14 mL tube to the 9 mL mark. Then collect 1 g of solid or 1 mL of liquid sample and add to the 14 mL tube containing the buffer.

A precise sample size is critical for accurate results. The provided scoop may be used to approximate 1 g or 1 mL. For increased accuracy, use a calibrated pipette or weigh balance.

**7.1.5.** Replace the lid tightly. Shake tube vigorously for two minutes.

**7.1.6.** For acidic samples, check the pH of the solution and if it is acidic, adjust the pH of the solution using NaOH as needed to make the pH neutral.

**7.1.7.** Allow sample to settle for 5 minutes. Alternatively, transfer sample to a suitable container and centrifuge.

**7.1.8.** Open a new lateral flow device and set it on a flat, level surface.

**7.1.9.** Open the extraction tube and withdraw sample extract using either a provided single-use dropper or a pipette calibrated to 100  $\mu\text{L}$ . If using the provided dropper, either angle the tube to access the extract directly, or carefully pour a small amount of extract into the cap.

**Note: Avoid withdrawing any of the sample particles that have settled to the bottom of the tube and avoid withdrawing foam that may be present on the surface.**

**To ensure correct volume of extract, hold the dropper horizontally, parallel to the lateral flow device.**

**7.1.10.** Apply either 5 drops from the single-use dropper or 100  $\mu\text{L}$  from the calibrated pipette of sample extract to the sample port of the lateral flow device.

**7.1.11.** Observe test results at 15 minutes. A valid positive result may be observed more rapidly.

**Note: To ensure accuracy and to avoid misinterpretation of the drying artifacts, analyze results promptly at 15 minutes.**

## 8. Environmental Sampling and Analysis

### 8.1. Sample Preparation and Analysis



**IMPORTANT:** Prior to starting, ensure ALL test kit components have been brought to room temperature!

OnSite® III Allergen Accessory Kit (PA-46) required, sold separately.

One sample may be used to analyze multiple allergens simultaneously, provided all LFD Devices are compatible with the same OnSite® III Allergen LFD Buffer (A or B).

**8.1.1.** Prior to beginning, thoroughly clean hands to reduce the risk of allergen contamination. Disposable gloves may be used. Open one of the 1.5 mL tubes and pour 1 mL of LFD buffer in. Open a new sampling swab and moisten the swab tip with RO water or, if not available, with LFD buffer that has been poured into the tube.

**8.1.2.** Collect a surface sample by swabbing a 4 x 4 inch (or 10 x 10 cm) area using a rolling crosshatch technique shown in **Figure 2**. Ensure that all sides of the swab bulb come into contact with the surface.

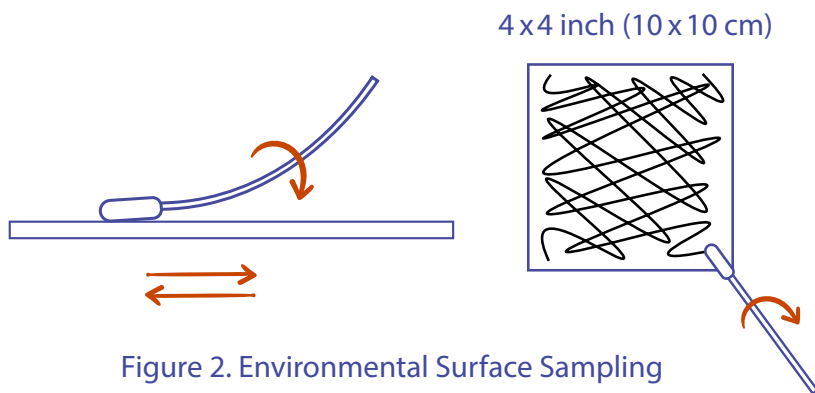


Figure 2. Environmental Surface Sampling

**8.1.3.** Open the 1.5 mL tube containing extraction buffer and place the swab tip into the extraction buffer. Carefully snap the swab at the breakaway point by pressing the swab against the wall of the vial.

**8.1.4.** Replace the lid tightly. Shake tube vigorously for two minutes.



**8.1.5.** Open a new lateral flow device and set it on a flat, level surface.

**8.1.6.** Open the extraction tube and withdraw sample extract using either a provided single-use dropper or a pipette calibrated to 100  $\mu\text{L}$ .

**8.1.7.** Apply either 5 drops from the single-use dropper or 100  $\mu\text{L}$  from the calibrated pipette of sample extract to the sample port of the lateral flow device.

**Note: Avoid withdrawing any of the sample particles that have settled to the bottom of the tube and avoid withdrawing foam that may be present on the surface.**

**To ensure correct volume of extract, hold the dropper horizontally, parallel to the lateral flow device.**

**8.1.8.** Observe test results promptly at 10 minutes.

## 9. Interpreting Test Results

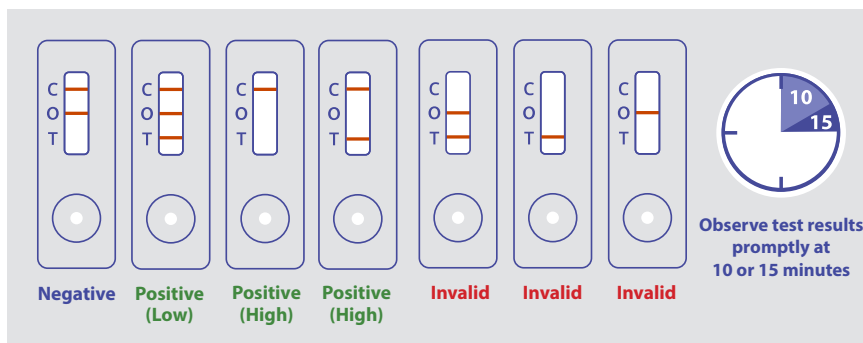


Figure 3. Interpreting Test Outcomes

**9.1.** A negative result is indicated by the development of a strong control line (C) and overload (O) line. An example of a negative result is depicted in **Figure 3** labeled “**Negative**”.

**9.2.** Positive test results at lower analyte concentrations will result in the appearance of the test line (T) in addition to the overload line (O) and/or the control line (C). At higher analyte concentrations, the test line (T) and/or the overload line (O) will disappear, leaving only the appearance of the control line (C). Thus at high analyte concentrations, the disappearance of the overload line (O) indicates that the sample is positive for the analyte regardless of the appearance of the test line (T). Examples of positive results are depicted in **Figure 3** labeled “**Positive**”.

**9.3.** Failure of the control line to appear regardless of test line or overload line development is an invalid result. In addition, any line malformation visible on the membrane denotes an invalid test. Examples of malformations would be dark spots, gaps or incomplete line development. In the event of an invalid test, the procedure should be repeated using a new test device.

## 10. Test Limitations and Validated Matrices

**IMPORTANT: Not all samples are suitable for use with this product.**



As with all test kits that rely on antibody-based detection methods, there are additives, matrices, and processing methods that may limit the ability to detect the target analyte. Contact your distributor for technical support regarding sample suitability or help in validating samples for testing.

The OnSite® III Allergen rapid tests are designed to detect native and processed allergen residues. However, detection of highly processed (autoclaved or extensively hydrolyzed) or high-fat samples may be less efficient. The test may underestimate target allergen in matrices that have extensive polyphenolic compound content (e.g. seasoning and spices) or those having undergone extensive chemical or thermal processing. Furthermore, foods that are sticky and/or starchy may require additional dilution measures to improve operation. Note that dilution will affect the detection limit of the test.

OnSite® III Allergen Mustard rapid tests do not exhibit cross reactivity with any of the commodities tested.

## 11. Best Practices and Troubleshooting

- **Timing is extremely critical.** When testing multiple samples, consider the amount of time required to process each sample. Variation in timing, at different stages of the testing procedure can produce varied results. Once a sample has been extracted, the protocol must be run to completion. Keep in mind that LFD strips should be interpreted promptly as indicated in sections 7 and 8.
- Do not remove LFD cassettes from foil pouch until the indicated time. Excess exposure to humidity or moisture may cause decreased performance or failure of the test strip.
- Prior to testing, ensure that all components are brought to room temperature.
- When testing highly acidic or basic samples, pH confirmation is suggested. The pH of the diluted sample should be between 6.8 and 7.4. If the pH is not in range, adjust accordingly.
- All tolerances for this assay are temperature  $\pm 5$  °C, volumes and weights  $\pm 1\%$ .
- Avoid using powdered gloves as this may introduce unwanted allergens.
- Do not re-use kit components.
- Store kit components as indicated.
- Do not use expired reagents.
- Read test under adequate lighting.

## 12. Warnings and Customer Support

For Laboratory use only, not intended for human diagnostic use. Testing results are only applicable to the portion of the sample product tested and to this extent, Microbiologique cannot guarantee that target allergen is, or is not present in the untested portions of the sample product. Strict adherence to the assay protocol is mandatory to ensure proper operation of the test kit.

All waste must be disposed of in compliance with federal, state, and local rules and regulations. SDS information can be obtained from your local distributor or by emailing: [info@onsitefoodsafety.com](mailto:info@onsitefoodsafety.com).

For additional information on using this kit, please call **866-256-1804** or email **[info@onsitefoodsafety.com](mailto:info@onsitefoodsafety.com)**.

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Manufactured by Microbiologique, Inc  
8315 Lake City Way NE  
Seattle, WA 98115  
USA