

AlerTox[®] Sticks Walnut

Immuno-chromatographic rapid test for qualitative detection of walnut antigen in food, kitchens and production facilities.

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AlerTox® Sticks

Walnut

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1. Intended use

AlerTox Sticks Walnut is an immunochromatographic rapid test for the qualitative detection of walnut antigen in food, kitchens and production facilities.

2. Introduction

English/common Walnut (*Juglans regia*) is a tree nut belonging to the Juglandaceae family.

Allergy to walnut can display the variety of symptoms from mild oral allergy or hives to severe life-threatening systemic reactions, i.e. anaphylactic shock or bronchial asthma. Allergy to walnut is one of the most common within tree nut allergies with estimated 0.1-1% prevalence in population.

Food Allergen Labeling and Consumer Protection Act (FALCPA) identified tree nut allergy as one of the 8 major food allergies, and the presence of specific types of nuts should be labeled on the package. In the EU, tree nuts are included in the list of allergens established by the European Food Safety Authority, whose presence must be indicated in foods according to Regulation (EU) No. 1169/2011 Annex II.

3. Test sensitivity and specificity

AlerTox Sticks Walnut uses a combination of monoclonal antibodies against a major English/common walnut antigen, the 11 S globulin seed storage protein known as allergen Jug r 4 from *Juglans regia*. AlerTox Sticks Walnut does NOT detect the antigens of cereals, legumes and other nuts, including pecan, black walnut, peanut, hazelnut, almond, macadamia, pistachio, cashew, brazil nut and coconut.

PLEASE NOTE: English/common Walnut (*Juglans regia*), pecan (*Carya illinoensis*) and black walnut (*Juglans nigra*) are closely related nuts of trees belonging to the family Juglandaceae. However, the test shows no cross-reactivity to pecan or black walnut (*Juglans nigra*). The test is 100% specific to English/common walnut.

The LOD (limit of detection) of AlerTox Sticks Walnut is 2.25 ppm of walnut protein. The range of detection (ROD) is 2.25-100'000 ppm. Above this range, the test can present a hook effect. The hook effect can appear as a negative result or as a test line with reduced intensity. If a false negative due to hook effect is suspected, repeat the test on a diluted sample.

On dry surfaces collected by a wet swab, the LOD is approximately 2 ug of walnut protein/16cm².

The sensitivity of the test decreases in an environment rich in fats (for example, in the presence of oil or creams).

If you need to quantify the amount of antigen, please acquire AlerTox ELISA Walnut (KIT3052)

4. Kit contents

- 10 immunochromatographic sticks individually packed in foil pouches
- 10 sample collection tubes (tube with yellow cap)
- 10 sample extraction buffer tubes, 10 mL (tube with blue cap)
- 10 spoons
- 10 pipettes (3 mL- only for testing liquid samples)
- 10 small pipettes
- 10 swabs (only for testing surfaces)
- Instructions for use (download from webpage)

5. Other materials not supplied

- Grinder, mortar or any other manual or automatic homogenization system to crush the sample
- Scissors
- Optional: digital scale sensitive to 0.1 g

6. Precautions

- The test sticks must be stored at a temperature between 10 °C and 30 °C (50 °F and 86 °F).
- Use the test within 10 minutes after opening the foil pouch.
- Do not touch the white end of the stick.
- Do not use the test stick when its pouch is torn, or the stick is broken or damaged.
- All the components of the test kit are disposable; do not reuse them.
- Do not use the test sticks beyond the expiry date.




7. Sample handling

The samples must be brought to a temperature between 18 °C and 35 °C (64.4 °F and 95 °F) before use. The test is designed to detect the target antigen in:

- Solid food.
- Liquid samples: beverages, washwater from cutting equipment and surfaces used in food processing and storage.
- Surfaces.

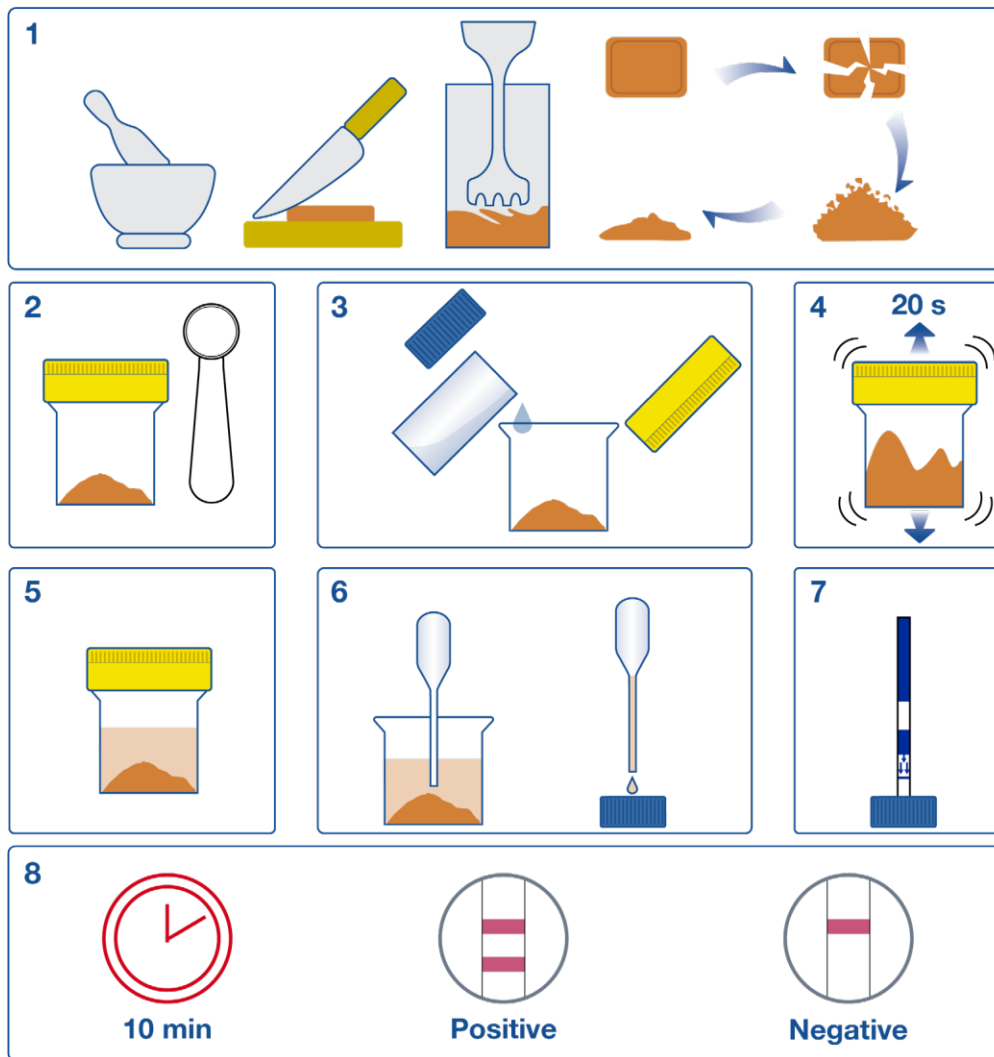
8. Test procedure for solid foods

- 8.1. Before opening the foil pouch containing the test stick, please leave it at room temperature while you process the samples.
- 8.2. Mash or crush the sample to obtain the finest crumbs possible. Use a mortar or a grinder if possible.
- 8.3. Use a scale to weigh 1 g of the sample, or follow the chart below to add an equivalent amount of sample to a yellow-capped tube, using one of the single-use spoons provided.

Food type	Examples	Spoonfuls
Flours, fine powders	Corn flour, rice flour, milk powder, spices, etc.	
Fine crumbs	Bread, cookies, cakes, snacks, etc.	
Meat, fish and cured meat	Meat, fish, sausage, black pudding, pate, canned meat and fish, etc.	

- 8.4. Pour the entire content of a blue-capped tube (10 mL) into the yellow-capped tube.
Keep the blue cap, as it will be used later on.
- 8.5. Close the yellow-capped tube and shake it vigorously for at least 20 seconds. Let it rest for 2 minutes so the solids settle.
- 8.6. With a small pipette, transfer supernatant to the blue cap until it is full.
- 8.7. Open the envelope and pull out the stick carefully, by holding its BLUE end. Do **NOT** touch the white end of the stick.
- 8.8. Place the white end of the stick in the blue cap and wait 10 minutes to read the result. Do not leave the stick longer than indicated, as the results may vary. Do not touch the stick while waiting.

Test procedure for solid foods



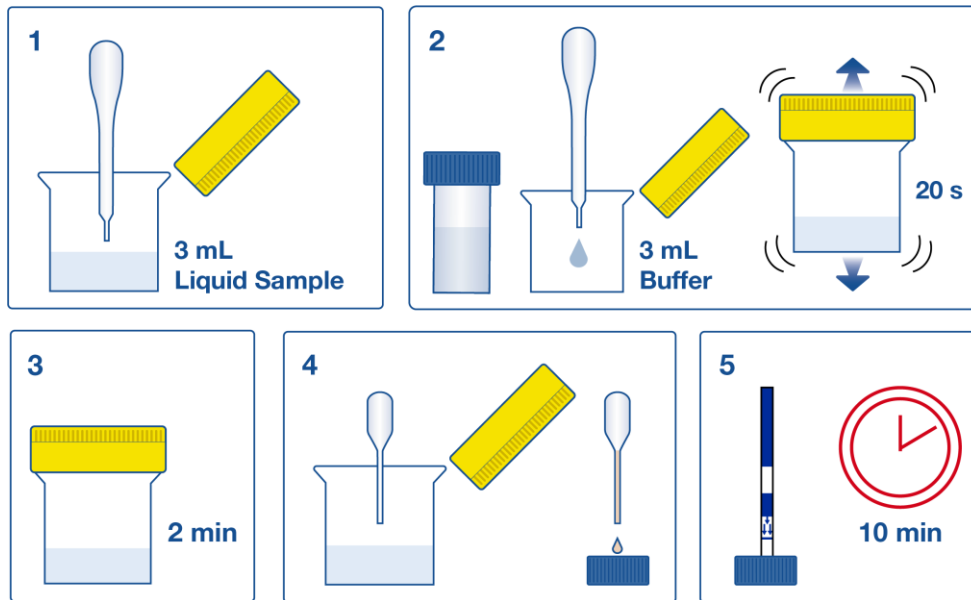
9. Test procedure for liquid samples

Liquid samples – beverages, wash waters from kitchen dishes, technological surfaces or cutting machines – may be tested directly. Turbid samples should be filtered (paper or textile filter) or allowed to settle.

- 9.1. Before opening the foil pouch containing the test stick, please leave it at room temperature while you process the samples
- 9.2. Using a provided 3 mL pipette, add 3 mL of your liquid sample to a yellow-capped tube. If the sample is thick (e.g., yogurt, sauce, etc.), follow the chart below to add an equivalent amount of sample to the yellow-capped tube, using one of the single-use spoons provided.
- 9.3. Add an equal volume of sample extraction buffer (3 mL) using the same pipette, screw the yellow cap and mix by gently shaking the tube for at least 20 seconds. If the liquid is cloudy, let it settle.
Keep the blue cap, as it will be used later on.
- 9.4. With a small pipette, transfer supernatant to the blue cap until it is full.
- 9.5. Open the envelope and pull out the stick carefully by holding its BLUE end. Do **NOT** touch the white end of the stick.
- 9.6. Place the white end of the stick in the blue cap and wait 10 minutes to read the result. Do not leave the stick longer than indicated, as the results may vary. Do not touch the stick while waiting.

Food type	Examples	Spoonfuls
Liquid and sauces	Milk, juice, condensed milk, yogurt, soup, gravy, sauce, cream, etc.	

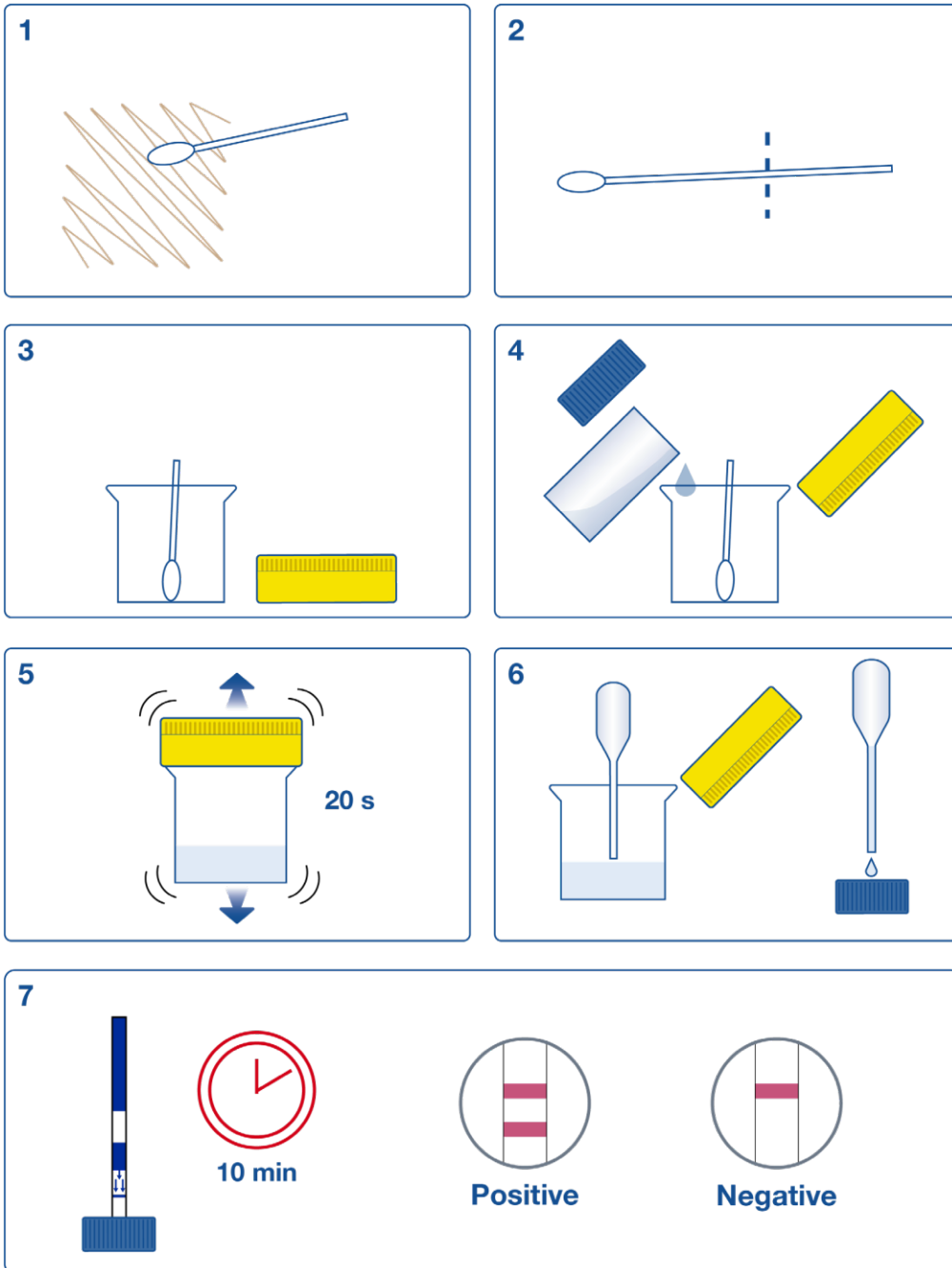
Test procedures for liquid samples



10. Test procedure for surface analysis

- 10.1.** Wet the swab by dipping it in the blue-capped tube. Firmly rub the swab on the surface that is going to be analyzed (at least 16 cm²/2.46 in², or a line of 40 cm/15.6 in. The area selected for analysis must be representative of the total area of interest.
- 10.2.** Introduce the swab into the sample collection tube and, using scissors, trim the swab so that it will fit in the yellow-capped tube with the cap closed.
- 10.3.** Pour the entire content of a blue-capped tube (10 mL) into the yellow-capped tube.
Keep the blue cap, as it will be used later on.
- 10.4.** Vigorously shake the tube for at least 20 seconds.
- 10.5.** With a small pipette, transfer supernatant to the blue cap until it is full.
- 10.6.** Open the envelope and pull out the stick carefully by holding its BLUE end. Do **NOT** touch the white end of the stick.
- 10.7.** Place the white end of the stick in the blue cap and wait 10 minutes to read the result. Do not leave the stick longer than indicated, as the results may vary. Do not touch the stick while waiting.

Test procedure for surface analysis



11. Interpretation of results

The result of the test is POSITIVE if TWO colored lines appear: One in the control zone (C) and one in the test zone (T).



The result of the test is NEGATIVE if only ONE colored line is clearly visible, in the control zone (C).



If NO colored line appears in the control zone (C), the test is INVALID.



In the case of an invalid test, repeat the test with another stick, check the correct specimen handling and test procedure, expiry date and storage conditions. Contact your distributor for further details.

IMPORTANT NOTE!

- **AlerTox Sticks is a qualitative test intended for the screening of samples for internal quality control. Under no circumstances can it replace the quantification test of the laboratory analysis.**

12. Validation

AlerTox Sticks Walnut has been validated for the following matrices:

- Protein bars
- Cereals, cereals snacks, muesli
- Chocolate and chocolate bars
- Nuts
- Cheese
- Spreads (nut butter)
- Bakery products
- Soft cakes and cakes
- Yogurt / dairy based desserts
- Ice cream
- Bread, bread sticks, biscuits and waffles
- Nut or Cereal drinks