

# Validation Report

## AlerTox Sticks Peanut

KIT3094

<b>Cat No.</b>	KIT3094 (10 tests)
<b>Test Name</b>	AlerTox Sticks Peanut
<b>Description</b>	Immunochromatographic rapid test for qualitative detection of peanut antigen in food, kitchens and production facilities.
<b>Specificity and Sensitivity</b>	<p>The LOD (limit of detection) of AlerTox Sticks Peanut is 1 ppm of peanut protein. The range of detection (ROD) is 1-10000 ppm of peanut protein. Above this range, reduction of test line intensity due to hook effect can result in a false negative result.</p> <p>AlerTox Sticks Peanut uses a monoclonal antibody against a major peanut antigen, the 11 S globulin seed storage protein known as allergen Ara h 3. AlerTox Sticks Peanut does NOT detect the antigens of cereals, other legumes and tree nuts including pecan, walnut, hazelnut, almond, macadamia, pistachio, cashew, brazil nut, chestnut, pine nut and coconut.</p>
<b>Storage</b>	Store at 10-30 °C (50-86 °F)

### Assay Procedure

Solid, liquid and surface samples: according to manual of use INS3028

## 1. Analytical Sensitivity

Reference material (roasted peanut butter) was extracted according to the kit instructions and the supernatant obtained was serially diluted in extraction buffer, to obtain dilutions 4:10<sup>3</sup>, 1:10<sup>4</sup>, 1:10<sup>5</sup>, 1:10<sup>6</sup>, 2:10<sup>6</sup> and 1:10<sup>7</sup>. These dilutions, together with a negative control sample (extraction buffer) were tested directly with the immunochromatographic sticks, according to the kit instructions, with the following results:

Reference material	Dilution	ppm (peanut)	ppm (peanut protein*)	Test result
Roasted peanut butter	4:10 <sup>3</sup>	4000	1000	weak positive
	1:10 <sup>4</sup>	100	25	positive
	1:10 <sup>5</sup>	10	2,5	positive
	1:10 <sup>6</sup>	1	0,25	positive
	2:10 <sup>6</sup>	0.5	0,125	positive
	1:10 <sup>7</sup>	0.1	0,025	weak positive
Extraction buffer	negative control	0	0	negative

(\*) assuming a protein content in peanut = 25 g protein / 100 g peanut.

## 2. Cross-reactivity and sensitivity in extracts of food commodities

Common food commodities were extracted according to the kit instructions and the supernatants obtained diluted 4:10<sup>3</sup> in extraction buffer (4000 ppm). Each extract, spiked with roasted peanut butter extract (0.5 ppm), or with extraction buffer (negative control) was tested directly with the immunochromatographic sticks, according to the kit instructions, with the following results:

Commodity (4000 ppm)	Test results	
	Spiking with extraction buffer (negative control)	Spiking with 0,5 ppm peanut (0,125 ppm peanut protein*)
Barley	negative	positive
Buckwheat	negative	positive
Wheat	negative	positive
Rice	negative	positive
Rye	negative	positive
Oat	negative	positive
Corn	negative	positive
Soybean	negative	positive
Green Pea	negative	positive
Lima (butter) bean	negative	positive
Chickpea	negative	positive
Sunflower seed	negative	positive
Pumpkin seed	negative	positive

Commodity (4000 ppm)	Test results	
	Spiking with extraction buffer (negative control)	Spiking with 0,5 ppm peanut (0,125 ppm peanut protein*)
Sesame seed	negative	positive
Poppy seed	negative	positive
Almond	negative	positive
Brazil Nut	negative	positive
Cashew	negative	positive
Chestnut	negative	positive
Coconut	negative	positive
Hazelnut	negative	positive
Macadamia	negative	positive
Pistachio	negative	positive
Pecan	negative	positive
Pine Nut	negative	positive
Walnut	negative	positive
Skim milk powder	negative	positive
Cocoa	negative	positive
Lecithin	negative	positive
Porcine gelatin	negative	positive

(\*) assuming a protein content in peanut = 25 g protein / 100 g peanut.

### 3. Sensitivity in spiked food matrices

Common food matrices were spiked with roasted peanut butter (5 ppm), previously dispersed in 0.9% NaCl, or with 0.9% NaCl (negative control), before extracting and testing them according to the kit instructions, with the following results:

Food matrix	Test results	
	Spiking with 0.9% NaCl (negative control)	Spiking with 5 ppm peanut (1,25 ppm peanut protein*)
Cow Milk	negative	positive
Yogurt	negative	positive
Vegan (non-dairy) ice cream	negative	positive
Soy milk	negative	positive
Soy sauce	negative	positive
Pâtè	negative	positive
Cornstarch	negative	positive

(\*) assuming a protein content in peanut = 25 g protein / 100 g peanut.

#### 4. Sensitivity in an incurred food matrix

A common food matrix (butter cookies), incurred with peanut was prepared by spiking peanut-free, butter cookie dough with different amounts of roasted peanut butter, and baking the dough at 140 °C for 3 minutes. Baked cookies were extracted and tested according to the kit instructions, with the following results:

Food matrix	ppm (peanut)	ppm (peanut protein*)	Test result
Butter cookies	1000	250	positive
	100	25	positive
	10	2,5	positive
	5	1,25	positive
	1	0,25	weak positive
	0	0	negative

(\*) assuming a protein content in peanut = 25 g protein / 100 g peanut.

#### 5. Swab Test

Roasted peanut butter was dispersed in water at different concentrations and 0,045 ml of each sample applied to a 16 cm<sup>2</sup> square of a standard surface material (stainless steel), including a negative control (water) and allowed to dry overnight at room temperature. Surface samples were collected and tested according to the kit instructions, with the following results:

Reference material	Spiking level in liquid sample (mg/ml)	Peanut in 16 cm <sup>2</sup> surface (mg)	Peanut protein* in 16 cm <sup>2</sup> surface (mg)	Test result
Roasted peanut butter	0,4	18	4,5	positive
	0,2	9	2,25	weak positive
	0,1	4,5	1,125	negative
Water	negative control	0	0	negative

(\*) assuming a protein content in peanut = 25 g protein / 100 g peanut.

#### 6. Notes

Some acidic matrices (e.g. lemon and orange juice) are not compatible with the test. Testing of these matrices can produce false positive results.