

Document: K384AIE
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 Cat.# K384A **Soybean antigen EIA**

Instruction version: 011

Format version 011

SUMMARY

Determination of soybean % (w/w) content in meat products also is a part of quality control procedure in food control instances. The first quality dividing step is determination of "soybean free" products, in some countries "soybean free" cut off is postulated on the level of <1% w/w. Current kit formulation detects thermo-stabile epitopes of Soybean antigens and shows completely zero level of background in pure meat samples. In some cases kit allows semi quantitative determination of soybean content.

PRINCIPLE OF THE TEST

This test is based on two-site sandwich enzyme immunoassay principle. Extract of tested specimen is placed into the microwells. Soybean antigen with other antigens from the specimen binds on the microwell surface. Unbound material is removed by washing procedure. Monoclonal antibodies directed towards SOYBEAN ANTIGEN, and labelled with peroxidase, are then added into the microwells. After subsequent washing procedure, the remaining enzymatic activity bound to the microwell surface is detected and quantified by addition of chromogen-substrate mixture, stop solution and photometry at 450 nm. Optical density in the microwell is directly related to the quantity of antigen in the specimen.

KIT CONTENTS

	Code	Description	Qty	Units	Colour code
1	P384A	Plates with maximal adsorbtion capacity, 8x12 wells	1	pcs	
2	N003	Plate sealing tape	2	pcs	
3	S005Z	Pill for extraction buffer preparation	1	pcs	
4	C384AZ	Set of positive and negative controls, 1 ml each*	1	pcs	dark blue
5	S008Z	Washing solution concentrate 21x, 22 ml	1	pcs	colourless
6	T384AZ	Conjugate, 11 ml	1	pcs	blue
7	R055Z	Substrate solution, 11 ml	1	pcs	colourless
8	R050Z	Stop solution, 11 ml	1	pcs	colourless
9	K384AI	Instruction SOYBEAN ANTIGEN EIA	1	pcs	
10	K384AQ	QC data sheet SOYBEAN ANTIGEN EIA	1	pcs	
*	The set contains 0 and 1% of soybean				

materials required but not provided

- Distilled or deionized water;

NECESSARY EQUIPMENT

Microplate photometer with 450 nm wavelength and OD measuring range 0-3.0.
 Dry thermostat for 37°C

HANDLING NOTES

1. INFECTION HAZARD: There are no available test methods that can absolutely assure that Hepatitis B and C viruses, HIV-1/2, or other infectious agents are not present in the reagents of this kit. All human blood products, including patient samples, should be considered potentially infectious. Handling and disposal of this material should comply with the rules defined by appropriate local biohazard safety guidelines.
2. Avoid contact with 5% H₂SO₄. It may cause skin irritation and burns.
3. Do not use reagents after expiration date.
4. Do not mix or use components from kits with different lot numbers.
5. Replace caps on reagents immediately. Do not swap caps.
6. Do not pipette reagents by mouth.

STORAGE CONDITIONS AND STABILITY

1. Store the whole kit at 2 to 8°C upon receipt until the expiration date.
2. After opening the pouch keep unused microtiter wells TIGHTLY SEALED BY ADHESIVE TAPE (INCLUDED) to minimize exposure to moisture.

DISPOSAL OF THE KIT

Kit components should be considered as potentially infectious material and discarded according to appropriate local biohazard safety guidelines.

SPECIMEN COLLECTION AND STORAGE

1. This kit is intended for use with serum or plasma (ACD- or heparinized). Grossly hemolytic, lipemic, or turbid samples should be avoided.
2. Specimens may be stored for up to 48 hours at 2-8°C before testing. For a longer storage, the specimens should be frozen at -20°C or lower. Repeated freezing/thawing should be avoided.

ASSAY PROCEDURE**Reagent Preparation**

1. All reagents (including the desired number of unsealed microstrips) should be allowed to reach room temperature (18 to 25°C) before use.
2. All reagents should be mixed by gentle inversion or vortexing prior to use. Do not allow foam formation.
3. Prepare working wash solution: dilute wash solution concentrate (S008Z) 1:21 in distilled water – e.g., 1 ml concentrate + 20 ml distilled water. *Stability after dilution:* 5 days at 18-25°C or 30 days at 2-8°C.
4. It is recommended to spin down shortly the tubes containing calibrators on a low speed centrifuge.

Procedural Note:

It is recommended that pipetting of all calibrators and samples should be completed within 3 minutes.

Samples preparation

*1	Using a disposable plastic spatula, take a sample, put 1.0 g of it into a pre-weighted sampling tube. Add 20 ml of extraction buffer, close the tube with a screw cap and mix thoroughly (either by inverting or by vortexing).
*2	Extract should be cleared from particles (by sedimentation for NLT 2 hrs, or by short (3-5 min) centrifugation at 300-500 g, or by filtering through a gauze tampon or paper filter. Supernatant could be used in analysis (see table below)

Test flowchart

1	Put the desired number of microstrips into the frame; allocate two wells for each unknown sample and 6 wells for the controls. DO NOT REMOVE ADHESIVE SEALING TAPE FROM UNUSED STRIPS.
2	Dispense 100 µl of calibrator or unknown sample supernatant into the wells
3	Incubate 60 minutes at 37°C.
4	Prepare washing solution by 21x dilution of washing solution concentrate (code S008Z) by distilled water. Diluted washing solution is stable for 2 weeks at +2-8°C. Wash strips 3 times
5	Dispense 100 µl of Conjugate into the wells.
6	Incubate 30 minutes at 37°C
7	Wash the strips 5 times.
8	Dispense 100 µl of substrate into the wells
9	Incubate 15 minutes at 20-25°C
10	Dispense 100 µl of stop solution into the wells.
	Measure OD (optical density) at 450 nm.
	Set photometer blank on first calibrator
*	Apply point-by-point method for data reduction.

See the example of calibration curve in Quality Control insert.

DATA REDUCTION:

If OD of unknown samples is higher than OD of corresponded control it means that soybean content is more than 1%, if OD of unknown samples is lower than OD of corresponded control it means that soybean content is less than 1%.

Attention! For calculation of soybean content in sausages and canned meat products use different control samples.

Quality control

Control sample(s) should fit into the ranges shown in QC insert (see attached).