**Intended Use**
Morinaga “Peanut Protein ELISA Kit” is a sandwich enzyme immunoassay for the quantitative determination of peanut protein in processed or unprocessed food. This kit is designed to be used by quality control personnel or others who monitor foods for the possible presence of peanut proteins.

**Improvement from previous kit**
New Extraction Buffer: We developed a unique extraction buffer containing high concentration surfactants and reducing agents facilitating higher recovery.

**Characteristics of Kit**
- **High extraction efficiency**
  Food products can be subjected to processing treatments such as heating, pressurization and sterilization, during which peanut proteins contained in food are often denatured in various manners. Denatured proteins are often less water-soluble than native proteins, which sometimes renders their extraction from food difficult. The extraction buffer of this kit contains high concentration surfactants and reducing agents in order to solubilize and extract insoluble or hardly soluble peanut proteins.
- **High recovery**
  The higher extraction efficiency reaches, the higher recovery and detectability.
- **High sensitivity**
  Limit of detection is 0.3 microgram peanut protein / g food.
- **High specificity**
  The polyclonal antibody reacts specifically with soluble peanut protein mixture.

**Assay flow**
1. **Preparation and extraction of food specimens**
   - Homogenize, weigh sample (1g), and add extraction buffer (19mL).
   - Shake overnight, centrifuge, and filter.
2. **First reaction**
   - Room temperature, 1 hour
3. **Second reaction**
   - Room temperature, 30 minutes
4. **Enzyme reaction**
   - Room Temperature, 30 minutes in dark.
   - Stop enzyme reaction.
5. **Measurement of absorbance**

**Specification**
- Measuring Range : 0.78ng/mL-50ng/mL (0.3ppm-20ppm)
- Sample preparation time : 30-60 minutes
- Sample extraction time : Overnight (12 hours)
- Time for analysis : 2.5 hours
- The intra-assay precision (C.V.) : ≤10%.
- The inter-assay precision (C.V.) : ≤10%.
【Kit component】

<table>
<thead>
<tr>
<th>Name of component</th>
<th>Content</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>A Antibody-coated microplate module (One module consists of 8 wells)</td>
<td>6 modules / pack</td>
<td>2 packs</td>
</tr>
<tr>
<td>B Peanut protein standard (50 ng/mL)</td>
<td>1 mL</td>
<td>2 vials</td>
</tr>
<tr>
<td>C Enzyme-conjugated antibody</td>
<td>13 mL</td>
<td>1 vial</td>
</tr>
<tr>
<td>D Enzyme substrate (TMB solution*)</td>
<td>13 mL</td>
<td>1 vial</td>
</tr>
<tr>
<td>E Stop solution (1 N Sulfuric acid)</td>
<td>13 mL</td>
<td>1 vial</td>
</tr>
<tr>
<td>F Sample buffer (20X concentrate)</td>
<td>50 mL</td>
<td>2 bottles</td>
</tr>
<tr>
<td>G Wash buffer (20X concentrate)</td>
<td>50 mL</td>
<td>1 bottle</td>
</tr>
<tr>
<td>H Extraction component (10X concentrate)</td>
<td>55 mL</td>
<td>2 bottles</td>
</tr>
<tr>
<td>Frame for mounting the microplate module</td>
<td></td>
<td>1 piece</td>
</tr>
<tr>
<td>Microplate cover</td>
<td></td>
<td>1 piece</td>
</tr>
</tbody>
</table>

TMB solution* : Avoid exposure to light.

【Other required reagents and instruments】

- 2-Mercaptoethanol
- Distilled water (or Deionized water)
- Micropipets/disposable tips ranging 0.01 - 1mL
- Volumetric cylinders
- Polypropylene centrifugation tube
- Polypropylene test tubes (1-2 mL size)
- Tube racks
- Clear-plastic wrap
- Homogenizer/blender
- Horizontal shaker
- Vortex mixer
- Aspirator for washing procedure
- Microplate reader covering 450nm, and 610 to 650 nm

【Recovery of Model processed food】

Model processed foods were prepared by spiking peanut protein at 10 ppm to the food before the usual processing. After processing, recovery of Model processed food was evaluated by Peanut protein ELISA kit.

<table>
<thead>
<tr>
<th>Model processed food</th>
<th>Recovery (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Miso soup</td>
<td>86.8</td>
</tr>
<tr>
<td>Meat ball</td>
<td>87.5</td>
</tr>
<tr>
<td>Canned jelly</td>
<td>89.1</td>
</tr>
<tr>
<td>Canned orange juice</td>
<td>84.6</td>
</tr>
<tr>
<td>Corn soup</td>
<td>104.7</td>
</tr>
<tr>
<td>Tomato souse</td>
<td>109.6</td>
</tr>
</tbody>
</table>

【Contact us】

Morinaga Institute of Biological Science, Inc.
2-1-16 Sachiura, Kanazawa-ku
Yokohama 236-0003, Japan
e-mail: info@miobs.com