



BMA BIOMEDICALS



Peninsula Laboratories

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## S-5000 Extraction Kit

### For the Extraction of Peptides from Biological Fluids

#### **Contents:**

Buffer A: 1% trifluoroacetic acid (Y-1040); 2x 500ml in PP bottles.

Buffer B: 60% acetonitrile with 1% trifluoroacetic acid (Y-1045); 1x 200ml in PP bottle.

Sep-Column (50 columns) - C18 sorbent (Y-1000)

The kit components should be enough to extract 50 samples.

#### **Storage:**

Buffers should be stored 0 – 4°C. Store Sep-columns at room temperature. Opened bag should be stored in a desiccator.

#### **Recommended Use:**

##### **Extraction of Peptide from Plasma**

1. Add an equal amount of Buffer A to the plasma. For example, if you are using 1ml of plasma, acidify with 1ml of Buffer A. Centrifuge at 6,000xg to 17,000xg for 20 minutes at 4°C. Discard any pellet that may be present.
2. Equilibrate a SEP-COLUMN containing 200mg of C18 (Cat No. Y-1000) by washing with Buffer B or Buffer D (100% Acetonitrile) (1ml, once) followed by Buffer A (3ml, three times).
3. Load the plasma solution onto the pre-treated C-18 SEP-COLUMN.

**Note:** For steps 4 and 5, a light vacuum (10 sec per drop) may be applied to the column.

4. Slowly wash the column with Buffer A (3ml, twice) and discard the wash.
5. Elute the peptide slowly with Buffer B (3ml, once) and collect eluant in a polypropylene tube.
6. Evaporate eluant to dryness using a centrifugal concentrator or by a suitable method; e.g. lyophilizer.
7. Dissolve the residue in RIA buffer for radioimmunoassay as follows: For a normal subject, dissolve in 250 µl RIA Buffer for a two-tube assay. Aliquot 100 µL into each tube (50 µl is left over). If each tube is found to contain 3.963 pg/tube, then the total level of peptide in plasma sample = 3.962 pg/tube x 2.5 tube = 9.9 pg. If upon assaying, the peptide value exceeds or does not fall in the range of detection, dilute or concentrate the samples accordingly.
8. The total time for extraction should be 0.5 days for extraction and 0.5 days for lyophilization. Once the sample is lyophilized, it can be stored at -70°C before assaying, but should be assayed as soon as possible.

**NOTE:** This is a generic extraction protocol, which can be used for multiple types of samples. If a more suitable extraction method is found in literature, we encourage the researcher to use it.

Related Products: Y-1000: SEP-Columns (200mg, 50 per box)  
Y-1010: SEP-Column Adapters (15 per box)  
Y-1040: Buffer A  
Y-1045: Buffer B

This product contains no preservative and is intended for laboratory use and research purposes only. Purchase of this product does not include authorization to use it in diagnostic or therapeutic applications.

S-5000

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