Peninsula Laboratories International, Inc.

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Cat. No. Y-1000
Sep-Column

NOTE:
This instruction/datasheet provides information for general care and use of the Sep-Column

SORBENT: 200 mg of C18

Storage and disposal of cartridges:

Cartridges stored in their original sealed pouch remain stable for long periods. To store unused cartridges in opened pouches, squeeze the air out of the pouch, fold over the opened end of the pouch twice (if possible), seal with tape and store in a dessicator. Store dessicator in a cool dry place.

Dispose of used cartridges safely in accordance with government and local regulations.

Recommended Use:

Extraction of Peptide from Plasma

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

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

4. Slowly wash the column with Buffer A (3 ml, twice) and discard the wash.
5. Elude the peptide slowly with Buffer B (3 ml, once) and collect eluant in a polypropylene tube.
6. Evaporate eluant to dryness using a centrifugal concentrator or by a suitable method;

   Ex: Lyophilizer. Recommended to freeze eluant with dry ice/methanol for fastest freeze.

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/100 µL, then the total level of peptide in plasma sample = 3.962 pg/100 µL 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 such methods.