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# Immobilized Enzyme Column

The IMER is an immobilized enzyme column used for the analysis of choline (Ch) and acetylcholine (ACh). The column contains two enzymes, choline oxidase and acetylcholinesterase. The conversion efficiency is 100% due to the amount of immobilized enzyme that is present.

#### Instructions

- 1. Do NOT connect the enzyme column at this point. First, connect the HPLC column and stabilize the system with mobile phase. Make sure that any traces of storage liquid from the system or HPLC is washed out and the system is fully equilibrated with mobile phase.
- 2. Run mobile phase (pH7.5) and turn on the detector. Wait for steady baseline.
- 3. After the baseline is stable, remove the two-way joint and connect the enzyme column. DO NOT stop flushing with mobile phase.
- 4. Wait for a steady baseline.
- 5. Begin analysis by injecting a standard to check for appropriate peak area and retention time.

## Storing

If you plan to use in less than a month, you make store the column with mobile phase at 4°C. If the column will not be used for more than a month, flush with 0.1M phosphate buffer (pH8.2) and then store at in 4°C. DO NOT store it in the freezer.

## Warning

The enzyme can be inactivated by antiseptic, acid conditions, and organic solvents such as methanol. Even if only mildly acidic buffer (ie. pH6.5) is used the enzymes may be inactivated. DO NOT flush the enzyme column with any of these substances.

### **Activity of IMER**

- Maximum activity at 33 35°C.
- Maximum activity at pH7.5 8.5. No activity below pH6.5.

#### Age

The lifetime for the column can vary depending on use, but generally, the activity does decline with age. You should examine the peak size of a standard over time. Generally, the column will tolerate 500 - 600 injections of tissue homogenate or 1000 - 5000 microdialysis samples.

## Warranty

Like any other column, the IMER is a consumable product with limited lifetime. The IMER has to be used (tested) within a week after receiving. Claims for 'dead on arrival' warranty beyond this period of time will not be accepted.