

Protocol: Desalting Oligonucleotide using C18 Sep-Pak Cartridge

CAUTION: *Wear gloves, use RNAase free materials.*

1. Prepare a C18 Sep-Pak cartridge (Waters# WAT020515) by passing 10 ml of HPLC grade acetonitrile through it, followed by 10 ml of distilled water. Use a syringe to do this. *Keep air out of the cartridge!*
2. Load 3 ml of oligo solution using a syringe attached to the cartridge. Pass the oligo solution slowly through the cartridge. Collect flow-through in the same tube.
3. Wash cartridge slowly with 5 ml distilled water and collect 1 ml fractions (3 or 4 fractions).
4. For short oligos (less than 40 mer): Wash cartridge slowly with 3 ml of 60% methanol (HPLC grade or better). Collect 3 to 4 of 1 ml fractions.

For long oligos (greater than 40 mer): Wash cartridge slowly with 3 ml of 70% acetonitrile (HPLC grade or better). Collect 3 to 4 of 1 ml fractions.

Note: The majority of the oligo should elute in the first and second fraction.

5. Measure A260 for each oligo fraction.
6. Concentrate oligo-containing fractions in the Speed-Vac and dissolve to an appropriate final concentration in distilled water. Aliquote and store at -20 C.