

## CA-cleaning

Start the Magnatrix Software and login as Facility. Select Illumina → Illumina\_CA purification.

Fill in the following specifications in the interactive screen:

Sample volume: 50 µL

CA beads: 17 µL/µg DNA to be recovered

Binding time: 10 minutes

Precipitation buffer: 100 µL

EB: as required for further applications, but no less than 10 µL

Press forward.

Take a 96-well plate and fill it according to the instructions. Dilute DNA in EB if necessary to achieve volume. The precipitation solution is composed of PEG in 1.5 M NaCl and the concentration of PEG determines the length of fragments selected. A useful guideline is as follows:

| PEG concentration<br>in solution | Final PEG<br>concentration | Lower size cut-off |
|----------------------------------|----------------------------|--------------------|
| 16.6                             | 10.4                       | 180-250            |
| 15                               | 9.4                        | 250-300            |
| 14.1                             | 8.8                        | 300-350            |
| 13                               | 8.1                        | 430-470            |
| 12                               | 7.5                        | 480-520            |
| 11                               | 6.9                        | 600-800            |

All steps downstream of the use of beads should be conducted on a magnetic stand, to avoid bead carry-over, which inhibits most reactions, as well as affecting measurements.

*Special precautions with reagents:*

CA beads should not be vortexed. When not in use, keep them refrigerated.

80% EtOH should not be older than a week. If EtOH is to be kept overnight, ideally it should be placed at -20°C. When pipetting EtOH, resuspend a few times until bubbles are no longer formed. This prevents dripping during pipetting.

PEG should be kept refrigerated and protected from light.