Application Note #26

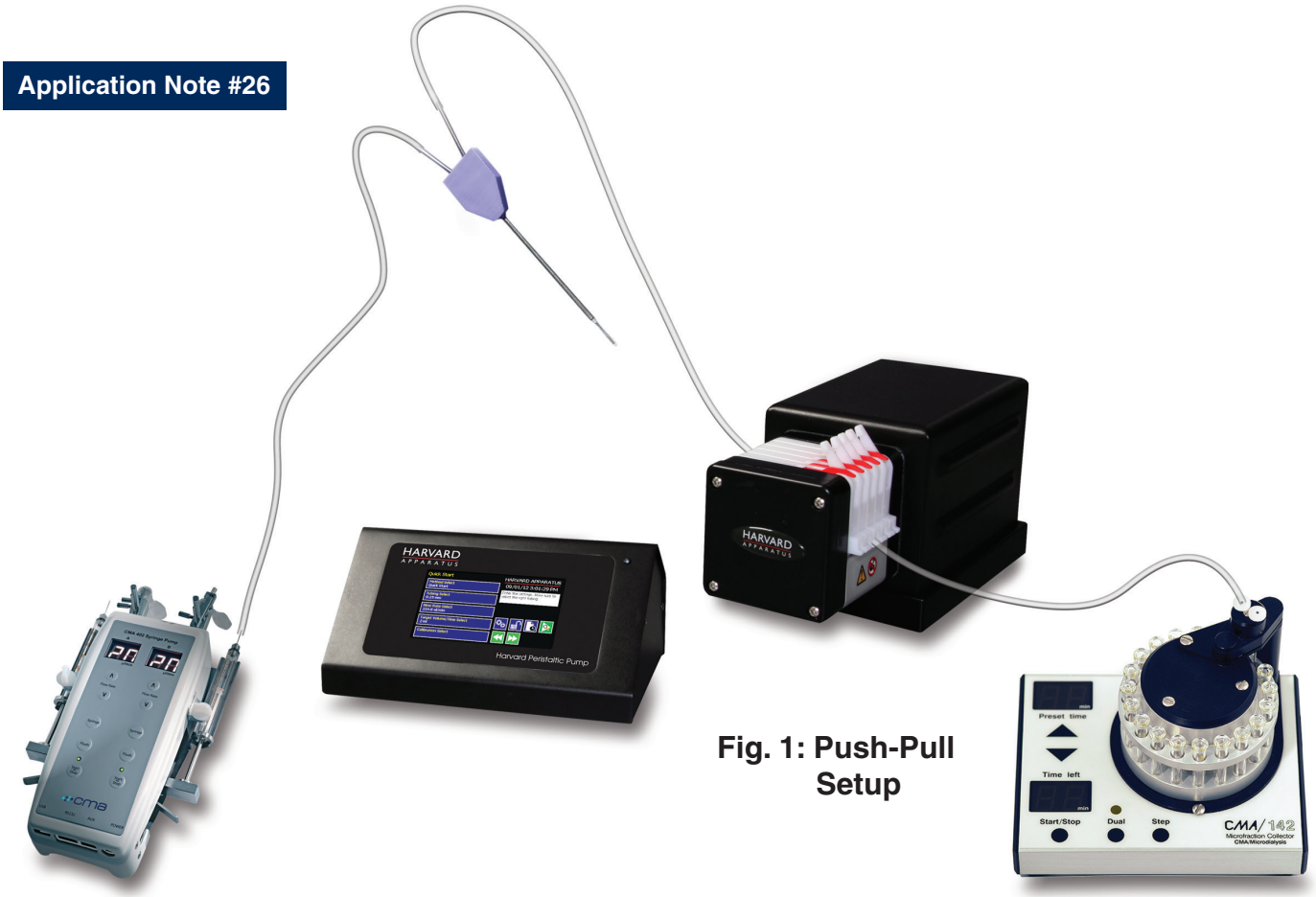

Fig. 1: Push-Pull Setup

Monitoring big molecules with Microdialysis: the use of a "push-pull" system with high molecular weight cut off probes

Microdialysis is a bioanalytical sampling technique that allows continuous sampling of compounds from the interstitial fluid of living tissue.

However the feasibility of using Microdialysis to sample big molecules (such as proteins, cytokines and neuropeptides), presents numerous challenges due to the loss of perfusion fluid through the high molecular weight cut off dialysis membrane. We refer to this loss of perfusion fluid through the large size pores of a high molecular weight Microdialysis probe as ultra-filtration.

One way to avoid ultra-filtration is to reduce the back pressure caused by the perfusion fluid coming from the probe inlet. We achieve this by performing Microdialysis in a "push-pull" mode using a syringe pump to perfuse the Microdialysis probe ("push") and a peristaltic pump to collect the sample coming from the probe outlet ("pull").

We developed a complete system to perform "push-pull" Microdialysis with our CMA 12 Ultra High probe that present a membrane with 1 million Daltons cut-off. The probe is perfused with a CMA 402 syringe pump and the samples are collected using a Harvard Apparatus peristaltic pump that significantly reduces the back pressure, thus avoiding ultrafiltration.

Fig. 1 shows a typical CMA-Harvard Apparatus "push-pull" setup. A CMA 402 syringe pump is used to perfuse the probe and a Harvard Apparatus peristaltic pump connected to the CMA 142 fraction collector to withdraw the sample.

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Instruction for use (see CMA 12 Ultra High Manual for details):

Syringe Pump:

1. Mount the probe in a Probe/Guide Clip on the CMA 130 In Vitro Stand and put the probe membrane into a vial filled with perfusion fluid.
   Connect the probe inlet with the syringe mounted on the CMA 402 syringe pump and flush the probe at 8-10 µL/min for 3-5 min to wash out air.
2. Set the pump to the required perfusion flow, usually 1-5 µL/min.

Peristaltic Pump:

1. Connect a CMA Tubing Connector to the inlet of the tubing on the Peristaltic Pump.
2. Connect a second Tubing Connector to the outlet of the tubing on the peristaltic pump.
3. Place the inlet end of the Tubing Connector in a beaker with Perfusion Fluid and flush the pump to fill all tubing with perfusion fluid.
4. Set the pump to the required perfusion flow, usually 1-5 µL/min. Make sure that the syringe pump and the peristaltic pump are running exactly at the same flow rate.

Final Connection:

1. Connect the inlet Tubing Connector at the peristaltic pump to the outlet of the probe.
2. Control the membrane to not ultrafiltrate.
3. Connect the peristaltic pump to the CMA142 fraction collector using the Tubing connector.
4. The system is now ready to use.

Ordering Information:

Probes	Ref.No.
CMA 12 Ultra High Cut-Off, 1 mm, pkg. of 3	**CMA 8011231**
CMA 12 Ultra High Cut-Off, 2 mm, pkg. of 3	**CMA 8011232**
CMA 12 Ultra High Cut-Off, 3 mm, pkg. of 3	**CMA 8011233**
CMA 12 Ultra High Cut-Off, 4 mm, pkg. of 3	**CMA 8011234**

Guides	
CMA 12 Guide Cannula, pkg. of 3	
CMA 12 Guide Cannula, pkg. of 30	

Accessories	Ref.No.
Tubing Adapter, pkg. of 10	**CMA 3409500**
FEP Tubing, 1 m, pkg. of 1	**CMA 3409501**
FEP Tubing, 1 m, pkg. of 10	**CMA 8409501**
Tubing Connector, pkg. of 3	**CMA P000113**
3-Stop Collared Tubing for P-70 Pump Head	**72-0654**
CMA 11 & 12 Probe Clip	**CMA 8309013**

Instruments	Ref.No.
CMA 402 Pump	**CMA 8003110**
Harvard Apparatus P-70 Peristaltic Pump	**70-7000**
CMA 142 Fraction Collector	**CMA 8381143**

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