



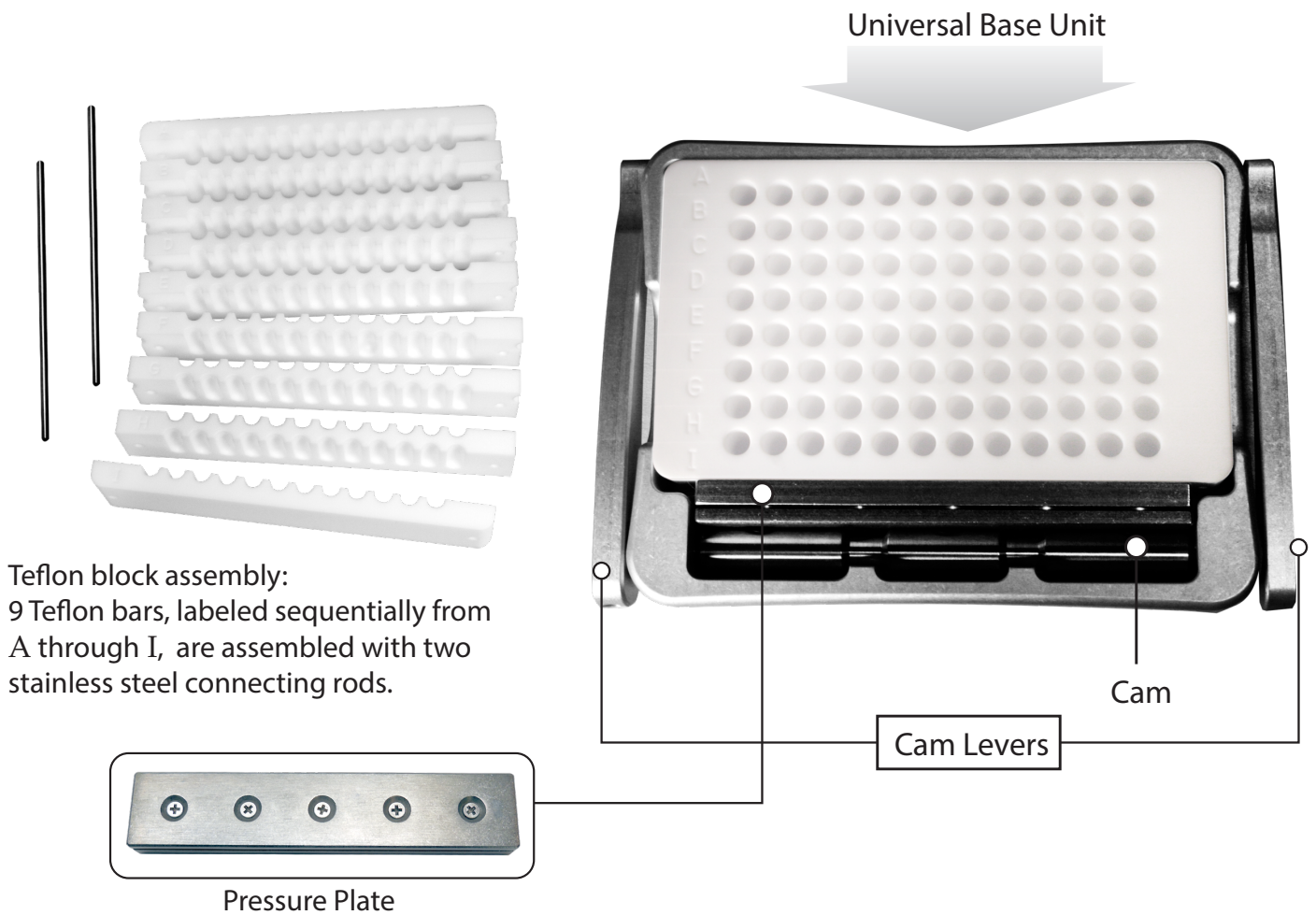
Operating Instructions

Model **HTD96B**



HTD96b : Operating Instructions

Components



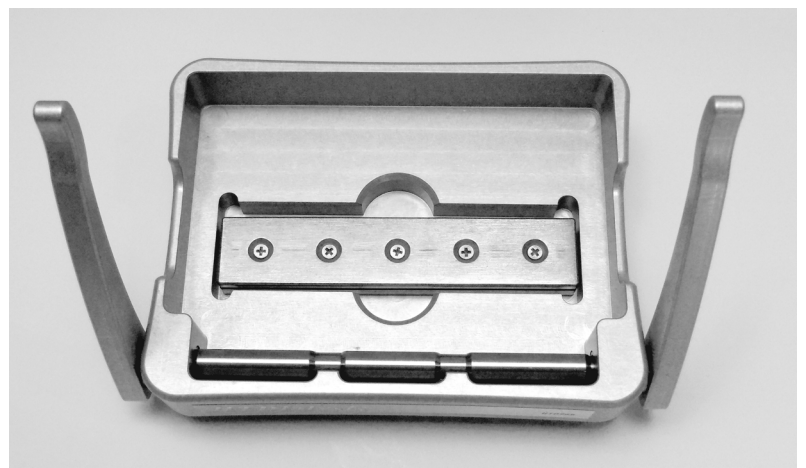
Clamp In Closed Position



Clamp In Open Position

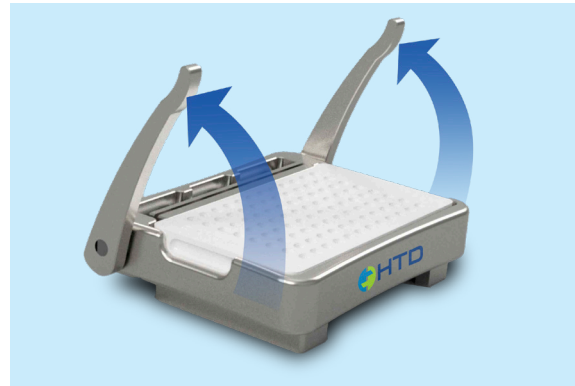


Place Stainless Steel Pressure Plate in Base Slot when unit is not in use

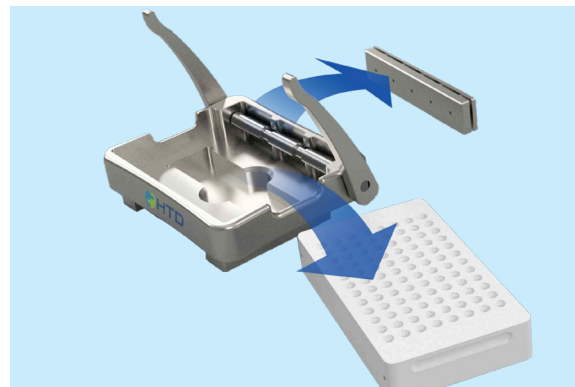


Cleaning the Teflon bars

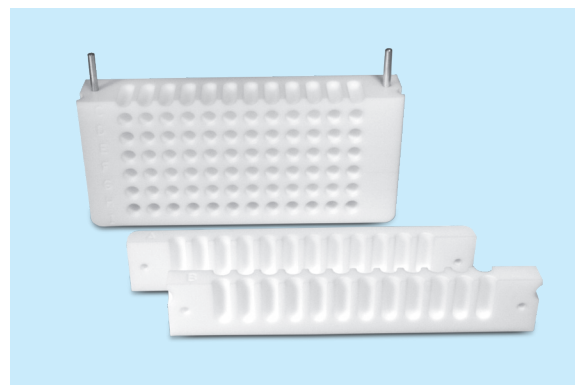
- 1 Open the clamp by rotating the cam levers 45 degrees. To ensure even pressure, always use 2 hands to operate the clamp.



- 2 Remove the entire Teflon block assembly from the clamp, then remove the stainless pressure plate.

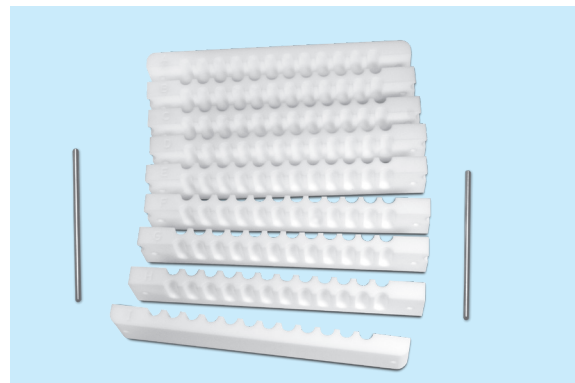


- 3 Remove one or two Teflon bars from one end of the assembly, then pull out both stainless steel connecting rods. (Teflon bars will disassemble.)



- 4 Clean separated Teflon bars with non-ionic laboratory detergent.

Note:
Always rinse away all detergent residue with distilled water before re-use.



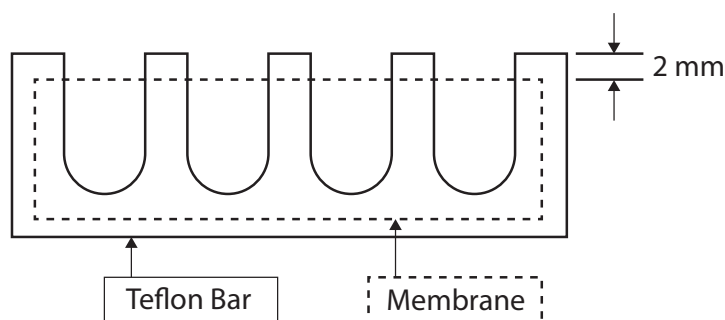
Hydrating the Dialysis Membranes

Each dry membrane strip consists of a pair of dialysis membranes that separate upon hydration. Hydrate the dialysis membranes by soaking in distilled H₂O or phosphate buffered saline (PBS) for 60 minutes. Add 20% by volume ethanol and soak for a further 20 minutes. Membranes are ready for use or may be left in the 20% ethanol for extended storage. Prior to use membranes must be rinsed twice in distilled H₂O or PBS.

- Make sure only a single sheet of dialysis membrane is placed between Teflon bars.
- Wipe off excessive buffer from the membrane.
- Flatten membrane before the next Teflon bar is put in place.

Assembling the Teflon Bars with Dialysis Membrane

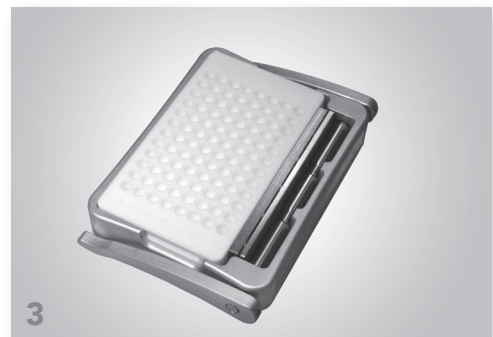
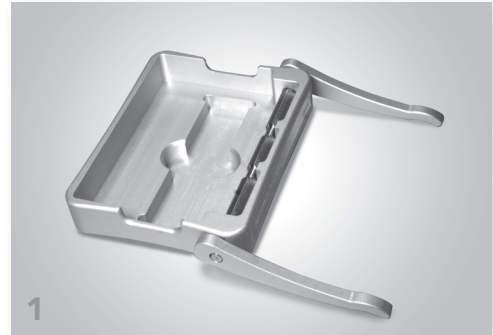
- 1** Lay the first Teflon bar (labeled A) flat on the bench (see picture), insert the two stainless steel connecting rods so they are perpendicular to the Teflon bar.
- 2** Place the membrane on the Teflon bar. Ensure that the membrane is approximately 2mm below the top edge of the bar and the lower membrane edge overlaps the bottom of all wells.
- 3** Repeat layering dialysis membranes and Teflon bars until the unit is fully assembled (assemble Teflon bars in alphabetical order).



Note: Although the number of membranes inserted can be varied to suit the experimental requirements, all the Teflon bars must be assembled before loading the Teflon block into the base.

Loading the Teflon Block into the Base

- 1 Make sure the clamp is in the open position before loading the Teflon block into the base.
- 2 Insert the Teflon block into the base, then place the stainless steel pressure plate between the Teflon block and the cam as shown.
- 3 Always tighten the assembled unit with even pressure by using both hands to rotate the cam levers.



Fully assembled unit

Important Note:

Immediately add buffer to the dialysate side of the wells to prevent dehydration of the membranes before the test samples are added.

Unit is ready for use!

Loading Samples into Dialysis Wells

Samples can be added to the dialysis wells using any standard 8- or 12-well multi-channel pipetting device or 96-well pipetting instrument. Test samples are most conveniently prepared in 96-well tubes or plates, since this facilitates preparation of dilutions, replicate samples and mixing. Volumes of 25 to 150 μL can be used in this apparatus, however it is essential that equal volumes are used on each side of the membrane.

Dialysis

After loading, cover the top surface of the Teflon block with an adhesive sealing film (e.g., HTD adhesive sealing film Catalog #1102) to prevent evaporation and maintain a constant pH. The dialysis block can be incubated at any desired temperature. Equilibrium is reached more rapidly if the block is placed on an orbital or reciprocating shaker. Although most compounds reach equilibrium in about four hours, the standard protocol recommends six hours dialysis to ensure equilibrium conditions are achieved.

Samples can be added or removed from the dialysis well at anytime during the experiment by merely removing the adhesive seal to gain access.

Care and Storage of the HTD96b

The HTD96b base and stainless steel pressure plate do not require routine washing and should merely be wiped clean with a paper towel. However, after each use, the Teflon block should be cleaned with a non-ionic laboratory detergent and rinsed thoroughly with distilled water. Care must be taken to ensure that all detergent has been rinsed from the Teflon block as residual detergent could compromise future binding studies. The Teflon block may also be disinfected with a 10% bleach solution if desired. It is essential to always store the stainless steel pressure plate in the base slot that is under the Teflon block to eliminate continual pressure on the Teflon block. Storing the HTD96b unit in the closed position with the pressure plate inserted in the clamp for extended periods may compress the Teflon and reduce the effectiveness of the base clamp.

Pressure Plate Maintenance

Normal operation of the HTD96b does not require washing of the pressure plate assembly and therefore no scheduled maintenance is required.

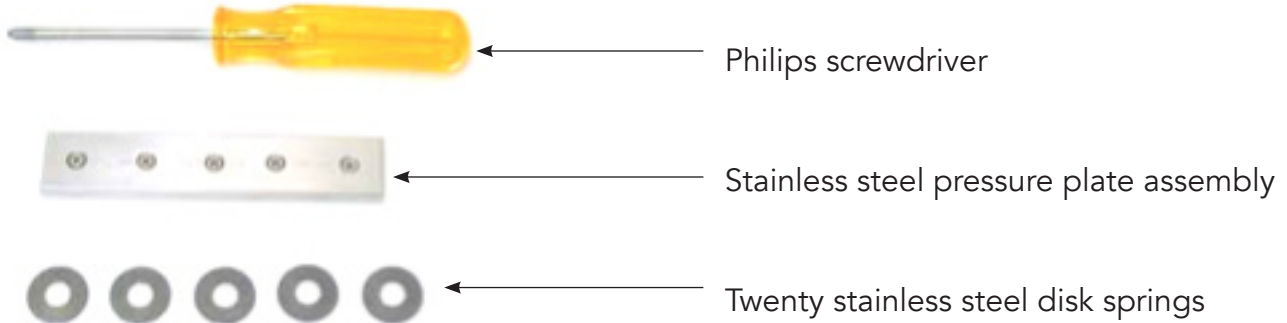
The two pressure plates are separated by disk springs that allow the application of sufficient force to prevent seepage or leakage between wells in the Teflon block. Although these disk springs are made of series 17 stainless steel, prolonged exposure to aqueous solvents or oxidizing cleaning agents may result in some rust. To minimize exposure to aqueous solvents the springs are coated with a silicone seal during factory assembly.

If the experimental protocol uses radioisotopes or other hazardous reagents we recommend the following options:

- A** Wrap the pressure plate assembly in a film of clear plastic wrap to isolate it from the liquids.
- B** Line the base with clear plastic wrap before inserting the Teflon block, thereby isolating the entire base from the Teflon block and all reagents.
- C** If it is necessary to wash the pressure plates, disassemble after every 25 uses, dry, coat with fresh silicone seal and reassemble.

Stainless steel disk springs replacement procedure to eliminate spring oxidation

Components



- 1** Disassemble the pressure plate assembly by removing five setscrews with a Phillips screwdriver.
- 2** Remove stainless steel disk springs, dry and wipe clean, coat with an appropriate silicone sealant. Place the stainless steel disk springs in sequence as shown in Figure 1 (four disks each column)
- 3** Reassemble pressure plate assembly by placing plate 2 over the disk springs and tightening the setscrews until the unit height is 0.46" everywhere. Figure 2

