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OPERATING MANUAL

BFS FREEZING STAGES
MODELS BFS-3, BFS-5, BFS-30

APPENDIX I

OPERATING INSTRUCTIONS FOR PHYSITEMP PUMP AND TANK UNIT, PTU-3

1. Unscrew cap. Fill reservoir with 5 gallons of distilled water.

2. Connect extension tubing to the freezing stage tubes and the connections on the tank. Direction of the flow is not important. All water fittings are automatically self-sealing when disconnected to prevent water spills.

3. Connect the AC line cord to the receptacle on the pump and to the outlet on rear of the power supply. This ensures that the water is always flowing when the power supply is on.

4. Switch on the power supply.

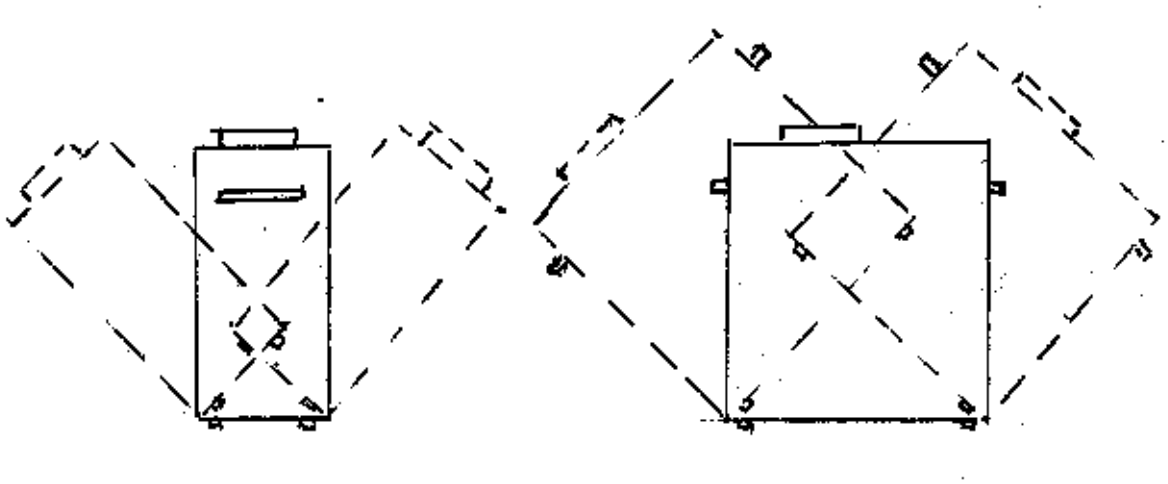
GENERAL INSTRUCTIONS

The use of distilled water is recommended. This avoids discoloration of the tubing due to organic matter in untreated water. A purification agent can be added to the water. Any commercially available dehumidifier treatment in liquid, powder or tablet form may be used.

If the stage will be operated for long periods, water temperature may gradually increase. Ice cubes can be added to the tank to maintain low temperature.

The tank should not be placed more than three feet below the stage itself.

When filling the reservoir for the first time, or when refilling it, a small amount of air may become trapped in the pump housing and cause intermittent noise, cavitation and a reduced flow rate. If this occurs, screw the cap firmly onto the tank and tip it an an angle of approximately 45 degrees to the horizontal in all four directions as shown below. This will eliminate the air in the pump housing and restore flow to its maximum.



8.0 MEASUREMENT AND CONTROL OF TISSUE TEMPERATURE

8.1 The optimum cutting temperature for a given type of tissue is best determined by experiment. Several Physitemp thermometers are available for such measurements; they also have many other laboratory uses. There are several sizes of needle probe, which can be inserted in the specimen either before or after freezing. The BAT-12 thermometer, which has a range from -100°C to +200°C, permits the user not only to find optimum cutting temperature, but to return to it subsequently.

8.2 One of the main advantages of these freezing stages is the ease with which the temperature of frozen tissue can be varied, simply by adjusting the CURRENT control. The following temperatures have been found satisfactory:

Tissue	Temperature °C
Kidney	-15
Brain	-18
Liver	-18
Thyroid	-20
Skin	-30
Keloid	-35
Patella	-40

9.0 MAINTENANCE, WARRANTY AND SERVICE

9.1 MAINTENANCE. The stage needs no maintenance at all. It may be cleaned as necessary with a soft cloth, water or detergent. DO NOT IMMERSE IN WATER. Because of expansion and contraction due to the wide temperature range, it is not possible to completely seal the stage.

9.2 WARRANTY. Physitemp Instruments Inc. warrants this instrument to be free from defects in material and workmanship for 12 months from date of shipment. Repair or replacement will be made at no charge at the discretion of Physitemp if the defect is not the result of misuse or abuse. Physitemp accepts no consequential liabilty for delay in delivery, alleged faulty performance of the product, or for any other cause.

9.3 REPAIRS AND RECALIBRATION. For technical or applications information on this instrument, contact us at:

Tel 973-779-5577
Fax 973-779-5954
e-mail physitemp@aol.com.

In the event that any part of this system is to be returned for repair or recalibration, please pack it with care and send it prepaid to:

Physitemp Instruments, Inc.
Service Department
154 Huron Avenue
Clifton, NJ 07013 USA

Please include with the instrument:

(1) A note describing any problems encountered.
(2) The name and telephone number of the user or other person we can contact.
(3) The complete return address for shipping.

For your protection, please pack returned items carefully, and insure them against possible damage or loss in transit. Physitemp will not be responsible for damage resulting from careless or inadequate packaging. Please return freight prepaid.

5.4 To return the stage to normal it is necessary only to restore the water flow. Within seconds, the stage will automatically switch on; this can be checked by noting that the meter is indicating current flow.

6.0 OPERATING THE EQUIPMENT

6.1 Check that cold water is flowing through the stage, as described above.

6.2 Plug the black connector plug into the socket of the right side of the Power Supply.

6.3 Place tissue specimen on the stage. If it is dry, first moisten the surface with a few drops of water to provide better heat conduction. The thinner the specimen, the faster it will freeze. These powerful stages will freeze a block of tissue over 1" thick, but it will take several minutes to do so. Whenever possible, tissue thickness should be limited to 2-3mm. With this thickness, sectioning can commence in less than two minutes.

6.4 SPECIMENS PRESERVED IN FORMALIN should be thoroughly washed in water before attempting to freeze them. Formalin depresses the freezing point of water to such an extent that the specimen may not freeze sufficiently for sectioning even though it may be below -30ºC,

6.5 If even quicker freezing is desired, a blast of CO_2 can be applied to the specimen when the Power Supply is switched on.

6.6 Switch on the Power Supply by turning the CURRENT control clockwise. Maximum cooling is achieved at a current of 10.5 Amps. There is no advantage in increasing current beyond this point.

6.7 When the specimen is completely frozen, commence sectioning using normal techniques. If specimen is too cold, reduce cooling current. See Section 8.

6.8 When sufficient sections have been made, remaining tissue is easily removed from the stage. Press the spring-loaded switch to THAW position and hold it down until the frost on the stage surface is seen to melt. This takes about ten seconds. Switch the Power Supply off, slide the tissue specimen from the surface of the stage. Wipe the stage.

6.9 If the stage is being used in a Cryostat, it is essential to remove the water from its tubing. This is most easily done by blowing gently into one of the tubes until air has replaced the water.

7.0 TYPICAL STAGE PERFORMANCE

7.1 The degree of cold obtained is dependent on the temperature of the cooling water. Thus, lower stage temperatures are achieved in winter than in summer. At 10.5 Amperes the stage temperature should reach 55-65ºC below that of the cooling water. Thus a stage temperature of -40ºC can be achieved under normal conditions. A 1/2" thickness of liver which covers the stage should freeze with 15-20 minutes.

Temperature throughout the tissue should drop to -20ºC in a further 10 minutes. Tissue under 1/16" in thickness should be ready for sectioning with 90 seconds after switching on.

3.0 THE POWER SUPPLY

3.1 The power supplies for all stages are physically alike. They plug into any standard three wire outlet or, with an adaptor, into older two wire outlets. The power supplies for the BFS-3 or 5 are capable of providing an output 5V DC at a maximum of 10 amps. The BFS-30 power supply provides 12V DC at a maximum of 10 amps.

Please note: The BFS-30 and BFS-3/5 supplies are not interchangeable and under no circumstances should a BFS-3 or 5 stage be used with the BFS-30 power supply.

3.2 BFS units have only two controls:

(1) The CURRENT control. This increases cooling when roated clockwise. When turned fully counter-clockwise, it clicks off.

(2) The FREEZE/THAW switch. As long as this spring-loaded switch is held down, the stage top plate will heat instead of cool. The fast warmup facilitates removal of frozen tissue from the stage.

4.0 SET-UP: MOUNTING THE STAGE

4.1 If the user specified the make and model number of his/her microtome when ordering the freezing stage, it should have been supplied with the correct mounting hardware already in place. Available hardware includes a 3/4" vise block and two 1 1/2" pillars of diameter 15/32" and 3/8". Special mountings can be made to order.

4.2 The stage has a 10/32" tapped hole in its underside. Into this hole can be screwed either of the two pillars or the square block, which is supplied with a stainless steel screw. If not already fitted, the correct pillar or vise block should be screwed into the underside of the stage and tightened. The stage should then be firmly clamped in the microtome and adjusted so that connecting leads and tubing are clear of the knife and other moving parts.

5.0 SET-UP: WATER COOLING

This section gives instruction for set-up if the equipment is to be used with tap water. If a Pump and Tank Unit is to be used to circulate the cooling water, see Appendix 1.

5.1 Thermoelectric modules are solid-state heat pumps: As one side gets lower in temperature, there is a corresponding rise in temperature on the other side. The head pumped through the module must be removed. This can be done either by air or water cooling. The former requires copper radiation fins, for which there is little space in most microtomes. In BFS stages, water cooling is used. PVC tubing carries a trickle of water from the lab faucet to the module and leads it away to waste disposal.

5.2 Connected to the stage are an electrical lead and two PVC tubes which terminate in male self-sealing connectors. A pair of 5' water tubes with mating female connectors is also provided. Connect the 5' tubes to the tubes on the stage. Attach one tube to the laboratory water supply, while the other goes to a convenient sink or drain. The direction of the water flow through the module is not important. The water supply should be adjusted until it flows steadily from the output tube without spurting.

5.3 If the water supply is accidentally reduced or turned off while the equipment is operating, the stage will tend to heat up. However, the electronic safety control will sense the rise in temperature and close down the equipment before damage can occur. When this closedown occurs, the meter on the power supply will indicate zero current flow; the red pilot lamp will continue to glow.

OPERATING INSTRUCTIONS FOR PHYSITEMP FREEZING STAGES

1.0 INTRODUCTION

1.1 This manual gives general information and detailed operating instructions for three freezing stages manufactured by Physitemp Instruments Inc for use in microtomes. It also cover the FRM systems which comprise an Erma microtome complete with BFS Freezing Stage.

1.2 All stages operate in the same manner. However, they differ in several ways, namely:

1) Surface area
BFS-3 has a 3.1 x 3.8 cm stage area
BFS-5 has a 3.8 x 3.8 cm stage area
BFS-30 has a 8 x 8 cm stage area

2) Minimum operating temperature under no load conditions
(cooling water temperature 20°C)

BFS-3	-38°
BFS-5	-35°
BFS-30	-40°

1.3 This series of freezing stages includes major improvements over our earlier equipment. There is an electronic control built into each stage which automatically closes down the equipment if the device over-heats. Overheating never occurs during normal use, but can be caused by two abnormal conditions:

a) seriously reduced water flow to the module, see Section 5.

b) the spring-loaded FREEZE/THAW switch being held at THAW for too long (see Section 6.8)

1.4 Every BFS stage is supplied complete with Power Supply which converts 115V AC house current to the non-lethal low-voltage current which operates the freezing stage. The stage itself is normally supplied with mounting hardware to fit the user's microtome.

1.5 BFS-3 and BFS-5 are intended for use in clinical or Minot type microtomes. The larger BFS-30 stage is designed for use in sliding or sledge microtomes. Mounting hardware is available to fit Physitemp freezing stage to almost every model of microtome.

2.0 FEATURES OF THERMOELECTRIC FREEZING

2.1 Thermoelectric cooling was discovered by Jean Peltier in 1834. However, practical uses for the Peltier Effect had to await developent of such efficient material as bismuth telluride. These materials have become available only in recent years as a by-product of transistor technology.

2.2 Thermoelectric freezing stages provide deep and continual cold when fed with low-voltage direct electric current. They consume no materials, need no maintenance and require only a supply of cooling water. Freezing at any desired temperature down to -40°C is maintained just as long as current is supplied to the stage. The electrical supply to the stage is perfectly safe and non-lethal, as it never exceeds 15V DC.

2.3 Thermoelectric freezing eliminates the need to use aerosol spray coolants or freon.

2.4 If the direction of current in a thermoelectric module is reversed, heat instead of cold is produced on the top plate. All BFS units make use of this characteristic, and include a FREEZE/THAW switch, which reverses the current. This provides a quick method of melting the stage-tissue interface, so that solid block of frozen tissue may be quickly removed from the stage.

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