Vibroslice NVSL & Vibroslice NVSLM1

Manual and motorized tissue slicers

INSTRUCTION MANUAL

Serial No._____

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World Precision Instruments, Inc.

Contents

Introduction

The **Vibroslice** was designed to cut 50-700 μm thick slices of fresh brain tissue, but it has proved useful with a wide range of other tissues such as kidney or liver. It is also useful with fixed tissue where sections down to 20 μm thick help with the penetration of reagents for histochemistry. Vibrating blade slicers such as the **Vibroslice** appear to cause less damage than some alternative methods such as tissue choppers. This has proved valuable in preserving neurones in certain brain regions.

As the **Vibroslice** contains exposed moving parts and a blade capable of cutting tissue, it is important to take adequate safety precautions.

The blades used with this instrument are extremely sharp and should be handled with care. Dispose of used blades carefully.

Spillage — If the cutting lubricant/preserving liquid (*e.g.,* physiological saline) spills over the instrument it is important for electrical safety reasons to ensure that the instrument remains safe to use. To avoid the possibility of electrical shock if a spillage occurs, the unit should be unplugged, inspected and, if necessary, tested by a qualified technician before being used again.

This instrument must not be operated unless it is properly grounded.

User servicing is limited to changing the drive belts when necessary; the instrument contains no other user-serviceable parts. Contact WPI if you require assistance. WPI will not accept equipment for service or repair unless it has been cleaned of all contaminates and has been adequately packaged.

Operation

Vibroslice NVSL with manually advanced tissue bath

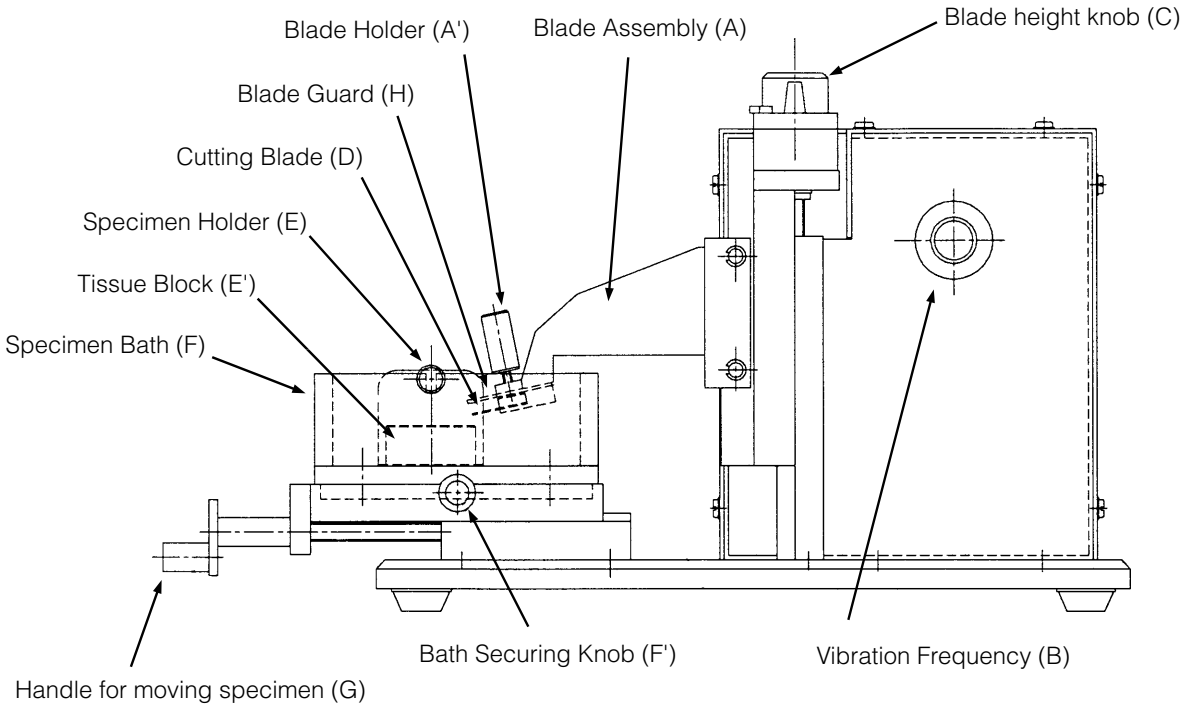

Figure 1

Fix a new blade (D) in the slot in the blade assembly (A, A') and fix the specimen holder (E) in the bath (F). It should be noted that the blade (D) is protected by a blade guard (H). Care should be taken when setting up the instrument and when replacing the blade. You may wish to pre-set the height of the first cut by lowering the blade until it just touches the stage (E'), and then raising it the required distance using knob (C).

Remove the brain or other tissue and trim as necessary.

Glue the tissue block to the specimen holder (E') using a thin film of cyanoacrylate (Super Glue) adhesive.

Pour cold, freshly oxygenated physiological saline into the bath (F) until the tissue block is immersed.

Set the blade vibrating by depressing the non-latching foot switch and adjust the vibration frequency (B) as required.

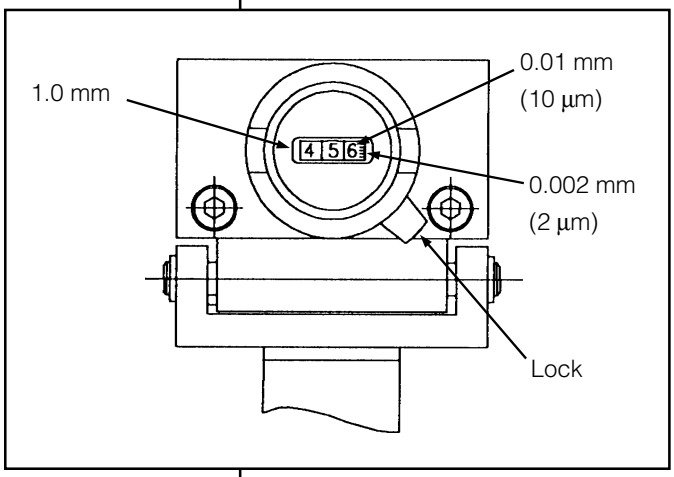

Figure 2

Advance the tissue block onto the blade using the handle (G). When the blade begins to cut you may find it necessary to adjust the blade frequency to a more suitable rate.

When the cut is finished, the tissue block should be retracted from the blade by reversing the rotation of knob (G). You may wish to remove the tissue slice before retracting the tissue block.

Lower the blade by the required slice thickness. The numerals on the knob (C) represent 10 µm, the small sub-divisions being 2 µm as shown in Figure 2.

Please note that the increments specified are nominal values only.

Repeat the above steps until you have sufficient slices, or you have run out of tissue.

Vibroslice NVSLM1 with motorized advance tissue bath

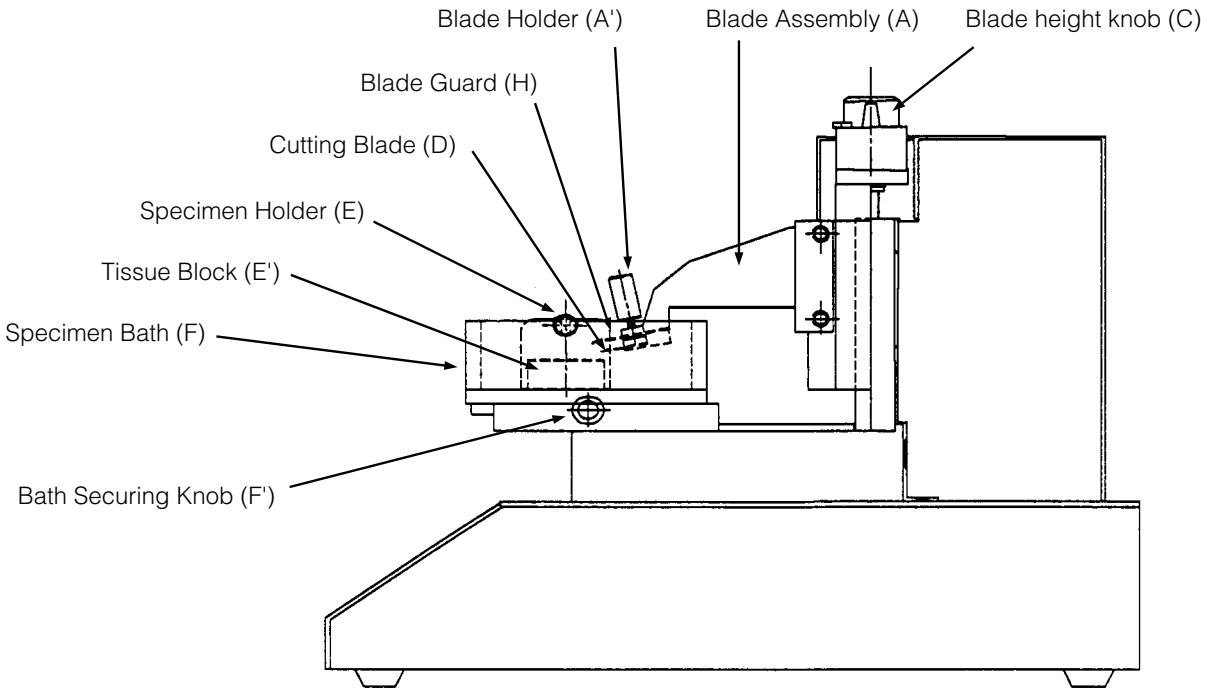

Figure 3

Operation of the **NVSLM1 Vibroslice** is generally the same as the **NVSL** except that an electric motor removes the chore of repeatedly advancing and retracting the tissue bath by hand. The bath feed motor is controlled by a three-position switch which controls bath feed direction and a knob (graduated 1-11) which varies the speed of the bath advance.

Push the switch upwards for “bath advance” to cut the tissue. When the cut is finished set the switch to the center “off” position. Reverse the bath by moving the switch down to “bath reverse”, and holding it down as this position is biased to “off”. Limit switches will stop the carriage automatically at the limit of travel.

The speed of the bath advance is continuously variable. The speed of the bath reverse is fixed.

Blades

The recommended blades are the “Valet” blades (WPI part no. **BLADES**). Care should be taken when handling, inserting or removing blades.

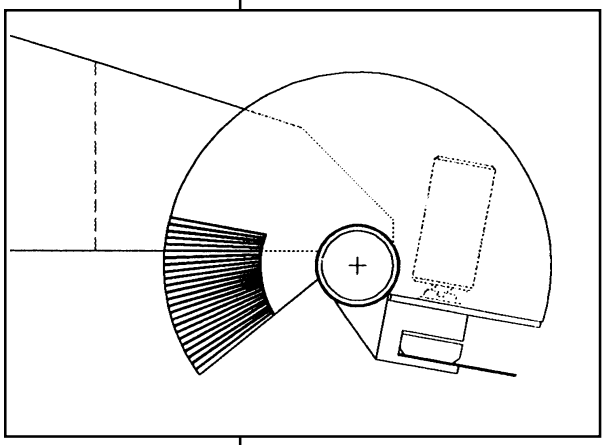

Figure 4:

Use of protractor for adjusting blade angle.

The protractor supplied with the instrument is designed to facilitate the setting of the blade angle. The protractor should be slipped over the inner hub of either the blade locking nut or locking screw and rotated so that its leading edge rests on the blade guard as shown above.

The blade angle may be set by loosening the locking screw and rotating the blade holder until the desired the graduated line is aligned with the lower edge of the nosepiece. The lines on the protractor are spaced at 2° (nominal) intervals. Experience has shown that an angle of approximately 12° is suitable for the blades supplied with the instrument. Should you wish to personalize the protractor to suit a particular blade or tissue type, additional protractors are available.

Preparing Tissue

Glued tissue should be immersed in the physiological saline quickly to minimize any anoxic damage.

Trimming the tissue block is important because the orientation of the slices is determined by the way the tissue is glued to the stage. Parasagital cuts have been found useful for transverse slices of rat dorsal hippocampus (glue down the cut midline) and for preserving the planar dendritic trees of Purkinje cells in folia of the cerebellum close to the midline (a Parasagital initial cut should be made which removes the lateral lobes). In some cases it is easier to dissect the target structure free before mounting it on the stage. In others leaving the adjacent tissues on can give useful mechanical support (*e.g.,* with transverse hippocampal slices). Tough connective tissue can cause trouble and should be cut off; or the block can be oriented to keep it out of the way of the cutting of the larger structure. In a few special cases, additional support may be needed from external agents, such as embedding in Agar, or in formalized albumin (fixed tissues only).

Fixing Tissue

The block of tissue must be fixed firmly to the stage. Apply a thin film of cyanoacrylate adhesive to the stage over an area large enough to accommodate the whole of the cut surface of the tissue block.

Two common problems associated with fixing tissue are:

i. The tissue block floats away during cutting. This means that the glue has not bonded properly. This is usually due to the specimen being too wet. Use filter paper to draw off the excess liquid from the block before placing it on the glued stage.

ii. The glue forms a rigid film up the side of the block, interfering with the cutting process. This probably means that too much glue has been used.

Preparing the Tissue Bath

Slices are cut under liquid to lubricate the blade as it cuts, and in the case of fresh tissue, to avoid anoxia. The tissue block should be covered to a depth of approximately one millimeter; much more than this reduces visibility due to rippling of the liquid surface.

Cutting fresh tissue under cold (<8 °C) physiological saline improves the quality of recordings, presumably by reducing the metabolic rate and avoiding anoxic damage. Keeping the saline on ice while oxygenating it may be enough. However, the saline warms up remarkably during the cutting process (to 10 °C in 5-15 minutes depending on conditions). An ice-cold stainless steel block in lhe bottom of the chamber can help, as can freezing the whole cutting chamber with a few millimeters of physiological saline in the bottom (spare chambers are useful in this case). Chilling can cause condensation on the stage interfering with gluing the tissue block; either wipe it dry at the last moment or keep it out of the freezer and insert it in the chamber just before slicing. In the latter case a silicone rubber insert can be used to leave a gap in the frozen saline. A simpler solution is to buy the **Peltier Cooling System** (WPI part no. **13856**)which has been designed to fit onto the Vibroslice.

Speed adjustment

Three factors govern the cutting action: The frequency of the blade vibration (oscillation), the speed of advance of the tissue block onto the blade and the amplitude of the blade movement. The vibration frequency rarely needs changing. The fastest setting is generally suitable for slices of fresh brain over 200 μm thick. Thinner slices may benefit from slower vibration.

The rate of cutting, which is determined by the speed of the tissue advance, is one of the most important factors in successful slice cutting. It should be relatively slow (one turn every few seconds). If it is too fast, the blade tends to deform the tissue, often pushing it over rather than cutting it cleanly. What is meant by “too fast” depends on the local consistency of the tissue; it tends to be slower in tougher tissues. Sometimes local areas of tougher tissues may require withdrawing the blade a little before resuming the cut. It always pays to monitor the tissue block as the cut progresses to check that all is well; a magnifier or dissecting microscope may help, as will good illumination.

The blade movement amplitude is fixed by an eccentric cam to 1 mm — this has proved suitable for most applications.

If the blade consistently deforms the tissue block rather than cuts it, several possibilities should be considered. The simplest is to use a shorter block of tissue so that the cuts are made closer to the rigid support of the stage. In extreme cases extra support may be needed, most simply by leaving adjacent tissues attached. In a few cases the tissue may need to be embedded in Agar, or perhaps fix a suitable support on the stage next to the tissue, such as cork or pith (not expanded polystyrene as this disintegrates in contact with cyanoacrylate adhesive). If tissue block deformation is a regular problem, it may be worth modifying the stage or blade holder, in which case contact WPI for advice.

Slice Thickness

Slice thickness depends on the application for which the tissue is being cut. For most physiological studies on brain slices, the thickness is a compromise between anatomical integrity (better in thicker slices), and oxygenation of the tissue (better in thinner slices). There are several experimental and theoretical accounts of the “limiting thickness” of tissue slices, which is the thickest slice with a non-anoxic center (*e.g.,* Warburg, 1930; Elliott, 1969; Harvey, Schofield & Brown, 1974). Most

researchers usually use 400 µm brain slices equilibrated with 95% O_2, 5% CO_2 at 35 °C. If thicker slices are essential, then consider reducing the temperature to reduce the metabolic rate, or use a smaller animal. Slices of up to 750 µm have been described (Halliwell, 1975). Thinner slices of around 100 µm may be needed for applications involving direct visualization of cells (Yamamoto & Chuko, 1978; Keenen *et al.,* 1988; Konnerth, 1990), or where the problem is to improve the penetration of reagents for horseradish peroxidase labeling.

Removing Slices

Slices can be transferred as they are cut, or accumulated on the stage until the end of the run. A small stainless steel or plastic spatula ground to a fine edge resembling a chisel is useful to help slide the slice on and off. Alternatives are fine paintbrushes to wrap the slices on or wide-mouthed Pasteur pipettes to suck the slice up in a column of saline.

Maintenance

The robust construction of the Vibroslice means that little maintenance is necessary apart from keeping it clean. The stage should be scraped clean after each use, removing all traces of glue. All the wetted parts, especially the blade holder, should be rinsed in distilled water and dried.

Do not use any solvents such as acetone or chloroform.

After a long period of use, the drive belts may deteriorate and break. To check or change the belts, disconnect the instrument from the electrical supply and remove the screws securing the top cover. Remove the top cover and inspect the condition of the exposed belts — change as required. Note that the NVSL manual version has two belts to drive the blade head and the NVSLM1 motorized version has two additional belts to drive the bath slide. Replace the top cover and securing screws.

Specifications

Section Thickness	Minimum 20 µm (fixed tissue), 50 µm (fresh tissue) Maximum 700 µm	
Step Size	10 µm	
Chamber Dimensions	55 mm x 82 mm x 31 mm	
Chamber Volume	140 ml	
Bath Advance Speed (NVSLM1)	0.14 to 2 mm/sec continuously variable	
Bath Reverse Speed	2 mm/sec	
Vibration Speed	60-4500 rpm	
Vibration Displacement	1 mm	
Power Requirements (factory set)	220V/240V, 50 Hz, 0.5 A 110V/130V, 60 Hz, 0.5 A	
Permissible Voltage Tolerance	+10%, -6%	
Current Consumption	NVSL0.07 A (average)	
NVSLM1	0.09 A (average)	
Power Ratings	NVSL	15 W
	NVSLM1	20 W
Fuse Ratings	NVSL	220V/240V ac, 250 mA, type T 20 mm
		110V/130V ac, 500 mA, type T 20 mm
	NVSLM1	220V/240V ac, 250 mA, type T 20 mm
		110V/130V ac, 500 mA, type T 20 mm

Each Vibroslice is supplied with a tissue holder and bath, blade angle protractor, 10 individual stainless steel blades, foot switch, instruction manual and mains lead.

References

1. Elliot, K. (1969) pp 103-114 in *Handbook of Neurochemistry* vol I 2 (Ed. Lajtha).
2. Halliwell, J.V. (1975) *J.Physiol.* 246, 91-'93P.
3. Harvey, J.A., Schofield, C.N. & Brown, D.A. (1974) *Brain Res.* 76, 235-245.
4. Konnerth, A. (1990) *Trends Neurosci.* 13:321-323 (1990).
5. Keenan, C.L., Chapman, P.F., Chang, V.C. & Brown, T.H. (1988) *Brain Res. Bull.* 21,373-383.
6. Warburg, O. (1930) pp 75-93 in *The Metabolism of Tumours*. New York.
7. Yamamoto, C.A. & Chujo, T. (1978) *Exp. Brain Res*. 31, 299-301.

Warranty

WPI (World Precision Instruments, Inc.) warrants to the original purchaser that this equipment, including its components and parts, shall be free from defects in material and workmanship for a period of one year* from the date of receipt. WPI's obligation under this warranty shall be limited to repair or replacement, at WPI's option, of the equipment or defective components or parts upon receipt thereof f.o.b. WPI, Sarasota, Florida U.S.A. Return of a repaired instrument shall be f.o.b. Sarasota.

The above warranty is contingent upon normal usage and does not cover products which have been modified without WPI's approval or which have been subjected to unusual physical or electrical stress or on which the original identification marks have been removed or altered. The above warranty will not apply if adjustment, repair or parts replacement is required because of accident, neglect, misuse, failure of electric power, air conditioning, humidity control, or causes other than normal and ordinary usage.

To the extent that any of its equipment is furnished by a manufacturer other than WPI, the foregoing warranty shall be applicable only to the extent of the warranty furnished by such other manufacturer. This warranty will not apply to appearance terms, such as knobs, handles, dials or the like.

The foregoing obligations set forth in this paragraph are in lieu of all obligations and liabilities, including all warranties of merchantability or otherwise, expressed or implied or statutory, and state WPI's entire and exclusive liability and purchaser's exclusive remedy for any claim of damages in connection with the sale or furnishing of all equipment, including design, suitability for use, operation, or installation. There are no warranties which extend beyond the description of the face hereof. In no event shall WPI be liable for any special or consequential damages.

Warning: This equipment is not designed or intended for use on humans.

** Electrodes, batteries and other consumable parts are warranted for 30 days only from the date on which the customer receives these items.*

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DECLARATION OF CONFORMITY

We: World Precision Instruments, Inc.
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USA

as the distributor of the apparatus listed, declare under sole responsibility that the product(s):

Title: NVSL Manual Vibroslice
NVSLM1 Motorized Vibroslice

to which this declaration relates is/are in conformity with the following standards or other normative documents:

EC Directive(s):
- The Machinery Directive 89/392/EEC as amended by:
  - Directive 91/368/EEC
  - Directive 93/44/EEC
- Electromagnetic Compatibility Directive 89/336/EEC
- The Low Voltage Directive 73/23/EEC

UK Regulations:
- The Supply of Machinery (Safety) Regulations 1992 (SI 1992/3073)
- Electricity at Work Regulations 1989

European Standards:
- EN 50081-1:1992 Electromagnetic compatibility generic emissions standard part 1
- EN 50082-1:1992 Electromagnetic compatibility generic emissions standard part 2

Additionally, the health and safety requirements of the following British and harmonized European Standards have been incorporated in the design of the above machines:
- BS 2771: part 1: 1986 (EN 60 204:part1:1985)
- BS5304:1988

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