

MICROTOME

- A microtome (from the Greek mikros, meaning “small”, and temnein, meaning “to cut”) is a tool used to cut extremely thin slices of material, known as sections.

The most common applications of microtomes are:

- **Traditional Histology Technique:**
- **Cryosectioning Technique:**
- **Electron Microscopy Technique:**
- **Botanical Microtomy Technique:**

Botanical Microtomy Technique:

- Hard materials like wood require a sledge microtome.
- These microtomes have heavier blades and cannot cut as thin as a regular microtome.

Various types of microtomes are available.

- **Rotary Microtome**
- **Sledge Microtome**
- **Cryomicrotome**
- **Ultramicrotome**
- **Vibrating microtome**
- **Saw microtome**
- **Laser microtome**

Rotary Microtome

- It is most commonly used microtome. This device operates with a staged rotary action such that the actual cutting is part of the rotary motion. In a rotary microtome, the knife is typically fixed in a horizontal position.

- The typical cut thickness for a rotary microtome is between 1 and 60 μm .
- For hard materials, such as a sample embedded in a synthetic resin, this design of microtome can allow for good “Semi-thin” sections with a thickness of as low as 0.5 μm .

Advantages of Rotary Microtome

- The machine is heavy, so it is stable and does not vibrate during cutting.
- Serial sections can be obtained.
- Cutting angle and knife angle can be adjusted.
- It may also be used for cutting celloidin embedded sections with the help of special holder to set the knife.

- <https://youtu.be/KnMdSgd5mts>

Sledge Microtome

- Sample is placed into a fixed holder (shuttle), the sledge placed upon a linear bearing, a design that allows for the microtome to readily cut many coarse sections.
- Applications for this design of microtome are of the preparation of large samples, such as those embedded in paraffin for biological preparations.
- Typical cut thickness achievable on a sledge microtome is between 10 and 60 micron.
- <https://youtu.be/5sq9szNtsal>

Cryomicrotome

- For the cutting of frozen samples, many rotary microtomes can be adapted to cut in a liquid nitrogen chamber, in a so-called cryomicrotome setup.
- The reduced temperature allows for the hardness of the sample to be increased, such as by undergoing a glass transition, which allows for the preparation of semi- thin samples.
- However the sample temperature and the knife temperature must be controlled in order to optimise the resultant sample thickness
- <https://youtu.be/5afOen2AVwg>

Ultramicrotome

- A ribbon of ultrathin sections prepared by room temperature ultramicrotomy, floating on water in the boat of a diamond knife used to cut the sections.
- The knife blade is the edge at the upper end of the trough of water.
- It can allow for the preparation of extremely thin sections
- These extremely thin cuts are important for use with transmission electron microscope (TEM) and Serial Block-Face Scanning Electron Microscopy (SBFSEM), and are sometimes also important for light-optical microscopy.

Serial Block-Face Scanning Electron Microscopy

Vibrating microtome

- The vibrating microtome operates by cutting using a vibrating blade, allowing the resultant cut to be made with less pressure than would be required for a stationary blade.
- The vibrating microtome is usually used for difficult biological samples.
- The cut thickness is usually around 30-500 μm for live tissue and 10-500 μm for fixed tissue.

Saw microtome

- The saw microtome is especially for hard materials such as teeth or bones.
- The microtome of this type has a recessed rotating saw, which slices through the sample.
- The minimal cut thickness is approximately 30 μm , and can be made for comparatively large samples.

Laser microtome

- The laser microtome is an instrument for contact free slicing. Prior preparation of the sample through embedding, freezing or chemical fixation is not required, thereby minimizing the artifacts from preparation methods.
- Alternately this design of microtome can also be used for very hard materials, such as bones or teeth as well as some ceramics. Dependent upon the properties of the sample material, the thickness achievable is between 10 and 100 μm .

Laser microtome

- The device operates using a cutting action of an infra-red laser.
- As the laser emits a radiation in the near infra-red, in this wavelength regime the laser can interact with biological materials.
- Through the non-linear interaction of the optical penetration in the focal region a material separation in a process known as photo-disruption is introduced.

Laser microtome

- By limiting the laser pulse durations to the femtoseconds range, the energy expended at the target region is precisely controlled, thereby limiting the interaction zone of the cut to under a micrometre.
- External to this zone the ultra- short beam application time introduces minimal to no thermal damage to the remainder of the sample.

MICROTOME KNIFE

- It is the important instrument used to cut uniform thin serial sections of the tissue. Various types of knives are used with different microtomes. For routine purpose wedge (C type) knife is used. It is plain on both sides. The size varies from 100 mm to 350 mm in length.
- Microtome knives are made of good quality of high carbon or steel which is tempered at the tip. Hardness of knife is essential to obtain good tissue sections.

Sharpening of microtome knife

- To achieve good sections knife should be very sharp.
- The knife is put in the knife back to sharpen.
- Knife can be sharpened manually or by the use of automatic machine.
- <https://youtu.be/12BVnDTy4xg>

Honing

- This is done to remove nicks and irregularity from the knife edge. Coarse and fine honing is done using different abrasives.

Stropping

- The purpose of stropping is to remove the “burr” formed during honing and to polish cutting edge.

Other types of knives

- Diamond and glass knives. These knives are very expensive and used for ultramicrotomy.

Disposable knife

- Nowadays these microtome blades are used. Two types of disposable blades are available.
- **1. Low profile blade** - Usually used for cutting small and soft biopsies like kidney and liver biopsies.
- **2. High profile blade**-Used for any tissue like myometrium, breast tumor or skin.