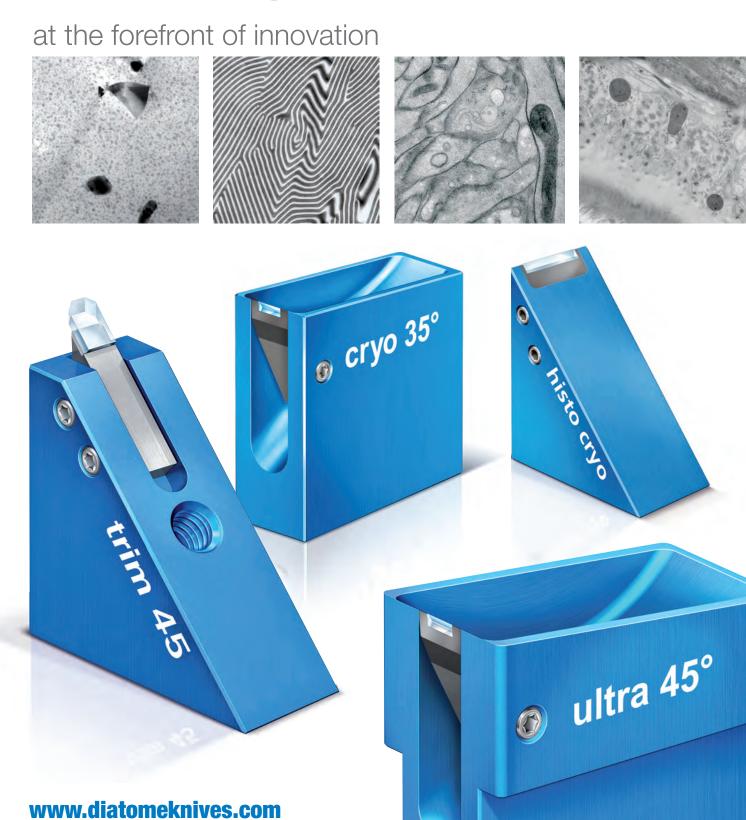
# DIATOME



# Introduction

Dear customer,

# **DIATOME** Diamond Knives – Development, manufacturing and customer service since 1970

## **Our developments in this period:**

our development	is in this period.
ultra 45°	the diamond knife with a hydrophilic cutting edge, allowing high quality ultrathin sections of epoxi resin embedded samples
ultra 35°	the low angle diamond knife for reduced compression and better structure preservation
ultra Maxi	similar to the ultra 35° but with a larger boat
ultra semi	for alternating sectioning, ultrathin/semithin
ultra ats	for placing sections on Si wafers
histo	the first diamond knife for cutting semithin sections for the observation in the optical microscope
cryo	the diamond knife for cutting in the cryo-ultramicrotome
Static Line Ionizer	for eliminating electrostatic charging
cryo immuno	the knife with a large diamond platform fascilitating pick-up of cryo sections from sucrose infiltrated samples (Tokuyasu)
cryo 25°	the low angle diamond knife for sectioning frozen hydrated samples (CEMOVIS)
ultra AFM and cryo AFM	the knives for generating absolutely smooth and flat surfaces of biological and technical samples for the observation in the AFM
ultra sonic	the oscillating diamond knife for eliminating compression and allowing best structure preservation
ultra sonic Maxi	a new wider ultra sonic knife specifically for serial sectioning in biological applications
trim 20, trim 45, trim 90	for quick, accurate trimming at room and cryo-temperatures



These developments were possible by the co-operation with you, our valued customers.

We are convinced that also in the future new developments are possible and that our knives may be adapted to the changing requirements.

Make use of our many years experience in perfecting our knives.

With a telephone call or an email we can inform you on any details you require.

We are looking forward to hear from you soon!

Your DiATOME team

Ultrastructure of the roundworm
Caenorhabditis elegans.
Thomas Müller-Reichert, EM Technology
Development, MPI Dresden, and Kent McDonald,
Electron Microscopy Laboratory, University of
California, Berkley.
Scale bar: 28 mm = 1 µm

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# Characteristics of **DiATOME** Diamond Knives

# DIATOME knives are compatible with all ultramicrotomes.

The boat is designed in such a manner that the water surface is horizontal when the clearance angle is set. This results in a stationary water surface and good reflection.

The shape of the boat offers you the advantage of easy pick-up of the floating sections.

The surfaces of the *DiATOME* diamond knives are hydrophilic. This property allows an easy wetting of the cutting edge, even

with a very low water level (important for sectioning Lowicryls and other hydrophilic embedding media, as well as for water sensitive samples.)

The diamond is fastened in a metal shank which is securely screwed into the boat.

The cementing material seals boat and knife. It is hydrophilic and aids the easy wetting of the cutting edge.

# **DIATOME** Resharpening and Exchange Service

Resharpened *DiATOME* knives undergo the same stringent optical checking and sectioning test as new knives.

A diamond knife resharpened by DiATOME is the same high quality as a new one!

During resharpening a knife may be reworked into another type (ultra to cryo, 45° to 35°, etc).

We let you choose: You may have your knife resharpened, or exchanged against a new one. In the exchange service a type change is possible (ultra to cryo, 45° to 35°, small knives to large ones, etc). This, of course includes changing the boat to your desired style.

When we resharpen our diamond knives we restore it to its original condition.

Your resharpened *DiATOME* knife will have the same length, the identical cutting edge and carry the same exact guarantee of quality as the day it first left our factory.

We are the only manufacturer that guarantees that after a resharpening, the knife will be the same flawless quality as the knife you originally purchased. Other manufacturers state that during their resharpening process the diamond may become smaller (up to 5% of its original length). With our advanced techniques in resharpening you will never, and we mean never, lose any diamond length.

Other companies "CAN NOT SUCCESSFULLY" resharpen a DiATOME Knife. We have found that when other companies try to resharpen our knives, the original parameters of our knives are either altered significantly or totally lost; returning to you an inferior diamond knife. Many times we have also found that other manufacturers ruin our diamonds during their resharpening process and end up replacing our diamond with one of inferior quality.

Any *DiATOME* knife can be resharpened an "UNLIMITED" number of times due to the following reasons:

- The cutting edge of the diamond knife is on the narrow side of the rectangular cut diamond, leaving the large part of the stone as a resharpening reserve.
- Special attention is given to minimizing the amount of grind off to the diamond during the resharpening process.

When you send a knife to us for resharpening we will automatically replace your boat at no charge if it is either damaged, discolored, or at your request.

Each *DiATOME* Diamond Knife, whether new or resharpened, is subjected to extensive testing for its ability to cut accurately without scoring or compression. Only if its performance passes our tests will we ship it to you.

# Sample Testing and Service of **DiATOME** Diamond Knives

The great experience obtained during long years of development, manufacturing and application of diamond knives allows us to offer a unique service:

You send us biological or industrial samples of any nature and let us know what you would like to obtain. The more information you give us the better results you will receive. We perform a sectioning test and

send you the resulting ultrathin section ( ie. the sample face for SEM, AFM, STM investigation), a report of how the results were obtained and recommendations for the most suitable knife. This service is free of charge as long as the number of samples is reasonable. You may take it for granted that we treat your "secret samples" with absolute discretion.

Please allow us to help you choose the appropriate knife from our large range for your specific application.

# Custom **DiATOME** Diamond Knives and Boats

Upon request, we can supply you with different color (Black) and shaped boats (Huxley for instance) at no extra cost to you.

Certain applications require special knives and or tools outside of the standard knives and tools that are available. For years *DiATOME* has been manufacturing custom diamond blades, tools, and knives for customers meeting all of their specific needs.

If you already have in mind the type of diamond knife or tool that you require, please submit a diagram of it to us as well as your sample blocks or specimens. We, in turn, will manufacture a prototype knife for you and test it in accordance to your requirements with your blocks. Only if the knife or tool passes all of our extensive testing will we send it to you for your evaluation and approval.

If you are not quite sure how to attack your sectioning problems but you believe that you may be a candidate for a custom diamond knife or tool please send us your sample blocks and a complete description of the work that you are doing and what you would like to achieve with your diamond knife. We will take it from there. We will manufacture a prototype knife, evaluate and test it, and then send it to you for your evaluation and approval.

There is absolutely no charge for this service outside of the regular cost of purchasing the blade once you have approved it.

# Quality Guarantee for **DiATOME** Diamond Knives

Before delivery, each knife, new or resharpened, is subject to extensive testing. We can therefore guarantee perfect sectioning (in the respective thickness range) over the entire cutting edge.

Our goal has always been to assure our customers satisfaction. Therefore, in the

unlikely event that you experience difficulties, or for any reason are unhappy with the performance of your new or resharpened knife, contact us immediately. You can be sure that any problem with your knife will be corrected.

We stand by our commitment to quality and customer satisfaction.

# **DIATOME** ultra knives and their applications

- Highest quality diamonds and optimal crystal orientation guarantee perfect ultrathin sections and a durable cutting edge
- Section pick-up is facilitated as the boat is horizontal allowing the water to completely fill the boat all the way round
- · A hydrophilic surface makes it easy to wet the cutting edge, even with low water level

Knife type	Knife angle	Size [mm]	Thickness range [nm]	Boat type	Code (new knife)*	Application
ultra 35°	35°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–200	Standard boat	15-UL 20-UL 25-UL 30-UL 35-UL 40-UL	<ul> <li>Soft industrial samples such as metals and polymers</li> <li>Hard and brittle samples such as semiconductors, superconducting oxides, nanocrystalline ceramics</li> </ul>
ultra semi	35°	3.0 mm	50–500	Standard boat	30-S	Alternating sectioning ultrathin/semithin
ultra AFM	35°	2.0mm 3.0mm	15–100	Small boat	20-AFM-UDL 30-AFM-UDL	Sample surfacing for AFM
ultra 35° Jumbo	35°	2.0mm 3.0mm	50–200	Jumbo boat	20-ULJ 30-ULJ	Section series for 3D reconstruction, STEM
ultra Maxi	35°	4.0mm	30–200	Large boat	40-ULM	• Similar to ultra 35, features a larger boat.
ultra sonic (with control unit) ultra sonic (knife only)	35°	3.0mm 4.0mm 3.0mm 4.0mm	15–100	Special boat	30-ULTSON 40-ULTSON 30-ULTSONK 40-ULTSONK	<ul><li>Compression-free sections</li><li>Best structure preservation</li><li>Biological and materials science specimens</li></ul>
ultra sonic Maxi (with control unit) ultra sonic Maxi (knife only)	35°	3.0mm 4.0mm 3.0mm 4.0mm	30–150	Special boat	30-ULTSONM 40-ULTSONM 30-ULTSONMK 40-ULTSONMK	<ul> <li>Compression-free sections</li> <li>Serial sectioning in biological applications</li> </ul>
ultra ats	35°	3.0mm	30–200	Special boat	30-UL-ATS	• Placing sections on Si wafers
ultra 45°	45°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–200	Standard boat	15-US 20-US 25-US 30-US 35-US 40-US	Routine sectioning of biological and materials science specimens
ultra 45° Jumbo	45°	2.0mm 3.0mm	50–200	Jumbo boat	20-USJ 30-USJ	Section series for 3D reconstruction, STEM
trim 20 trim 45 trim 90	20° 45° 90°			Triangular holder	TT-20 TT-45 TT-90	Trimming biological and materials science samples

<sup>\*</sup> Product Codes shown are for new knives.

To order a resharpened knife, add "R" to the Product Code that corresponds to your knife.

**Example:** Product Code 15-ULR = Resharpened ultra 35° 1.5mm Knife.

Ultra 35°, 45°, and ultra semi can be exchanged. To order an exchanged knife, add "E" to the Product Code.

**Example:** Product Code 15-ULE = Exchange of an ultra 35° 1.5mm Knife.

# ultra 35°

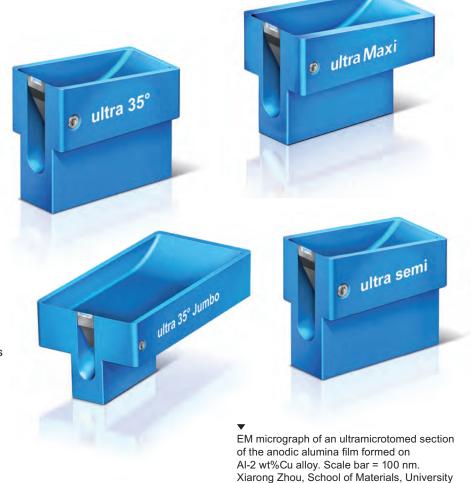
In 1989 considerably reduced compression, smoother section surfaces and improved structural preservation thanks to the use of our ultra 35° knives was demonstrated (J. C. Jésior, Scanning Microscopy Supplement 3, pp. 17 – 153, 1998).

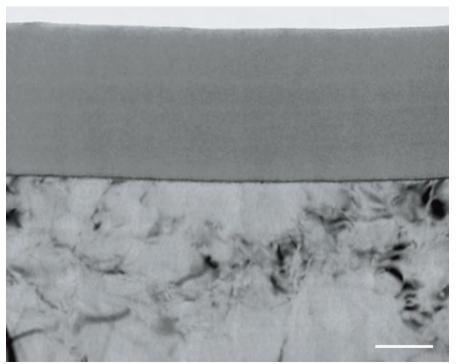
In the meantime, a large number of researchers have recognized the advantages of 35° knives, in particular for sectioning biological specimens of all kind, non-homogenous specimens, non decalcified bone, dental material, etc.

The ultra 35° knives are perfect for sectioning relatively soft materials research specimens including metals and polymers, as well as mixed specimens such as polymers filled with nanoparticles, brittle materials such as catalysts, crystals, semiconductors, etc (G. Mahon et al., Microscopy Research and Technique, Vol. 31, pp. 267 – 274, 1995, S. R. Glanvill, Microscopy Research and Technique, Vol. 31, pp. 275 – 284, 1995, P. Swab et al., Mat. Res. Soc. Symp. Proc. Vol. 115, pp. 229 – 234, 1989, P. Schubert-Bischoff et al., Microscopy and Microanalysis, proceedings, page 359, 1997).

The ultra 35° knife has demonstrated it's usfulness as a standard knife for the majority of applications in both biological and materials research.







of Manchester.

# ultra sonic

- · Thinner sections
- · No compression
- · Best structure preservation

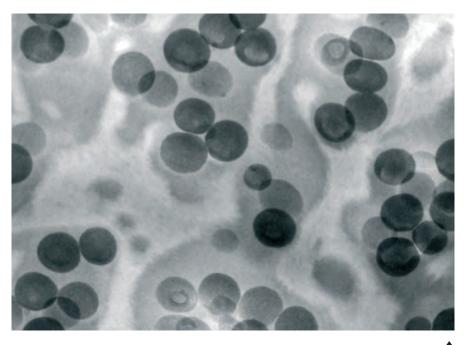
The patented ultra sonic knife allows the cutting of ultrathin sections free of compression (D. Studer et al., Journal of Microscopy, Vol. 197, pp. 94 – 100, 2000).

With correct setting of frequency and amplitude the sections become as long as the height of the sample.

Best results are not only achieved with biological samples, but also with polymers (J. S. Vastenhout et al., Microscopy and Microanalysis 8, 2001. J. S. Vastenhout et al., Microscopy Today, pp. 20–21, 2006).

We have tested the ultra sonic knife with the following samples:

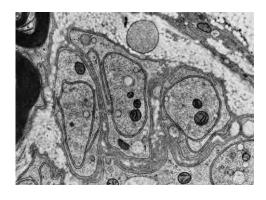
- Biological samples in Epon, Araldite, EM Bed, etc.
- Biological samples in acrylic resins (Lowicryl, LR White).
- Rigid polymers such as PS, PMMA, ABS, HIPS, modified PP, blends of various kind.



Polycarbonate modified with rubber Jens Sicking, Bayer Technology Services, Leverkusen.

—— 200 nm





Peripheral nerve (rat), HP frozen, freeze substituted, Epon embedded, cut with the ultra sonic knife, section thickness 50nm. W. Graber, Institut of Anatomy, University of Bern, Switzerland

# ultra sonic

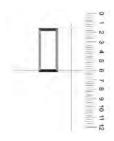
# Handling

### Sample preparation

• Trim the sample with a trim 45 or a trim 20 diamond blade. The sample width should be a maximum of 0.5 mm.

# Measuring the sample height

With the use of an eyepiece graticule in one of the stereomicroscope oculars (graticule 10450336 for the Leica M80). Sample block (fixed in the sample holder), is mounted in the trimming plate. Measure the height with the graticule.



## **Ultramicrotome Settings**

- Set the clearance angle to 6° as shown on the guarantee card.
- · Set the desired section thickness.
- Set the sectioning speed (0.4 0.6 mm/sec)
- · Tighten all set screws.

### Installing the knife

- Mount the knife in the knifestage of the ultramicrotome and tighten the set screw.
- · Connect the control unit to the power supply.
- Connect the control unit OUTPUT and the knife with the blue cable.
- Switch on the control unit (switch ON the back side).
- Set the resonance frequency (Toggle switch on AUTO). After a few seconds the display will show «Peak locked».
- · Adjust the amplitude to approx. 2 V.
- Approach the sample with the knife (settings as shown in our diamond knife handling manual.
- · Start sectioning as usual.

### Measuring the section length

- Measure the section length with the graticule and compare it with the sample height.
- If sections are too short, increase the the amplitude (turn the button clockwise).
- If sections are too long, decrease the amplitude (turn the button counterclock wise).
- Too high an amplitude may lead to drifting of the sections.

### **Drifting of the sections**

When working in resonance, the sections may drift slightly to the right or to the left. If this is the case, the following procedure helps: Switching from AUTO to MAN. Now increase or decrease the the frequency a few hundred Hertz, until the sections float straight on the water surface.

If sections drift to the right: decrease the frequency. If sections drift to the left: increase the frequency.

# **Control Unit**

Frontside:



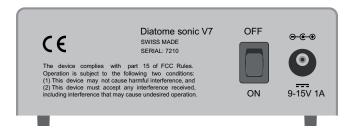
Output: Connection with the knife (blue cable)

Amplitude: 0 - 25.5 V

Frequency: Toggle switch MAN: Manual setting of

frequency, range 15 – 35kHz Toggle switch AUTO: Automatic setting of the resonance frequency.

### Backside:



# ON/OFF Switch

Connection for the power supply adapter.

# **Specifications**

Knife: ultra sonic, ultra sonic Maxi

Knife angle: 35°

Cutting range: 20 – 100 nm Cutting edge length: 3.0mm, 4.0mm

Order number: DUS3530 (for ultra sonic)

DUSM3530 (for ultra sonic Maxi)

Control unit

Frequency range: 15 – 35kHz, or automatic setting of

the resonance frequency

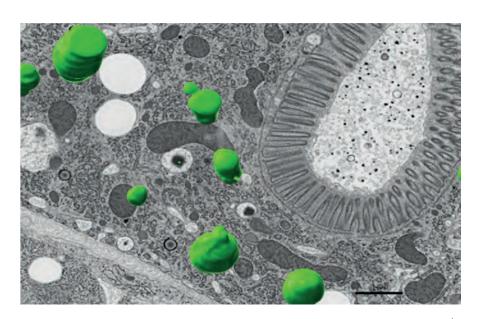
Amplitude: variable (voltage 0 - 25.5V)

Mains voltage: 230 V, 110 V

# ultra sonic Maxi

The ultra sonic Maxi is a new wider ultra sonic knife specifically for serial sectioning in biological applications. The knife comes in 3.0mm and 4.0mm sizes with 35° angle.

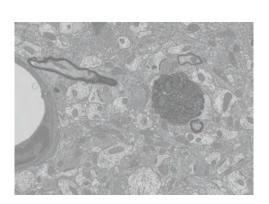
- Ultrathin sections without compression
- All embedding resins (Epoxies and Acrylates)
- The same control unit as for the ultra sonic usable
- Automatic setting of the resonance frequency
- Knife angle 35°
- Boat (inside 17 x 25 mm) made from titanium
- Easy wetting of the cutting edge



Section obtained with the ultra sonic Maxi knife. Scale bar 1 µm.

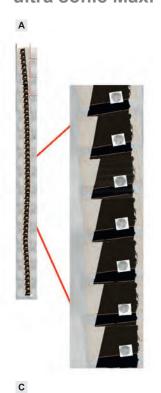
More details on the next page.

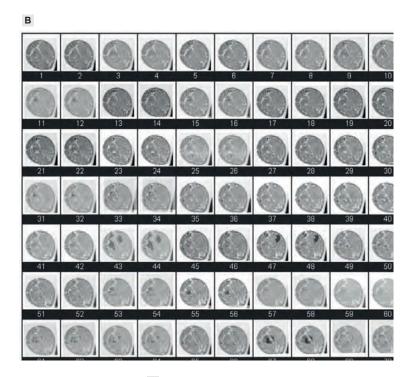


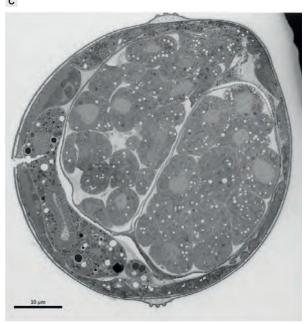


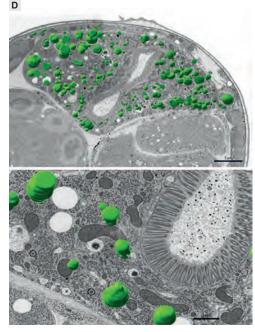
Biocytin-labeled giant bouton from the auditory cortex of a Mongolian gerbil acquired with an SEM.
Saldeitis et al., 2019 Eur. J. Neurosci.
Vol.50-9:3445-3453.

# ultra sonic Maxi









Serial sections obtained with the ultra sonic Maxi knife at 55 nm from a high pressure frozen nematode sample. **A)** Long ribbons collected on silicon wafers imaged using a SEM **B)** Gallery of serial sections

- C) Single section from the series. Scale bar 10  $\mu m$
- D) Yolk granules and lysosomes were segmented shown in green. Scale bar 5  $\mu$ m; detail image: 1  $\mu$ m Center for Microscopy and Image Analysis, University of Zürich, Switzerland

# ultra ats

The ultra ats Diamond Knife is perfect for placing sections on Si wafers to view under the SEM. The knife comes in 3.0mm size with 35° angle. Please see our Technical Data Sheet for an illustration showing the technique of placing sections on a wafer.

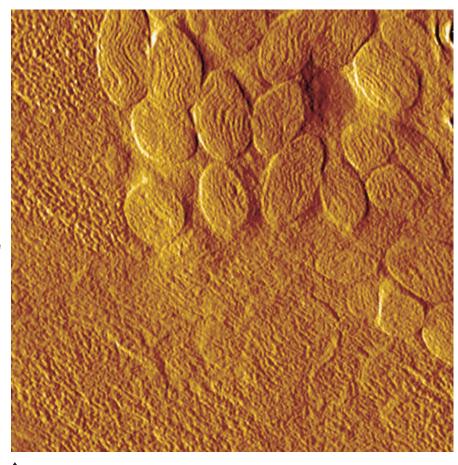


# ultra AFM

In order to achieve the best results for AFM investigation, only the highest quality diamond knives should be used (P. H. Vallotton et al., J. Biomater. Sci. Polymer Edn., Vol. 6, No. 7, pp. 609 – 620, 1994. N. Matzko et al., Journal of Structural Biology 146, pp. 334 – 343, 2004).

Our ultra AFM knives are made of highest quality to ensure the increased quality requirements of AFM investigation. They preduce extremely smooth sample surfaces and guarantee the best possible structure preservation.





AFM amplitude image of the muscle of cat's mite Otodectes cynotis. The contrast covers amplitude variation in the 1 – 3 nm range. Size of the whole image equals 4.6 microns. Nadejda Borisovna Matsko, Institut für angewandte Physik, ETH Zürich.

# ultra 45°

Acknowledged as the appropriate knife angle for routine sectioning of both biological and materials research specimens, it represents a balanced compromise between section quality and durability.

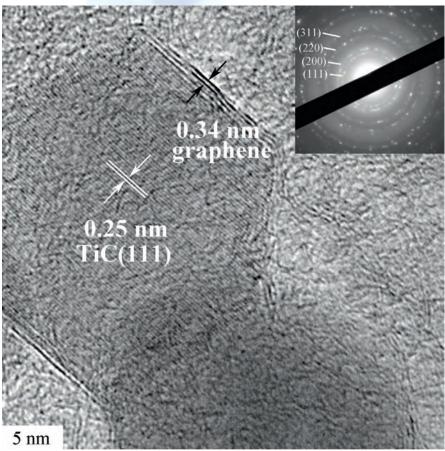
For the sectioning of a number of hard materials such as ceramics, semiconductors, oxides etc, with the use of the ultra 45° knife a longer service time may be expected. Kindly contact us and make use of our long years experience in all ultramicrotomy applications.

# ultra 45° Jumbo

The Ultra Diamond knife is now available in Jumbo with 35° and 45° angles and in 2.0mm and 3.0mm sizes.

These knives are well suited for serial sectioning for 3D reconstructions and STEM.





HRTEM image of a TiC microsphere showing nanocrystalline carbon coating on the TiC grain (confirmed by the electron diffration pattern). J. Zhong et al, Journal of the European Ceramic Society 32, pp. 3407 – 3414, 2012.

# trim 20 trim 45 trim 90

Finally, one trimming tool for all of your trimming needs, be it at room or cryo temperatures. The trimtool series supercedes the cryotrim and ultratrim lines of trimming tools. Resharpening is still available for these lines.

For successful ultramicrotomy in biology and materials science, precise trimming is mandatory.

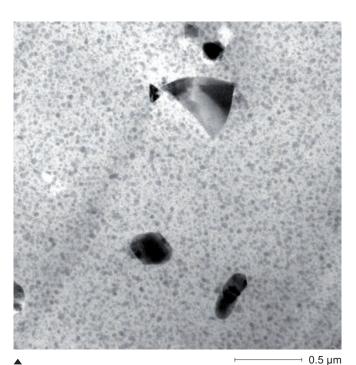
The DiATOME trimming blades trim 90, trim 45, and trim 20 will fulfill all your trimming requirements, allowing quick, easy and accurate trimming at both room and cryo-temperatures.

A well trimmed sample is a precondition for perfect section ribbons. Due to the extreme sharpness of our diamond blades, less mechanical damage is applied to the sample during trimming.

Very shiny sample faces and precise sides are the result.

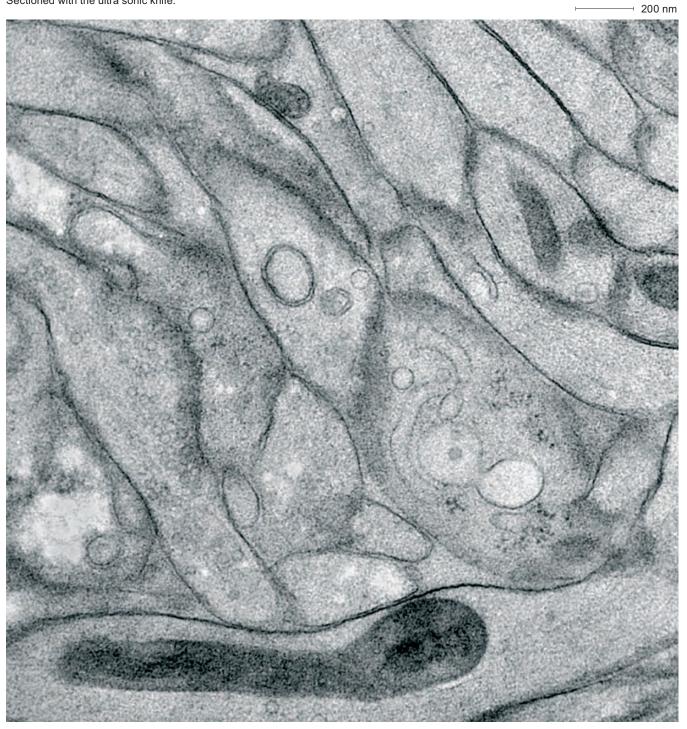
The trim 45 produces pyramidal sides with an inclined angle of 45° and the trim 20 produces pyramidal sides with an inclined angle of 20°. The trim 90 aids in the FIB cutting of biological and technical sample blocks, trimming the 90° inclined blocks sides and well as the surface of the block.





SiC, SiO2, TiO2 and AlO3 nanoparticles in polymer matrix.
Claudia Mayrhofer, TU Graz.

Neuropil of a HP frozen / freeze substituted hippocampus slice culture, Daniel Studer and Werner Graber, Inst. of Anatomy, University of Bern. Sectioned with the ultra sonic knife.



# **DIATOME** cryo knives and their applications

- Thinner cryo sections
- Perfect cryosections from ultrathin to semi with the same knife
- Minimal compression and best structure preservation
- Highest quality diamonds and optimal crystal orientation guarantee perfect ultrathin sections and a durable cutting edge

Knife type	Knife angle	Size [mm]	Thickness range [nm]	Boat type	Code (new knife)*	Application
cryo 25°	25°	3.0mm	30–150	Triangular holder	30-CD25	<ul> <li>Frozen hydrated samples (CEMOVIS)</li> </ul>
cryo immuno	35°	2.0mm 3.0mm	30–300	Triangular holder	DCIMM3520 DCIMM3530	<ul> <li>Sucrose infiltrated samples (Tokuyasu)</li> <li>Frozen hydrated samples (CEMOVIS)</li> </ul>
<b>cryo 35°</b> (dry)	35°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–300	Triangular holder	15-CDL 20-CDL 25-CDL 30-CDL 35-CDL 40-CDL	Polymers, rubber, paints etc.
cryo 35° (wet)	35°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–300	Triangular holder	15-CWL 20-CWL 25-CWL 30-CWL 35-CWL 40-CWL	Wet sectioning of polymers with DMSO/water mixture
<b>cryo 45°</b> (dry)	45°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–300	Triangular holder	15-CDS 20-CDS 25-CDS 30-CDS 35-CDS 40-CDS	Routine dry cryo sectioning of polymers
cryo 45° (wet)	45°	1.5mm 2.0mm 2.5mm 3.0mm 3.5mm 4.0mm	30–300	Small cryo boat	15-CWS 20-CWS 25-CWS 30-CWS 35-CWS 40-CWS	<ul> <li>Routine wet cryo sectioning of polymers with DMSO/ water mixture</li> </ul>
cryo AFM	35°	2.0mm 3.0mm	20–100	Triangular holder	20-AFM-CDL 30-AFM-CDL	<ul> <li>Sample planing for AFM imaging</li> </ul>
trim 20 trim 45 trim 90	20° 45° 90°			Triangular holder	TT-20 TT-45 TT-90	Trimming biological and materials science samples

<sup>\*</sup> Product Codes shown are for new knives.

To order a resharpened knife, add "R" to the Product Code that corresponds to your knife.

**Example:** Product Code 30-CD25R = Resharpened cryo 25° 1.5mm Knife.

Cryo 35° and 45° can be exchanged. To order an exchanged knife, add "E" to the Product Code.

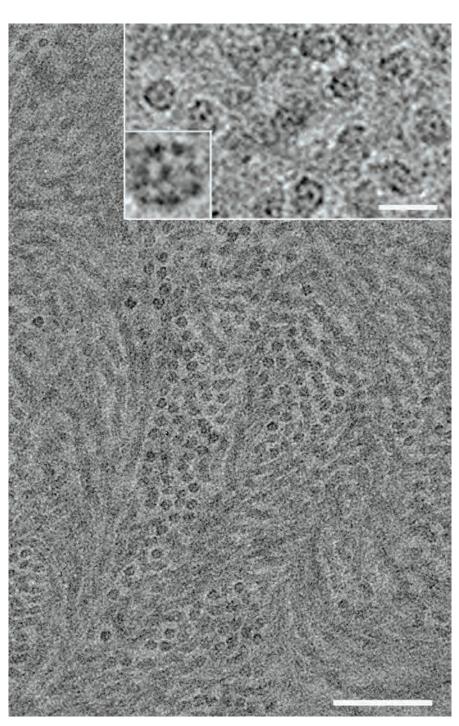
**Example:** Product Code 15-CWLE = Exchange of an cryo 35° (wet) 1.5mm Knife.

# cryo 25°

The cryo 25° knife is designed for sectioning frozen hydrated specimens. The 25° angle results in the least possible compression and the best structure preservation (H.M. Han et al., Journal of Microscopy, Vol. 230, Pt. 2, pp. 167–171, 2007).

Please note: best results are achieved at low humidity, when the cryo-ultramicrotome is placed in a glovebox and the sections attached by electrostatic force (J. Pierson et al., Journal of Structural Biology 169, pp. 219 – 225, 2010).





▲ High resolution electron micrograph of vitreous section of keratin intermediate filaments in the midportion of stratum corneum of human epidermis. The fine structure of the keratin filaments is well resolved and their molecular organisation is seen in favourable cases (inset). Scale bar = 100 nm. Scale bar inset = 20 nm. Ashraf Al-Amoudi, Laboratoire d'Analyse Ultrastructurale, Lausanne.

# cryo immuno

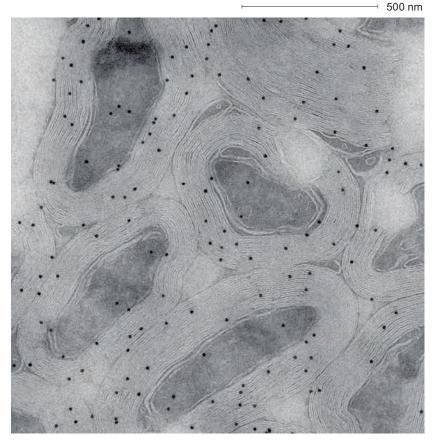
The first cryo knife with a diamond platform, guarantees the best possible sectioning for sucrose infiltrated samples (Tokuyasu).

The diamond platform guarantees an easy and gentle section pick-up.

The sections are collected directly from the diamond surface using a loop and a sucrose/methyl-cellulose droplet (W. Liou et al., Histochemistry and Cell Biology, Vol. 106, pp. 41-55, 1996. P. J. Peters et al., Current Protocols in Cell Biology, pp. 4.7.1-4.7.18, 2006).

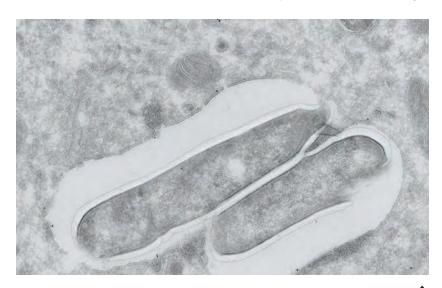
The  $35^{\circ}$  angle leads to a considerable reduction in mechanical stresses and therefore to improved structure preservation in sucrose infiltrated samples (E. Bos et al., Journal of Structural Biology 175, pp. 62-72, 2011).

We recommend the cryo immuno knife also for sectioning frozen hydrated samples (CEMOVIS). The 35° angle is a good compromise between durability and cutting performance (A. Al-Amoudi et al., Journal of Structural Biology 150, pp. 109 – 121, 2005).



Mouse optic nerve, immunolabeling of the major myelin protein proteolipid protein (PLP), 10 nm gold. Wiebke Möbius, Dept. of Neurogenetics, EM Core Facility, MPI of Experimental Medicine, Göttingen.





Fusion of late endosome/lyosome with the Mycobacteria tuberculosis phagosome.

Aldehyde fixed cryo-section labeled with anti- CD63 and protein-A gold 10nm.

Scale bar = 100nm. Nicole van der Wel and Peter J. Peters,

Netherlands Cancer Institute, Amsterdam.

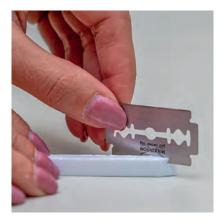
# CIYO V

# Cleaning procedure for our cryo Diamond Knives

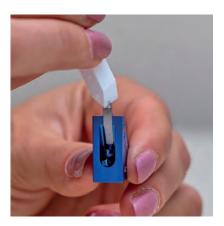
This procedure serves for the cleaning of our cryo immuno and cryo 25° knives. We are at your disposal for any further assistance you might require.



Remove the knife stage from the cryo chamber, leave the knives in the stage, wash under tap water for warming up.



Bevel a polystyrene stick with a perfectly clean razer blade.



Clean the knife with 15% ethanol, keep wet.



Rinse with 100% ethanol.



Dry with a dustblower or canned air.

# cryo 35° (wet/dry) cryo 45° (wet/dry)

The cryo 35° knife has demonstrated its usefulness as a standard knife for the low temperature sectioning of polymers, rubber, paints, etc.

The 35° angle represents a balanced compromise between section quality and durability.

The cryo 35° and cryo 45° knife mounted in the triangular holder is suitable for dry cryosectioning.

The cryo 35° and cryo 45° knife mounted in the trough are used for sectioning with fluids such as a DMSO/water mixture.

The cryo 45° knife is well suited for routine cryo sectioning.



▲ Styrene-butadiene block copolymer x 25'000 Ronald Walter, BASF Aktiengesellschaft, Polymer Physics, D-67056 Ludwigshafen.

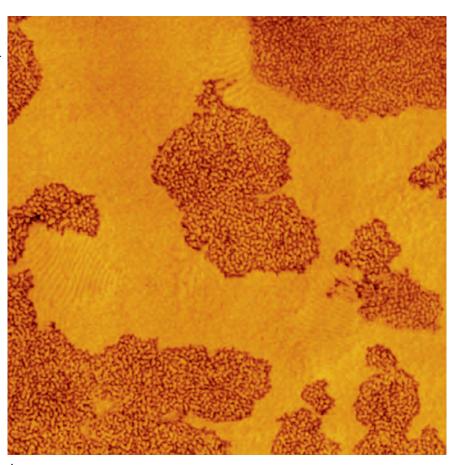


# olylo

# cryo AFM

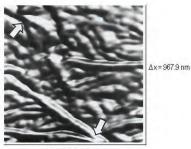
Our cryo AFM knives are made of highest quality to ensure the increased quality requirements of AFM investigation. They produce extremely smooth sample surfaces and guarantee the best possible structure preservation.





Morphology of a blend of two SBS block copolymers with different chain-architecture. AFM tapping mode, phase image, image size = 3 x 3 μm. Rameshwar Adhikari, Institut für Werkstoffwissenschaft, Martin-Luther-Universität, Halle-Wittenberg.

▶ Top-view, light shaded AFM image of a cryomicrotomed surface of ultrahigh molecular weight polyethylene. The arrows indicate zones with lamellae splitting. P.H. Vallotton, Materials Science Division, Lawrence Berkley Laboratory. References Ref 1: P.H. Vallotton, M.M. Denn, B.A. Wood and M.B. Salmeron: Comparison of medical-grade ultrahigh molecular weight polyethylene microstructure by atomic force microscopy and transmission electron microscopy. J. Biomater. Sci. Polymer Edn., Vol 6, No. 7, 609-620, 1994. Top of Page



 $\Delta x = 967.9 \text{ nm}$ 

# trim 20 trim 45 trim 90

Finally, one trimming tool for all of your trimming needs, be it at room or cryo temperatures. The trimtool series supercedes the cryotrim and ultratrim lines of trimming tools. Resharpening is still available for these lines.

For successful ultramicrotomy in biology and materials science, precise trimming is mandatory.

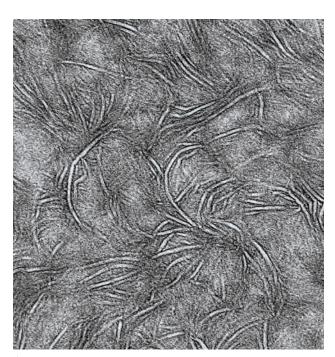
The DiATOME trimming blades trim 90, trim 45, and trim 20 will fulfill all your trimming requirements, allowing quick, easy and accurate trimming at both room and cryo-temperatures.

A well trimmed sample is a precondition for perfect section ribbons. Due to the extreme sharpness of our diamond blades, less mechanical damage is applied to the sample during trimming.

Very shiny sample faces and precise sides are the result.

The trim 45 produces pyramidal sides with an inclined angle of 45° and the trim 20 produces pyramidal sides with an inclined angle of 20°. The trim 90 aids in the FIB cutting of biological and technical sample blocks, trimming the 90° inclined blocks sides and well as the surface of the block.





Polyethylene x120'000
Ronald Walter, BASF Aktiengesellschaft, Polymer Physics, D-67056 Ludwigshafen.

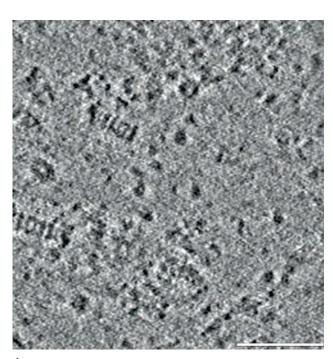
# olylo

# Diamond Knives for CEMOVIS

For the cryo Electron Microscopist, we are pleased to present the DiATOME knives for sectioning vitrified cells and tissues.

Our application specialist is at your disposal for any further information or assistance you might require.





26S proteosomes within cell nucleus of a Drosophila melanogaster neuron. M. Eltsov, IGBMC, Strasbourg, Scale bar: 50nm



Drosophila embryonic brain axons. Defocus -4μm, no phase plate Tomographic reconstruction, computational 5nm slice.

M. Eltsov, IGBMC, Strasbourg Scale bar: 100nm

# **DIATOME** histo knives and their applications

- High quality diamonds guarantee perfect sections and a durable cutting edge
- Easy wetting cutting edge
- · Optimised serial sectioning
- Large boat for easy pick-up

Knife type	Knife angle	Size [mm]	Thickness range [µm]	Boat type	Code (new knife)*	Application
histo	45°	4.0 6.0 8.0	0.2-2	Standard histo boat	40-HIS 60-HIS 80-HIS	Sectioning biological and industrial materials specimens for optical microscopy
histo Jumbo	45°	6.0 8.0	0.2-2	histo Jumbo boat	60-HISJ 80-HISJ	Large boat for serial sectioning
histo cryo (dry)	45°	4.0 6.0 8.0	0.2-2	Triangular histo holder	40-HDC 60-HDC 80-HDC	Sectioning biological and materials specimens for optical microscopy
histo cryo (wet)	45°	4.0 6.0 8.0	0.2-2	Small histo boat	40-HWC 60-HWC 80-HWC	Wet sectioning of polymers (with DMSO/water mixture)

# \* Product Codes shown are for new knives.

To order a resharpened knife, add "R" to the Product Code that corresponds to your knife.

**Example:** Product Code 40-HISR = Resharpened histo 45° 4.0mm Knife.

Histo, histo Jumbo, and histo cryo can be exchanged. To order an exchanged knife, add "E" to the Product Code.

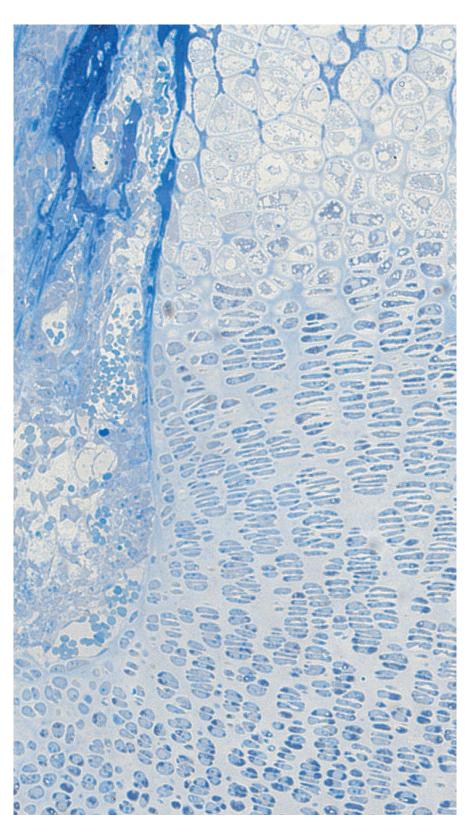
**Example:** Product Code 60-HISJE = Exchange of a histo Jumbo 45° 6.0mm Knife.

# histo

The histo knife is designed for the sectioning of hard and soft biological and material research specimens, non embedded or embedded in acrylic or epoxy resins (O. L. Reymond, Bas. Appl. Histochem. 30, pp. 487 – 494, 1986).

Our histo knives can be used on all ultramicrotomes as well as on microtomes with a retraction of the specimen arm in the return phase.

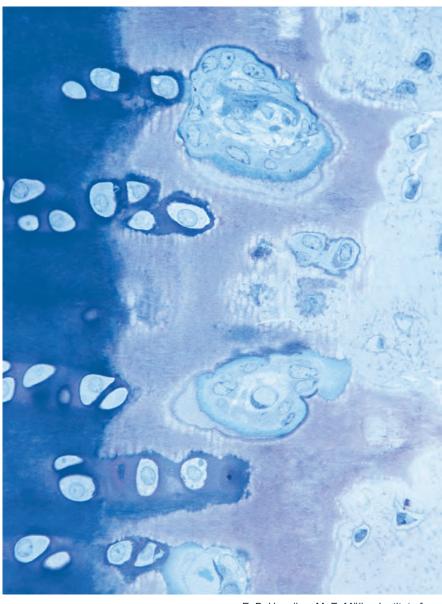




Nondecalcified rat bone.
Scale: 35 mm = 100 μm.
Daniel Studer, Anatomisches Institut, Bern.

# histo cryo

Our histo cryo knives are delivered with a boat for wet sectioning using a DMSO/ water mixture or in triangular holders for dry sectioning.







▲ E. B. Hunziker, M. E. Müller, Institute for Biomechanics, University of Bern. Rabbit joint, calcified cartilage/bone. X760

# histo Jumbo

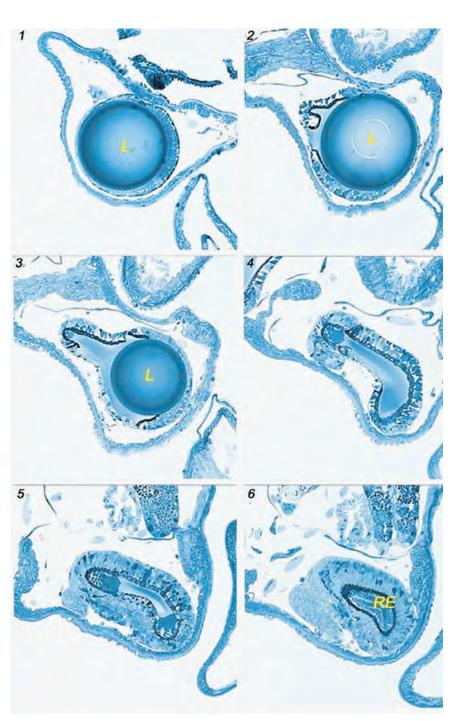
For 3D reconstruction it is imperative not to loose a single section (M. J. F. Blumer et al., Journal of Neuroscience Methods 120, pp. 11-16, 2002).

The large Jumbo boat as well as the adhesive (Pattex compact by Henkel) applied to the upper side of the sample block increase the distinct advantages of our histo knives.

## They allow:

- easy production of section ribbons (0.5 – 2 μm)
- no section loss
- no folding
- the same orientation of all sections
- · easy collection of section ribbons
- multiple ribbons on one glass slide
- · perfect for immuno-histo-chemistry.





Eye of A. peroni: photographic sequence of some semithin sections of a complete serie through the eye.

L = lens; RE = retina.

Michael J. F. Blumer, Institut für Zoologie, Universität Wien.

Reprinted from: Ribbons of semithin sections: an advanced method with a new type of diamond knife. Journal of Neuroscience Methods 120, pp. 11-16, 2002, with permission from Elsevier.

▼
Rat brain x 18'000
Werner Graber, Anatomisches Institut, Bern.



# Static Line II Ionizer

The Static Line II Ionizer is an antistatic device which emits negative and positive ions, neutralizing the electrostatic charging in the cryochamber.

The sections no longer stick to the knife edge, or bunch up one on top of the other, but float in a nice ribbon over the knife surface. The pickup of the sections can be done easily.

We recommend the use of the Static Line II lonizer for trimming, sectioning and section collection.

# **Applications**

- · Sucrose protected specimens.
- Cell cultures in elatine/ sucrose.
- · Frozen hydrated specimens.
- Dry cut Lowicryls at room temperature.
- Polymers and rubber.



Description	Code
Static Line II ionizer	SLI

# **EM Trim Diamond Cutter** for the Leica "EM-Trim"

These cutters are used exclusively in the Leica EM Trim and are available in either 6 or 12 mm diameter. These blades allow for parallel edges of the complete block which is mandatory for successful sectioning.

Description	mm	Code
EM Trim Diamond Cutter	6 12	DEMT DEMT-12

# Handling and Use

## Introduction

DIATOME is the leading manufacturer of diamond knives for ultramicrotomy in biological and materials research, with many years experience. The expertise obtained in collaboration with our customers enables us to offer you the highest quality knives available in the market today. This manual was written in order to allow you an easy handling and use of our knives. Please contact us, if you should need any assistance or if you have any special requirements in your ultramicrotomy applications. We are only a phone call or an email away and look forward to hearing from you.

### Handling

A diamond knife is not as fragile as you might think. Taking into account the vast array of soft and hard specimens it cuts successfully, the diamond knife cutting edge is quite durable.

With proper care and handling, a *DiATOME* knife can withstand a great deal of use. However, in order to avoid any unfortunate mishaps, the following precautions should be taken when receiving the knife:

- Inspect the two securing stickers on the knife box and inform us if they have been cut or damaged.
- · Ask your receiving department not to open the box.
- When unpacking the knife, care should be taken not to touch the cutting edge.
- Once the knife is unpacked it is ready for use and does not need initial cleaning.

## Preparation before sectioning

A perfect section ribbon can only be obtained from a well trimmed block. The upper and the lower side of the block must be parallel to the knife edge. The block should not be too wide because this would substantially increase the cutting pressure. This may result in «chatter».

We have found that the best trimming results of biological and materials research samples, at room- and cryo temperatures, are obtained with our diamond trim blades trim 45 (ord. no. DTB45), trim 20 (ord. no. DTB20), and trim 90 (ord. no DTB90). See our knife brochure for more information.

### Caution

If trimming is done with a razor blade, always use a fresh, degreased blade of good quality. If the razor blade is old and damaged, it has tendency to leave steel particles on the sample block. Upon sectioning, these particles will cause damage to the diamond knife's cutting edge. If a sample block is precut with a glass knife, use only a clean new portion of the cutting edge. This will reduce the risk of glass particles sticking on the sample block. Avoid during the entire preparation of the sample blocks the embedding of hard particles from pipettes, razor blades, glass knives, etc.

# Sectioning at room temperature

The quality of sections will be determined by a number of factors. The following points should be checked before commencing sectioning:

- Set the clearance angle and the cutting speed as indicated on the guarantee card.
- Tighten all of the screws in the sample block holder, the knife holder, etc.
- Align the block (with the backlight on) parallel to the cutting direction.
- · Align the block (with the backlight on) parallel to the cutting edge.
- Align the lower side of the block (with the backlight on) parallel to the cutting edge.

The water in the boat should be level with the cutting edge and give a good reflection. **Exception:** For the sectioning of hydrophilic embedding materials we recommend lowering the water level to a concave shape.

To avoid difficulties during sectioning and extend the life of a diamond knife, the following should be kept in mind:

- The sample block should be fully polymerized.
- Do not add solvents (i.e. acetone) to the distilled water in the boat. They may dissolve the sealing material between the knife and the boat. If the adding of solvents is unavoidable, ask us for sealing the knife with an epoxy resin.
- Avoid cutting thicker sections than the limits given for each type (ultra 150nm, cryo 500nm, histo 2µm).
- Exercise care when picking-up sections. Do not touch the cutting edge with any solid objects (grids, loops, tweezers, etc).
- · Do not allow the sections dry on the cutting edge.

# Sectioning at low temperatures

# **Trimming**

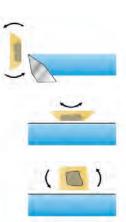
For successful cryosectioning of biological and materials research specimens trimming is imperative. Our diamond trim blades trim 45, trim 20, and trim 90 allow you to quickly and easy fulfill your trimming requirements.

For trimming we recommend the same temperatures as set for the sectioning. For trimming it is advisable to use an ionizer. It eliminates section debris sticking to the specimen and on the trim blade.

### Sectioning

For dry cryo sectioning the following temperatures are recommended for the cryo chamber, knife and specimen:

- Sucrose infiltrated biological samples: semithin sections – 80°C, ultrathin sections – 110° to – 120°C
- Frozen hydrated biological samples: 150°C
- Polymers, rubber: 120°C (or below the glass transition temperature).



For dry cryo-ultramicrotomy the use of an ionizer allows the cutting of perfect section ribbons and an easy section collection.

For the cryo sectioning with boat liquid (i.e. DMSO/water 50/50%) set the following temperatures:

- Cryochamber 120°C
- Specimen 120°C
- Diamond knife 30°C

These figures usually allow best cutting results. Please contact us for more detailed information.

At low temperature the handling of the diamond knives is the same as for room temperature. Be careful when picking up the dry cryosections from the diamond surface. The cutting edge may not be touched with any solid objects (i.e. loops, tweezers or grids)!

# Cleaning

### Method 1

Immediately after picking up the sections, remove all unused sections with a hair curl on an eyelash. Then clean the knife edge as follows:

- Empty the boat, dry carefully with filter paper (without touching the knife edge!) and remount it in the ultramicrotome knife stage.
- Take one of our polystyrene sticks and bevel it to an angle of approx.
   60° using an oil free razor blade.
- Dip the rod into 100% ethanol and shake off the excess.
- Pass the rod over the cutting edge without applying pressure.

Mechanical cleaning as mentioned above is absolutely necessary after cutting poorly polymerized blocks (i.e. Lowicryl polymerized in a freeze substitution apparatus).

### Method 2

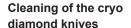
If you do not feel comfortable using method 1, there is an alternative. Immediately after picking up the sections, proceed as follows:

- Remove all unused sections with a hair curl or an eyelash.
- · Rinse the knife thoroughly with distilled water.
- Take a can of clean pressurized air and blow the water off the cutting edge.

# Method 3

If sections or debris dry on the knife edge we recommend the following cleaning procedure:

- Place the knife in distilled water, adding one or two drops of mild liquid dishwashing detergent.
- · Let the knife sit overnight.
- · Remove the knife and rinse with distilled water.
- · Now proceed to clean the knife using method 1.



- Remove the knife from the cryo chamber (before heating the chamber up!).
- Rinse the knife under tap water to warm it up.
- Clean the knife using method 1, use 50% ethanol for cryo knives used in biological applications, use 100% ethanol for cryo knives used for polymer sectioning.



### Caution

We do not recommend sonification for the cleaning of any of our diamond knives. Solvents and acids should never be used under any circumstances.

# Resharpening

*DiATOME* diamond knives can be resharpened an unlimited number of times due to special attention given to minimizing the amount of grinding during the sharpening procedure. The cutting edge length is not affected.

Our sophisticated manufacturing and resharpening methods are unique and allow us to guarantee that your resharpened DiATOME knife will be of the same superior quality as your new DiATOME knife.

# Guarantee

DIATOME

Before delivery, each knife, new or resharpened, is subject to extensive testing. Only if its performance is to the highest standards, we will ship it.

In the unlikely event that you experience difficulties, or for any reason are unhappy with the performance of your knife, contact us immediately. Your short description will allow us to solve the problem to your full satisfaction.

# **DIATOME U.S.**

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