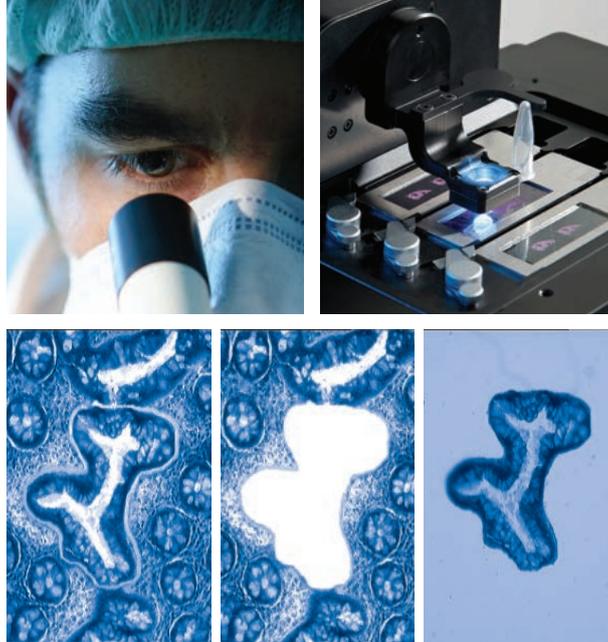




**Molecular Machines & Industries**

# **Solutions for Molecular Pathology**

Laser microdissection technologies for life sciences research and clinical applications



**Microscopy based Single Cell Isolation**

# Solutions for Molecular Pathology

From routine to most complex laser microdissection	4-5
MMI CellCut Plus: Unique features & benefits	6-7
MMI CellCut Plus: A high degree of expandability	8-9
MMI CellCut Plus: A wide range of applications	10-11
Customer report and application protocols	12-13
Customer support and high-quality consumables	14-15

## Contents





# Introduction

In molecular pathology and life sciences research, laser microdissection is an essential technology for the isolation of cells and tissues for further downstream analysis. Laser microdissection enables early diagnosis of cancers and neurological disorders, and it is now established as a step towards personalized medicine.

MMI has developed a laser microdissection technology with unmatched precision and speed where sample isolation is absolute contamination free. The result is a verified sample collection with a high degree of efficiency combined with reproducibility and least sample damage. This creates unique prerequisites for successful downstream applications. Furthermore, MMI instruments are highly modular and can be mounted on numerous microscope brands.

# From routine to most complex laser microdissection

## MMI CellCut

The MMI CellCut was designed for the quick and precise isolation of cells and tissue and so is an essential tool for molecular pathology. A wide variety of sample types including fresh frozen, paraffin embedded, archived slides, cytospins, smears and live cells can all be used for diagnostic purposes.

The MMI CellCut enables cell selection quickly and easily directly on the touch screen. The cells of interest are marked and cut automatically using a precisely focused UV-Laser. The microdissected samples are collected for downstream analysis. This results in a pure cell population for reliable analysis and diagnosis.

The MMI CellCut is highly modular and can be mounted on numerous brands of microscope from entry level, mid range to high end, suitable for the most routine to the most complex research applications.

## Setup

- Based on an inverted or upright, manual or motorized, entry level or high end microscope
- High precision motorized scanning stage
- Electronically controlled solid-state laser (optional with high power for thick, hard, or wet tissue), laser beam delivery and transfer optics
- MMI CapHolder with MMI Single CapLift, MMI MultiSlide insert for parallel use of up to 3 slides, MMI LiveCell insert for applications with living cells, optional with MMI Multi CapLift for the parallel use of up to 8 caps
- High-resolution MMI CellCamera
- Modular and upgradeable workstation
- MMI CellTools software for full control of the laser, image capture, and scanning stage actions combined with MMI CellExplorer software for automatic cell recognition in fluorescence or bright field
- Interactive LED pen display

*MMI CellCut on Olympus IX83 motorized inverted microscope*



*Interactive pen display and intuitive MMI CellTools software*



*MMI controller unit*





Fast and easy isolation of cells from up to three slides simultaneously: the unique MMI CapLift technology provides an automated and contamination-free transfer to the adhesive lid of the downstream reaction tube

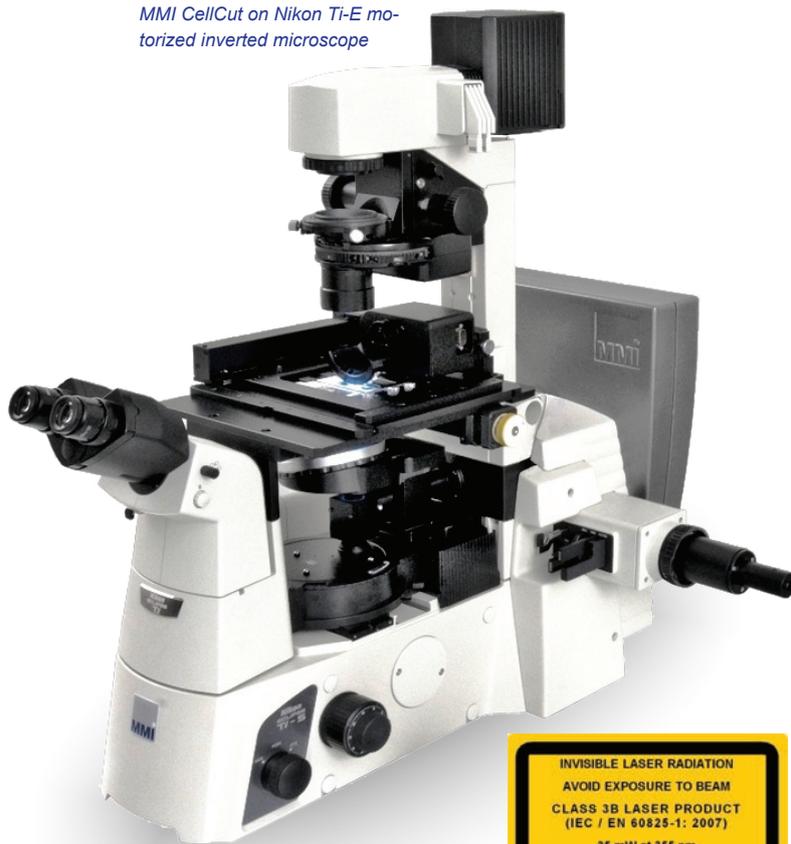
### Expandability:

- High power laser for microdissection of harder material, e. g. plants, bone (marrow), teeth, forensic tapes
- Digital remote laser microdissection
- MMI CellEctor for single cell sorting
- MMI CellManipulator optical tweezers
- Range of imaging configurations (Fluorescence, DIC, Phase contrast)
- Others on request

### Compatibility:

- Nikon Ti-S, Ti-E, Ni-U, Ni-E, A-1
- Olympus IX53, IX73, IX83, FV1000
- Zeiss Axio Observer, LSM 780
- Others on request

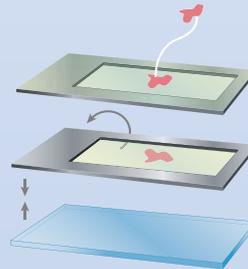
MMI CellCut on Nikon Ti-E motorized inverted microscope



INVISIBLE LASER RADIATION  
 AVOID EXPOSURE TO BEAM  
 CLASS 3B LASER PRODUCT  
 (IEC / EN 60825-1: 2007)  
 35 mW at 355 nm

## Technology

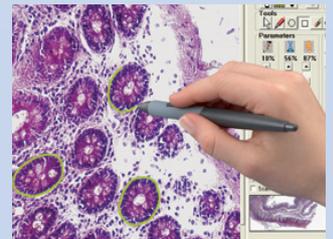
### 1. Sample preparation



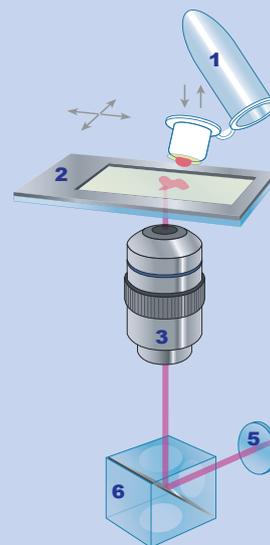
The section is placed on the MMI MembraneSlide, a frame slide covered with a thin membrane that is inert and has negligible auto fluorescence. Afterwards the MMI MembraneSlide is inverted and placed onto a glass slide for protection against contamination. Now the sample is sandwiched between the membrane and the glass.

### 2. Easy cell selection

The cells of interest can be selected on the display using either the mouse, by freehand or predefined geometrical shapes which can be modified. Any number of cells across the slide can be identified as targets within one screening process. The stage is moving to trace the path drawn and the laser is fixed and focused from below.



### 3. Automated laser cutting



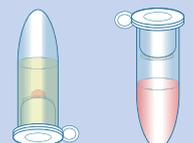
- 1 Adhesive MMI IsolationCap
- 2 MMI MembraneSlide and glass slide
- 3 Phase contrast objectives
- 4 UV solid state laser
- 5 Focusing lenses
- 6 Polarization beam combiner

Computer-controlled movements  
 Software-controlled laser focus and energy

The thin laser cutting path enables a precise and gentle extraction of the selected cell at an outstanding speed. The isolated target cell is collected by lowering and lifting of the adhesive cap held from above. The sample morphology remains 100% intact.

### 4. Selected target

After cutting the sample can be visualised on the cap. Lysis buffer is added and the tube inverted for approx. 10 mins. The cells are now in suspension ready for downstream processing.



# MMI CellCut: Unique features & benefits

## Contamination free cutting

The sample is sandwiched between a membrane slide and a glass slide and so it is not exposed to the air. This ensures a safer working environment and better sample integrity.

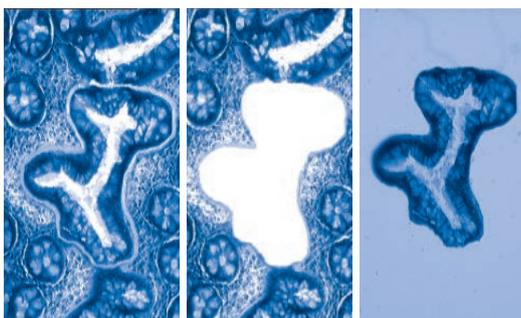
## Maximum sample collection

The unique MMI CapLift technology enables highest sample collection efficiency and minimal sample damage. The yield can be verified during and after isolation.

## Contamination free collection

The MMI IsolationCap is only in contact with the membrane and never in direct contact with the tissue.

## Positive sample inspection



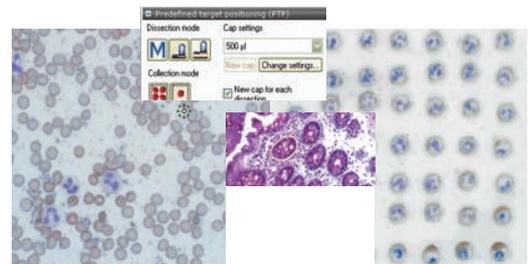
This feature ensures the sample collection after every laser cut. The isolation of the target area occurs in the same relative position as on the slide. The morphology of the sample remains 100% intact and damages of the surrounding tissue are prevented. Reproducible results are achieved easily and effectively. This is excellent for auditability and accurate quantitative results.

## The thinnest cleanest cut

MMI uses a solid state UV-laser with higher pulse rate and lower power. The result is a sharper and cleaner ultra precise cutting line of 0.3 microns and least sample damage.

## Predefined Target Positioning (PTP)

PTP is a patented product feature that allows for the precise, predefined, contamination free visually controlled collection of individual cells on the cap. PTP is excellent to maximize the number of samples collected into a single reaction tube. It guarantees that the isolated samples are not compromised by subsequent cutting which is essential for auditability.

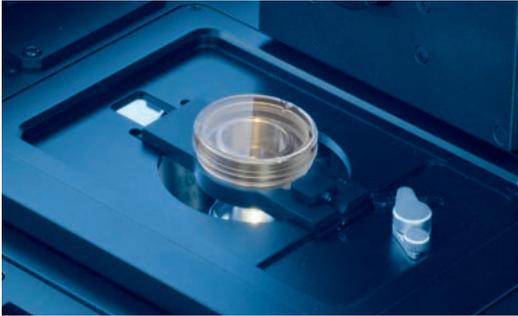


*Firstly an overview of the entire sample is produced. Secondly visual inspection and selection of the target cells is performed. This step can be fully automated, using the MMI CellExplorer software. Switch to PTP mode and cells are dissected and collected on the adhesive cap in a spiral pattern starting from the centre of the cap.*

## Autodocumentation

The MMI CellCut allows full verification and traceability of every cut. The autodocumentation feature records images showing the position of the cut and of the collection (PTP) on the cap together with the time, date and method details.

## MMI LiveCell Chamber



The low power laser allows a gentle cut. In specially designed MMI LiveCell Chamber even live cells can be microdissected, for unrivalled success in re-culturing and cloning.

## MMI MultiCap & MMI MultiSlide



The MMI MultiSlide allows for the isolation of cells from up to three slides and the MMI MultiCap for the isolation of cells into up to eight individual caps without the need to reload. This is essential for diagnostic workflows involving RNA isolation, where speed is of the essence.

## Serial section

This feature maximises the RNA recovery by minimising the use of staining. The visible position of target cells is used on a stained section to detect, mark, cut and isolate regions of interest on an unstained section.

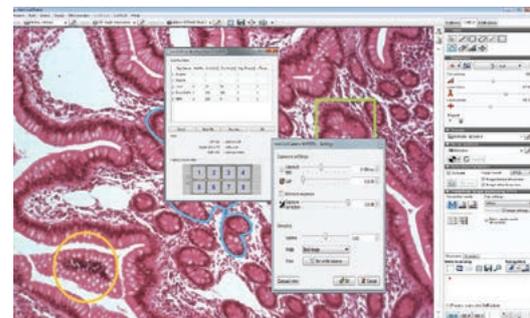
## Z-drill

By this feature, thick and wet tissue can be cut without using a high power laser. Refocus the laser on different z-levels whilst cutting to minimise the use of excessive laser power and so preserve your sample and maximise its usability for downstream analysis. An optional high power laser is available for working with harder to cut material such as teeth, bones, or forensic tapes.

## MMI CellTools software

All MMI instruments come with the intuitive MMI CellTools software package which generates a live view of the entire sample. High quality components such as the MMI CellCamera ensure high resolution and real time application success when working with a wide range of delicate sample material.

The MMI CellTools software and corresponding Plug-Ins provide full control of the system. Adjusting the xy-stage, camera, laser and lighting conditions is easy and convenient. Automated microscope functions such as objective or fluorescence turret changes can quickly be initiated directly from the software making full remote control possible.

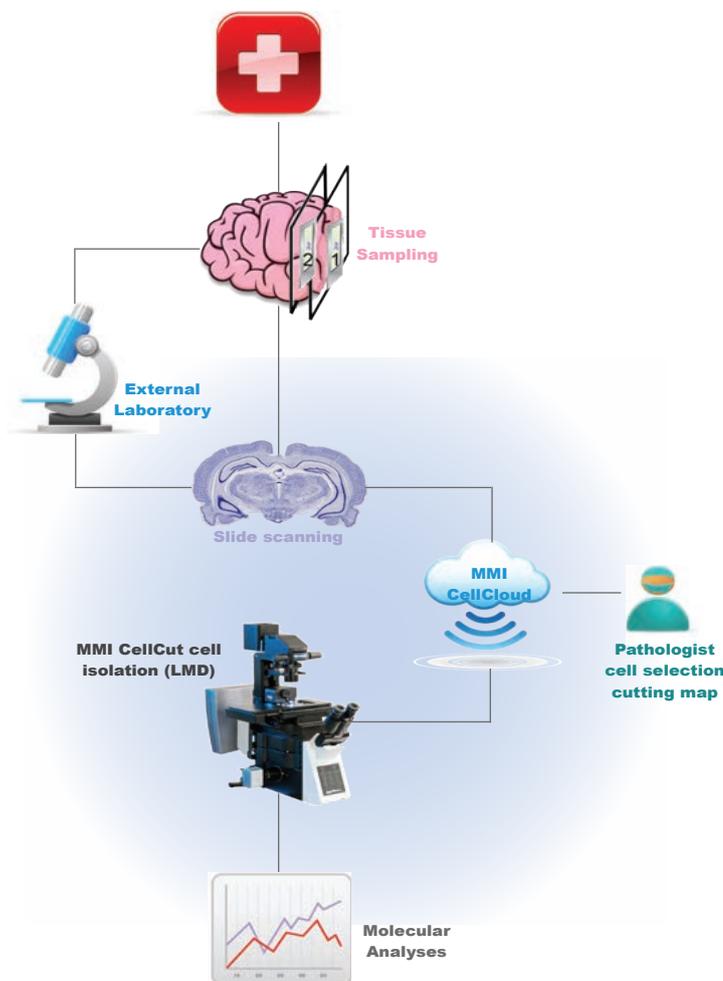


*The MMI CellTools software for easy and convenient laser microdissection provides a live view of the whole sample*

# MMI CellCut Plus: A high degree of expandability

## Digital remote laser microdissection

Laser microdissection plays an important role in modern pathology and cancer research. Now it is possible to combine the advantages of digital and molecular pathology. In collaboration with 3DHistech MMI has established the entire process from digital slide scanning to remote laser microdissection via internet based cloud technology. Transfer of the “cutting map” directly to the MMI CellCut facilitates cell isolation and collection following remote digital diagnosis.



MMI presents the entire process from digital slide scanning to remote laser microdissection via internet based cloud technology

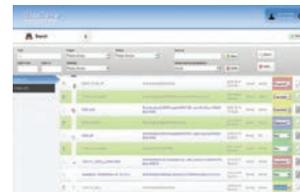
## Workflow

### 1. Scan your slides



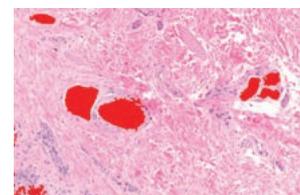
Direct scanning to the CaseCenter, for example with the Pannoramic MIDI from 3DHistech, a 12-slide digital slide scanner which combines brightfield and fluorescence scanning in one and delivers superb image quality.

### 2. Share your digital slides



Slides are stored in the CaseCenter, a web based slide and case database with HIS/LIS integration possibility, and can now be shared with the remote specialist.

### 3. Select cells or group of cells



The areas of interest can be found automatically with the image analysis software, a multi-purpose solution which runs on local and remote slides. The annotations on the digital slide serve as a cutting map for the MMI CellCut Plus.

### 4. Automatic cell isolation



The MMI CellCut precisely cuts and isolates the cells on the imported cutting map and transfers them to the PCR tube for downstream analysis.



Designed for fast and reliable detection and isolation of single cells, stem cells and CTCs by fully controllable microscopic collection:  
MMI CellEctor

## MMI CellEctor for integrated workflows

MMI's fully modular platforms can be used either alone or in combination with other technologies or micromanipulation tools. As an example, the MMI CellCut can be combined with the MMI CellEctor for the detection and isolation of single cells in suspension. This creates unique flexibility for a wide range of applications and for the development of integrated workflows for single cell isolation.

The MMI CellEctor is a capillary based single cell sorting system. The completely software manipulated 3D CellRobot controls the precise landing position of the capillary and generates a highly reproducible contact point. The high precision of the MMI CellPump regulates the acquisition and deposition in a variety of modes, giving the user complete control in manual and automated cell recognition, acquisition and deposition.

All accessories were directly developed in consultation with MMI CellEctor users to reflect their exacting needs. They allow for the deposition of single cells onto reaction slides or directly into PCR tubes, IBIDI style chambers, microfluidic devices or well plates. User defined liquid handling programmes enable the development of dedicated cleaning and service cycles for capillary cleaning and maintenance.

*Combi solution:  
MMI CellCut and MMI CellEctor (here with  
Olympus IX83 microscope)*

## Features & benefits

- Microscope based software controlled movements of the capillary
- Easy to achieve accurate and precise cell isolation and deposition
- No contamination from unwanted cells
- Faster and easier than manual systems
- Nanoliter pump allows isolation of cells in small volumes
- Brightfield or fluorescence automated detection of cells
- Full process control of cell identification, acquisition and deposition
- Work manually or automatically as your workflow demands
- Compatibility with a wide range of accessories and microfluidic devices

## Applications

- Detection and isolation of single and rare cells in suspension, e.g. Circulating Tumor Cells (CTCs)
- Cancer research and oncology
- Detection and isolation of monoclonal antibodies
- Cell sorting on a wide range of biochips
- Lab-on-a-chip technologies
- Stem cell research and cellular diagnostic
- Single Cell PCR and gene expression analysis

For more information, please have a look into our brochure *MMI Single Cell Solutions* or visit our website

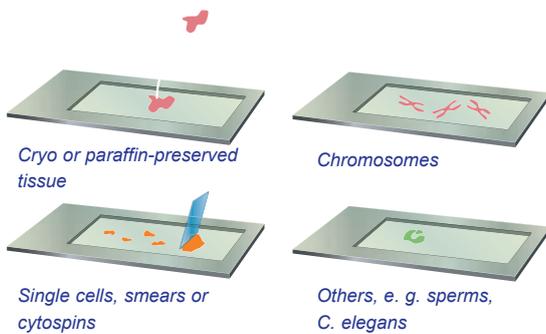
[www.molecular-machines.com/products/mmi\\_cellector\\_plus/](http://www.molecular-machines.com/products/mmi_cellector_plus/)



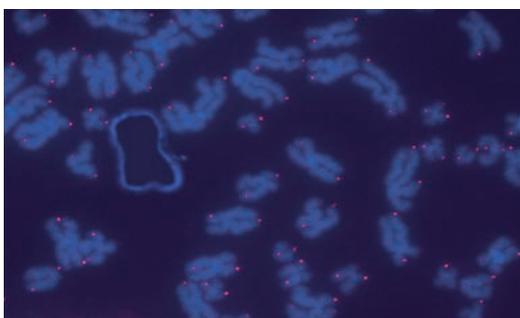
# MMI CellCut Plus: A wide range of applications

## The starting materials

The MMI CellCut is an extremely versatile instrument and therefore the best choice for clinical and research microdissection. Whether isolating cells from an entire tissue, maintaining a pluripotent stem cell line, or extracting cells for forensic investigation, the MMI CellCut performs efficiently and reproducibly.



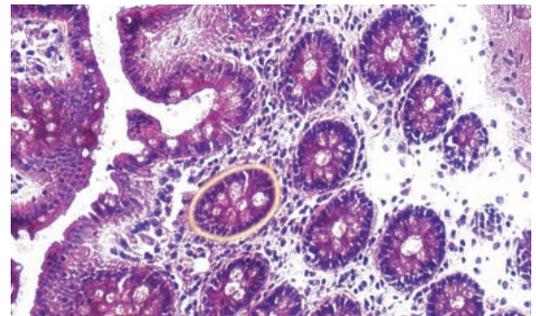
## Fluorescence-labeled cell components



*Fluorescence-labeled chromosomes*

The precision of the MMI CellCut enables the simple location and microdissection of sub-cellular structures. For example, chromosomes with detectable fluorescence in situ hybridization (FISH) signals can be isolated for subsequent investigation.

## Pathological samples



*Isolation of intestinal gland from transverse tissue sections of colon (H&E stained)*

Targeting specific cells in healthy and diseased tissues is of particular importance in elucidating the molecular mechanisms that leads to cancer and other life threatening diseases. Differences in genomic (DNA/RNA) and proteomic expression of any tissue type can be analyzed easily. For example, transverse sections of intestinal glands from colon tissues can be identified and quickly isolated for the study of specific genes and the corresponding hormonal response.

## Live cells

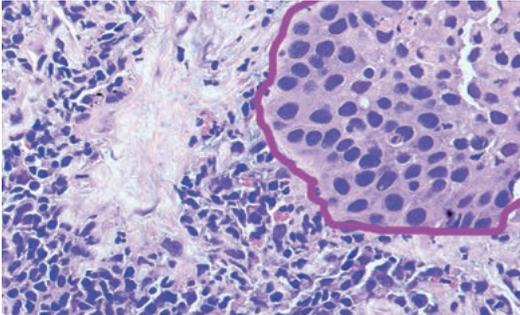


*Cell culture in phase contrast observation*

Most stem cell lines are grown at high densities on mouse fibroblast “feeder layers”. Therefore, isolating specific pluripotent cells from the surrounding fibroblasts and differentiating cells needs to be fast and precise. The MMI CellCut offers the perfect balance between speed, precision and ease of use, allowing the target stem cells to be isolated and re-cultured without any side effects, such as karyotype changes.

The MMI CellCut is widely used in traditional research, for cancer and personalized medicine

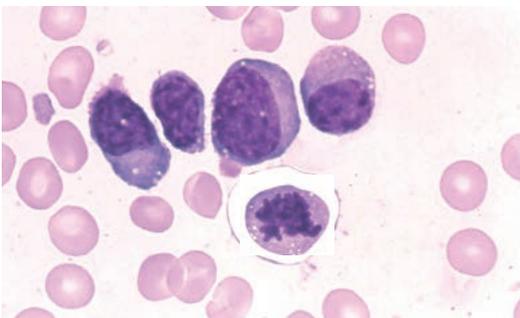
## Cancer research & Oncology



Detection and isolation of cancer tissues from tissues or cell cultures

The enrichment of cell populations from tumors extracted from tissue or blood samples is essential for the development of truly effective treatment programmes. The key to success in cancer treatments is fast and accurate diagnosis, rapid implementation of specific treatments and treatment effectiveness monitoring. The MMI CellCut is predestinated to work with a wide range of sample types for the isolation of cancer tissue from sections facilitating fast and effective diagnosis.

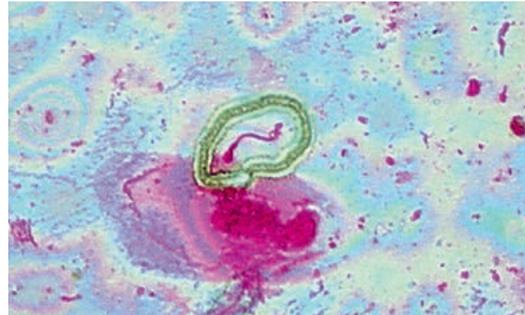
## Haematological and cytological samples



Blood smear with a typical plasmablast

Single cells, such as a plasmablast from a blood smear can be identified, cut, and isolated automatically. The dissected cells are now ready for downstream analysis such as molecular profiling for proteomics or genomics purposes.

## Forensic analysis



Detection and isolation of sperm cells from case work smears

The MMI CellCut allows the location, cutting, isolation and collection of in excess of 300 sperm cells on a single cap, all under full audited visual control. This maximises the chances of obtaining a high quality full DNA profile that can be loaded and searched on appropriate crime databases. The MMI CellCut can also be utilised for the isolation of cells from archived slides making it a powerful tool in the investigation of old unsolved cases.

## Others



Isolation of living *C. elegans*

*C. elegans* are extensively used as a model organism in molecular research and developmental biology and can also be examined with the MMI CellCut Plus. The living organism can be placed or fixed between the membrane and a glass slide, in order to isolate and extract areas of interest as required by the individual experiment.

# Customer report and application protocols

## Testimonial

Dr. med. Danny D. Jonigk, Pathological Institute, Medical School Hannover, Germany

*"We appreciate the resistant MMI product quality, the professional consulting, and the competent and quick service. MMI instruments are an important basis for our in-situ analysis in cellular tissue. Laser microdissection followed by gene expression analysis is complementary for further routine methods like conventional optical microscopy (fluorescence), in-situ hybridization, and immunohistochemistry."*

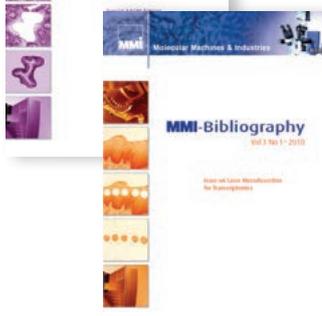
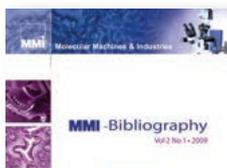
Dr. Jonigk and his team work in the field of lung tumor diseases, fibrous lung remodeling under different circumstances, and pulmonary hyper-tension. The focus is on the combination of clinically relevant issues and the morphology of the tissue, and on the expression of DNA, RNA, and proteins in sub-compartments of the lung. With the MMI CellCut laser microdissection system, Dr. Jonigk is able to retrieve high quality samples from the starting material. This significantly improves the accuracy of his results.

## MMI Bibliographies



First issue on laser microdissection in cancer research and clinical oncology, featuring technologies and references

Second issue on laser microdissection in cancer proteomics, featuring customer reports, technologies and references



Third issue on laser microdissection for transcriptomics, featuring customer and MMI reports, publications and references

## Contamination free staining: MMI Staining Kit Plus



The MMI H&E Staining Kit is designed for users with a need to quickly stain only a few samples and need to ensure that the result is clean, clear of RNase and contamination free. The staining solutions are supplied in MMI SafeStain ampoules, allowing quick handling without the need for pipetting or the preparation of staining jars.

The MMI SafeStain ampoule guarantees a uniform drop size, and ensures the solutions remain contamination free. The staining solutions have been rigorously tested for the demanding needs of laser microdissection users. The clear staining they need is reproducibly of the samples together when viewed with the MMI IsolationCap. Each kit contains 15 MMI SafeStain ampoules, designed for 30-60 staining sessions. Ampoules are designed for the staining of 2-4 slides.

Improved sample Lift-Off success rate due to quicker drying. By choosing the MMI H&E Staining Kit you help to reduce the amount of water hazardous stain waste and cut your laboratory running costs. The whole workflow is illustrated in the application protocol:



H&E Staining with the MMI H&E Staining Kit



# MMI -Bibliography

## Live cell dissection: MMI LiveCell Chamber

The isolation and enrichment of individual live cells for culture and differentiation experiments, or for their proteomic and genomic analysis is of increasing interest in stem and cancer cell research, and molecular pathology.

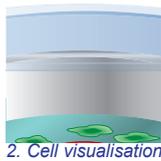
The MMI CellCut in combination with the approved MMI LiveCell Chamber enables contamination free isolation of live cells in living culture. Enzyme treatments (i.e. trypsin), other potentially harmful selective reagents as well as time consuming repetitive enrichments can be avoided. The benefit is an increase in reliability, effectiveness and accuracy of your research.

### MMI LiveCell Chamber

Prod. No. 50301



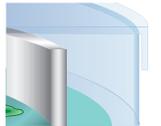
1. Cell seeding and cultivation



2. Cell visualisation



3. Cell selection and automated laser cutting



4. Cell isolation and reculturing

5. Further downstream application

Components of the MMI LiveCell Chamber are a membrane ring for the initial cultivation of cells, a cell culture dish to house the membrane ring with seeded cells, and a UV-permeable microdissection chamber. All components can be sterilized ready to use. The whole workflow is illustrated in the application protocol:



Live Cell Microdissection

Download the MMI Bibliographies and all protocols from [www.molecular-machines.com/support/information\\_material](http://www.molecular-machines.com/support/information_material)



Zebrafish Application Note



Tissue Transfer from Archived Samples



Smears and Cytospins



Embedding and Freezing



FFPE Sectioning



FFPE Embedding



Cryosectioning



Chromosome LCM



ScreenCell® Application Note



General Staining



Director™ Slides



Tips for working with RNA

# Customer support and high-quality consumables

You make a substantial investment by buying a microdissection system and the right choice by selecting a MMI product. To ensure you always get the best from your instrument and longterm continuous use, we recommend all customers consider the benefits of service contracts, regular upgrades, application trainings and our high-quality consumables for better results.

## Keep your system running smoothly, reliably and ready for use:

Choose between different service contracts over a time frame to fit your needs. They can include annual preventative maintenance by one of our service engineers and also emergency callout cover. Please contact MMI directly for availability and options in your region.

## Stay up-to-date with new developments and upgrades:

We regularly provide various upgrades for all our instrument platforms, for example a more powerful workstation with interactive pen display, a new laser or camera, the latest software version with new features and benefits.

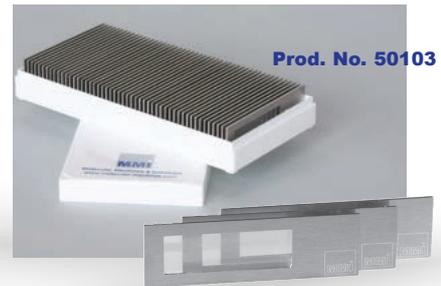
## Ensure trouble-free daily operation:

We offer protocol development and instrument trainings either in-house or on-site with your own equipment.

## MMI TeamViewer for remote service available by default:

The MMI TeamViewer function allows our service engineers to connect with your MMI system via remote service. This makes world-wide troubleshooting fast and effective. Even the MMI CellTools software can be updated this way. The MMI TeamViewer function operates very easily without the need for configuration by the end user, even through firewalls and proxy servers. If the TeamViewer is not yet installed on the MMI computer, you can download it from our website. Please call us after the download to facilitate the connection.

## MMI MembraneSlides



Prod. No. 50102  
(RNase free)

The 26 mm x 76 mm metal frame is coated with a 1.5  $\mu\text{m}$  thin PEN membrane. You can choose between standard MMI MembraneSlides that are packed into a slide rack (Prod. No. 50103). Or, If you need the highest standards of purity the MMI MembraneSlides are available RNase and nucleic acid free (Prod. No. 50102). They are packaged into boxes of five for better contamination control. Every sales unit comes with 50 MMI MembraneSlides in total.

## MMI SupportSlide



To support the transfer process from a cryo section to a membrane slide. The MMI SupportSlide is designed to give rigidity to the membrane during sample preparation. It also acts as a heat sink to aid reproducibility and ensure you correctly place the sample on the correct side of the membrane.

## MMI ServiceSlide

Prod. No. 50108



MMI ServiceSlide and 18-well IBIDI® Slide are used for liquid handling on the MMI CellEctor Plus.

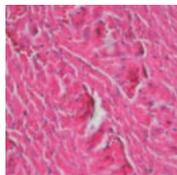
## MMI IsolationCaps

For the isolation and collection of the target cells from the MMI MembraneSlide. Available in three sizes (0.2 ml, 0.5 ml and 1.5 ml) and two types (either filled with nontransparent silicone, diffused, or with transparent silicone). Diffuser caps are recommended for stained or coloured samples.



### Diffuser caps

**Prod. No.**  
**50206 (0.2 ml)**  
**50202 (0.5 ml)**  
**50210 (1.5 ml)**

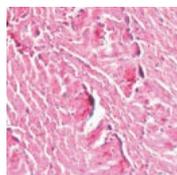


Transparent caps are used for fluorescent applications. Available in boxes of 50 pieces.

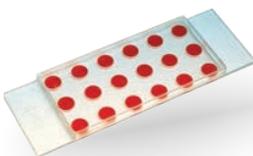


### Transparent caps

**Prod. No.**  
**50208 (0.2 ml)**  
**50204 (0.5 ml)**  
**50212 (1.5 ml)**



## 18-well IBIDI® Slide



**Prod. No. 50109**

If you are working with small cell populations an alternative to the MMI LiveCell Chamber is the 18-well membrane IBIDI slide. The cells are cultivated within the wells. Afterwards, the slide is turned upside down and placed on the stage. The cells can now be cut and isolated with a standard MMI IsolationCap (0.2 ml recommended). If still filled with liquid the MMI CellEctor can, instead, be used to transfer the cells directly from the wells.

**Starter Kit**  
**MMI CellEctor Plus**  
**Prod. No. 70303**

## MMI 8-CapStrip (0.2 ml per cap)

**Prod. No.**  
**50214 (transparent)**  
**50213 (with diffuser)**



Used together with the MMI Multi CapLift for the collection of cells in up to 8 different caps simultaneously. A box per type contains 5 strips.



## MMI CapStrip with 8 empty caps (0.2 ml per cap)

**Prod. No. 50215**

For the collection of cells with the MMI CellEctor Plus, 0.2 ml per cap, 5 strips per box.



## MMI Capillaries

**Prod. No. 80105 (40 µm)**  
**Prod. No. 80103 (20 µm)**  
**Prod. No. 80102 (10 µm)**

Designed for cell collection with MMI CellEctor Plus, 54 mm long, very sharp 45° beveled tip to collect even adherent cells, available in 3 sizes, 10 pieces per box. Also available: MMI Oil and MMI CapillaryClean. MMI CapillaryClean keeps the capillary free of dirt and contamination and avoids blockage. Cleaning of capillaries is recommended as part of all workflows.

## MMI Oil Prod. No. 80108



**Prod. No. 80107**  
**MMI CapillaryClean**

## MMI Starter Kits

Recommended for first application use after instrument purchase. Contain all available consumables for MMI CellEctor or MMI CellCut Plus.

**Starter Kit**  
**MMI CellCut Plus**  
**Prod. No. 70304**



Please note that some illustrated products or features might be unavailable in some countries. Please contact MMI or your local distributor for availability and options in your country.

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Published by Molecular Machines & Industries GmbH,  
Eching, Germany | 3rd edition, 2016