

MACSima™ Imaging Platform

The easy way to ultrahigh-content imaging





MACSima™ Imaging

A new platform for fully automated, ultrahigh-content imaging

Biological systems and cellular processes are inherently complex due to the interaction of thousands of proteins involved in proper functioning of the entire organism. Thus, an in-depth analysis of biological systems requires the examination of a plethora of parameters in order to decipher the underlying principles. Currently available techniques can provide only a very limited perspective on the complexity of biological systems.

Miltenyi Biotec's new MICS (multiparameter imaging cell screen) technology impressively overcomes these limits as it allows the microscopic analysis of an unprecedented number of proteins or other antigens on a single sample.

Based on this technology, Miltenyi Biotec developed the MACSima™ Imaging Platform which enables fully automated high-content imaging. The possibility of evaluating localization, expression, and potential interaction of a multitude of different proteins allows scientists to explore new horizons in basic and biomedical research.

nti-HLA-ABC+- lgD-TSPAN-8+CD3-CD4-CD8-CD18-CD29+CD45RO-C 38-CD46+ CD47+- CD183+- CD27- CD31- CD49f+- CD107a+- CD162- CD274-F ·CD54+- CD59+- HLA-DR+- HLA-DQ- CD45- CD20+ CTLA4-CD152- CD86-51+ CD56+ CD80+ CD94+ CD95L-CD178-FasL+- CD104-Integrinb4+ CD117-Oct-4- CD23- CD24+ CD58+ CD34+- CD73+- CD90+ CD95-Fas+- CD133+ C -ICOS-CD326+ Anti-CLA+ Podoplanin-CD49c-ITGA3+ CD55-DAF+ CD68 9+ CD45RO- CD49e+- CD71+- CD146+- CD147+ CD166+ CD204- CD227+ CD D162⁻ CD274-PD-L1+- CD163⁻ IgA+- CD276+- CD45RA-Anti-HLA-A2-A28+ C ·CD152- CD86- GITR-CD357- ki-67+- OX40-CD134- SSEA-1+- Vimentin- CD 4-Integrinb4+ CD117- CD138+ CD141- CD142+ CD155+ CD171- CD309- CD318 CD133+ CD156c-ADAM10+- CD184-CXCR4+ CD195+ CD223-LAG3- CD240D CD227+ CD239- CD279-PD-1- CD298- CD38-CD46+ CD47+- CD183+- CD27-·A2-A28+CD66acde-CD66c-CD44+ICAM-1-CD54+-CD59+-HLA-DR+-I ntin- CD11c- CD14- CD19- CD28- CD49B+ CD51+ CD56+ CD80+ CD94+ CD9 09- CD318-CDCP1+ Anti-SSEA-4+- lgG- Oct-4- CD23- CD24+ CD58+ CD34+ 3-LAG3⁻ CD240DCE+- CD273-PDL-2⁻ CD278-ICOS⁻ CD326+ Anti-CLA+ Pod - lgD- TSPAN-8+CD3- CD4- CD8- CD18- CD29+ CD45R0- CD49e+- CD7 CD183+- CD27- CD31- CD49f+- CD107a+- CD162- CD274-PD-L1+- CD163- Id - HLA-DR+- HLA-DQ- CD45- CD20+ CTLA4-CD152- CD86- GITR-CD357-CD80+ CD94+ CD95L-CD178-FasL+- CD104-Integrinb4+ CD117- CD138+ CD CD58+ CD34+- CD73+- CD90+ CD95-Fas+- CD133+ CD156c-ADAM10+- CD18 Anti-CLA+ Podoplanin- CD49c-ITGA3+ CD55-DAF+CD68- CD105-CD206 CD71+- CD146+- CD147+ CD166+ CD204- CD227+ CD239- CD279-PD-1- CD2 D163⁻ lgA+- CD276+- CD45RA-Anti-HLA-A2-A28+ CD66acde- CD66c- CD4)357 ki-6 D138+C SSEA-4+

CD105-CE CD279-PI CD266c-D19- CD2 CP1+ Ant E+- CD27: 3- CD4- C - CD49f-O+- HLA-E CD80+ C

-ADAM

D58+ C

· Anti-Cl CD71+- (

lgA+-CD

ki-67+- (

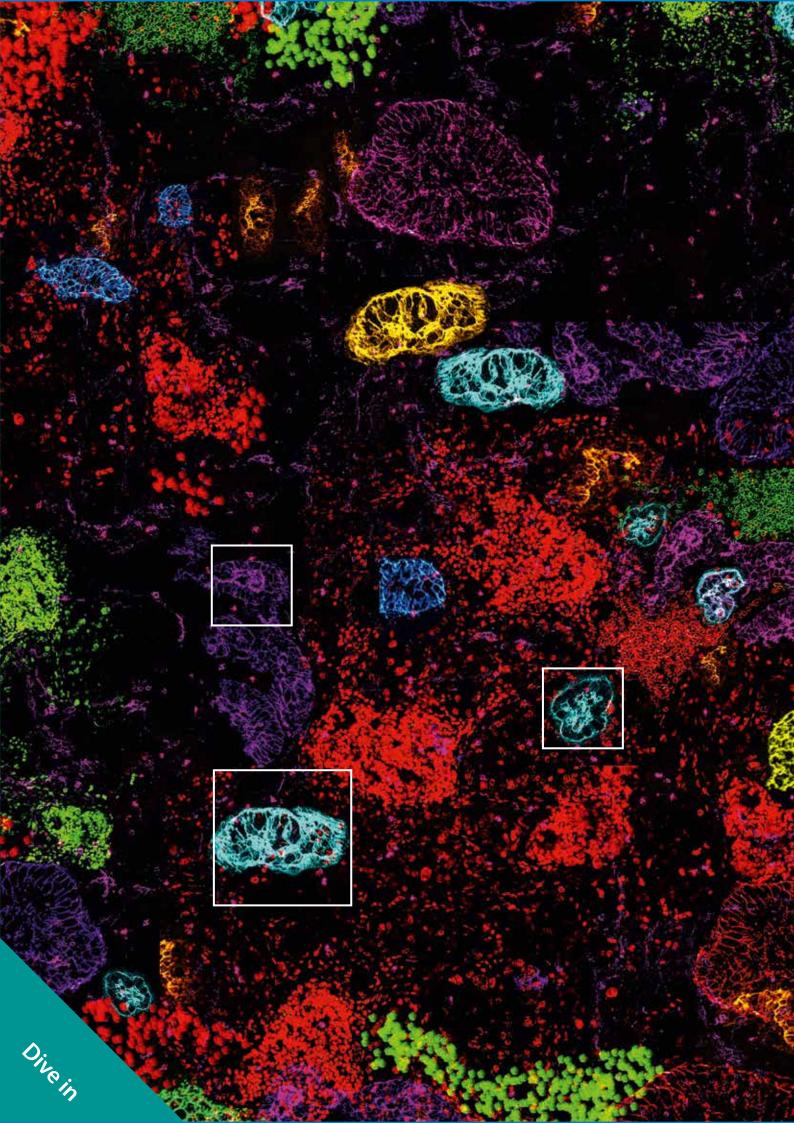
+- CD90+ D49c-IT0 47+ CD16 D45RA-7 D357- ki D138+ CD D+- CD18--CD206-D-1- CD20 D44+ ICA 28- CD49

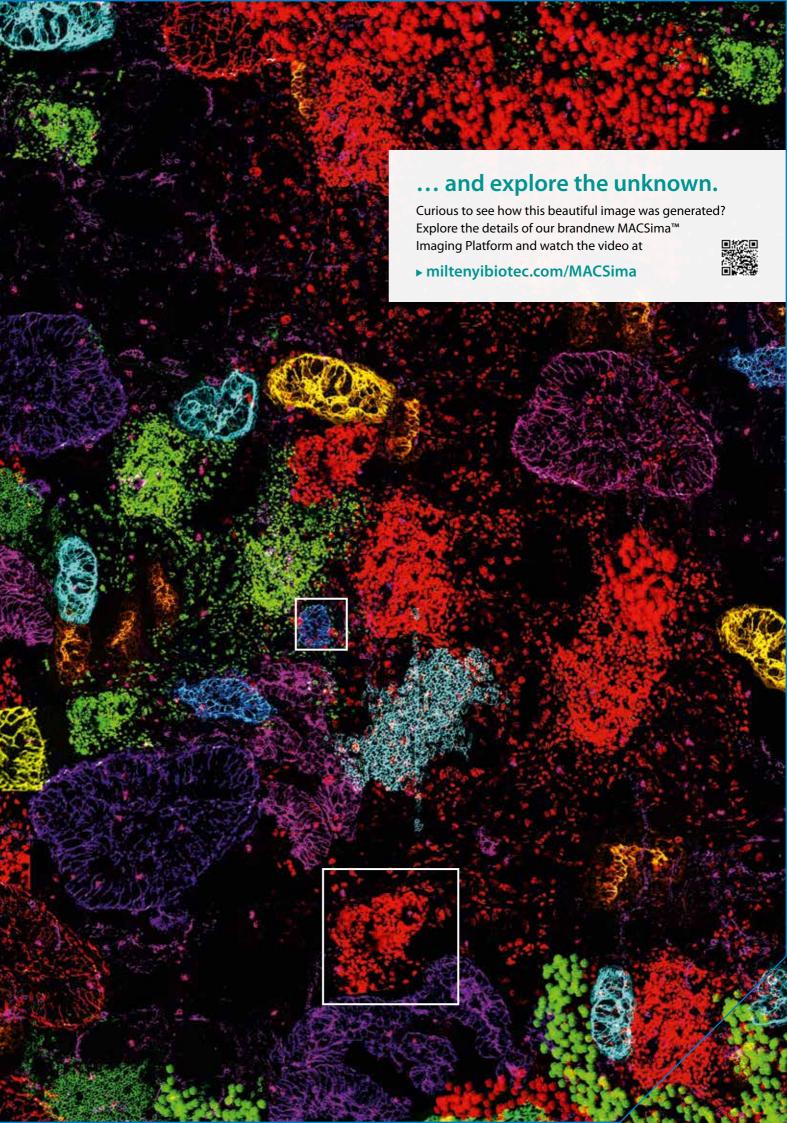
D273-PE

CD4-CD

49f*- CE 5- CD20*

:-ADAM10+- CD184-CXCR4+ CD195+ CD223-LAG3- CD240DCE+- CD273-P CD66c- CD44+ ICAM-1-CD54+- CD59+- HLA-DR+- HLA-DQ- CD45- CD20 D19- CD28- CD49B+ CD51+ CD56+ CD80+ CD94+ CD95L-CD178-FasL+- CD CP1+ Anti-SSEA-4+- lgG- Oct-4- CD23- CD24+ CD58+ CD34+- CD73+- CD90 - CD273-PDL-2- CD278-ICOS- CD326+ Anti-CLA+ Podoplanin- CD49c-IT - CD4- CD8- CD18- CD29+ CD45RO- CD49e+- CD71+- CD146+- CD147+ CD CD49f+- CD107a+- CD162- CD274-PD-L1+- CD163- IgA+- CD276+- CD45RA - HLA-DR+- HLA-DQ- CD45- CD20+ CTLA4-CD152- CD86- GITR-CD357-CD80+ CD94+ CD95L-CD178-FasL+- CD104-Integrinb4+ CD117- CD138+ CD :D58+ CD34+- CD73+- CD90+ CD95-Fas+- CD133+ CD156c-ADAM10+- CD18 Anti-CLA+ Podoplanin-CD49c-ITGA3+CD55-DAF+CD68-CD105-CD206-CD71+- CD146+- CD147+ CD166+ CD204- CD227+ CD239- CD279-PD-1- CD29 lgA+- CD276+- CD45RA-Anti-HLA-A2-A28+ CD66acde- CD66c- CD44+ IC/ ki-67+- OX40-CD134- SSEA-1+- Vimentin- CD11c- CD14- CD19- CD28- CD4 CD138+CD141-CD142+CD155+CD171-CD309-CD318-CDCP1+Anti-SSEA-4 CD105-CD206- CD317-BST2-Anti-HLA-ABC+- lgD- TSPAN-8+CD3- CD4- C CD279-PD-1⁻ CD298⁻ CD38⁻CD46⁺ CD47⁺⁻ CD183⁺⁻ CD27⁻ CD31⁻ CD49f⁺⁻ (CD66c- CD44+ ICAM-1-CD54+- CD59+- HLA-DR+- HLA-DQ- CD45- CD20 D19- CD28- CD49B+ CD51+ CD56+ CD80+ CD94+ CD95L-CD178-FasL+- CD +- CD273-PDL-2- CD278-ICOS- CD326+ Anti-CLA+ Podoplanin- CD49c-IT - CD4- CD8- CD18- CD29+ CD45RO- CD49e+- CD71+- CD146+- CD147+ CD



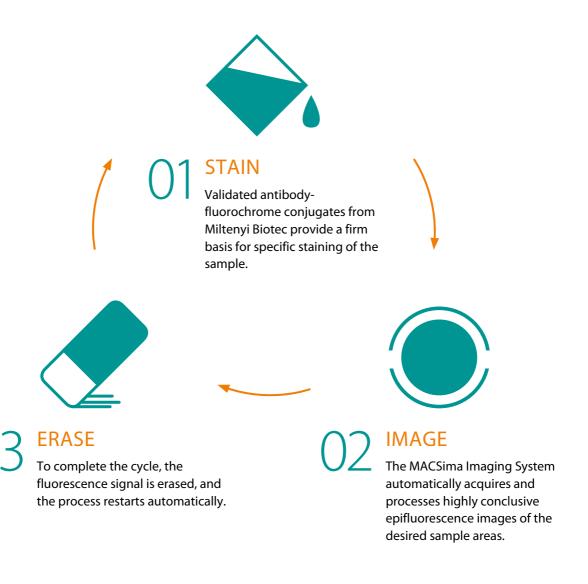


MICS technology – the basic principle

Miltenyi Biotec's MICS technology enables the simultaneous analysis of hundreds of markers on a single sample based on fluorescence microscopy. It uses the principle of iterative staining with different fluorochrome-conjugated antibodies to acquire microscopy data for a multitude of parameters without harming the sample. The iterative process comprises three main steps: fluorescent staining, image acquisition, and erasure of the fluorescence

signal, all of which are conducted by the MACSima™ Imaging System in a fully automated manner.

The resulting stack of potentially hundreds of marker images provides an unprecedented insight into the physiological or pathological characteristics of the sample. Due to on-the-fly processing, data analysis can start at any time, even when the iterative process is still running.



Simplicity demystifies complexity

With its automated processes and optimized components, the MACSima™ Imaging Platform reduces the effort for the generation of complex

high-content microscopy data to the basic essentials. The benefits speak for themselves.

Hundreds of markers on one sample

Obtain high-content microscopy data to analyze hundreds of proteins and other antigens on a single sample.

Automated sample processing and imaging

Plan your experiment and leave the execution to the MACSima Imaging System.

Analyze any kind of fixed sample

Image all types of fixed samples, whether tissue or single cells, and perform any analysis imaginable.

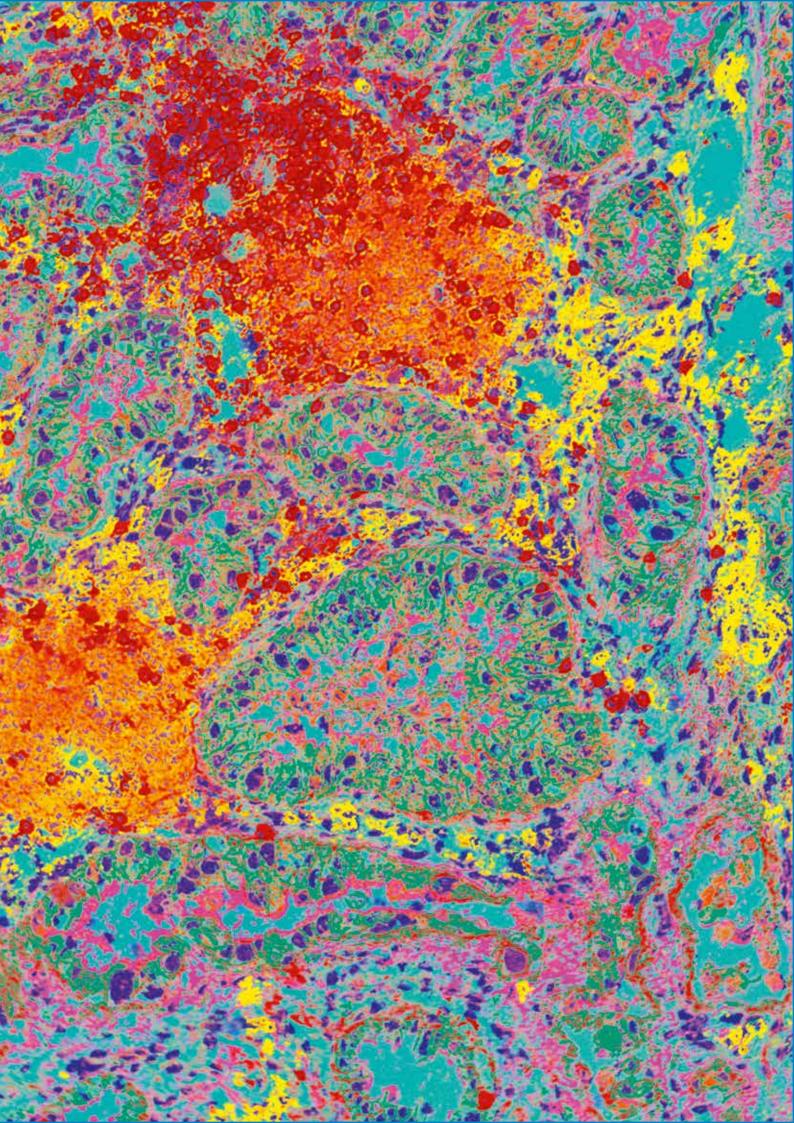
Complete imaging system

Count on the MACSima Imaging Platform which provides a complete, harmonized system including instrument, optimized sample carriers, validated antibodies, and intuitive software that allows on-the-fly data processing.

Effortless experiment preparation

Benefit from ready-to-use reagent plates containing pre-defined sets of antibodies from Miltenyi Biotec for an effortless yet comprehensive analysis.





The easy way to ultrahigh-content microscopy data

Setup experiment

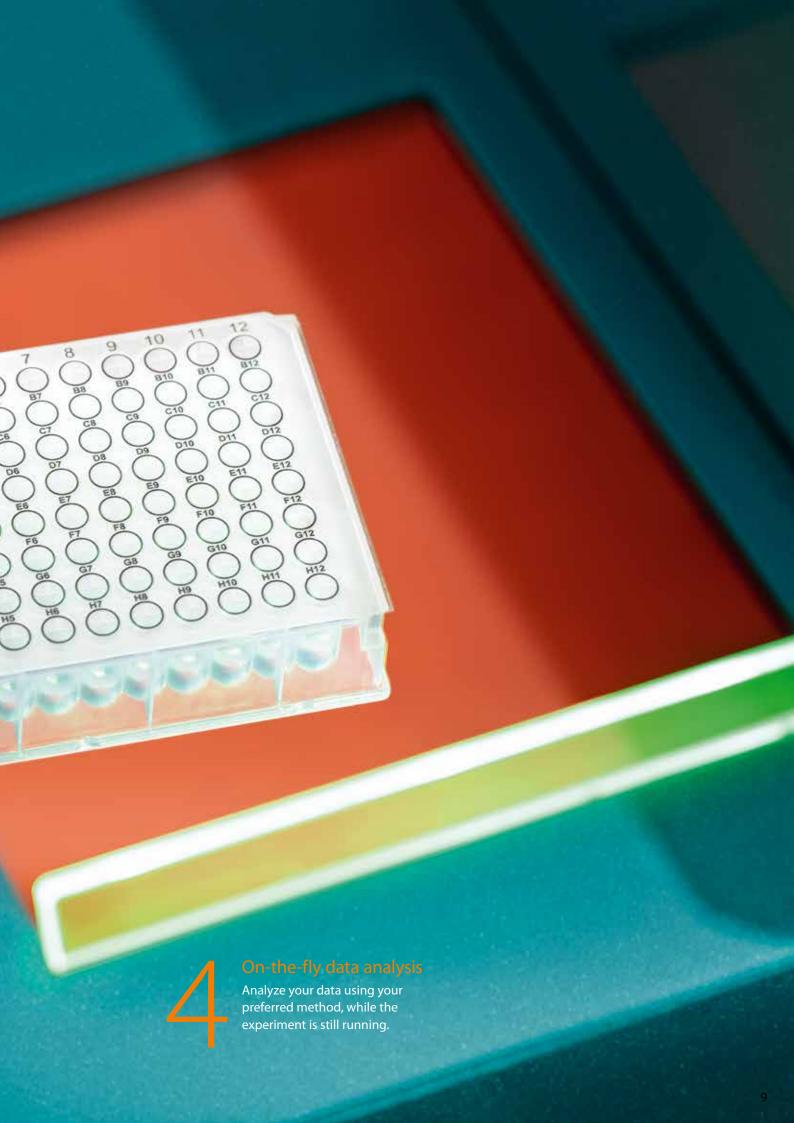
Prepare your sample and antibody panel taking your preferred fixation method into account.

Experiment start

Place your sample and reagent plate in the MACSima™ Imaging System, define your preferred imaging area, and start the experiment.

Fully automated process
The MACSima Imaging System pe

The MACSima Imaging System performs the iterative staining, imaging, and erasing process without further input or handling by the operator.



Unlimited options

Analyze any kind of fixed sample

To answer complex scientific questions one cannot afford to be restricted by technical limitations. To give you complete flexibility in the type of fixed sample you want to analyze with the MACSima™ Imaging System, we have developed the MACSwell Sample Carriers.

To support either tissue, adherent or suspension cells,

we have designed three different types of carriers, MACSwell One, Two, and Four Imaging Frames, MACSwell 24 Imaging Plates, and MACSwell Micro Slides. Each of them contains a well-defined reaction cavity to perform iterative staining easily and safely and give you the ease of failure-free experimentation.



- Formalin-fixed paraffin-embedded (FFPE)
- ✓ Paraformaldehyde (PFA)
- Acetone

TISSUE

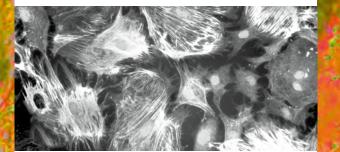


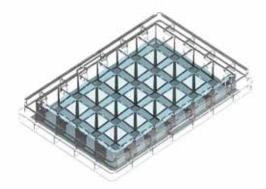


MACSwell One, Two, and Four Imaging Frames

can be mounted around any standard microscope slide and provide the reaction cavity needed for a MACSima Imaging experiment. MACSwell Imaging Frames are provided in three different sizes of reaction cavities to perfectly fit the size of your tissue sample.

ADHERENT CELLS

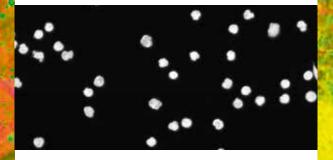


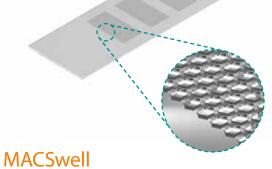


MACSwell 24 Imaging Plates

are pre-assembled and contain 24 rectangular wells with a clear 170 μm thick cover glass bottom. Simply pipet your cell suspension onto the plate, culture as usual and fix the sample according to your standard protocol directly on the plate.

SUSPENSION CELLS





MACSwell Micro Slides

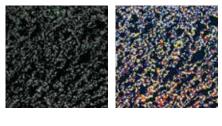
contain over 1.5 million hexagonal microcavities with a diameter of 20 μ m and are therefore perfectly sized to fit exactly one cell. Simply mount your choice of MACSwell Imaging Frame around the slide and pipet your suspension cells in the resulting reaction cavity.

One experiment. Multiple readouts.

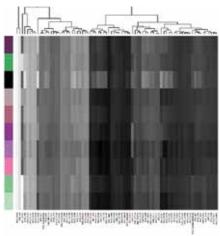
Glioblastoma multiforme is a highly malignant, thus far incurable type of brain tumor. To identify novel types of therapy, classification schemes have been developed that allow the categorization of several distinct subtypes of the disease. The high-content

data generated by the MACSima™ Imaging Platform is perfectly suited to apply these classification schemes and subsequently identify novel glioblastoma-specific markers that might be promising for future CART cell-based therapies.







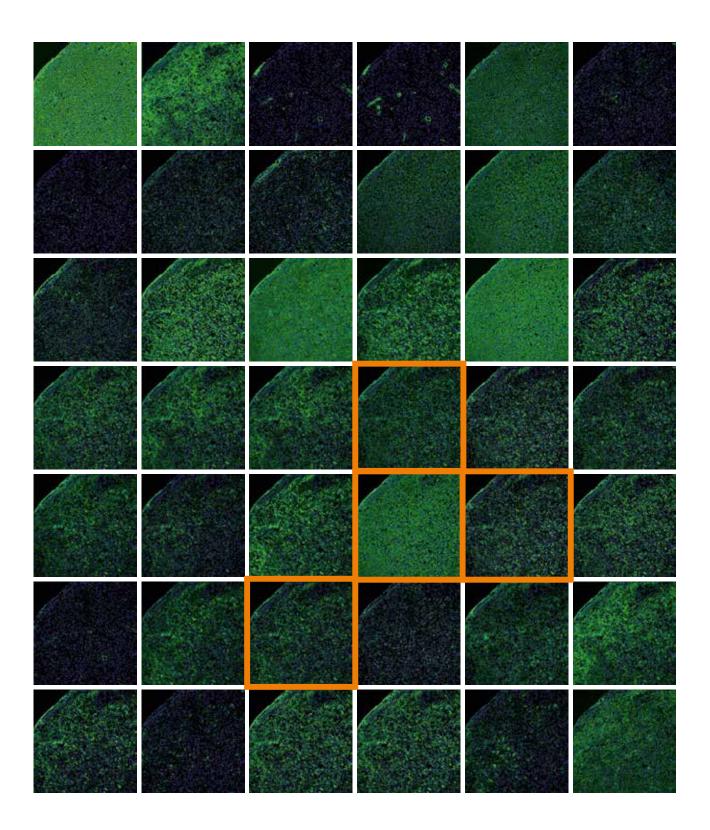


O3 CORRELATION

04 CLUSTERING

Deep phenotyping of glioblastoma subtypes

2D-image stacks were obtained by the MACSima Imaging System and analyzed in a pixel- and segmentation-based approach. The resulting expression profile and pattern recognition data were used for in-depth classification of different glioblastoma specimen.



Identification of novel glioblastoma marker candidates

Selection of immunofluorescence images of 96 antibodies that were used with the MACSima Imaging System. Clustering and correlation analysis were performed to identify novel glioblastoma-specific markers. For more information visit www.miltenyibiotec.com/MACSima

Integrated system allows for effortless and reliable processing

MICS technology is based on the well-established and straightforward technique of immunofluorescence staining. However, to translate this technique into comprehensive and effortless multiparameter imaging a functional, well-orchestrated system is a prerequisite. The MACSima™ Imaging Platform relies on four essential pillars that are combined seamlessly to ensure the easy generation of conclusive high-content microscopy data.

MACSima™ IMAGING SYSTEM

At the heart of the system is the MACSima™ Imaging System, a benchtop device that performs the iterative staining cycle and data processing in a fully automated fashion.

MILTENYI BIOTEC ANTIBODIES

A broad and continuously growing portfolio of validated antibodies from Miltenyi Biotec, including REAfinity™ Recombinant Antibodies, ensures specific staining, and thus the reliable analysis of hundreds of parameters.





All you need for your ultrahigh-content experiment

MACSima™ Instrument specifications		
Instrument type	Epifluorescence microscope	
Camera	Latest sCMOS technology with high-resolution, thermoelectrically cooled sensor	
Filters	5 different emission filters	
Autofocus	Dual approach of hardware and image-based autofocus mechanism	
Illumination	6 different excitation LEDs	
Objectives	Set of 3 lenses from overview to resolution, optimized for all supported sample carriers	
Liquid handling	 Washable stainless-steel needle with minimized spill-over Syringe pump drive for accurate volumetric staining of samples 	
Automation	 Fully automated liquid handling Fluorescent staining, image acquisition, and erasing 1.5 L fluid containers for automatic operation over several days without the need to exchange containers 	
Stage	100 nm resolutionConvenient loading and clamping of all supported sample carriers	
Sample carriers	Miltenyi Biotec's MACSwell Sample Carriers for slide-based and plate-based sample handling	
Computer	 Integrated redundant storage for multiple experiments Tiltable integrated touch display (Full HD) Trackball keyboard 	
Operating conditions	Non-condensing humidity, air-conditioned laboratory	
Software	Software package allowing for experiment planning, execution and analysis of high dimensional data sets	
Weight	~165 kg	
Dimensions	1,210 mm × 750 mm × 650 mm (L×W×H)	

Product type Product type	Order no.
Instrument	
MACSima Instrument	130-121-164
Antibodies	
REAscreen, MAX, human, FFPE, version 01	130-124-376
REAscreen, MAX, human, PFA, version 01	130-124-377
REAscreen, MAX, human, Acetone, version 01	130-124-378
REAscreen, MAX, mouse, PFA, version 01	130-124-379
REAscreen, MAX, mouse, Acetone, version 01	130-124-380
REAfinity® Recombinant Antibodies	*
REAlease® Releasable Antibodies	*
MACS® Antibodies	*
Sample carriers	
MACSwell One Imaging Frame	130-124-673
MACSwell Two Imaging Frame	130-124-675
MACSwell Four Imaging Frame	130-124-676
MACSwell 24 Imaging Plate	130-124-677
MACSwell Micro Slide	130-124-678
Buffers	
MACSima Running Buffer 1×1.5 L	130-121-564
MACSima Running Buffer 6×1.5 L	130-121-565
MACSima System Buffer 1×1.5 L	130-122-183
MACSQuant®/MACSima Storage Solution 6×1.5 L	130-092-748

 $^{{}^* \, \}text{For details on the wide range of Miltenyi Biotec antibodies, visit} \, \textbf{www.miltenyibiotec.com/antibodies}$

Miltenyi Biotec provides a wealth of antibodies validated for imaging, including REAfinity $^{\text{\tiny{M}}}$ Recombinant Antibodies and REAlease $^{\text{\tiny{R}}}$ Releasable Antibodies, which ensure specific staining and highly reproducible results.

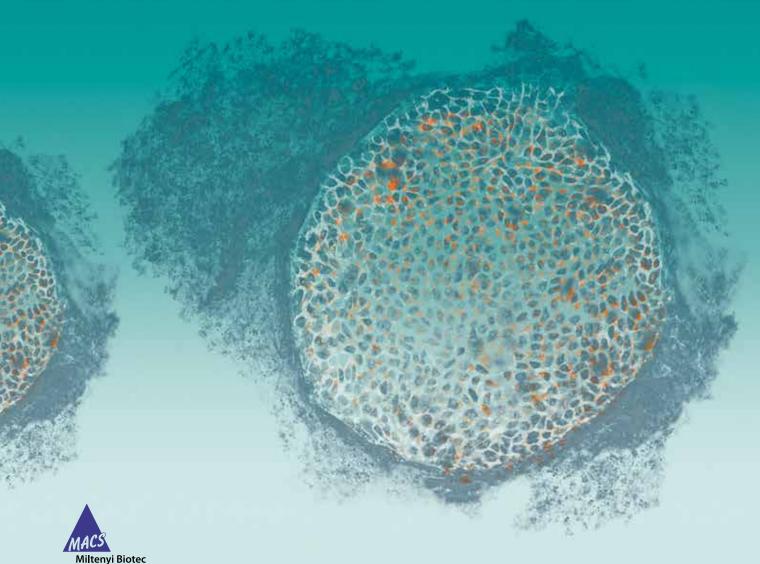
For more information visit www.miltenyibiotec.com/antibodies







miltenyibiotec.com



Germany/Austria Miltenyi Biotec GmbH Friedrich-Ebert-Straße 68 51429 Bergisch Gladbach Germany Phone +49 2204 8306-0 Fax +49 2204 85197 macs@miltenyibiotec.de

USA/Canada

Miltenyi Biotec Inc. 2303 Lindbergh Street Auburn, CA 95602, USA Phone 800 FOR MACS Phone +1 530 888 8871 Fax +1877 591 1060 macs@miltenyibiotec.com

Australia

Miltenyi Biotec Australia Pty. Ltd. Unit 16A, 2 Eden Park Drive Macquarie Park NSW 2113 Australia Phone +61 2 8877 7400 Fax +61 2 9889 5044 macs@miltenyibiotec.com.au

Benelux

Miltenyi Biotec B.V. Sandifortdreef 17 2333 ZZ Leiden The Netherlands macs@miltenyibiotec.nl Customer service

The Netherlands Phone 0800 4020120

Fax 0800 4020100 **Customer service Belgium** Phone 0800 94016 Fax 0800 99626

Customer service Luxembourg Phone 800 24971 Fax 800 24984

China

Miltenyi Biotec Technology & Trading (Shanghai) Co., Ltd. Rooms 2303 and 2309 No. 319, Xianxia Road Changning District 200051 Shanghai, P.R. China Phone +86 21 62351005 Fax +86 21 62350953 macs@miltenyibiotec.com.cn

Miltenyi Biotec SAS 10 rue Mercoeur 75011 Paris, France Phone +33 1 56 98 16 16 Fax +33 1 56 98 16 17 macs@miltenyibiotec.fr

Italy

Miltenyi Biotec S.r.l. Via Paolo Nanni Costa, 30 40133 Bologna Italy Phone +39 051 6 460 411 Fax +39 051 6 460 499 macs@miltenyibiotec.it

Miltenyi Biotec K.K. Nittsu-Eitai Building 5F 16-10 Fuyuki, Koto-ku, Tokyo 135-0041, Japan Phone +81 3 5646 8910 Fax +81 3 5646 8911 macs@miltenyibiotec.jp

Nordics and Baltics

Miltenyi Biotec Norden AB Scheelevägen 17 223 70 Lund Sweden macs@miltenyibiotec.se Customer service Sweden Phone 0200-111 800

Fax 046-280 72 99 Customer service Denmark Phone 80 20 30 10 Fax +46 46 280 72 99

Customer service Norway, Finland, Iceland, and Baltic countries Phone +46 46 280 72 80 Fax +46 46 280 72 99

Singapore

Miltenyi Biotec Asia Pacific Pte Ltd. 100 Beach Road #28-06 to 28-08 Shaw Tower Singapore 189702 Phone +65 6238 8183 Fax +65 6238 0302 macs@miltenyibiotec.com.sg

South Korea

Miltenyi Biotec Korea Co., Ltd Arigi Bldg. 8F 562 Nonhyeon-ro Gangnam-gu Seoul 06136, South Korea Phone +82 2 555 1988 Fax +82 2 555 8890 macs@miltenyibiotec.co.kr

Spain

Miltenyi Biotec S.L. C/Luis Buñuel 2 Ciudad de la Imagen 28223 Pozuelo de Alarcón (Madrid) Spain Phone +34 91 512 12 90 Fax +34 91 512 12 91 macs@miltenyibiotec.es

Switzerland

Miltenyi Biotec Swiss AG Gibelinstrasse 27 4500 Solothurn Switzerland Phone +41 32 623 08 47 Fax +49 2204 85197 macs@miltenyibiotec.ch

United Kingdom

Miltenyi Biotec Ltd. Almac House, Church Lane Bisley, Surrey GU24 9DR, UK Phone +44 1483 799 800 Fax +44 1483 799 811 macs@miltenyibiotec.co.uk

www.miltenyibiotec.com

Miltenyi Biotec provides products and services worldwide. Visit www.miltenyibiotec.com/local to find your nearest Miltenyi Biotec contact.