

We are pleased to share with you the first edition of our international newsletter, based on a new concept. Below, you can find key updates, from our core product categories: **Cell Control Arrays** and **Immunohistochemistry**.

Cell Control Arrays (CCA)

Suitable for immunohistology, *in situ* hybridization and special stainings

Cell Control Arrays (CCA) are designed for the qualitative control of immunohistochemical (IHC) staining, *in-situ*-hybridization (ISH) and special staining. In addition to entire FFPE blocks, in which cell and tissue punches are homogeneously melted with the surrounding paraffin using a patented method, Zytomed Systems also offers individual slide sets. Control blocks can be cut according to standard procedures and mounted on coated slides. A core of myocardial tissue serves as an easy orientation

during mounting and microscopy. Each block is around 5 mm high and allows a yield of around 150 sections. Depending on usage and FFPE block, up to 350 sections are possible. The small size of the slices enables simultaneous mounting of patient sample and control material on the same slide (On-slide control).

Zytomed offers the following arrays:

- ▶ The **Cell Control Array ALK (IHC)** contains one core of ALK positive cells and one core of ALK negative cells. It is suitable as qualitative control for immunohistochemical stains on ALK-positive tissues. RNA can be extracted and gene fusions as e. g. EML4-ALK (E13:A20) can be detected using RT-PCR.
- ▶ The **Cell Control Array Receptor** contains 4 cores of breast carcinoma cell lines. They show different expression levels of estrogen receptor (ER), progesterone receptor (PR) and HER2 (ERBB2). Thus, a differentiation between high and low staining intensity is possible, indicating the sensitivity of your stains. The system is suitable for immunohistochemistry and *in-situ*-hybridization.
- ▶ The **Cell Control Array Virus** is designed for the qualitative control of immunohistochemical staining and *in-situ*-hybridization of virus infected tissue. The paraffin block contains cell line cores of CMV, HSV type 1 and type 2, EBV and Polyomavirus/SV40 infected cell lines.
- ▶ The **Cell Control Array Bacteria plus Fungi** contains 4 cores of different germs and fungi. These are *Mycobacterium bovis*, gram-positive and gram-negative bacteria and *Aspergillus fumigatus*. Antibodies against *Mycobacterium tuberculosis* react positive on the core of *Mycobacterium bovis*. DNA can be extracted and used as positive control for the detection of mycobacterium using PCR.
- ▶ The **Cell Control Array ROS1 (IHC)** includes two ROS1-positive cell lines showing weak and medium ROS1 expression, one ROS1-negative cell line, and one core of heart muscle tissue. RNA can be extracted and be used for detection of CD74-ROS1-fusion mRNA via RT-PCR.

▶ CCA FFPE blocks and Cell Control Slides sets

Cell Control Arrays (homogenous paraffin blocks)

Product	Description	Amount	Status	Order no.
Cell Control Array ALK (IHC)	1 core of ALK positive cell line + 1 core of ALK negative cell line	1 Block	RUO	MB-CC ALK
Cell Control Array Bacteria plus Fungi	3 cores of different bacteria + 1 core of fungi	1 Block	RUO	MB-CC BAC
Cell Control Array Receptor	4 cores with different expression levels of ER, PR and HER2	1 Block	RUO	MB-CC REZ
Cell Control Array ROS1 (IHC)	3 cores with different expression of ROS1	1 Block	RUO	MB-CC ROS1
Cell control Array Virus	5 cores of virus-infected cell lines	1 Block	RUO	MB-CC VIR

Cell Control Slides sets (precut sections that are mounted on coated slides and already baked)

Product	Description	Amount	Status	Order no.
Cell Control Slides HPV	3 cores of different HPV infected cell lines + 1 core of a HPV negative cell line	5 Slides	RUO	MB-CC HPV-S

Custom made

We offer the preparation of tissue arrays from your own laboratory tissues or cell lines for control and research purposes.

▶ Selection of corresponding Zytomed System antibodies

Description	Clone	Host	Dilution	Amount	Status	Order No.
ALK (p80)	5A4	Mouse	1:100 - 1:200	0.5 ml	CE/IVD	MSK096-05
Rabbit anti-Estrogen Receptor	SP1	Rabbit	Ready-to-use	16 ml	CE/IVD	BRB053
			1:200	1 ml	RUO	RBK018
				0.5 ml		RBK018-05
Rabbit anti-HER2 (c-erbB-2)	SP3	Rabbit	Ready-to-use	6 ml	CE/IVD	RBG026
			1:100 - 1:200	1 ml		RBK026
				0.5 ml		RBK026-05
Rabbit anti-Progesterone Receptor	SP42	Rabbit	Ready-to-use	6 ml	CE/IVD	BRB038
			1:200 - 1:400	1 ml		RBK020
				0.5 ml		RBK020-05
ROS1	EPMGHR2	Rabbit	Ready-to-use	6 ml	RUO	RBG071
			1:100	0.5 ml		RBK071-05
				0.1 ml		RBK071-01
Cytomegalovirus (CMV Cocktail)	DDG9 + CCH2	Mouse	Ready-to-use	6 ml	RUO	MSG121
			1:10 - 1:25	0.5 ml		MSK121-05

Abbreviations

CE/IVD: for in vitro diagnostic use; RUO: research use only

! Note that the use of a non CE/IVD-labeled reagent will result in a LDT. This must be validated by the user to meet the regulatory requirements of the In-vitro Diagnostics Regulation (EU) 2017/746 (IVDR).



CAL10: monoclonal rabbit antibody against PD-L1

PD-L1 (Programmed Cell Death Ligand 1) serves as a biomarker for the identification of patients who are likely to benefit from immune checkpoint therapy with PD1 or PD-L1 antibodies. Immunohistochemically, PD-L1 is detected either with diagnostic companion kits (*Companion Diagnostics*) or with in-house developed methods using freely available antibodies (*Laboratory Developed Test* or *LDT*). In the USA, the fact that a single target molecule is to be detected with 4 or more different methods (and possibly on different immunostaining machines) as part of companion or complementary diagnostics, depending on the therapy in question, presents pathology with major, previously unknown challenges, according to several experts [1,2].

In Germany, pathologists have a free choice of method. However, it is required to „use an appropriately validated and robust method to minimize false negative or false positive determinations“ [3]. Harmonization studies aim to simplify PD-L1 testing and clinical decision making [4,8] and „in princi-


ple allow the use of all approved anti-PD1/PD-L1 therapies in a patient, regardless of the PD-L1 assay used“ [5].


In addition to the PD-L1 kit systems investigated in larger comparative studies, several non-kit-based antibodies are available [6]. The costs of the immunohistochemical PD-L1 test with „free“ antibodies are 3 to 5 times lower than the costs of the test with a diagnostic companion kit [5]. With the CAL10 clone, Zytomed Systems offers a free, CE/IVD-classified rabbit monoclonal antibody that is not a companion diagnostic. Validation of the product's companion diagnostic performance is the responsibility of the user.

▶ suggested protocol

Dewaxing: Conventional via xylene and the descending alcohol series
HIER: Steam pressure, 7 Min. / 110 °C, HIER Citrate Buffer pH 6,0*
PD-L1 Antibody: Clone CAL10, diluted in Antibody Diluent C**

Detection: HRP-Polymer & DAB High Contrast*

* Label: Zytomed Systems,  ZytoVision


** Label: Zytomed Systems,  Zytomed System

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▶ Gene information and terminology

Gene	Description	Synonyms	Gen-ID	Gen-Lokalisation
PDCD1	Programmed Cell Death 1	CD279, hSLE1, PD-1, PD1, SLEB2	MIM600244	2q37.3
CD274	CD274 Molecule	B7-H, B7-H1, B7H1, PD-L1, PDL1, PDCD1LG1	MIM605402	9p24.1
PDCD1LG2	Programmed Cell Death 1 Ligand 2	B7-DC, bA574F11.2, Btdc, CD273, PD-L2, PDL2	MIM605723	9p24.1

▶ Antibody against PD-L1

Description	Amount	Format	Dilution	Status	Order No.
PD-L1 (CD274)  Zytomed Systems GmbH Clone: CAL10 Host: Rabbit	6 ml	Ready-to-use	-	CE/IVD	RBG063
	0,5 ml	Concentrate	1:100 - 1:200		RBK063-05

▶ Literature

- [1] Kerr KM, Nicolson MC. Non-Small Cell Lung Cancer, PD-L1, and the Pathologist. Arch Pathol Lab Med 140:249-254, 2016
- [2] Kerr KM and Hirsch FR. Programmed Death Ligand-1 Immunohistochemistry: Friend or Foe? Arch Pathol Lab Med 140:325-331, 2016
- [3] Fachinformation KEYTRUDA® 50 mg, MSD SHARP & DOHME GMBH, Juli 2016
- [4] Scheel AH *et al.* Harmonized PD-L1 immunohistochemistry for pulmonary squamous-cell and adenocarcinomas. Mod Pathol 29:1165-1172, 2016
- [5] Harlt S. Das Dilemma der PD-L1-Testung – eine Herausforderung an die Pathologen. krebs.hilfe! 11:2-7, 2016
- [6] Hutarew G. PD-L1 testing, fit for routine evaluation? From a pathologist's point of view. memo 9:201-206, 2016
- [7] Karnik T, Kimler BF, Fan F, Tawfik O. PD-L1 in Breast Cancer: Comparative Analysis of Three Different Antibodies (Poster Session II/13, USCAP-Meeting März 2017, San Antonio, Texas)
- [8] Tsao MS *et al.* PD-L1 Immunohistochemistry Comparability Study in Real-Life Clinical Samples: Results of Blueprint Phase 2 Project. J Thorac Oncol 13:1302-1311, 2018



Highlights

We are thrilled to share an update of our International Distribution Meeting held in Frankfurt, co-organized with our sister company ZytoVision GmbH.

This event marked a significant milestone in our journey, bringing together our valued distributors from around the world. It was a dynamic and collaborative event with plenty engaging discussions, insightful presentations, and networking sessions. We used this opportunity to strengthen our relationships, foster collaboration, and our partnership.

Moving forward, we are more motivated than ever to continue this remarkable journey together, further strengthening our partnerships, and exploring new avenues for international growth.

To all our distributors, thank you for your unwavering support, commitment, and dedication. We are excited to shape the future together!



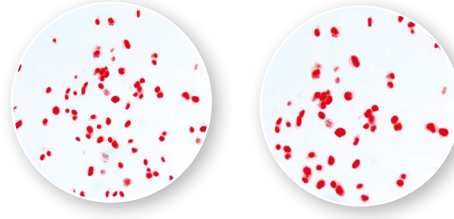
For questions or requests regarding our full product portfolio, write an email to international@zytomed-systems.de

CCA features:

- ▶ Suitable as on-slide controls next to test samples.
- ▶ No loss of cell cores. Homogenous paraffin block ensures integrity of the cell cores after cutting.
- ▶ Dyed paraffin and myocardial tissue core ensure easy handling and orientation.
- ▶ Consistent high quality.

CCA applications:

- ▶ New marker establishment
- ▶ Assay optimization
- ▶ Protocol validation
- ▶ Routine IHC runs



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” Kerr & Nicolson, 2016
“The biology of PD-1/PD-L1 is complex, the clinical data for these drugs show considerable variation, the selection performance of the PD-L1 biomarker test is not perfect, and the existence of 4 drug/test combinations adds significantly to the problems faced”.