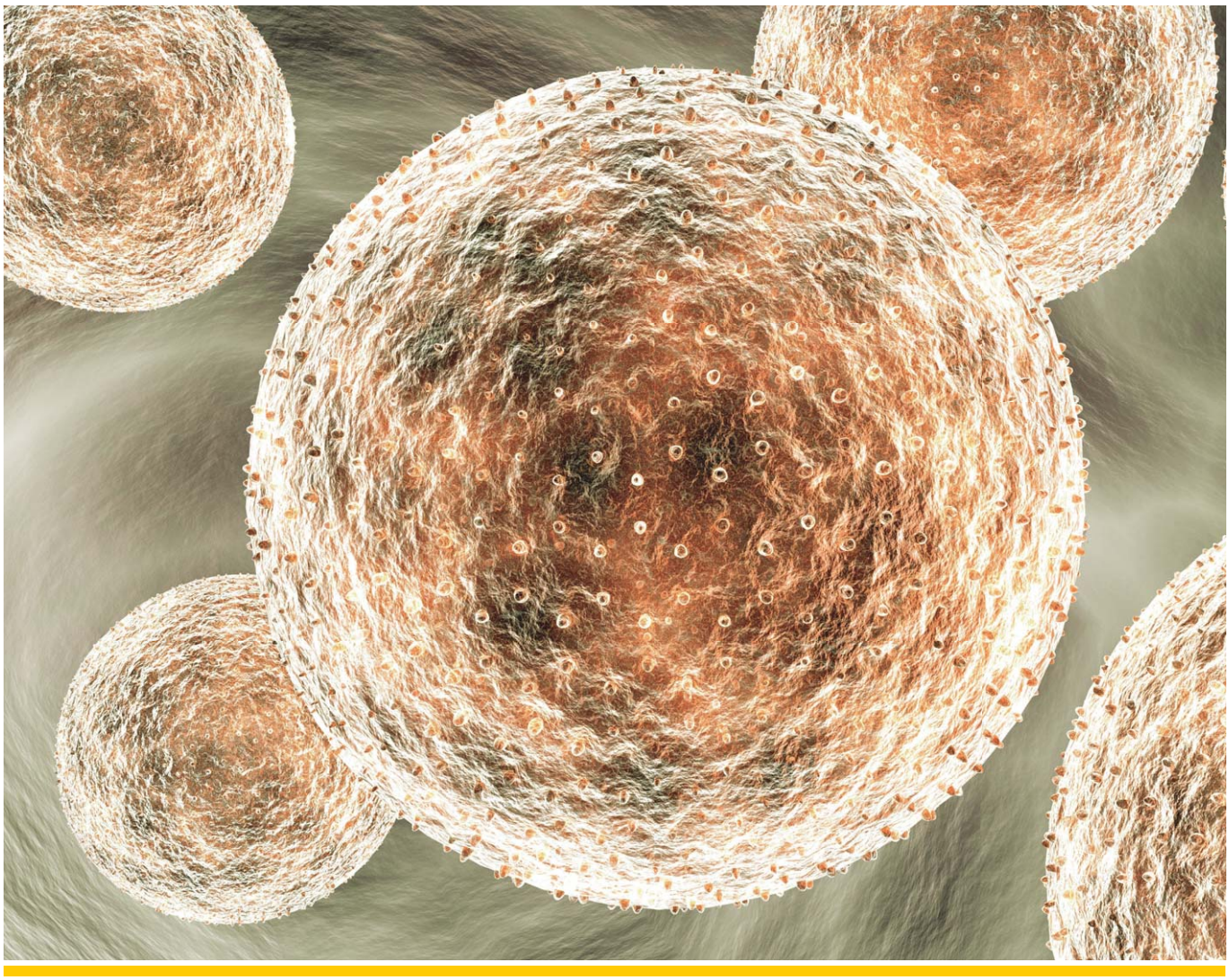
EUROIMMUN Medizinische Labordiagnostika AG

Chlamydia infections

Comprehensive diagnostics with EUROIMMUN test systems



- Detection of infections with *C. trachomatis, C. pneumoniae* and *C. psittaci*
- A broad range of products for direct detection (PCR microarray) and serology (microimmunofluorescence, immunoblot and ELISA)
- EUROIMMUN automation solutions for all test systems

Chlamydia

Chlamydia are the smallest gram-negative, obligatory intracellular bacteria. They show a complex parasitic reproduction cycle (see info box). The three human pathogenic *Chlamydia* species encompass *Chlamydia trachomatis*, *Chlamydia pneumoniae* and *Chlamydia psittaci*. So far, there is no vaccine available against *Chlamydia*. However, infections can be well treated with antibiotics.

C. trachomatis

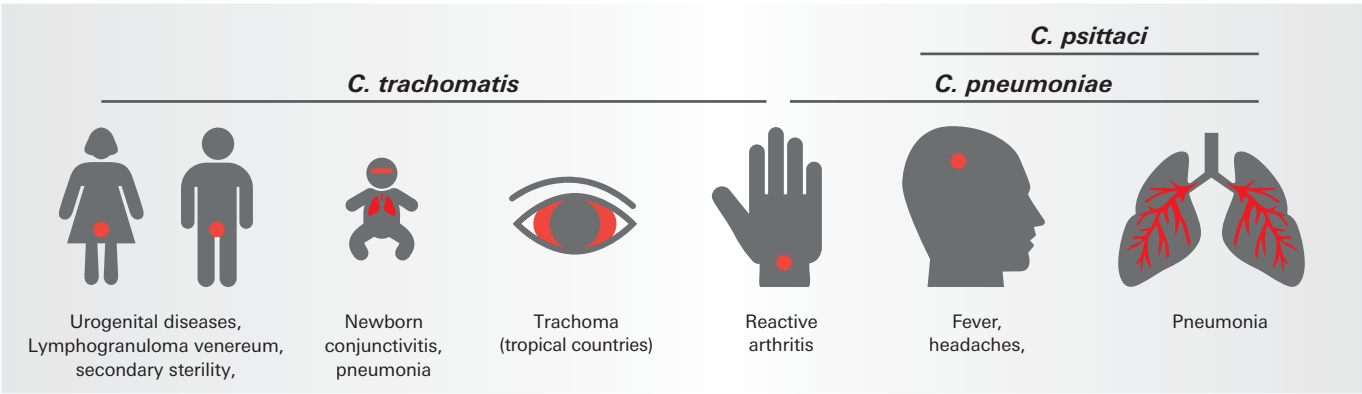
The bacterium belongs to the worldwide most frequent pathogenic agents of sexually transmitted infections. Infection can cause Lymphogranuloma venereum (serotypes L1-3), trachoma (serotypes A-C, tropical regions) and urogenital diseases (serotypes D-K) – women typically develop cervicitis, men urethritis. However, infections generally proceed asymptomatically. Chronic infections are frequently associated with secondary sterility and reactive arthritis. Babies of infected mothers are especially at risk since the pathogen is transmitted during birth in approximately 70% of cases. Usually the newborn develops conjunctivitis, less frequently pneumonia or otitis media.

C. pneumoniae

The pathogen is transmitted by aerosols and causes infections of the upper respiratory tract and pneumonia. 50% of courses are asymptomatic. In manifest cases, patients complain about non-productive cough, headaches and fever. The infection rate in adults amounts to 50 to 70%. Some chronic diseases are also associated with *C. pneumoniae* infections, especially reactive arthritis, which, as opposed to rheumatoid arthritis, may be treated by antibiotics.

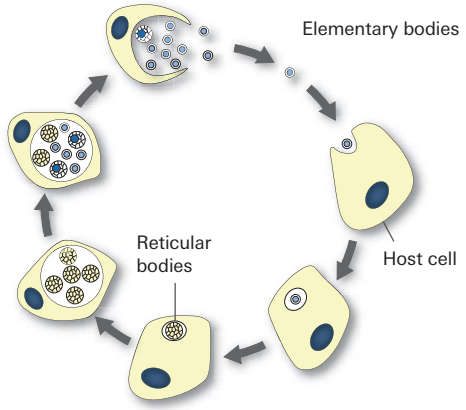
C. psittaci

The pathogenic agent of psittacosis is transmitted to humans via the secretions and excrements of infected cage or breeding birds. Mainly pet owners, traders and people working in poultry farming are at risk. The clinical symptoms encompass mostly flu-like symptoms. Over the course of the disease, life-threatening pneumonia can develop, which is often associated with organ manifestations (esp. liver, kidneys, brain, heart).



Chlamydia development cycle

During their reproduction cycle *Chlamydia* take two different life forms: They present as an extracellular, germ-like elementary body and as an intracellular, reproductive reticular body. First, elementary bodies are taken up by their host cells through endocytosis, e.g. epithelial cells of the urogenital or respiratory tract. Within the endosomes, they develop into metabolic reticular bodies and reproduce depending on the energy balance of their hosts and under inhibition of the host's immune reaction. The reticular bodies convert back into elementary bodies after some divisions. The elementary bodies are released to infect further cells. Elementary bodies are also harnessed for application in *Chlamydia* diagnostics.



INFO

Chlamydia diagnostics

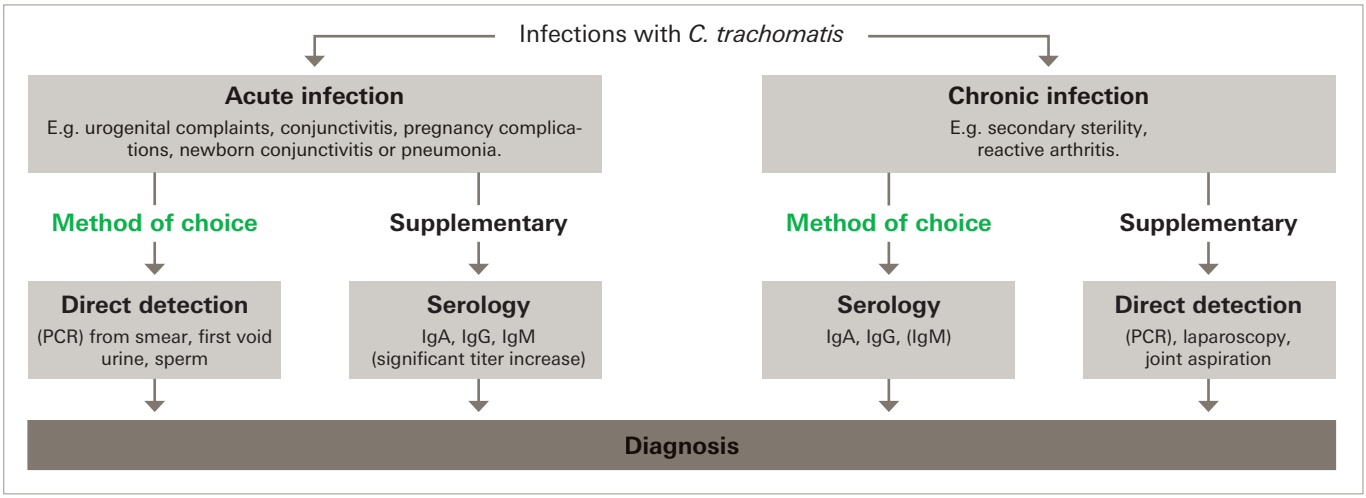
The selection of the best suitable diagnostic method for the detection of *Chlamydia* infections depends on the clinical image of the patient and the disease stage. PCR-based direct detection offers the highest sensitivity during the acute phase. Since antibodies are only detectable some days/weeks after the infection, serology plays an important part mainly in chronic diseases, for the retrospective determination of acute infections and in epidemiological studies.

	Direct detection	Serological detection		
	PCR	IgA	IgG	IgM
C. trachomatis	In acute infections	In acute* and chronic infections or reinfections	In acute*, past and chronic Infections or reinfections	Not recommended
C. pneumoniae	In acute infections	In acute*, past and chronic infections or reinfections	With acute*, past and chronic infections or reinfections	With primary infection
C. psittaci	Not commercially available	Relevance unclear	In acute* or past infections, observe anamnesis	In primary infection; Observe anamnesis

* Titer increase

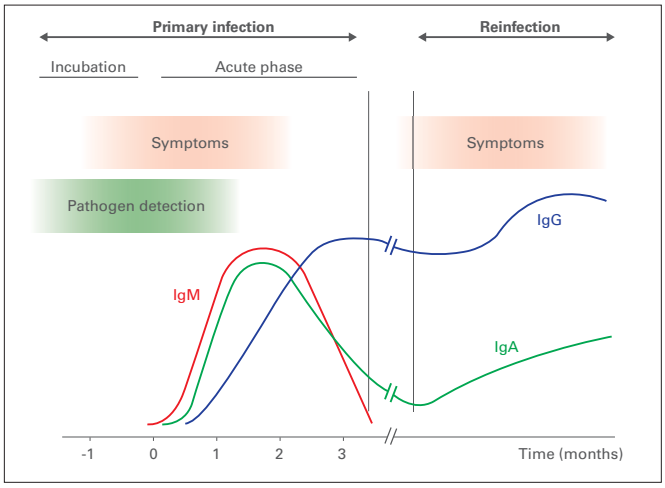
C. trachomatis

Direct pathogen detection in acute infections should be performed with PCR-based methods. With longer courses and suspected chronic diseases (e.g. reactive arthritis, tube sterility), detection of specific antibodies against *C. trachomatis* is recommended. Determination of antibodies against *C. trachomatis*-specific antigen MOMP (major outer membrane protein) enables delimitation from infections with *C. psittaci* and *C. pneumoniae*.



C. pneumoniae

Since diagnosis of *C. pneumoniae* infections by means of symptoms or radiography is not entirely reliable, laboratory diagnostics play a significant role. Serology represents a useful supplement to direct detection. A positive IgM and/or IgA result together with a significant increase in the IgG titer of a follow-up sample taken after an interval of at least two weeks indicate an acute infection. Reinfections are characterised by an increase in the IgA and/or IgG titer in the absence of an IgM response.



Titer course using the example of a *C. pneumoniae* infection

C. psittaci

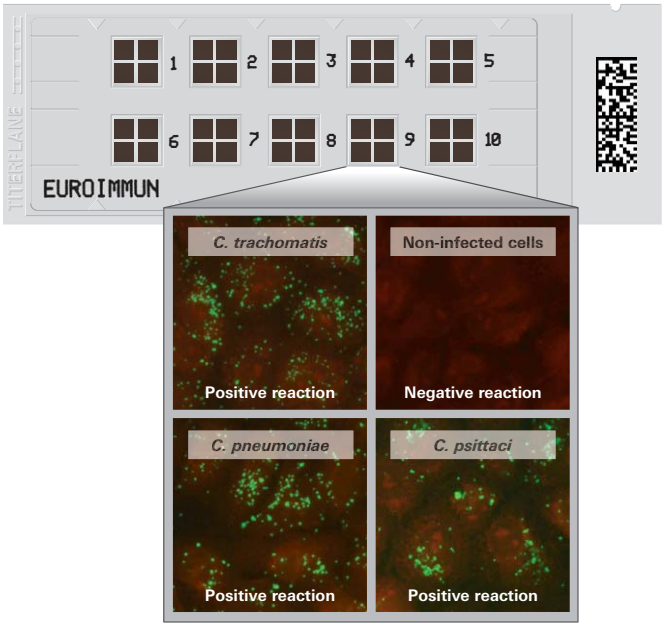
Direct detection (e.g. from sputum) is only performed in a safety laboratory (L3) due to the high contagiosity. The finding is mainly based on the anamnesis (cf. risk group) and can be supported by serology. Here, the species-specific microimmunofluorescence assay (MIF) is especially well suited.

Test systems for the diagnosis of *Chlamydia* infections of all three human pathogenic (HP) species

Anti-Chlamydia MIF*, BIOCHIP Mosaic (IgA, IgG, IgM)

- Parallel determination of antibodies against all three *Chlamydia* species in one incubation.
- This method uses purified elementary bodies of the species *C. trachomatis*, *C. pneumoniae* and *C. psittaci* as the antigen.
- Differentiation between unspecific and specific fluorescence by means of uninfected cells
- Clear reduction of cross reactivity by inactivation of the LPS antigen (LPS: lipopolysaccharide)
- Simplified finding of the focusing plane at the microscope with cell-based substrates as opposed to the yolk sac matrix.

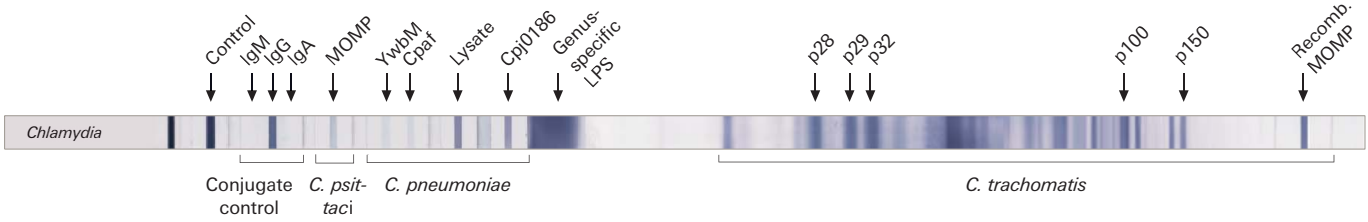
* MIF: Microimmunofluorescence assay



EUROIMMUN Anti-Chlamydia MIF		n	Correlation with target values from the QA institute **
C. trachomatis	IgA	35	94 %
	IgG	35	100 %
	IgM	30	100 %
C. pneumoniae	IgA	36	97 %
	IgG	36	100 %
	IgM	28	100 %

** INSTAND e. V., Germany; for *C. psittaci* there is no quality assessment scheme available yet; borderline samples excluded.

Anti-Chlamydia HP EUROLINE-WB (IgA, IgG)



- Multiparameter analysis for sensitive and specific detection of infections with *C. trachomatis, C. pneumoniae* and *C. psittaci*.
- Electrophoretically separated extract from *C. trachomatis* and selected individual antigens (on membrane chips).
- Unique combination of native and recombinant antigens: Species-specific MOMP (esp. for *C. trachomatis*), YwbM and Cpaf (*C. pneumoniae*) and genus-specific LPS

	EUROIMMUN Anti-Chlamydia HP EUROLINE-WB		Approved blot test			Correlation***
			positive	borderline	negative	
C. trachomatis	IgA (n = 94)	positive	10	5	2	97 %
		borderline	4	6	2	
		negative	0	4	61	
	IgG (n = 100)	positive	42	4	3	95 %
		borderline	4	2	1	
		negative	1	8	35	

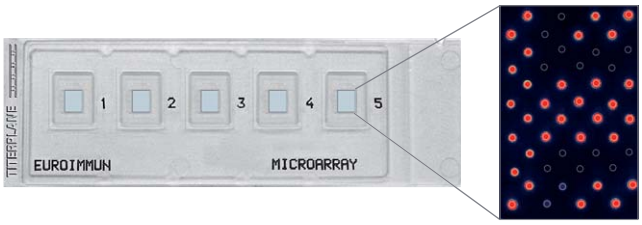
	EUROIMMUN Anti-Chlamydia HP EUROLINE-WB		Approved blot test			Correlation***
			positive	borderline	negative	
C. pneumoniae	IgA (n = 91)	positive	9	7	5	91 %
		borderline	0	0	0	
		negative	2	9	59	
	IgG (n = 92)	positive	56	10	1	94 %
		borderline	0	0	0	
		negative	3	10	12	

*** Borderline samples excluded

Test systems for the diagnosis of *C. trachomatis* infections

EUROArray STI – PCR-based direct detection using microarray

- Parallel detection of up to 11 sexually transmitted pathogens (e.g. *Chlamydia trachomatis, Neisseria gonorrhoeae, Treponema pallidum* and HSV-1/2)
- Suitable for pathogens which cannot be cultured or are difficult to culture (e.g. *C. trachomatis*).
- Reliable detection of infections with low germination numbers by amplification of the pathogen DNA.
- Ready-to-use reagents, integrated controls
- Identification of multiple infections (also asymptomatic)



Sensitivity and specificity: Within the framework of an evaluation study, 325 smear samples and 134 urine samples were investigated for 11 different pathogenic agents using the EUROArray STI-11. A high agreement with the precharacterisation was achieved. In many cases, additional pathogens that were not included in the precharacterisation were detection. Their presence in the sample was confirmed by independent tests.

Pathogen	Smear samples				Urine samples			
	Precharacterisation		EUROArray STI-11		Precharacterisation		EUROArray STI-11	
	Positive	Negative	Sensitivity	Specificity	Positive	Negative	Sensitivity	Specificity
Chlamydia trachomatis	53	196	91-98%	99%	74	58	94-97%	100%
Neisseria gonorrhoeae	33	216	97%	99%	11	121	100%	100%
Herpes simplex virus 1	35	148	94%	98-99%	0	5	ND	100%
Herpes simplex virus 2	18	165	94-100%	100%	1	4	100%	100%
Haemophilus ducreyi	1*	0	100%	ND	1	0	100%	ND
Mycoplasma genitalium	10	141	100%	100%	2	89	50% [1]	100%
Mycoplasma hominis	22	131	100%	99%	12	79	92%	94%
Treponema pallidum	5	46	100%	100%	0	0	ND	ND
Trichomonas vaginalis	10	3	100%	100%	8	8	63% [2]	100%
Ureaplasma parvum	77	102	90%	99%	38	54	100%	97%
Ureaplasma urealyticum	19	135	84%	100%	9	82	100%	100%

ND: Not determined; * Synthetic sample with *H. ducreyi* type strain; [1] In a weak positive sample of a total of two samples, *M. genitalium* was not detected; [2] For the detection of *T. vaginalis*, the use of smear samples is recommended

Anti-Chlamydia trachomatis ELISA

- Recombinant MOMP as the antigen ensures highly specific detection of antibodies (IgA/G/M) against *C. trachomatis*.

Sensitivity and specificity: Clinically precharacterised (INSTAND e.V., Germany) were investigated using the EUROIMMUN Anti-Chlamydia trachomatis ELISAs (IgA, IgG and IgM). The tests showed a sensitivity of 100% at a specificity of 97.4% (IgA), 97% (IgG) and 100% (IgM) (borderline sera excluded).

EUROIMMUN Anti-C. trachomatis ELISA		Targets from QA institutes		
		positive	borderline	negative
IgA (n = 53)	positive	9	1	1
	borderline	1	4	0
	negative	0	0	37
IgG (n = 53)	positive	17	0	1
	borderline	0	0	2
	negative	0	1	32
IgM (n = 53)	positive	2	0	0
	borderline	0	1	1
	negative	0	0	49

Test systems for the diagnosis of *C. pneumoniae* infections

Anti-Chlamydia pneumoniae ELISA

- Sensitive determination of antibodies (IgA/G/M) against *C. pneumoniae*, based on purified cell lysate.

Sensitivity and specificity: Clinically and/or serologically precharacterised patient samples from quality assessment institutes (INSTAND e.V., Germany, and Labquality, Finland) were investigated with the EUROIMMUN Anti-Chlamydia pneumoniae ELISAs (IgA, IgG and IgM). The sensitivity amounted to 95.5% (IgA) and 100% (IgG, IgM) at a specificity of 96.8% (IgA), 97.5% (IgG) and 100% (IgM) (borderline sera excluded).

*Cross reactions with anti-*C. trachomatis* antibodies cannot be excluded.

EUROIMMUN Anti-C. pneumoniae ELISA		Quality assessment target		
		positive	borderline	negative
IgA (n = 102)	positive	21	0	2
	borderline	4	5	5
	negative	1	4	60
IgG (n = 111)	positive	70	0	1
	borderline	0	0	1
	negative	0	0	39
IgM (n = 108)	positive	12	0	0
	borderline	0	0	5
	negative	0	0	91

Automation

PCR microarray: EUROArrayScanner

The PCRs are incubated in the thermocycler and then, using the TITERPLANE technique, on EUROArray slides containing microarray BIOCHIPs. The EUROArrayScanner including EUROArrayScan software enables fully automated evaluation of the EUROArray analyses and detailed result documentation.

- Fully automated and standardised evaluation, interpretation and archiving of results by fluorescence scanner – less than 20 seconds per slide
- Result output as detailed individual view or overview
- LIS connection and networkability for optimal data communication and integration

MIF: EUROPattern

State-of-the-art, fully automated immunofluorescence microscopy and convenient creation of result reports on the computer screen:

- Automatic identification of slides via Data Matrix codes
- Rapid processing of up to 500 analyses automatically in sequence – up to 250 fields in just one hour
- Archiving of images and results and direct access to the patient history
- User-friendly operation and result compilation for each patient, data management/communication through EUROLabOffice 4.0
- Bidirectional LIS connection
- Fully automated sample dilution, incubation and washing with the IF Sprinter

Immunoblot: EUROBlotOne

The compact stand-alone tabletop instrument processes immunoblots fully automatically – from sample identification through to the evaluation:

- Barcode identification of samples
- Up to 44 samples / strips in one run
- Convenient digitisation of the incubated immunoblot strips and subsequent automated evaluation by means of the EUROLineScan software
- EUROLineScan enables the bidirectional data transfer with LIS or EUROLabOffice 4.0

ELISA: EUROIMMUN Analyzer I and I-2P

Fully automated ELISA processing; the EUROIMMUN Analyzer I (photo) is designed for medium throughputs, the EUROIMMUN Analyzer I-2P for small sample throughputs:

- Analyzer I: up to 7 plates and 180 strips per run
- Analyzer I-2P up to 3 plates and 144 strips per run
- Heatable and shakeable incubators
- Integrated barcode reader for samples and reagents

ELISA: EUROLabWorkstation

Efficient complete solution for fully automated ELISA processing for large sample throughputs:

- Parallel processing of up to 15 microplates with over 700 samples
- Intuitive software guides through all work steps, convenient operation via touch screen user interface
- Complete traceability of samples, reagents, dilution and ELISA plates by barcode scanning

Order information

Tests for *Chlamydia* diagnostics	Order number	Methods
EUROArray STI	MN 2830-####-#	DNA microarray
Anti-Chlamydia pneumoniae ELISA (IgA, IgG, IgM)	EI 2192-9601-1 A, G, M	Enzyme immunoassay
Anti-Chlamydia trachomatis ELISA (IgA, IgG, IgM)	EI 2191-9601-1 A, G, M	Enzyme immunoassay
Anti-Chlamydia MIF, BIOCHIP Mosaic (IgA, IgG, IgM)	FI 2191-####-3 A, G, M	Microimmunofluorescence
Anti-Chlamydia MIF EUROPattern (IgA, IgG, IgM)	FR 2191-####-3 A, G, M	Microimmunofluorescence
Anti-Chlamydia HP EUROLINE-WB (IgA, IgG)	DY 2190-1601-1 A, G	Immunoblot
Anti-Chlamydia HP EUROLINE-WB (IgA)*	DY 2190-0148-1 A	Immunoblot
Anti-Chlamydia HP EUROLINE-WB (IgG)*	DY 2190-4801-1 G	Immunoblot

* Immunoblot-PreQ (pre-equipped individual channels, 48 strips); compatible only with the EUROBlotOne

At a glance

- The EUROArray STI is available for detection of acute *C. trachomatis* infections by PCR-based direct pathogen detection.
- MIF enables parallel testing for antibodies against all three *Chlamydia* species in one reaction – very conveniently with fully automated microscopy and on-screen diagnostics.
- Parallel detection of antibodies against all three *Chlamydia* species can also be performed with the Anti-Chlamydia HP EUROLINE-WB. The combination of native antigen extract and specific single antigens guarantees high sensitivity and specificity.
- EUROIMMUN ELISAs provide detection of specific antibodies against *C. trachomatis* or *C. pneumoniae*. With automated processing, they are excellently suited for large sample throughputs.