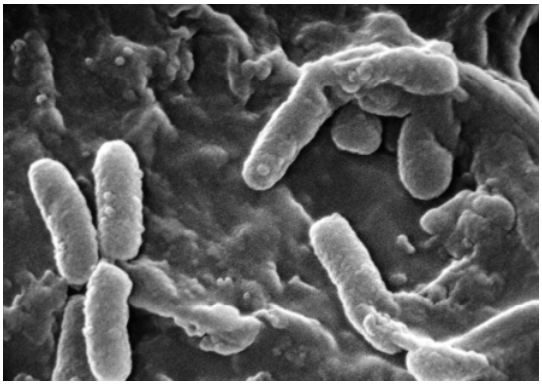
anti-Pseudomonas aeruginosa Antibody ELISA E15

Pseudomonas aeruginosa, a Gram-negative bacterium ubiquitously distributed in moist environment, causes about 10% of all nosocomial infections. This opportunistic pathogen leads to acute and chronic types of infection within various organs of susceptible patient groups, e.g. in patients with cystic fibrosis (CF). Early detection of infection and therewith early therapy improve the outcome significantly.



Pseudomonas aeruginosa Source: CDC/Janice Carr

P. aeruginosa infection provokes a rapid production of antibodies to a large number of *P. aeruginosa* antigens in CF patients.

Mediagnost sensitive antibody detection system detects a *P.aeruginosa* infection very early at the onset, **when microbiological information is not yet available.** The assay allows the commencement of anti-Pseudomonas chemotherapy early after onset of the infection and also serves as a tool to control the efficiency of antibiotic therapy (Ratjen F et al.:Lancet. 2001 Sep 22;358(9286):983-4). Due to the use of three *P.aeruginosa* antigens

Elastase
Exotoxin A
alkaline Protease

which are highly immunogenic and present in different parts from nearly all *P.aeruginosa* strains, this test has an extremly **high sensitivity and predictive value** (Kappler, M et al.: Thorax. 2006 61(8):614-618). Most patients immune systems react with at least one of the three antigens and therefore **false-negative** results are almost **impossible**, as well as false-positive.

Based on antibody titers the following diagnostic interpretation of test results is used in practice (already one positive titer indicates a pseudomonas infection) :

Titre	Interpretation
< 1:500	negative
1:500 to 1: 1.250	border-line
> 1:1.250	positive

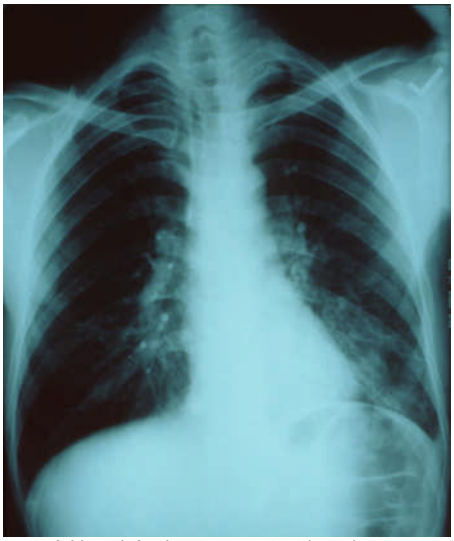
Mediagnost Anti-Pseudomonas aeruginosa ELISA E15

This sandwich enzyme immunoassay detects antibodies against Pseudomonas aeruginosa antigens. Serum or plasma samples are diluted and added to the wells of a microtitre plate, which has been previously coated with the *Pseudomonas aeruginosa* antigens **alkaline protease, elastase or exotoxin A.** Specific antibodies in the sample bind to the antigens, by Peroxidase-coupled anti-human IgG antibody bound antibodies are detected.

Assay Features E15

- ✓ Easy sample winning
- ✓ Inter-assay variance of 6.55%
- ✓ Intra-assay variance of 4.73%
- ✓ In comparison to pharyngeal swab 100% same results were obtained (104 samples)
- ✓ Control sera included

Mediagnost ELISA E15 has a **European Approval for Clinical Diagnostics**:EC Directive 98/79EG **(CE).**



Bacterial lung infection Source: CDC/Dr. Thomas Hooten

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Mediagnost anti-Pseudomonas aeruginosa ELISA E15

Dilution of Reagents and Samples		
Conjugate concentrate KK	in Dilution Buffer VP	1:100
Washing buffer WP	in Aqua dest. (e.g. add 100 ml WP in to a graduated flask and fill with A.dest to 2000 ml)	1:20
Dilute samples 1:1000 with dilution buffer (qualitative test). For quantitative antibody assays it is recommended to perform dilutions of 1:1.000, 1:10.000 and 1:100.000.		

Pipette	Reagents	Plate 1AP	Plate 2Ela	Plate 3Exo
2 x 100 µl	Negative Control NK	A1/A2	A1/A2	A1/A2
2 x 100 µl	Positive Control PK1	B1/B2		
2 x 100 µl	Positive Control PK2		B1/B2	
2 x 100 µl	Positive Control PK3			B1/B2
2 x 100 µl	Control Serum KS	C1/C2	C1/C2	C1/C2
2 x 100 µl	Sample dilution	Pipette in the rest of the wells according to requirements		
Seal the wells with the seeling tape				
Incubation: 2 h at 37°C				
3 x 300 µl	Aspirate the contents of the wells and wash 3x with 300 µl each Wash Buffer WP/ well	in each well		
100 µl	1:100 diluted Conjugate concentrate KK	in each well		
Seal the wells with the seeling tape				
Incubation: 2 h at 37°C				
3 x 300 µl	Aspirate the contents of the wells and wash 3x with 300 µl each Wash Buffer WP/ well	in each well		
100 µl	Substrate Solution S	in each well		
Incubation: 30 min in the dark at RT				
100 µl	Stopping Solution SL	in each well		
Measure the absorbance within 30 min at 450 nm with ≥ 590 nm as reference wavelength.				

Literature

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