Anti-Desmoglein 3 ELISA (IgG)

Indication: Test system for the in vitro determination of antibodies against desmoglein 3 in human serum or plasma for the diagnosis of the following disease: pemphigus vulgaris.

Clinical significance: Autoantibodies against desmoglein 1 and 3 are markers for pemphigus diseases, which can be clinically and immunopathologically subdivided into 4 different forms: pemphigus vulgaris, pemphigus foliaceus, paraneoplastic pemphigus and IgA pemphigus.

Pemphigus vulgaris always affects the mucous membranes. The majority of patients initially only develop lesions in the mucosa of the mouth. In the course of the disease some patients show flaccid blisters at the integument, particularly on parts of the body which are exposed to pressure and friction. Patients with pemphigus vulgaris who show damage exclusively to the mouth mucosa exhibit IgG antibodies only against desmoglein 3, whereas patients with lesions of the skin and mucosa produce antibodies against desmoglein 1 and 3. In patients with **pemphigus foliaceus**, blisters are rarely found due to a very superficial cleft formation in the stratum granulosum. The disease is rather characterised by scaly crusts, especially in the seborrheic areals. The mucosa is never affected. Congruently, pemphigus foliaceus is only associated with desmoglein 1. **Paraneoplastic pemphigus** is always associated with neoplasia. The resulting immune response is not only directed against desmoglein 3 but also against other proteins of desmosomal plaques such as envoplakin, periplakin, desmoplakin and plektin. **IgA pemphigus** is characterised by IgA antibodies against desmocolin 1 and desmoglein 3. Desmoglein 1 and 3 are cadherins, which are calcium-dependent transmembrane glycoproteins of epidermal desmosomes. They are components of the maculae adherentes and permit the cell-to-cell contact in the epidermis and the surface mucosa via homophilic and heterophilic extracellular binding. The pathogenetic relevance of autoantibodies against desmoglein 1 and 3 is well proven. For example, the injection of serum from pemphigus patients into neonatal mice leads to blister formation. The exact mechanisms which cause the blister formation, however, are unknown.

Panel	n	Anti-Desmoglein 3 positive
Pemphigus vulgaris (PV)	**71**	**71 (100%)**
Pemphigus foliaceus (PF)	50	0 (0%)
Asymptomatic blood donors (BD)	401	1 (0.2%)
Bullous pemphigoid (BP)	48	1 (2.1%)
Linear IgA dermatosis (LAD)	21	0 (0,0%)
Sensitivity for pemphigus vulgaris	**71**	**71 (100%)**
Specificity for pemphigus vulgaris	**470**	**2 (99.6%)**

Clinical sensitivity and specificity: Sera from 71 patients with pemphigus vulgaris, 50 patients with pemphigus foliaceus, a control panel of 69 patients with other autoimmune diseases and 401 healthy blood donors were investigated using the EUROIMMUN Anti-Desmoglein 3 ELISA. The sensitivity of the ELISA for pemphigus vulgaris was 100%, with a specificity of 99.6%. In the pemphigus foliaceus panel all patients were found negative.

Application of the Anti-Desmoglein 3 ELISA: In the diagnosis of pemphigus, determination of circulating autoantibodies using indirect immunofluorescence (on primate oesophagus as sensitive substrate) has proven successful. However, it does not allow differentiation between antibodies against desmoglein 1 and desmoglein 3. ELISA using desmoglein 1 and 3 offers the same sensitivity and specificity as IIFT. In most cases the Anti-Desmoglein 1 ELISA and Anti-Desmoglein 3 ELISA are sufficient to diagnose pemphigus. In suspected pemphigus cases with a negative ELISA result IIFT should be carried out in addition. The Anti-Desmoglein 3 ELISA is of particular importance in lesions of the mouth mucosa to differentiate pemphigus vulgaris from Lichen ruber mucosae, benign aphtha, Behcet's disease and Steven-Johnson syndrome.

The Anti-Desmoglein 1 ELISA and Anti-Desmoglein 3 ELISA are highly sensitive and specific test systems for the diagnosis of pemphigus diseases. In untreated patients a positive result in the Anti-Desmoglein 3 ELISA alone suggests the presence of pemphigus vulgaris with only mucosa involvement. If both the Anti-Desmoglein 3 ELISA and the Anti-Desmoglein 1 ELISA are positive, this indicates pemphigus vulgaris with mucosa and skin involvement. A positive Anti-Desmoglein 1 ELISA result alone is indicative of pemphigus foliaceus. The antibody levels of desmoglein 1 and 3 in the serum generally correlate with the severity and activity of the disease and the therapy success.

EUROIMMUN Microplate ELISA

Autoantibody determination:
AMA M2-3E (IgG)
ANCA Profile (IgG)
ANA Screen (IgG)
ANA Screen 9 or 11 (IgG)
ANA VarioProfile (IgG)
BP180-NC16A-4X (IgG)
BP230-CF (IgG)
C1q (IgG)
cardiolipin (IgA, IgG, IgM, IgAGM)
circulating immune complexes (CIC)
cyclic citrullinated peptide (CCP; IgG)
centromere protein B (IgG)
desmoglein 1 (IgG)
desmoglein 3 (IgG)
double-stranded DNA (dsDNA, nDNA; IgG)
dsDNA-NcX (IgG)
ENA Pool (IgG)
ENA PoolPlus (IgG)
ENA ProfilePlus 1 or 2 (IgG)
ENA SLE Profile 1 or 2 (IgG)
GAD
GAD/IA-2 Pool
glomerular basement membrane (GBM; IgG)
ß2-glycoprotein 1 (IgA, IgG, IgM, IgAGM)
histones (IgG)
IA-2
intrinsic factor (IgG)
Jo-1 (IgG)
liver cytosolic antigen type 1 (LC-1; IgG)
liver-kidney microsomes (LKM-1; IgG)
myeloperoxidase (MPO; IgG)
nRNP/Sm (IgG)
nucleosomes (IgG)
p53 (IgG)
parietal cells (PCA; IgG)
PM-Scl (PM-1; IgG)
phosphatidylserine (IgA, IgG, IgM, IgAGM)
proteinase 3 (IgG)
PR3 hn-hr (IgG)
PR3 capture (IgG)
rheumatoid factor (IgA, IgG, IgM)
ribosomal P-proteins (IgG)
Sa (IgG)
Scl-70 (IgG)
single-stranded DNA (ssDNA; IgG)
SLA/LP (IgG)
Sm (IgG)
SS-A (Ro; IgG)
SS-B (La; IgG)
thyroglobulin (TG; IgG)
thyroid peroxidase (TPO; IgG)
tissue transglutaminase (endomy.; IgA, IgG)
TSH receptor (TBII; IgG)
TRAk Fast (IgG)

Further autoimmune diagnostics:
gliadin (GAF-3X; IgA, IgG)
Saccharomyces cerevisiae (IgA, IgG)

Infectious serology:
Adenovirus (IgA, IgG, IgM)
Borrelia (IgG, IgM)
Borrelia VlsE (IgG)
Chlamydia pneumoniae (IgA, IgG, IgM)
Chlamydia trachomatis (IgA, IgG, IgM)
Cytomegalovirus (IgG, IgM)
Diphtheria toxoid (IgG)
Epstein-Barr virus capsid ag (IgA, IgG, IgM)
Epstein-Barr virus early ag (IgA, IgG, IgM)
Epstein-Barr virus nuclear ag, EBNA-1 (IgG)
Helicobacter pylori (IgA, IgG)
Helicobacter pylori CagA (IgA, IgG)
HSV-1 (glycoprotein C1; IgA, IgG, IgM)
HSV-2 (glycoprotein G2; IgA, IgG, IgM)
HSV-1/2 Pool (IgA, IgG, IgM)
Influenza virus type A (IgA, IgG, IgM)
Influenza virus type B (IgA, IgG, IgM)
Legionella pneumophila (IgA, IgG, IgM)
Measles virus (IgG, IgM)
Mumps virus (IgG, IgM)
Mycoplasma pneumoniae (IgA, IgG, IgM)
Parainfluenza virus Pool (IgA, IgG, IgM)
Parvovirus B19 (IgG, IgM)
RSV (IgA, IgG, IgM)
Rubella virus (IgG, IgM)
SARS-CoV (IgG)
TBE virus (IgG, IgM)
Tetanus toxoid (IgG)
Toxoplasma gondii (IgG, IgM)
Treponema pallidum (IgG, IgM)
Varicella zoster virus (IgG, IgM)
Yersinia enterocol. virulence fact. (IgA, IgG)

Allergology:
total IgE
Allercoat™ 6-ELISA (600 different allergens and allergen mixtures)

Serum proteins and tumour markers:
anti-p53

* Currently not available as IVD in the EU.

Made in Germany

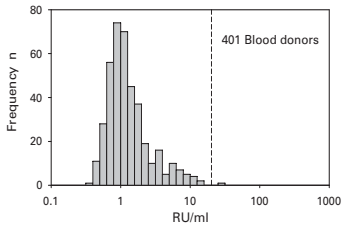
Test characteristics
Anti-Desmoglein 3 ELISA (IgG)

Linearity: The linearity of the ELISA was determined by assaying 4 serial dilutions of 6 serum samples. The linear regression was calculated, R2 amounting to > 0.95 in all samples. The Anti-Desmoglein 3 ELISA (IgG) is linear in at least the tested concentration range (14-195 RU/ml).

Reproducibility: The reproducibility of the test was investigated by determining the intra- and inter-assay coefficients of variation using 3 sera. The intra-assay CVs are based on 20 determinations and the inter-assay CVs on 4 determinations performed in 6 different test runs.

	Intra-assay variation, n = 20		*Intra-assay variation, n = 4 x 6*	
Serum	Mean value (RU/ml)	CV (%)	Mean value (RU/ml)	CV (%)
1	42	4.4	44	6.1
2	63	2.6	69	5.3
3	155	5.9	163	3.3

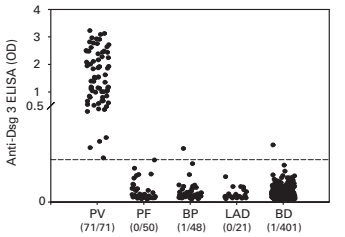
Reference range: Levels of anti-desmoglein 3 antibodies were determined in 401 sera from healthy blood donors of between 18 and 68 years of age (151 women, 250 men) using the EUROIMMUN ELISA. The mean concentration of antibodies against desmoglein 3 was 1.8 RU/ml and the values ranged from 0.4 to 25.3 RU/ml. With a cut-off of 20 RU/ml, 0.2% of the blood donors were anti-desmoglein 3 positive.



ROC analysis: In an analysis of 71 samples from patients with pemphigus vulgaris and 470 control samples the following results were achieved:

Blood donors, n = 401		
Percentile	99th	100th
Cut-off	11.4 RU/ml	25.3 RU/ml

Cut-off	Specificity	Sensitivity
15.9 RU/ml	99.4 %	100 %
21.8 RU/ml	99.6 %	100 %



Correlation of the EUROIMMUN and MBL Anti-Desmoglein 3 ELISAs: The antibody concentration was determined in 71 sera from patients with pemphigus vulgaris using the Anti-Desmoglein 3 ELISAs from EUROIMMUN and MBL. The qualitative results of the ELISAs were 99% in agreement.

PV patients, n = 71		EUROIMMUN positive	EUROIMMUN negative
MBL	positive	70	0
MBL	negative/ borderline	1	0

The antibody concentration was measured in 69 patients with other autoimmune diseases (bullous pemphigoid, linear IgA dermatosis) using the Anti-Desmoglein 3 ELISAs from EUROIMMUN and MBL. The qualitative results of the ELISAs were 97% in agreement.

Controls, n = 69		EUROIMMUN positive	EUROIMMUN negative
MBL	positive	0	1
MBL	negative/ borderline	1	67

Technical data:

Antigen	Recombinant, expression in mammalian cells, extracellular domain of desmoglein 3 (5 subdomains).
Calibration	Quantitative, in relative units per milliliter (RU/ml). Calibration serum 1: 200 RU/ml Calibration serum 2: 20 RU/ml; cut-off Calibration serum 3: 2 RU/ml
Sample dilution	Serum or plasma; 1:101 in sample buffer.
Reagents	Ready for use, with the exception of the wash buffer (10x). Colour-coded solutions, in most cases exchangeable with those in other EUROIMMUN ELISA kits.
Test procedure	30 min / 30 min / 15 min. Room temperature.
Measurement	450 nm. Reference wavelength between 620 nm and 650 nm.
Test kit format	48 break-off wells. Kit includes all necessary reagents.
Order no.	EA 1496-4801 G

EUROIMMUN Immunoblots

Autoantibody determination:

EUROASSAY:

flexible profiles of up to 7 antigens from:

ENA and related antigens: nRNP/Sm, Sm, SS-A, Ro-52, SS-B, Scl-70, Jo-1, dsDNA, histones, nucleosomes, CENP B, PM-Scl, ribosomal P-proteins, AMA M2

liver antigens: LKM-1, LC-1, SLA/LP, AMA M2, M4, M9

ANCA antigens: MPO, PR3

thyroid antigens: TG, TPO

EUROLINE:

ANA Profile 1: nRNP/Sm, Sm, SS-A, Ro-52, SS-B, Scl-70, Jo-1, CENP B, dsDNA, nucleosomes, histones, ribosomal P-proteins

ANA Profile 3: nRNP/Sm, Sm, SS-A, Ro-52, SS-B, Scl-70, PM-Scl, Jo-1, CENP B, PCNA, dsDNA, nucleosomes, histones, ribosomal P-proteins, AMA M2

Anti-ENA Profile 1: nRNP/Sm, Sm, SS-A, Ro-52, SS-B, Scl-70, Jo-1

Myositis Profile: Mi-2, Ku, PM-Scl, Jo-1, PL-7, PL-12, Ro-52

Liver Profiles: AMA M2, 3E (BPO), Sp100, PML, gp210, LKM-1, LC-1, SLA/LP, Ro-52

Neuronal Antigens Profile 2: amphiphysin, CV2.1** PNMA2 (Ma-2/Ta), Ri, Yo, Hu

Anti-Ganglioside Profile 1: GM1, GD1b, GQ1b

Anti-Ganglioside Profile 2: GM1, GM2, GM3, GD1a, GD1b, GT1b, GQ1b

ANCA Profiles: MPO, PR3, GBM

EUROLINE-WB:

neuronal antigens (+ recomb. Hu, Yo, Ri)
HEp-2 cell antigens (+ SS-A and Ro-52, CENP B)

Infectious serology:

EUROLINE:

Bordetella pertussis (IgA, IgG)
Borrelia-RN-AT (p18, p19, p20, p21, p58, OspC, p39, p83, LBb, LBa, VlsE Bg, VlsE Bb, VlsE Ba)
EBV Profile (IgG, IgM, VCA gp125, VCA p19 and EBNA-1, p22, EA-D)
Hanta virus (IgG, IgM)
Rubella virus (IgG)
TORCH Profile* (T. gond., rubella, CMV, HSV-1, -2)

Westernblot:

Borrelia burgdorferi (IgG, IgM)
Borrelia afzelii (IgG, IgM)
Borrelia garinii (IgG, IgM)
Epstein-Barr virus (IgG, IgM)
Helicobacter pylori (IgA, IgG)
Treponema pallidum (IgG, IgM)
Yersinia enterocol. virulence fact. (IgA, IgG)

EUROLINE-WB:

Anti-Borrelia (B. afzelii + rec. VlsE)
Anti-HSV (HSV-1 + HSV-2 gG2)
Treponema pallidum + cardiolipin

Allergology:

EUROASSAY:

Domestic Animal Profile (IgE)
Food Profile (IgE)
Inhalation Profile (IgE)
Insect Venom Profile (IgE)
Latex Profile (IgE)
Latex plus Profile (with ficus and fruit; IgE)

EUROLINE:

Atopy Profile (IgE)
Food Profile (IgE)
Inhalation Profile (IgE)
Paediatric Inhalation Profile
Pollen–Food Cross Reaction Profile (IgE)

Software/Automation:

EUROLineScan
camera system EUROBlotCamera
scanner system EUROBlotScanner
incubation processor EUROBlotMaster

EUROIMMUN Radioimmunoassays

Autoantibody determination:

thyroid peroxidase (TPO; IgG)
thyroglobulin (TG; IgG)
TSH receptor (IgG)
acetylcholine receptor (ACHR; IgG)
glutamic acid decarboxylase (GAD; IgG)
insulin (IAA; IgG)
P/Q calcium channel* (VGCC; IgG)
tyrosine phosphatase (IA2; IgG)
dsDNA (IgA/IgG/IgM)

Antigen determination:

thyroglobulin (TG)

Hormone determination:

free triiodothyronine (FT3)
free thyroxine (FT4)
thyrotropin (TSH)
calcitonin

* Currently not available as IVD in the EU.
** CV2 partial protein, which only contains the N-terminally localised epitopes of the antigen.

Made in Germany

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