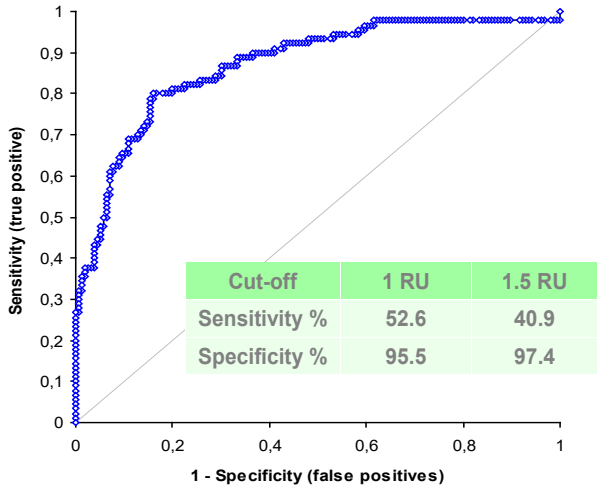
dsDNA ELISA

The dsDNA ELISA contributes to the diagnosis of systemic lupus erythematosus (SLE) and related autoimmune diseases

A characteristic feature of systemic autoimmune diseases such as systemic lupus erythematosus (SLE) is the occurrence of circulating antibodies against defined intra-cellular targets including double stranded desoxyribonucleic acid (dsDNA), U1-ribonucleoproteins (RNPs), Smith antigen (Sm), histones, proliferating cell nuclear antigen (PCNA) and ribosomal phosphoproteins (Rib-P). Currently, more than 100 different antigens have been described as the target of autoantibodies in sera of SLE patients. Anti-dsDNA antibodies are known as sensitive and specific marker for SLE. Anti-dsDNA antibodies can be detected in 20-70% of SLE patients depending on the test system, the genetic background of the patients and the make-up of the patient group under investigation. The titer of anti-dsDNA antibodies correlates with the disease activity of the patients. In rare cases, anti-dsDNA antibodies are found in patients with related connective tissue diseases or with viral infections.

The dsDNA ELISA is intended for the semi-quantitative determination of antibodies specific for dsDNA. The results of the dsDNA ELISA aid to the diagnosis of SLE and related diseases.

dsDNA ELISA REF 25005 96



Cut-off	1 RU	1.5 RU
Sensitivity %	52.6	40.9
Specificity %	95.5	97.4

Figure 1
Receiver operating characteristics (ROC) analysis. 171 serum samples from patients with SLE and 156 controls were tested for anti-dsDNA antibodies using the dsDNA ELISA (REF: 25005). The results show a good differentiation between SLE patients and controls as revealed by an area under the curve value (AUC) of 0.87. The sensitivity and specificity of the test in this patient group was determined as 40.9 % and 97.4 %, respectively.

dsDNA ELISA Specifications

- Highly purified plasmid antigen
- Colored reagents
- Assay time: < 1.5 h at RT (30min /30min/ 15min)
- 3 µL serum or plasma per test
- Detection System: HRP/TMB ($OD_{450nm\ /620\ nm}$)
- Wide measuring range
- Low detection limit

Assay performance

- Good correlation to reference ELISA systems and indirect immunofluorescence on *Crithidia luciliae* substrate
- Excellent "lot to lot" correlation R^2 > 0.95
- Low intra- and inter-assay variation CV% < 10
- Excellent linearity over the entire range

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ID	Target	RU	Interpretation
CDC 1	DNA	6.7	positive
CDC 2	SS-B/La	0.4	negative
CDC 3	RNP/Sm, SS-A/Ro, SS-B (La)	0.3	negative
CDC 4	U-1 RNP	0.4	negative
CDC 5	Sm	0.7	negative
CDC 6	Fibrillarin	0.2	negative
CDC 7	SS-A/Ro	0.6	negative
CDC 8	Centromere	0.4	negative
CDC 9	Scl-70	0.7	negative
CDC 10	Jo-1	0.2	negative
CDC 11	PM/Scl (PM 1)	0.4	negative
CDC 12	Rib-P	0.2	negative

Figure 2
Results of the CDC ANA reference sera. 12 reference serum samples, available from the "Center for Disease Control and Prevention (CDC)" were tested in the dsDNA ELISA (REF:25005). Only the anti-dsDNA positive sample (CDC 1) was found to be positive.

Table 1
Anti-dsDNA reactivity in different disease groups

	No. (%) of anti-dsDNA positive sera
SLE (n=171)	70 (40.9)
Rheumatic disease controls (84)	4 (4.7)
RA (22)	2 (2.3)
MCTD (7)	2 (2.3)
SSc (2)	0 (0)
DM (2)	0 (0)
UCTD (7)	0 (0)
Other rheumatic diseases (37)	0 (0)
Healthy individuals (72)	0 (0)

		dsDNA ELISA (25005)		
		neg	pos	
Reference	neg	27	5	32
	pos	6	29	35
		33	34	67

Figure 3
Agreement to reference ELISA. 67 serum samples from patients with SLE tested in the dsDNA ELISA (REF:25005) and in a validated reference method (CLIFT) demonstrated a good agreement (83.6%) between the two assays.

Literature

1. Tan EM: **Antinuclear antibodies: diagnostic markers for autoimmune diseases and probes for cell biology.** Adv Immunol 1989, 44:93-151.
2. James K, Carpenter AB, Cook L, Marchand R, and Nakamura RM for the Association of Medical Laboratory Immunologists Standards Committee: **Development of the Antinuclear and Anticytoplasmic Antibody Consensus Panel by the Association of Medical Laboratory Immunologists.** Clin Diagn Lab Immunol 2000, 7:436–443.
3. Hochberg MC: **Updating the American College of Rheumatology Revised Criteria for the Classification of Systemic Lupus Erythematosus.** Arthr Rheum 1997, 40: 1725-1734.
4. Bootsma H, Spronk PE, Ter Borg EJ et al.: **The predictive value of fluctuations in IgM and IgG class anti-dsDNA antibodies for relapses in systemic lupus erythematosus. A prospective long term observation.** Ann Rheum Dis 1997, 56: 661-666.
5. Tzioufas AG, Tergoglou C, Stavropoulos ED et al.: **Determination of anti-dsDNA antibodies by three different methods: Comparison of sensitivity, specificity and correlation with lupus acticity index (LAI).** Clin Rheumatol 1990, 9:186-192.
6. Mahler M, Waka A, Hiepe F, Fritzler MJ: **Effect of dsDNA binding to SmD-derived peptides on clinical accuracy in the diagnosis of systemic lupus erythematosus.** Arthritis Res Ther 2007, 9: R68.
7. Mahler M, Fritzler MJ: **Anti-dsDNA antibody testing in the clinic: Farr or ELISA?** Nat Clin Pract Rheumatol. 2007 Feb, 3:72-73.

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