Ro52 ELISA

The Ro52 ELISA contributes to the diagnosis of systemic lupus erythematosus (SLE), Sjögren Syndrome (SjS), systemic sclerosis (SSc) and related autoimmune diseases

Circulating antibodies to intra-cellular structures especially to nuclear antigens represent a characteristic feature of systemic autoimmune diseases. One of those antigens, the Ro52 could recently be identified as a RING dependent E3 Ligase. Autoantibodies specific for Ro52 occur in patients with systemic lupus erythematosus (SLE), Sjögren Syndrome (SjS), mixed connective tissue disease (MCTD) and systemic sclerosis (SSc). The Ro52, together with Ro60 is often termed as SS-A antigen (Sjögren Syndrome antigen-A). Anti-Ro52 antibodies are frequently accompanied by anti-La and anti-Jo-1 antibodies. Moreover, anti-Ro52 antibodies seem to play an important role for neonatal lupus and congenital heart block, especially those directed against certain Ro52 epitopes. Certain epitops seem to be associated with complications during pregnancy. Noteworthy, the prevalence of anti-Ro52 in PM and SSc is significantly higher than of anti-Ro60 antibodies. Therefore seperate determination of anti-Ro52 and anti-Ro60 antibodies is required.

The Ro52 ELISA is intended for the semi-quantitative determination of anti-Ro52 antibodies. The results of the Ro52 ELISA aid to the diagnosis of SLE, SjS, SSc and related autoimmune disorders and should be used as prognostic marker for the disease progression.

Ro52 ELISA Specifications

- Recombinant 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

Ro52 ELISA REF 25006 96

ID	Target	RU	Interpretation
CDC 1	DNA	0.4	negative
CDC 2	SS-B/La	2.1	positive
CDC 3	RNP/Sm, SS-A/Ro, SS-B/La	1.3	borderline
CDC 4	U-1 RNP	0.3	negative
CDC 5	Sm	0.3	negative
CDC 6	Fibrillarin	0.2	negative
CDC 7	SS-A/Ro	2.2	positive
CDC 8	Centromere	0.1	negative
CDC 9	Scl-70	0.2	negative
CDC 10	Jo-1	7.1	positive
CDC 11	PM/Scl (PM 1)	0.3	negative
CDC 12	Rib-P	0.2	negative

Figure 1
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 Ro52 ELISA (REF: 25006). Sample CDC2, CDC 7, CDC 10 were positive and CDC 3 borderline for anti-Ro52 antibodies.

Assay performance

- Good correlation to reference methods
- Excellent "lot to lot" correlation $R^2 > 0.95$
- Low intra- and inter-assay variation CV% < 10
- High sensitivity and specificity
- Excellent linearity over the entire range

Dr. Fooke-Achterrath Laboratorien GmbH - Habichtweg 16 - 41468 Neuss - Germany
Phone: + 49 2131 2984-0 - Fax: + 49 2131 2984-184 - email: information@fooke-labs.de

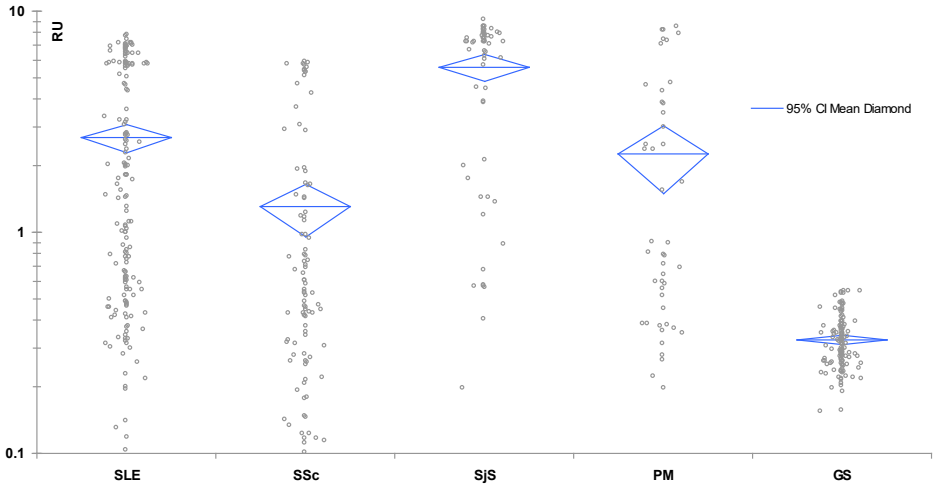

Figure 2
Anti-Ro52 reactivity in different cohorts of connective tissue diseases and healthy donors. Serum samples from patients with systemic lupus erythematosus (SLE, n=168), systemic sclerosis (SSc, n=100), Sjögren Syndrome (SjS, n=57), Polymyositis (PM, n=48) and healthy donors (HD, n=85) were tested for anti-Ro52 antibodies by Ro52 ELISA (REF: 25006). Highest prevalence and titers of anti-Ro52 antibodies were found in patients with SLE and SjS. The prevalence of anti-Ro52 antibodies in patients with SSc and PM was significantly higher as the prevalence of anti-Ro60 antibodies.

		Ro52 ELISA (25006)		
		pos	neg	
Reference	pos	90	2	92
	neg	20	76	96
		110	78	188

Figure 3
Agreement with a reference method. 188 samples from patients with SLE (n=131) and Sjögren Syndrome (n=57) were tested by Ro52 ELISA (REF: 25006) and by a validated, FDA approved reference system (multiplex assay). The results show good concordance (88%) between both methods. The sensitivity of the ELISA is significantly higher compared to the multiplex assay at 100% specificity against unrelated disease controls and healthy donors.

Table 1
Prevalence of anti-Ro52 antibodies in different disease cohorts and healthy donors

Disease	% pos Ro52 ELISA (REF: 25006)	% pos Literature
SLE (n=168)	49.4	40-60
SSc (Scl, n=100)	23	~ 20
SjS (n=57)	78.9	70-90
Myositis (n=48)	41.7	20-40
HD (n=85)	0	0

Literature

1. Tan EM: Antinuclear antibodies: diagnostic markers for autoimmune diseases and probes for cell biology. Adv Immunol 1989, 44:93-151.
2. Rutjes SA, Vree Egberts WT, Jongen P, Van Den Hoogen F, Pruijn GJ, Van Venrooij WJ: Anti-Ro52 antibodies frequently co-occur with anti-Jo-1 antibodies in sera from patients with idiopathic inflammatory myopathy. Clin Exp Immunol 1997, 109:32-40.
3. Fritsch C, Hoebeke J, Dali H, Ricchiuti V, Isenberg DA, Meyer O, Muller S: 52-kDa Ro/SSA epitopes preferentially recognized by antibodies from mothers of children with neonatal lupus and congenital heart block. Arthritis Res Ther 2005, 8:R4
4. Schulte-Pelkum J, Fritzler MJ, Mahler M: Latest Update on the Ro/SS-A Autoantibody System. Autoimmun Rev 2009, 8:632-637.

Dr. Fooke-Achterrath Laboratorien GmbH - Habichtweg 16 - 41468 Neuss - Germany
Phone: + 49 2131 2984-0 - Fax: + 49 2131 2984-184 - email: information@fooke-labs.de