Arthritis & Rheumatism (Arthritis Care & Research)
Vol. 47, No. 5, October 15, 2002, pp 546–555
DOI 10.1002/art.10558

SPECIAL ARTICLE

Guidelines for Immunologic Laboratory Testing in the Rheumatic Diseases: Anti-DNA Antibody Tests

ARTHUR F. KAVANAUGH, DANIEL H. SOLOMON, AND THE AMERICAN COLLEGE OF RHEUMATOLOGY AD HOC COMMITTEE ON IMMUNOLOGIC TESTING GUIDELINES

Introduction

This paper is part of a series on immunologic testing guidelines. The series introduction (1) outlines the full methodology for obtaining data, grading the literature, combining the information from multiple sources, and developing recommendations. Briefly, MEDLINE and Healthstar were searched using a variety of search terms, and all relevant available literature was reviewed. All papers were critically reviewed using published standards for studies of diagnostic tests. Test use was categorized as primarily diagnostic or prognostic (which also included monitoring). Information was extracted from each paper to allow for calculation of a weighted average for sensitivity and specificity; likelihood ratios (LRs) were then derived from these values (positive LR = sensitivity/[1 − specificity]; negative LR = [1 − sensitivity]/specificity). Recommendations for use of tests were based on the LRs, where a test was considered to be "very useful" for a given disease if the weighted average positive LR was >5 or negative LR was <0.2. A test was considered "useful" if the weighted average positive LR was >2 and ≤ 5 or negative LR was >0.2 and ≤ 0.5. A test was considered "not useful" if the positive LR was ≤ 2 or the negative LR was >0.5.

Antibodies capable of binding double-stranded DNA (dsDNA), single-stranded DNA (ssDNA), Z-DNA, and various other proteins associated with DNA (e.g., histones) can be detected in the serum of patients with systemic lupus erythematosus (SLE). In this analysis, the term anti-DNA antibody will refer only to antibodies specific for dsDNA (also referred to as native DNA). A number of studies have shown that antibodies to ssDNA (also referred to as denatured DNA) are less specific for the diagnosis of SLE than anti-dsDNA antibodies. While anti-ssDNA antibodies may be a useful research tool, they are an infrequently used clinical test, and will not be addressed further in this analysis.

Members of the American College of Rheumatology Ad Hoc Committee on Immunologic Testing Guidelines are as follows. Arthur F. Kavanaugh, MD (chair): University of California at San Diego; Daniel H. Solomon, MD, MPH, Peter Schur, MD: Brigham and Women's Hospital, Boston, Massachusetts; John D. Reveille, MD: University of Texas Health Science Center, Houston, Texas; Yvonne R. S. Sherrer, MD: Center for Rheumatology, Immunology and Arthritis, Fort Lauderdale, Florida; Robert Lahita, MD, PhD: St. Vincent's Medical Center, New York, New York.

Address correspondence to Arthur Kavanaugh, MD, Division of Rheumatology, Allergy, and Immunology, University of California at San Diego, 9310 Campus Point Drive, Suite A111, San Diego, California, 92037-0943. E-mail: akavanaugh@ucsd.edu.

Submitted for publication September 19, 2000; accepted in revised form November 1, 2001.

Background

Beginning in the 1950s, it was discovered that antibodies reactive with DNA could be detected in the sera of patients with SLE (2–10). It soon became appreciated that anti-DNA antibodies might be of value for the diagnosis of SLE, because elevated levels were detected infrequently in the sera of normal controls or patients with other autoimmune diseases. Because they were considered useful in diagnosing SLE, anti-DNA antibodies became part of the American College of Rheumatology classification criteria (11,12). In addition to serving as a laboratory marker for SLE, anti-DNA antibodies may directly contribute to pathologic processes, such as lupus glomerulonephritis (5–8,10,13,14). The stimuli driving the production of anti-DNA antibodies in SLE patients remain unknown.

Anti-DNA antibodies have been detected by various laboratory techniques over the years. Some methods such as immunodiffusion, hemaglutination, and complement fixation are no longer generally used and will not be considered in detail in this analysis. Currently, the most commonly used techniques for the detection of anti-DNA antibodies are enzyme-linked immunosorbent assay (ELISA) and immunofluorescence (e.g., using *Crithidia luciliae* as substrate). Radioimmunoassay (e.g., the Farr assay) is still available, but its use has decreased sharply.

Although each of these assays can detect anti-DNA antibodies, there are important differences between them. The Farr assay, which measures the precipitation of radiolabeled dsDNA by anti-dsDNA antibodies under stringent (i.e., high salt concentration) conditions, detects primarily

high affinity antibodies to dsDNA. However, the assay may also detect other proteins capable of precipitating dsDNA, it may on occasion be contaminated by ssDNA in the test preparation, and it does not distinguish between isotypes (e.g., IgG versus IgM anti-dsDNA).

The Crithidia assay detects binding of anti-dsDNA to the kinetoplast of the organism, which contains circular dsDNA unassociated with histone proteins. It can be used to detect IgG anti-dsDNA, IgM anti-dsDNA, or all isotypes of anti-dsDNA. In the ELISA, dsDNA is adhered to the wells of a plate, the test serum is added as a source of anti-dsDNA, and this anti-DNA antibody is detected by a second antibody.

Although the ELISA can be used to detect various isotypes of antibodies, the detection of IgG anti-dsDNA is most commonly used clinically, and will be focused on in this report. The ELISA detects low as well as high affinity antibodies, potentially making it less specific than the other assays. Another theoretical concern with the ELISA is that ssDNA may contaminate the dsDNA and give false positive results. In many commercial kits the preparation is highly purified and/or enzyme digested to remove ssDNA, in order to ensure that only anti-dsDNA are measured. Other techniques may also be used to ensure specificity. Even so, dsDNA can spontaneously denature, yielding ssDNA and producing false positive results in the assay. Based in part on these types of technical considerations, the Farr and Crithidia assays are generally considered somewhat more specific and less sensitive for SLE than the ELISA.

A number of studies have directly compared the performance of these assays, using replicate sera from SLE patients and healthy controls (15–39). Analysis of the data from these studies shows that overall there is a statistically significant correlation among the results obtained using the different types of assays. In addition, several studies assessing longitudinal determinations have found that results using different assays correlate over time (25–29). Although the results for anti-DNA testing using the different types of assays correlate for populations, there may be substantial discrepancies in the results for individual patient sera (23,27–39). For example, in some studies discrepant results were found for more than a third of the tested sera (e.g., results were positive in one assay yet negative in another). Discrepant results may also be found when using different commercial kits that are based on the same technique (27). Moreover, the close correlations among the different assays have typically been noted when they are applied predominantly to populations of SLE patients. These correlations may not hold up when applied to other populations (28).

Therefore, when interpreting the results of anti-DNA antibody testing for patients, the clinician should be aware of the technique used, the laboratory in which the test was performed, and the ranges for that test in that laboratory, both for healthy controls and for SLE patients. This information becomes even more important as clinicians' ability to choose the technique and laboratory becomes reduced. There may be considerable variation in results obtained from different laboratories.

In this analysis of the utility of anti-DNA antibody testing, methodologic considerations were taken into account in the grading of each article. Where relevant, statements concerning the specific techniques utilized in individual articles are included. However, results obtained using all 3 currently available techniques are included and will be grouped together.

Indications for clinical use of the anti-DNA antibody test

Diagnosis. An initial literature search was conducted and 168 articles were retrieved. These articles were graded according to the criteria reported in the Introduction article for this series (1). From this group of 168 articles, 43 that assessed the prevalence of anti-DNA antibodies in patients with SLE, patients with various other diseases, and healthy controls were considered for further review. Of these 43 studies, 11 were graded "A" (Table 1), and they form the basis of the recommendations (13 studies were graded as "B" and the remainder were graded "C" or "D").

In almost all studies, the prevalence of elevated levels of anti-DNA antibodies in healthy controls was zero or very low. Of note, in some studies the threshold for a positive test was 2 standard deviations above the mean of the controls; therefore 2.5% of the controls would have a positive test result by definition. It is not surprising that healthy persons may occasionally be found to have detectable anti-DNA antibodies in their serum, because normal B cells have been shown to be capable of producing such antibodies upon stimulation (B cells from SLE patients tend to produce such antibodies spontaneously) (40).

Anti-DNA antibodies have been reported in patients with a variety of rheumatologic diseases and other conditions including: rheumatoid arthritis, Sjögren's syndrome, scleroderma, drug induced lupus, Raynaud's phenomenon, mixed connective tissue disease, discoid lupus, myositis, chronic active hepatitis, other liver diseases, uveitis, relatives of SLE patients, patients hospitalized for nonrheumatologic diseases, Graves' disease, Alzheimer's disease, juvenile rheumatoid arthritis, certain laboratory workers, anticardiolipin antibody syndrome, and persons with silicone breast implants (41–60). The frequency of elevated levels of anti-DNA antibodies in conditions other than SLE is uniformly low ($\leq$5% of patients), and when present, they are often present in low titer. Therefore, outside of a research setting, ordering tests for anti-DNA antibodies is not useful for the diagnosis of any condition other than SLE. However, in a patient without SLE, a positive test result for anti-DNA, particularly at low levels, may be explained by the presence of one of these conditions.

Although anti-DNA antibodies have been reported in a number of other conditions, with rare exception their prevalence was also very low. Therefore in this analysis, healthy subjects and patients with other diseases have been combined, and will be contrasted with SLE patients.

Anti-DNA antibody testing is very useful for the diagnosis of SLE. As can be seen in Table 1 (15,16,18,38,44–46,61–64), weighted means (weighted according to numbers of patients assessed) for the use of anti-DNA in diagnosing SLE were 57.3% for sensitivity and 97.4% for

Table 1. Population statistics for anti-DNA antibodies in diagnosis: Systemic lupus erythematosus vs. healthy controls and other diseases*

Reference	# of patients	Technique	Sen	Spec	+LR	−LR	Grade
61	304	ELISA	0.5	0.96	14.7	0.52	A
14	315	Crithidia	0.57	0.97	18.4	0.58	A
		Farr	0.4	0.96	10.5	0.63	
17	130	ELISA	0.33	0.96	7.86	0.69	A
		Crithidia	0.13	1.0	>18.5	0.87	
		Farr	0.57	0.9	5.7	0.64	
15	216	ELISA	0.82	0.98	35.0	0.19	A
		Farr	0.6	0.98	26.0	0.41	
44	208	Farr	0.77	0.93	11.6	0.35	A
45	312	Farr	0.72	0.96	19.6	0.39	A
64	42	Farr	1.0	0.7	2.3	<0.07	A
46	158	ELISA	0.19	0.99	21.6	0.82	A
62	2500	Crithidia	ND	1.0	ND	ND	A
37	124	Crithidia	0.97	0.93	13.8	0.03	A
		Farr	0.68	0.85	4.5	0.38	
63	62	Farr	ND	0.95	ND	ND	A
Weighted means	–	–	0.573	0.974	16.4	0.49	–

* Sen = sensitivity; spec = specificity; LR = likelihood ratio; ELISA = enzyme-linked immunosorbent assay; ND = not determined (e.g., only 1 population, either SLE or controls, was assessed).

specificity. The positive likelihood ratios (LR) for anti-DNA antibodies in the diagnosis of SLE are very high. Most LR were >10, and the weighted mean positive LR was 16.3 for the grade A articles. This indicates that a positive test result will likely have a large impact on the pretest probability; consequently a positive test result will substantially increase the posttest probability of the diagnosis being SLE. Thus, in the setting of some clinical suspicion of SLE, a positive anti-DNA strongly supports the diagnosis.

The sensitivity of the anti-DNA varies substantially among the studies, with a mean sensitivity of 57.3%. This probably depends on a number of factors, particularly the specific population assessed. The negative LR also vary, but most cluster about 0.5 or higher. The weighted mean negative LR was 0.49. Given this small negative likelihood ratio, a negative test for anti-DNA antibodies does not offer strong support to exclude the diagnosis of SLE.

Interestingly, the development of anti-DNA antibodies has been shown to antedate the clinical diagnosis of SLE in some cases (65).

In studies reporting titers or units, the specificity increases with higher concentrations of anti-DNA antibodies. Although uncommonly found in other conditions, clinicians should be aware that a positive anti-DNA may be rarely seen in patients with other conditions, particularly when present in low titer. This is reflected in the range of specificities in Table 1, which overall are close to, but not equal to 100%. Therefore, a positive anti-DNA is not diagnostic of SLE, and it must be interpreted in the context of the clinical presentation.

Several studies have assessed the prevalence of anti-DNA antibodies in racially distinct populations of SLE patients (66–70). Although the reported prevalence of anti-DNA antibodies has varied among the populations, those studies that actually assessed different races in a single study have found comparable prevalences of anti-DNA antibodies among SLE patients (67,69,70).

Several studies have addressed the potential utility of testing for anti-DNA in patients without a positive antinuclear antibody (ANA) test (41,64,71–73). Using the Hep-2 substrate for ANA, the prevalence of patients with a positive anti-DNA assay despite a negative ANA test has been reported to be 0–0.8% (41,71). In older studies that used less sensitive rodent substrates, frequencies of anti-DNA among ANA negative patients has been reported as 3–8% (64,72,73). Therefore, unless there is reasonable suspicion that the ANA may be falsely negative, anti-DNA antibody testing is not generally indicated in ANA-negative patients.

Recommendations. Anti-DNA antibodies are very useful for the diagnosis of SLE. They are particularly useful to confirm the diagnosis for a patient whose clinical presentation already suggests a reasonable pretest likelihood of the diagnosis of SLE being present (e.g., 5% or more). While offering very strong support in the correct clinical setting, anti-DNA antibodies have been rarely described in a variety of other conditions; therefore a positive anti-DNA is not diagnostic of SLE by itself. Not all patients with SLE have positive anti-DNA antibodies; therefore a negative anti-DNA does not exclude the diagnosis of SLE. Anti-DNA antibodies are not useful for the diagnosis of other conditions. In general, anti-DNA antibody testing should be reserved for patients with a positive ANA.

Prognosis. From the literature search, 31 studies assessing the correlation between anti-DNA antibodies and some aspect of prognosis in patients with SLE met the criteria for further review. Of these 31 studies, 8

Table 2. Use of anti-DNA antibodies for prognosis among systemic lupus erythematosus patients

		Overall SLE active vs inactive				Renal involvement present vs. absent				Renal disease active vs. inactive				
Reference	Technique	Sen	Spec	+LR	−LR	Sen	Spec	+LR	−LR	Sen	Spec	+LR	−LR	Grade
61	ELISA	0.75	0.75	3.0	0.33	NA	NA	NA	NA	NA	NA	NA	NA	A
36	ELISA	0.69	0.77	3.05	0.4	NA	NA	NA	NA	NA	NA	NA	NA	A
	Farr	0.98	0.97	25.2	0.02	NA	NA	NA	NA	NA	NA	NA	NA	
	Crithidia	0.56	0.97	24.1	0.45	NA	NA	NA	NA	NA	NA	NA	NA	
14	Crithidia	0.62	0.75	1.8	0.5	NA	NA	NA	NA	0.92	0.55	2.1	0.14	A
17	ELISA	0.32	0.64	0.88	1.06	NA	NA	NA	NA	NA	NA	NA	NA	A
	Crithidia	0.14	0.91	1.55	0.94	NA	NA	NA	NA	NA	NA	NA	NA	
	Farr	0.41	0.73	1.5	0.81	NA	NA	NA	NA	NA	NA	NA	NA	
15	ELISA	0.92	0.44	1.6	0.18	NA	NA	NA	NA	0.97	0.44	1.7	0.07	A
	Farr	0.73	0.72	2.6	0.38	NA	NA	NA	NA	0.76	0.72	2.7	0.33	
74	ELISA	0.71	0.33	1.05	0.88	0.91	0.08	0.99	1.12	NA	NA	NA	NA	A
	Crithidia	0.43	0.6	1.07	0.95	0.46	0.6	1.15	0.9	NA	NA	NA	NA	
75	Farr	0.89	0.25	1.2	0.44	NA	NA	NA	NA	NA	NA	NA	NA	A
37	Crithidia	1.00	0.13	1.15	0.38	NA	NA	NA	NA	NA	NA	NA	NA	A
	Farr	0.89	0.4	1.48	0.28	NA	NA	NA	NA	NA	NA	NA	NA	
44	Farr	NA	NA	NA	NA	0.82	0.18	1.0	0.97	0.91	0.33	1.4	0.26	B*
50	Crithidia	0.74	0.95	14.8	0.27	0.88	0.41	1.5	0.29	NA	NA	NA	NA	B
77	Crithidia	NA	NA	NA	NA	0.85	0.33	2.6	0.45	NA	NA	NA	NA	B
76	Farr	0.76	0.66	2.28	0.36	NA	NA	NA	NA	0.82	0.32	1.2	0.56	B
78	RIA	0.68	0.82	3.76	0.39	NA	NA	NA	NA	NA	NA	NA	NA	B
79	Crithidia	NA	NA	NA	NA	0.53	0.64	1.46	0.73	NA	NA	NA	NA	B
80	ELISA	0.76	0.54	1.65	0.44	NA	NA	NA	NA	NA	NA	NA	NA	B
82	RIA	0.33	0.68	1.03	0.97	0.44	0.57	1.02	0.98	NA	NA	NA	NA	B
81	RIA	0.93	0.78	3.48	0.1	NA	NA	NA	NA	NA	NA	NA	NA	B
83	Farr	0.2	0.88	1.6	0.9	0.25	0.93	3.6	0.8	NA	NA	NA	NA	B
Weighted means		0.66	0.66	4.14	0.51	0.65	0.41	1.7	0.76	0.86	0.45	1.7	0.3	

* For studies assessing several facets of DNA testing (e.g., diagnostic and prognostic use) grades were assigned independently. This study was considered an "A" study for diagnosis, but a "B" study for prognosis.
NA = not assessed. See Table 1 for additional definitions.

were graded "A" and 11 were graded "B" (Table 2) (15,16,18,37,38,44,50,61,74–83), and form the basis of the recommendations.

After review and grading of the literature, it was considered that there were sufficient studies to assess the utility of anti-DNA antibodies in the measurement of several aspects of prognosis in patients with SLE. Measures of prognosis that were considered include: active versus inactive overall disease, presence of renal involvement, and active versus inactive renal disease.

There are several important caveats relevant to the interpretation of these studies. The presence of anti-DNA antibodies was often included as part of the criteria by which active disease was defined. This tautology makes it harder to separately assess the correlation of anti-DNA antibodies with disease activity. Also, there has been no single, universally accepted definition of active disease for SLE patients. Because of the substantial variability on definitions of activity, the authors' definitions of activity were accepted, provided they were explicitly defined. In many studies, any potential effect of immunomodulatory therapy or other treatment on the disease activity and/or laboratory test results is impossible to extract, as such information is often not specifically provided. Therefore, this has not been specifically addressed. Many studies provide limited information on patient selection criteria for the SLE patients reported, thus raising the potential for bias in the results.

Considering overall activity of SLE, as variously defined among the different studies, the data from these studies indicate that the presence of anti-DNA antibodies is associated with increased disease activity. However, the sensitivity and specificity vary among studies (weighted mean sensitivity and specificity were both 0.66). Positive LRs varied from approximately 0.88 to more than 10. This indicates that in some studies the presence of anti-DNA antibodies was strongly predictive of active disease while in others it had no prognostic significance whatsoever. This may result from heterogeneity in the populations of SLE patients assessed in the various studies. Thus, although anti-DNA antibodies can be associated with disease activity, there are clearly populations of SLE patients who have persistently elevated anti-DNA antibodies but do not have active disease (84,85).

The weighted mean positive LR was 4.14. This implies that the results of the test may alter the pretest likelihood of the determination of disease activity, but that the effect will be relatively small. A positive anti-DNA would be anticipated to be useful in assessing prognosis only in the correct clinical setting (i.e., if there are indications of disease activity from other clinical assessments). Of note, with one exception (75), higher titers of anti-DNA antibod-

ies were more strongly associated with active SLE in the studies reporting titers (15,41,44,50,76,86). Therefore, a higher threshold for a positive anti-DNA would be expected to increase its specificity, and perhaps be more predictive of SLE disease activity.

Also, in the 2 grade "A" studies that directly compared all 3 methods for anti-DNA determination (19,38), the performance characteristics for the Farr and Crithidia assays were superior to that of the ELISA (Table 2). The weighted mean negative LR for the association between anti-DNA and overall SLE disease activity was 0.51. An interpretation of this would be that although a negative test result does not exclude disease activity, it has a small but potentially relevant association with the lack of disease activity. Therefore, a negative test for anti-DNA would be most helpful in the setting of a low pretest probability of disease activity.

Studies analyzing the correlation of anti-DNA antibodies with the presence of renal disease had weighted mean sensitivity of 0.65, specificity of 0.41, positive LR of 1.7, and a negative LR of 0.76. Therefore, although there was some variability among the studies, the presence of a positive test for anti-DNA antibodies only slightly increases the likelihood that a SLE patient has renal disease; a negative anti-DNA test, by itself, does little to exclude the presence of renal disease. By extension, testing for anti-DNA in order to assess whether renal involvement was present in an SLE patients would be of most value (and should be limited to) patients with a reasonable pre-existing suspicion of renal involvement.

Several studies have assessed the association of anti-DNA antibodies with specific histopathologic measures of renal disease in SLE (some of these studies report only correlation coefficients or other summary statistics, but not primary data). Anti-DNA antibodies have been reported to correlate with renal activity (but not chronicity) index (83,87), and to have some correlation with WHO type IV (diffuse proliferative) glomerulonephritis (87,90). However, although correlations may have been statistically significant, anti-DNA antibody positive patients were noted among all histopathologic types.

In the one study (87) from which data could be clearly extracted according to WHO pathologic changes, the statistics for the association of anti-DNA antibodies with type IV glomerulonephritis (GN) (as compared to all other types) were: by Crithidia sensitivity 0.93, specificity 0.6, positive LR 2.3; and by ELISA sensitivity 1.0, specificity 0.04, positive LR 1.04. Higher titers of anti-DNA were more clearly associated with type IV GN. While determinations of serum complement protein concentrations were not specifically considered in this analysis, it has been suggested that the combination of increased titers of anti-DNA in conjunction with depressed levels of complement proteins may be more predictive of active lupus nephritis (91,92). This issue was not specifically addressed in this review.

Considering the activity of known renal disease in SLE patients, weighted means for anti-DNA were 0.86 sensitivity, 0.45 specificity, 1.7 positive LR, and 0.3 negative LR. Thus, as for overall disease activity, the presence of anti-DNA antibodies may increase the pretest-to-posttest likelihood of active renal disease in a patient with SLE, but the effect will be small. Although the number of studies was small, the negative LR was more relevant for determining activity of known renal disease as opposed to determining whether renal involvement was present at all. In the studies reporting titers (15,44,76,86), higher titers were often seen in patients with more active disease.

Several studies assessed the impact of anti-DNA antibodies on overall outcome or survival of SLE patients (75,79,93–96). Although the numbers of patients in each group in some of the studies were small, there was no significant effect of anti-DNA antibody on outcome or survival.

Recommendations. Anti-DNA antibodies correlate with overall activity of disease in patients with SLE, and they are useful for this purpose. However, the LRs for this correlation are relatively small. This implies that the finding of anti-DNA will have a limited impact on the pretest likelihood of active disease for a given patient with SLE. The anti-DNA should optimally be used for patients with some pretest likelihood of active SLE (i.e., those patients for whom active SLE is suspected on clinical grounds otherwise, placing the patient at a pretest likelihood of disease activity of 10% or greater). In SLE patients with no other evidence of disease activity, a positive anti-DNA is unlikely to make important changes on the overall impression of disease activity. In patients with a positive anti-DNA, higher titers were more closely associated with disease activity. As a guide to overall disease activity, anti-DNA should be interpreted in SLE patients only in conjunction with suspicion of disease activity based on history, physical examination, or the results of other laboratory tests.

Anti-DNA antibodies correlate with the presence and activity of renal disease in patients with SLE, and they are useful for this purpose. However, the LRs for this association are very small, implying that the finding of a positive anti-DNA may add little to the overall impression of renal disease or its activity. Although anti-DNA antibodies have also been associated with histopathologic type of lupus nephritis as well as a pathologic activity index, the literature in this area is not extensive, and precludes analysis that would be of the greatest assistance to the practitioner. In patients with a positive anti-DNA, higher titers were more closely associated with the presence and activity of renal disease. As a guide to renal disease and its activity in SLE patients, anti-DNA may be useful; however, it must be interpreted in conjunction with other measures of renal disease.

Anti-DNA antibodies do not correlate with overall survival or outcome in SLE, and are not useful for this purpose.

Longitudinal Assessment

From the literature search, 33 studies assessing anti-DNA antibodies and some measure of disease activity longitudinally in patients with SLE met the criteria for further review. Of these 33, 3 studies were graded "A" and 5 were graded "B" (Table 3) (17,75,91,97–107). A number of published studies have attempted to evaluate the potential use

Table 3. Use of anti-DNA antibodies for longitudinal assessment of SLE patients*

Reference	Technique	Data/Comments	Grade
97	Farr	156 patients; anti-DNA was checked monthly; 102 patients who were negative for anti-DNA or had no change in titer had 8 flares; patients with increasing anti-DNA were divided into standard followup or treatment with steroids; 20 flares (13 minor, 7 major) seen in 22 conventional patients, 2 major flares in 16 patients steroid treated; 17% in conventional group had increased anti-DNA but did not flare	A
16	Farr, Crithidia, ELISA	72 patients; anti-DNA tested monthly by 3 methods; 17 patients had 33 flares, 27 of which were accompanied by an increase in anti-DNA; 9 episodes of increased anti-DNA were not followed by a flare; 9 patients had flares without increases in anti-DNA	A
107	Crithidia, ELISA	53 SLE patients followed monthly for 1 year. 5 different measures of disease activity were used to define flare; rate of flare varied from 12% of visits to 25% of visits using different instruments; decrease in anti-DNA at the time of the visit correlated with flare using 3 instruments; previous increase in anti-DNA correlated with flare with 2 instruments; strength of correlation between changes in anti-DNA and flare was modest, even when statistically significant (e.g., positive likelihood ratios for previous increase in anti-DNA and flare varied from 1.0 to 2.7 using the different instruments)	A
91	Crithidia	27 patients followed over 47 cycles defined according to activity and end-organ; anti-DNA titers correlated only slightly with variations in disease activity	B
75	Farr	99 patients, 8 had flares; anti-DNA increased in 1, decreased in 2 and did not change in 4 preceding the flare.	B
98	Farr	78 patients, anti-DNA checked every 6 weeks or more frequently; 28 patients who were anti-DNA negative had 9 flares; 50 anti-DNA positive patients all had flares; major symptoms occurred mostly in the setting of decreasing anti-DNA (43/46 episodes); increasing anti-DNA was seen in quiet disease (2 patients), minor symptoms (7 patients) and major symptoms (3 patients); constant anti-DNA was seen in quiet disease (10 patients) and minor symptoms (8 patients)	B
99	Farr	130 patients; anti-DNA checked every 6 weeks or more; of 106 patients who had no increase in anti-DNA (including 50 who had persistently high anti-DNA) none flared; 24 patients had increases in anti-DNA, 15 flares occurred in 13 patients, all preceded by increasing anti-DNA; anti-DNA increased in 4 patients without flare	B
100	Farr	202 patients; 83 flares seen among 53 patients; anti-DNA at 3 month intervals modeled statistically to see if changes predicted flare; changes in anti-DNA were poor predictors of flare	B

* See Table 1 for definitions.

of anti-DNA antibodies in the longitudinal assessment of patients with SLE; however, the articles presented data from single or "representative" patients. Because of the substantial risk of reporting bias, such reports presenting only partial data were not considered suitable for this analysis.

There are additional caveats that are critical to the interpretation of studies showing longitudinal data for a more complete population of patients with SLE. Several concerns noted above in the section on prognosis also apply to studies addressing longitudinal followup, including 1) the use of diverse definitions of disease activity, 2) the inclusion of anti-DNA antibodies in the definition of disease activity, 3) the potential effects of therapy on disease activity and/or test results, and 4) possible selection bias in the populations studied. In addition, just as there has been no universal definition of disease activity, there is no universal definition of a flare of SLE. This has considerable impact on the interpretation of data from longitudinal studies; for example, rates of flare vary substantially among the studies. Many studies purportedly showing longitudinal data are actually cross-sectional studies (101). Because, as noted above, the presence of anti-DNA antibodies can correlate with disease activity, such studies would be confirming this relationship rather than providing information concerning flares or longitudinal followup.

Although differences in trial design preclude compilation of the data, it appears that in most of the studies, there was some correlation between changes in titers of anti-DNA antibodies and flare of disease activity. However, despite this association, it is clear that changes in anti-

DNA antibodies are not by themselves diagnostic of a flare. Thus there are individual patients who have flares without changes in anti-DNA antibody titer and vice versa. Because the number of articles is limited, there is a need for further research in this area.

A number of other studies have addressed the issue of longitudinal use of anti-DNA in SLE, but were not included in the table because they presented predominantly summary or derivative statistics. In one study (102), 21 patients were divided according to the pattern of anti-DNA antibodies over time; of the 14 patients with titers that remained low or fell to low levels over time, 12 had a favorable prognosis, whereas all 7 of those with persistently high or fluctuating anti-DNA titers deteriorated. In another study (103), 25 patients with elevated anti-DNA were treated with prednisone and/or immunosuppressive drugs in an attempt to decrease the titers of anti-DNA (and increase levels of complement proteins). Although they were able to normalize the anti-DNA level in 16 patients, they were unable to achieve this in 9 other patients. There was no substantial or consistent difference in creatinine or urinary protein between the groups.

As noted, SLE patients with serologic activity (elevated anti-DNA and/or decreased complement) but no clinical activity have been described (84). Longitudinal followup of a group of these patients revealed that high anti-DNA titers did not predict flare (85). In a study of 198 patients followed over 1,654 visits, no single laboratory value (including anti-DNA) predicted flare (104). In a study of 48 patients followed over 6–18 months, anti-DNA antibodies were not predictive of changes in disease activity (82). In a prospective study (105) of 53 patients, asymptomatic patients with high levels of anti-DNA experienced flares of disease activity more commonly than other patients (odds ratio 3.2; 95% confidence interval 1.7–5.3). In a study of 16 patients with childhood onset SLE followed over time, anti-DNA had a statistically significant linear correlation with disease activity (106). Characteristics other than rise in anti-DNA titer that have been suggested to correlate with flare include the rate of change of the rise in anti-DNA and anti-DNA titers that increase but subsequently decrease.

Recommendations. The presence of a positive anti-DNA antibody does not predict subsequent flares of disease activity in SLE. Although the number of relevant studies is small, it does appear that rising titers of anti-DNA antibodies can antedate or be associated with an increased risk of flares of disease activity. Therefore, longitudinal assessment of anti-DNA antibody titers may be useful in the care of SLE patients. However, there are clearly subsets of SLE patients who have flares without increases in anti-DNA, as well as patients who have increases in anti-DNA titers without flares of disease activity. In addition, the correlations between alterations in anti-DNA and flares of disease tend to be modest. Thus, the data at present do not support the concept of using alterations in anti-DNA titer to predict or diagnosis flare independent of clinical evaluation. Nor are the data sufficiently strong to endorse changes in therapeutic regimens based solely on alterations in anti-DNA (i.e. independent of clinical evaluation). Changes in titers of anti-DNA should be optimally interpreted in the context of information obtained from the history, physical examination, and other laboratory investigations. There are insufficient data to make recommendations concerning the optimum frequency of testing anti-DNA in patients with established disease in order to assess disease activity longitudinally. However, if such testing is performed, the results should be interpreted in the overall clinical context.

There are a number of important questions related to the optimal use of anti-DNA antibodies in the longitudinal assessment of SLE patients that cannot be answered on the basis of the available literature. Although these areas could benefit substantially from additional research, at present no recommendations can be given for these questions. Areas that require further investigation include: 1) What is the optimal frequency for the determination of anti-DNA antibodies (e.g., monthly, every 3 months, no fixed interval)? 2) Are there subsets of patients for whom serial determination of anti-DNA antibodies would be particularly useful? Recently, some studies related to serial anti-DNA determinations have introduced therapeutic arms (i.e., altering therapy based upon changes in anti-DNA titers). Additional research in this and other areas should include a complete analysis of all of the potential risks and benefits of such an approach.

Conclusions

Anti-DNA testing can be very useful for the diagnosis of SLE. Whereas a positive test for anti-DNA offers strong support for the diagnosis of SLE, a negative test result does not exclude the diagnosis. Anti-DNA testing should be reserved for patients who have a positive ANA. Anti-DNA antibodies do correlate with overall disease activity in SLE. However, as the correlations are at best modest, test results must be interpreted in the overall clinical context. Similarly, anti-DNA antibodies correlate with the activity of renal disease in SLE, but to a limited extent. Higher titers of anti-DNA have a stronger association with disease activity. Concerning longitudinal assessment, the presence of a positive test for anti-DNA does not predict a flare of disease. Increasing titers of anti-DNA may antidate or be associated with flares of disease activity. However, the number of high quality studies addressing this issue is limited, and a number of important questions concerning the optimal use of anti-DNA testing longitudinally remain to be answered.

REFERENCES

1. American College of Rheumatology Ad Hoc Committee on Immunologic Testing Guidelines. Guidelines for immunologic laboratory testing in the rheumatic diseases: an introduction. Arthritis Rheum (Arthritis Care Res) 47;4:429–33.
2. Robbins W, Holman H, Deicher H, Kunkel H. Complement fixation with cell nuclei and DNA in lupus erythematosus. Proc Soc Exp Biol Med 1957;96:575–89.
3. Deicher H, Holman H, Kunkel H. The precipitin reaction between DNA and a serum factor in systemic lupus erythematosus. J Exp Med 1959;109:97–114.
4. Casals SP, Friou GJ, Myers LL. Significance of antibody to DNA in systemic lupus erythematosus. Arthritis Rheum 1964;7:379–90.

5. Tan E, Schur P, Carr R, Kunkel H. Deoxyribonucleic acid (DNA) and antibodies to DNA in the serum of patients with systemic lupus erythematosus. J Clin Invest 1966;45:1732–40.
6. Koffler D, Schur P, Kunkel H. Immunological studies concerning the nephritis of systemic lupus erythematosus. J Exp Med 1967;126:607–24.
7. Harbeck R, Bardana E, Kohler P, Carr R. DNA, Anti-DNA complexes: their detection in systemic lupus erythematosus sera. J Clin Invest 1973;52:789–95.
8. Maini R, Holborow E. Detection and measurement of circulating soluble antigen-antibody complexes and anti-DNA antibodies. Ann Rheum Dis 1977;36 Suppl:S1–142.
9. Schur PH, Sandson J. Immunologic factors and clinical activity in systemic lupus erythematosus. N Engl J Med 1968; 278:533–8.
10. Hahn BH. Antibodies to DNA. N Engl J Med 1998;338:1359–68.
11. Tan EM, Cohen AS, Fries JF, Masi AT, McShane DJ, Rothfield NF, et al. The 1982 revised criteria for the classification of systemic lupus erythematosus. Arthritis Rheum 1982;25: 1271–7.
12. Edworthy S, Zatarain E, McShane D, Bloch D. Analysis of the 1982 ARA lupus criteria data set by recursive partitioning methodology: new insights into the relative merits of individual criteria. J Rheumatol 1988;15:1493–8.
13. Emlen W, Pisetsky DS, Taylor RP. Antibodies to DNA: a perspective. Arthritis Rheum 1986;29:1417–26.
14. Isenberg DA, Ehrenstein MR, Longhurst C, Kalsi JK. The origin, sequence, structure, and consequences of developing anti-DNA antibodies: a human perspective. Arthritis Rheum 1994;37:169–80.
15. Chubick A, Sontheimer RD, Gilliam JN, Ziff M. An appraisal of tests for native DNA antibodies in connective tissue diseases: clinical usefulness of Crithidia luciliae assay. Ann Intern Med 1978;89:186–92.
16. Miller TE, Lahita RG, Zarro VJ, MacWilliam J, Koffler D. Clinical significance of anti–double-stranded DNA antibodies detected by a solid phase enzyme immunoassay. Arthritis Rheum 1981;24:602–10.
17. Ter Borg EJ, Horst G, Hummel EJ, Limburg PC, Kallenberg CGM. Measurement of increases in anti–double-stranded DNA antibody levels as a predictor of disease exacerbation in systemic lupus erythematosus: a long-term, prospective study. Arthritis Rheum 1990;33:634–43.
18. Isenberg D, Dudeney C, Williams W, Addison W, Charles S, Clarke J, et al. Measurement of anti-DNA antibodies: a reappraisal using five different methods. Ann Rheum Dis 1987; 46:448–56.
19. Feltkamp T, Kirkwood T, Maini R, Aarden L. The first international standard for antibodies to double stranded DNA. Ann Rheum Dis 1988;47:740–6.
20. Cronin M, Leair D, Jaronski S, Lightfoot R. Simultaneous use of multiple serologic tests in assessing clinical activity in systemic lupus erythematosus. Clin Immun Immunopathol 1989;51:99–109.
21. Sommerfield S, Roberts M, Booth R. Double-stranded DNA antibodies: a comparison of four methods of detection. J Clin Pathol 1981;34:1032–5.
22. Davis P, Russell A, Percy J. A comparative study of techniques for the detection of antibodies to native deoxyribonucleic acid. Am J Clin Pathol 1977;67:374–8.
23. Monier J, Sault C, Veysseyre C, Bringuier J. Discrepancies between two procedures for Ds-DNA antibody detection: Farr test and indirect immunofluorescence on Crithidia lucillae. J Clin Lab Immunol 1988;25:149–52.
24. Smeenk R, van der Lelij G, Aarden L. Avidity of antibodies to dsDNA: comparison of IFT on Crithidia luciliae, Farr assay, and PEG assay. J Immunol 1982;128:73–8.
25. Ward M, Pisetsky D, Christenson V. Antidouble stranded DNA antibody assays in systemic lupus erythematosus: correlations of longitudinal antibody measurements. J Rheumatol 1989;16:609–13.
26. Lachman P. Measurement of anti-DNA antibodies: report on the organization and results of an Arthritis and Rheumatism Council workshop study (1976). Ann Rheum Dis 1977;36 Suppl:S67–75.
27. Kadlubowski M, Jackson M, Yap P, Neill G. Lack of specificity for antibodies to double stranded DNA found in four commercial kits. J Clin Pathol 1991;44:246–50.
28. Smeenk R, Brinkman K, van den Brink H, Swaak T. A comparison of the assays used for the detection of antibodies to DNA. Clin Rheumatol 1990;9 Suppl:63–72.
29. Tipping P, Buchanan R, Riglar A, Dimech W, Littlejohn G. Measurement of anti-DNA antibodies by ELISA: a comparative study with Crithidia and a Farr assay. Pathology 1991; 23:21–4.
30. McMillan S, Fay A. Evaluation of five commercial kits to detect dsDNA antibodies. J Clin Pathol 1988;41:1223–8.
31. Takeuchi Y, Ishikawa O, Miyachi Y. The comparative study of anti-double stranded DNA antibody levels measured by radioimmunoassay and enzyme-linked immunosorbent assay in systemic lupus erythematosus. J Dermatol 1997;24: 297–300.
32. Tzioufas A, Terzoglou C, Stavropoulos E, Athanasiadou S, Moutsopoulos H. Determination of anti-ds-DNA antibodies by three different methods: comparison of sensitivity, specificity, and correlation with lupus activity index. Clin Rheumatol 1990;9:186–92.
33. Eaton RB, Schneider G, Schur PH. Enzyme immunoassay for antibodies to native DNA: specificity and quality of antibodies. Arthritis Rheum 1983;26:52–62.
34. Clarke MC, Carr R, Burdash NM, Chen ZY, Ainsworth SK. A comparison of three anti-double stranded DNA antibody assays on sera from SLE and other diseases. Diagn Immunol 1986;4:288–93.
35. Wong K, Lawton J, Cheng S, Lau C. Measurement of anti-dsDNA: a comparative study of two ELISA and the Crithidae assay. Pathology 1998;30:57–61.
36. Emlen W, Jarusiripipat P, Burdick G. A new ELISA for the detection of double-stranded DNA antibodies. J Immunol Methods 1990;132:91–101.
37. Bootsma H, Spronk P, Hummel E, de Boer G, ter Borg E, Limburg P, Kallenberg C. Anti-double stranded DNA antibodies in systemic lupus erythematosus: detection and clinical relevance of IgM-class antibodies. Scand J Rheumatol 1996;25:352–9.
38. Whiteside T, Dixon J. Clinical usefulness of the Crithidia luciliae test for antibodies to native DNA. Am J Clin Pathol 1979;72:829–35.
39. Halbert S, Karsh J, Anken M. Studies on autoantibodies to deoxyribonucleic acid and deoxyribonucleoprotein with enzyme-immunoassay (ELISA). J Lab Clin Med 1981;97:97–111.
40. Hoch S, Schur P, Schwaber J. Frequency of anti-DNA antibody producing cells from normals and patients with systemic lupus erythematosus. Clin Immunol Immunopathol 1983;27:28–37.
41. Juby A, Johnston C, Davis P. Specificity, sensitivity and diagnostic predictive value of selected laboratory generated autoantibody profiles in patients with connective tissue diseases. J Rheumatol 1991;18:354–8.
42. Koffler D, Carr R, Agnello V, Fiezi T, Kunkel H. Antibodies to polynucleotides: distribution in human serums. Science 1969;166:1648–9.
43. Grennan DM, Sloane D, Behan A, Dick WC. Clinical significance of antibodies to native DNA as measured by a DNA binding technique in patients with articular features of rheumatoid arthritis. Ann Rheum Dis 1977;36:30–3.
44. Pincus T, Schur PH, Rose JA, Decker JL, Talal N. Measurement of serum DNA-binding activity in systemic lupus erythematosus. N Engl J Med 1969;281:701–5.
45. Weinstein A, Bordwell B, Stone B, Tibbetts C, Rothfield NF. Antibodies to native DNA and serum complement (C3) levels: application to diagnosis and classification of systemic lupus erythematosus. Am J Med 1983;74:206–16.
46. Le Page SH, Williams W, Parkhouse D, Cambridge G, MacKenzie L, Lydyard PM, et al. Relation between lymphocyto-

toxic antibodies, anti-DNA antibodies and a common anti-DNA antibody idiotype PR4 in patients with systemic lupus erythematosus, their relatives and spouses. Clin Exp Immunol 1989;77:314–8.
47. Williams WM, Isenberg DA. A cross-sectional study of anti-DNA antibodies in the serum and IgG and IgM fraction of healthy individuals, patients with systemic lupus erythematosus and their relatives. Lupus 1996;5:576–86.
48. Koffler D, Carr R, Agnello V, Thoburn R, Kunkel HG. Antibodies to polynucleotides in human sera: antigenic specificity and relation to disease. J Exp Med 1971;134:294–312.
49. Hasselbacher P, LeRoy EC. Serum DNA binding activity in healthy subjects and in rheumatic disease. Arthritis Rheum 1974;17:63–71.
50. Ballou SP, Kushner I. Anti-native DNA detection by the Crithidia luciliae method: an improved guide to the diagnosis and clinical management of systemic lupus erythematosus. Arthritis Rheum 1979;22:321–7.
51. Mecocci P, Ekman R, Parnetti L, Senin U. Antihistone and anti-dsDNA autoantibodies in Alzheimer's disease and vascular dementia. Biol Psychiatry 1993;34:380–5.
52. Hughes GR, Cohen SA, Christian CL. Anti-DNA activity in systemic lupus erythematosus: a diagnostic and therapeutic guide. Ann Rheum Dis 1971;30:259–64.
53. Carr RI, Hoffmann AA, Harbeck RJ. Comparison of DNA binding in normal population, general hospital laboratory personnel, and personnel from laboratories studying SLE. J Rheumatol 1975;2:178–83.
54. Notman DD, Kurata N, Tan EM. Profiles of antinuclear antibodies in systemic rheumatic diseases. Ann Intern Med 1975;83:464–9.
55. Epstein WV, Tan M, Easterbrook M. Serum antibody to double-stranded RNA and DNA in patients with idiopathic and secondary uveitis. N Engl J Med 1971;285:1502–6.
56. Davis P, Read AE. Antibodies to double-stranded (native) DNA in active chronic hepatitis. Gut 1975;16:413–5.
57. Lange A. Evaluation of the simultaneous estimation of anti-dsDNA and anti-ssDNA antibodies for clinical purposes. Clin Exp Immunol 1978;31:472–81.
58. Ruffatti A, Calligaro A, del Ross T, Bertoli M, Doria A, Rossi L, Todesco S. Anti-double-stranded DNA antibodies in the healthy elderly: prevalence and characteristics. J Clin Immunol 1990;10:300–3.
59. Ehrenstein M, Swana M, Keeling D, Asherson R, Hughes G, Isenberg D. Anti DNA antibodies in the primary antiphospholipid syndrome. Br J Rheumatol 1993;32:362–5.
60. Cuellar M, Scopelitis E, Tenebaum S, Garry R, Silveira L, Cabrera G, et al. Serum antinuclear antibodies in women with silicone breast implants. J Rheumatol 1995;22:236–40.
61. Froelich CJ, Wallman J, Skosey JL. Clinical value of an integrated ELISA system for the detection of 6 autoantibodies (ssDNA, dsDNA, Sm, RNP/Sm, SSA, and SSB). J Rheumatol 1990;17:192–200.
62. Fritzler MJ. Antinuclear, anticytoplasmic, and anti-Sjögren's syndrome antigen A (SS-A/Ro) antibodies in female blood donors. Clin Immunol Immunopathol 1988;36:120–8.
63. Rochimis PG. Native DNA binding in rheumatoid arthritis. Ann Rheum Dis 1974;33:357–60.
64. Davis P, Atkins B, Hughes GR. Antibodies to native DNA in discoid lupus erythematosus. Br J Derm 1974;91:175–81.
65. Swaak T, Smeenk R. Detection of anti-dsDNA as a diagnostic tool: a prospective study in 441 non-systemic lupus erythematosus patients with anti-dsDNA antibody. Ann Rheum Dis 1985;44:245–51.
66. Al-attia H. Clinical laboratory profile of 33 Arabs with systemic lupus erythematosus. Postgrad Med J 1996;72:677–9.
67. Ward M, Studenskis S. Clinical manifestations of systemic lupus erythematosus: identification of racial and socioeconomic influences. Arch Intern Med 1990;150:849–53.
68. Boey ML, Peebles CL, Tsay G, Feng PH, Tan EM. Clinical and autoantibody correlations in Orientals with systemic lupus erythematosus. Ann Rheum Dis 1988;47:918–23.
69. Hochberg MC, Boyd RE, Ahearn JM, Arnett FC, Bias WB, Provost TT, et al. Systemic lupus erythematosus: a review of clinico-laboratory features and immunogenetic markers in 150 patients with emphasis on demographic subsets. Medicine (Baltimore) 1985;64:285–95.
70. Molina JF, Molina J, Garcia C, Gharavi AE, Wilson WA, Espinoza LR. Ethnic differences in the clinical expression of systemic lupus erythematosus: a comparative study between African-Americans and Latin Americans. Lupus 1997;6:63–7.
71. Manoussakis M, Garalea K, Tzioufas A, Moutsopoulos H. Testing for antibodies to ENA and to dsDNA is not indicated in FANA-negative sera. Clin Rheumatol 1988;7:465–9.
72. Rigby R, Dawkins R, Kay P, Matz L, Papadimitriou J, Quintner J, et al. The contribution of immunopathology to the diagnosis of systemic lupus erythematosus: relationship between serum anti-DNA, complement and renal biopsy changes. Aust N Z J Med 1978;8:29–38.
73. Maddison PJ, Provost TT, Reichlin M. Serological findings in patients with "ANA-negative" systemic lupus erythematosus. Medicine (Baltimore) 1981;60:87–94.
74. Kalmin N, Bartholomew W, Wicher K. Relative values of laboratory assays in systemic lupus erythematosus. Am J Clin Pathol 1981;75:846–51.
75. Cameron JS, Lessof MH, Ogg CS, Williams BD, Williams DG. Disease activity in the nephritis of systemic lupus erythematosus in relation to serum complement concentrations. DNA-binding capacity and precipitating anti-DNA antibody. Clin Exp Immunol 1976;25:418–27.
76. Weitzman R, Walker S. Relation of titered peripheral pattern ANA to anti-DNA and disease activity in systemic lupus erythematosus. Ann Rheum Dis 1977;36:44–9.
77. Garcia CO, Molina JF, Gutierrez-Urena S, Scopelitis E, Wilson WA, Gharavi AE, et al. Autoantibody profile in African-American patients with lupus nephritis. Lupus 1996;5:602–5.
78. Miniter MF, Stollar BD, Agnello V. Reassessment of the clinical significance of native DNA antibodies in systemic lupus erythematosus. Arthritis Rheum 1979;22:959–68.
79. Ballou SP, Kushner I. Lupus patients who lack detectable anti-DNA: clinical features and survival. Arthritis Rheum 1982;25:1126–9.
80. Isenberg DA, Shoenfeld Y, Schwartz RS. Multiple serologic reactions and their relationship to clinical activity in systemic lupus erythematosus. Arthritis Rheum 1984;27:132–8.
81. Davis P, Percy JS, Russell AS. Correlation between levels of DNA antibodies and clinical disease activity in SLE. Ann Rheum Dis 1977;36:157–9.
82. Abrass CK, Nies KM, Louie JS, Border WA, Glassock RJ. Correlation and predictive accuracy of circulating immune complexes with disease activity in patients with systemic lupus erythematosus. Arthritis Rheum 1980;23:273–82.
83. Feldman MD, Huston DP, Karsh J, Balow JE, Klima E, Steinberg AD. Correlation of serum IgG, IgM, and anti-native-DNA antibodies with renal and clinical indexes of activity in systemic lupus erythematosus. J Rheumatol 1982;9:52–8.
84. Gladman DD, Urowitz MB, Keystone EC. Serologically active clinically quiescent systemic lupus erythematosus: a discordance between clinical and serologic features. Am J Med 1979;66:210–5.
85. LeBlanc B, Gladman D, Urowitz M. Serologically active clinically quiescent systemic lupus erythematosus—predictors of clinical flares. J Rheumatol 1994;21:2239–41.
86. Bernstein KA, Kahl LE, Balow JE, Lefkowith JB. Serologic markers of lupus nephritis in patients: use of a tissue-based ELISA and evidence for immunopathogenic heterogeneity. Clin Exp Immunol 1994;98:60–5.
87. Okamura M, Kanayama Y, Amastu K, Negoro N, Kohda S, Takeda T, et al. Significance of enzyme linked immunosorbent assay (ELISA) for antibodies to double stranded and single stranded DNA in patients with lupus nephritis: correlation with severity of renal histology. Ann Rheum Dis 1993;52:14–20.
88. Steinman CR, Grishman E, Spiera H, Deesomochok U. Binding of synthetic double-stranded DNA by serum from pa-

tients with systemic lupus erythematosus: correlation with renal histology. Am J Med 1977;62:319–23.
89. Tron F, Bach JF. Relationships between antibodies to native DNA and glomerulonephritis in systemic lupus erythematosus. Clin Exp Immunol 1977;28:426–32.
90. Houssiau FA, D'Cruz D, Vianna J, Hughes GR. Lupus nephritis: the significance of serological tests at the time of biopsy. Clin Exp Rheumatol 1991;9:345–9.
91. Lloyd W, Schur P. Immune complexes, complement, and anti-DNA in exacerbations of systemic lupus erythematosus. Medicine 1981;60:208–17.
92. Schur P, Sandson J. Immunologic factors and clinical activity in systemic lupus erythematosus. N Engl J Med 1968;278: 533–8.
93. Gulko PS, Reveille JD, Koopman WJ, Burgard SL, Bartolucci AA, Alarcon GS. Survival impact of autoantibodies in systemic lupus erythematosus. J Rheumatol 1994;21:224–8.
94. Ludivico CL, Zweiman B, Myers AR, Hebert J, Green PA. Predictive value of anti-DNA antibody and selected laboratory studies in systemic lupus erythematosus. J Rheumatol 1980;7:843–9.
95. Fries JF, Weyl S, Holman HR. Estimating prognosis in systemic lupus erythematosus. Am J Med 1974;57:561–5.
96. Gripenberg M, Heive T. Outcome of systemic lupus erythematosus. A study of 66 patients over 7 years with special reference to the predictive value of anti-DNA antibody determinations. Scand J Rheumatol 1991;20:104–9.
97. Bootsma H, Spronk P, Derksen R, de Boer G, Wolters-Dicke H, Hermans J, et al. Prevention of relapses in systemic lupus erythematosus. Lancet 1995;345:1595–9.
98. Swaak AJG, Aarden LA, Statius van Eps LW, Feltkamp TEW. Anti-dsDNA and complement profiles as prognostic guides in systemic lupus erythematosus. Arthritis Rheum 1979;22: 226–35.
99. Swaak AJ, Groenwold J, Aarden LA, Statius van Eps LW, Feltkamp EW. Prognostic value of anti-dsDNA in SLE. Ann Rheum Dis 1982;41:388–95.
100. Esdaile JM, Abrahamowicz M, Joseph L, MacKenzie T, Li Y, Danoff D. Laboratory tests as predictors of disease exacerbations in systemic lupus erythematosus: why some tests fail. Arthritis Rheum 1996;39:370–8.
101. Esdaile J, Joseph L, Abrahamowicz M, Li Y, Danoff D, Clarke A. Routine immunologic tests in SLE: is there a need for more studies? J Rheumatol 1996;23:1891–6.
102. Adler MK, Baumgarten A, Hecht B, Siegel NJ. Prognostic significance of DNA-binding capacity patterns in patients with lupus nephritis. Ann Rheum Dis 1975;34:444–50.
103. Appel AE, Sablay LB, Golden RA, Barland P, Grayzel AI, Bank N. The effect of normalization of serum complement and anti-DNA antibody on the course of lupus nephritis: a two year prospective study. Am J Med 1978;64:274–83.
104. Petri M, Genovese M, Engle E, Hochberg M. Definition, incidence, and clinical description of flare in systemic lupus erythematosus: a prospective cohort study. Arthritis Rheum 1991;34:937–44.
105. Zonana-Nacach A, Salas M, Sanchez M, Camargo-Coronel A, Bravo-Gatica C, Mintz G. Measurement of clinical activity of SLE and laboratory abnormalities: a 12 month prospective study. J Rheumatol 1995;22:45–9.
106. Ting C-K, Hsieh K-H. A long term immunologic study of childhood onset SLE. Ann Rheum Dis 1992;51:45–51.
107. Ho A, Magder LS, Barr SG, Petri M. Decreases in anti-double-stranded DNA levels are associated with concurrent flare in patients with systemic lupus erythematosus. Arthritis Rheum 2001;44:2342–9.