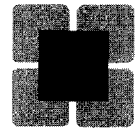

The ANA Profile: quality and cost-effective laboratory utilization

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■ The ANA Profile was introduced in 1981 and computerized in 1984 as a means of facilitating follow-up testing for specific antibodies (anti-nDNA, anti-Sm, anti-RNP, anti-La/SS-B) in sera found to contain antinuclear antibodies (ANA). A second purpose was to avoid unnecessary specific antibody testing on negative or low-titer sera. This study was done to evaluate the effectiveness of the computerized ANA Profile reporting system in accomplishing these purposes. The authors compared ordering practices during two two-week periods, one in 1984 and a second in 1988, and found that follow-up testing on positive sera had improved from 27% to 70% with a reduction in unnecessary specific-antibody testing of ANA-negative or low-titer sera from 11% to 1.6%. In 1988 dollars, the annual savings from eliminating unnecessary testing was calculated to be $12,000. The ANA Profile has been partially successful in accomplishing the purposes for which it was introduced.

□INDEX TERM: ANTINUCLEAR FACTORS □ CLEVE CLIN J MED 1989; 56:245–248

THE DIAGNOSTIC utility of autoantibody testing in systemic lupus erythematosus (SLE) and related conditions is well established.[1] The fluorescent antinuclear antibody (ANA) test is highly sensitive (>0.99) for SLE, but the specificity is low (0.69)[2]; therefore, when the result is positive, follow-up testing is required to determine the clinical significance of the positive result. Anti-native DNA (anti-nDNA) and anti-Sm are highly specific (>0.99 in each case) but relatively insensitive (0.57 and 0.22, respectively) for SLE[2]; these tests are only rarely positive when ANA is negative. A maximally efficient testing strategy would make diagnostic testing for anti-nDNA and anti-Sm dependent on a prior positive ANA result.

The ANA Profile was introduced at the Cleveland Clinic in 1981. This test profile calls for the initial performance of an indirect immunofluorescent assay for ANA. If the result is positive at a titer ≥1:40, the serum is additionally tested for the presence of anti-nDNA by the Farr radioimmunoassay (and, if this is positive, by *Crithidia luciliae* immunofluorescence for confirmation of specificity) and for precipitating antibodies against ribonucleoprotein (RNP, or more properly U1RNP), Sm, and La/SS-B. Neither anti-nDNA nor any of the precipitating antibodies is likely to be positive if ANA is <1:40.

The reasons for introducing the ANA Profile were:

1. To improve diagnostic accuracy by following up on all positive ANA results with specific antibody testing and
2. To eliminate unnecessary antibody testing in patients with negative or low-level ANA test results.

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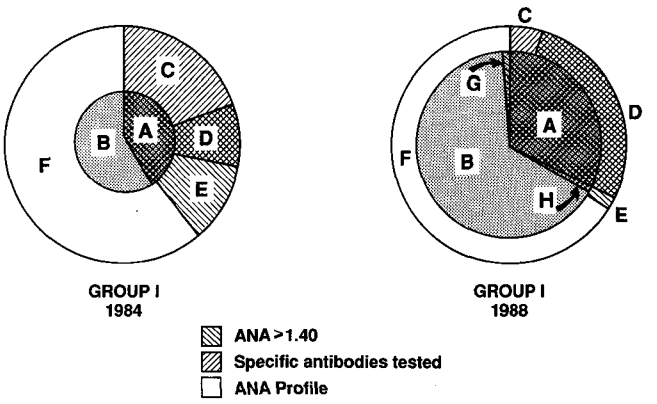

FIGURE 1. Comparison of appropriateness of specific antibody testing between two-week periods in 1984 (Group I) and 1988 (Group II). The large circles represent all ANA testing for the periods reviewed; the inner (shaded) circles represent ANA testing as a part of the ANA Profile. Positive ANA results are seen in areas A + D + E (+ H for Group II) and negative results in areas B + C + F (+ G in Group II). The crosshatched areas (A + D) denote appropriate follow-up testing of positive ANA results with specific antibody testing, and the unhatched areas (B + F) represent appropriate absence of such testing when ANA is negative or low titer. Inappropriate follow-up testing on such sera is represented by area C (and area G for Group II), and failure to follow up positive ANA results is represented by area E (and area H for Group II); these areas were found to decrease greatly between 1984 and 1988 with a commensurate increase in the appropriate areas.

The ANA Profile was computerized in 1984, allowing the automated generation of an interpretive report and the establishment of an easily accessible database. The interpretive report was based on two studies of these assays at this institution,[2,3] and the introduction of this report greatly improved the acceptance and utilization rate of the ANA Profile. The database makes it possible to develop lists of patients with particular serological characteristics for research purposes.

The purpose of this study is to evaluate the success of the computerized ANA Profile reporting system in facilitating accomplishment of the purposes for which the ANA Profile was originally intended. By comparing freestanding ANA and ANA Profile test results at the time of introduction of the computerized report with those three years later, we found that both purposes were partially accomplished, resulting in cost savings as well as improved quality of patient care.

MATERIALS AND METHODS

Serologic testing

ANA tests were performed by indirect immunofluorescence with a standard method[4] using frozen sections of rat kidney as substrate. Screening of sera was carried out at a 1:20 dilution. Polyvalent fluorescein-labelled goat anti-human immunoglobulins (Behring, catalog number 643901), reconstituted as directed, were used to detect binding of human immunoglobulin to the nucleus under epi-illumination using a Zeiss 9901 ultraviolet microscope. Sera yielding positive results at 1:20 were then titrated at 1:20, 1:40, 1:80, 1:160, and 1:320 dilutions.

Anti-nDNA was measured by the Farr assay,[5] modified as previously described.[6] *Crithidia luciliae* immunofluorescence testing[7] was carried out on 1:10 dilutions of unheated aliquots of samples giving a positive result in the modified Farr assay using a commercial test kit (Kallestad Quantafluor) with results reported as positive or negative but not titrated further.

Precipitating antibodies (anti-RNP, anti-Sm, and anti-La/SS-B) were assayed by double diffusion in agarose, as previously described,[6] using undiluted, unheated serum samples.

Data analysis

Laboratory records in the Immunopathology Department were reviewed, and 100 consecutive ANA results for the first two weeks of July 1984 (Group I) and 189 consecutive ANA results for the first two weeks of January 1988 (Group II) were recorded. Results of testing for antibodies against native DNA, RNP, Sm, and La/SS-B (whether or not such testing was done as a part of the ANA Profile) on sera from Group I patients at any time during July 1984 or on sera from Group II patients at any time during January 1988 were recorded. Comparisons were then made between the two time periods using the following standards:

1. ANA results ≥1:40 should be followed by testing for specific antibodies.
2. ANA results <1:40 should not be followed by testing for specific antibodies.

These standards have been incorporated into the ANA Profile, and to the extent that use of the ANA Profile increases, the standards will be more closely met.

Statistical comparisons were made using Fisher's exact test.[8]

RESULTS

The results are displayed graphically in *Figure 1*.

Of 100 consecutive ANA tests in 100 patients (Group I) obtained during the first two weeks of July 1984 immediately following introduction of the computerized reporting system, 37 (37%) were "positive" (titer ≥1:40). Twelve (12%) were ordered as ANA Profiles, and of these, five were positive. Overall, 10 of 37 patients (27%) with positive ANA results during this time had testing for either anti-nDNA or precipitating antibodies (against RNP, Sm, La/SS-B), or both (including all of those done as ANA Profiles). Of the 63 patients with negative or low-titer ANA results, seven (11%) had specific antibody testing, which was always negative as would be expected; none of these was from the ANA Profile subset.

Of 189 consecutive ANA tests in 184 patients (Group II) obtained during an equivalent time period in January 1988, 63 (33%) were positive. One hundred twelve (59%) were ordered as ANA Profiles, and of these, 38 (34%) were positive. Overall, 44 of 63 (70%) sera with positive ANA results during this time (including all but one of those done as ANA Profiles) had specific antibody testing. Of the 126 sera with negative or low-titer ANA results, only two (1.6%) had specific antibody testing, both of which were negative; one of these was from the ANA Profile subset but was tested further by special request.

ANA PROFILE

Dear Dr. Watson

The following is a summary of the ANA Profile results on your patient. The routine lab reports have been forwarded to the chart in the usual manner. This report is for your own records.

Name: Holmes Sherlock
Clinic number: 1234-567-8
Date drawn: 3/1/88
Date reported: 3/7/88

Results

ANA titer (normal < 1:40)	1:320
Anti-native DNA	
Farr assay:	35%
Crithida assay:	+
Anti-ENA	
Anti-RNP:	-
Anti-Sm:	+
Anti-SS-B (La, HA):	-
Anti-other:	-

Comment

This patient almost certainly has systemic lupus erythematosus. The predictive value of positive anti-DNA for SLE (true pos/all pos) is .974. The predictive value of positive anti-Sm for SLE (true pos/all pos) is .958. With both anti-DNA and anti-Sm positive, the predictive value for SLE approaches 1.00.

If you have any questions regarding these results or comments, please call either of the undersigned. We are especially interested in hearing about results that seem inconsistent with the clinical picture.

*Abbreviations:
+ = positive
- = negative
ND = not done

Record VII-438
Revised 12/7/87

FIGURE 2. The ANA Profile report.

DISCUSSION

In an attempt to encourage greater use of the ANA Profile for ANA testing at the Cleveland Clinic, a computerized reporting system was introduced in July 1984. This system produces a report that tabulates results of testing for various antinuclear antibodies and generates an interpretation based on data previously reported from our laboratory.[2,3] An example is shown (*Figure 2*). Clearly, this was successful, since use of the ANA Profile increased from 12% of all ANA tests ordered at the time the computerized report was introduced (three years after introduction of the ANA Profile) to 59% in January 1988 ($P = 1.5 \times 10^{-15}$).

Since the ANA test has a low positive predictive value for SLE (only about 0.04 in our laboratory[2]), a positive result cannot be interpreted without more specific testing, and in the absence of this testing, a posi-

tive ANA result is a "loose end," that is, the meaning cannot be easily determined. The ANA Profile automatically orders the additional tests when the ANA titer is ≥1:40 and eliminates this problem. The physician could do the same in the traditional way, but the patient would need to be contacted to return for an additional venipuncture, and the results would be delayed. In practice, the data suggest that, when a freestanding ANA test is positive, the follow-up tests are frequently not ordered as good practice would dictate. In July 1984, 73% of positive ANA results were not followed up within two weeks with specific testing, while in January 1988, this figure was reduced to 30% ($P = 5.9 \times 10^{-5}$). We did not determine whether appropriate follow-up may have occurred later than two weeks following the ANA result, so both of these percentages may be high, but at the very least the ANA Profile appears to improve the timeliness of testing.

On the other hand, it is equally important to conserve resources by avoiding unnecessary testing. Since the specific tests done as a part of the ANA Profile are only rarely positive when ANA <1:40, it is usually unnecessary to perform them in this situation. One important SLE-related serological test (anti-Ro/SS-A) was specifically not included in the ANA Profile because it does not fit this criterion, occasionally being positive in spite of negative or low-titer ANA. The use of a human substrate (such as Hep-2) for routine ANA testing might reduce the likelihood of this, but the cost of the profile would be increased out of proportion with the increase in sensitivity, which is already quite high with rat kidney substrate (>0.99 in our laboratory, as we previously reported.[2,3]) Between the two time periods examined in this study, there was a nearly sevenfold reduction in specific antibody testing when the ANA titer was <1:40. In 1988 dollars, the cost savings by eliminating unnecessary specific antibody testing based on these data was calculated to be $501 for a two-week period—an annual savings of approximately $12,000.

An additional benefit of computerization of the ANA Profile was the establishment of an easily accessible database of test results, containing more than 7200 records by March 1988. This is a useful research tool and also lends itself to assessment of quality.

CONCLUSION

The ANA Profile has been partially successful in achieving the goals for which it was designed. It has served an educational function as well as facilitating appropriate follow-up of positive ANA results and reducing unnecessary testing, thus leading to improved quality of patient management and efficiency of resource utilization.

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