ANA and Antibody Series

False Positive Scl-70 Results – A Major Problem in Laboratory Testing

Scl-70 antibody testing can be done by a number of different methods. Historically, Scl-70 antibody testing was mostly done by a technique called double immunodiffusion (ID), which is considered to be the most reliable Scl-70 antibody testing method [1]. ID testing is time-consuming and expensive. Because of the difficulty and cost of doing Scl-70 antibody testing using ID, almost all labs have switched to ELISA or Multiplex testing.

While Scl-70 antibodies are considered to be highly specific to systemic sclerosis (SSc), a number of studies [2,3,4,5] have documented that patients without a clear diagnosis of SSc often consistently test positive for Scl-70 antibodies when testing is done by ELISA or Multiplex testing methods. One example of this occurs in some patients with a diagnosis of systemic lupus erythematosus (SLE). While the reasons for this are not fully known, some researchers have suggested that dsDNA antibodies may be cross-reactive with the antigen used in Scl-70 ELISA testing assays [3,4].

Clinicians may be aware of the false-positive Scl-70 antibody testing problem; as a result, when they see an initial positive Scl-70 result, they will often order a repeat test at the same lab, thinking that the problem is a testing precision error. Unfortunately, because of the nature of the Scl-70 testing issue, the repeat test will frequently also be positive, leading the clinician to (incorrectly) assume that the Scl-70 test result is accurate and that the patient has SSc.

Unfortunately, recent research suggests that the false positive Scl-70 antibody testing problem is much worse than previously realized.

Last week, a study [6] was published online that compared Scl-70 antibody testing of 129 patients by three different testing methods: Multiplex, ELISA, and ID. Without going into a lot of detail, here are the key findings from this study:

- Of the 129 patients who tested positive by Multiplex, only 51 (39.5%) of the patients were positive when tested again using ELISA
- If you then took the group of 51 patients positive by Multiplex and ELISA and retested them using the much more accurate ID testing method, only 21 (41.2%) were positive by ID
- Since ID testing is considered to be the "gold standard" for commercial Scl-70 antibody testing, this suggests that **out of the 129 patients who tested positive by Multiplex test, 108 (83.7%) of these were false positive results!**

Another way to look at the data:

- Of the 129 patients who tested positive for Scl-70 antibodies using Multiplex testing, only 33 of the 129 patients (25.6%) had a formal diagnosis of systemic sclerosis (SSc)
- Of the 51 patients who tested positive by Multiplex and ELISA testing, 23 of the 51 patients (45.1%) had a formal diagnosis of SSc
- However, if you look at the 21 patients who tested positive by all three testing methods, 19 of the 21 patients (90.5%) had a formal diagnosis of SSc

While there was no data in this paper that looked at Multiplex result level versus diagnosis, the paper does indicate that a cutoff of 110 ELISA units was highly predictive of an SSc diagnosis. ELISA units for the patients who were also positive by ID ranged from 70 to 129. The normal range for this particular ELISA assay is 0 to 20, so the 110 SSc diagnosis cutoff is more than five times the positive cutoff of 21 units.

There is a second paper about to be published on this same topic that shows similar results. We can't go into the details on that paper since it is not yet formally published, but that paper does show a correlation between how high the Scl-70 result is versus a formal diagnosis of SSc. In that paper, there was a similar cutoff – if the Scl-70 result was at least five times the minimum cutoff for positive, it was highly correlated with a formal diagnosis of SSc.

(One key limitation of both of these recent studies. You cannot generalize the results to other Multiplex or ELISA testing kits. It is possible that other testing kits might be either better or worse than these recently published results.)

In summary, if you test positive for Scl-70 antibodies by Multiplex or ELISA at a high level (at least five times the positive cutoff), it is highly likely that the result is not a false positive, according to two recent studies. If the Scl-70 result is in the low to moderate range, then additional testing is needed to determine if the result is a false positive. The best way to do this is to retest for Scl-70 antibodies at a lab that still does testing using the more accurate double immunodiffusion method. In the US, we are only aware of one lab that currently offers ID testing for Scl-70 antibodies, RDL Reference Lab in Los Angeles. We do not currently have any information on the availability of ID testing for Scl-70 antibodies outside of the US, unfortunately.

References

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