Correspondence on 'Standardisation of myositis-specific antibodies: where are we today?'

We have read with great interest the recent article from Espinosa-Ortega *et al*¹ and the commenting letter by Mahler *et al*² on the reliability of line immunoassay (LIA) versus immunoprecipitation (IP) in the detection of myositis-specific autoantibodies (MSAs) for the diagnosis of idiopathic inflammatory myopathies (IIM). Even if the search for MSA by dot immunoassay (DIA) or LIA has been used for over a decade and represents a 'non-criteria' test to assist clinicians in IIM diagnosis, MSAs have been included in classification criteria only for a couple of years.³ However, initial studies on the clinical utility of the DIA/LIA methods in diagnosing IIM were conducted on a restricted panel of MSA and mainly on selected IIM patients.⁴

Only recently have real-life studies on larger series of patients and using a larger panel of MSA-related antigens been published, highlighting on one side a great intra-method analytical variability of DIA/LIA in detecting MSA,⁵ and on the other side their weak correlation with IP.⁶ Considering the controversial data between Espinosa-Ortega's¹ and Cavazzana's^{7,8} studies, it is emblematic that there is still no concordance among LIA and IP even for anti-Jo1, the most common MSA and the first discovered in this group of diseases. The low agreement of IP versus other methods, evidenced by recent studies,^{2,6,7,9} raises the question of whether IP should still be considered the reference method for detecting MSA.

Another study that has opened Pandora's box is Vulsteke's¹⁰ which compared three different DIA/LIA assays showing significant differences in diagnostic performance which, however, varied according to the MSA considered. This great variability clearly demonstrates the urgent need to harmonise methods, and that their clinical validation against the reference IP method remains an issue.^{8,9} In addition, studies conducted so far are retrospective, including patients diagnosed using previous criteria,¹¹ and mostly performed on cohorts in which there was a very small number of some MSA, representing a further bias for comparative analysis.^{12,13} Nevertheless, today some tools have emerged that may help to improve the specificity of MSA detection by DIA/LIA and to confirm the diagnosis of MSA-associated IIM (table 1). Among these tools, our group has observed the importance of the agreement between DIA/LIA results and a compatible HEp-2 IIF pattern, showing a concordance of around 50%

in IIM patients.¹⁴ Recently, Piette *et al* have confirmed these data, suggesting caution in interpreting the results in case of low-positive MSA signal intensity.⁵ This may be due to a cut-off that is not well set in some cases, since combining different antigens in a single assay may produce suboptimal performance for each MSA. Creating MSA-related cut-off values could help to improve this issue. Moreover, since MSA are usually mutually exclusive, the simultaneous detection of two or more MSA might indicate possible false-positive results and the need for MSA positive findings to be confirmed by another method.

Even if DIA/LIA are promising MSA-detection technologies, their use in IIM diagnostics is still a challenge. Prospective and multicentre studies are needed to validate these new methods and clarify whether they can be reliably used instead of the reference IP method.

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Table 1 Tools to improve specificity of MSA detection by dot blot (DIA) or line immunoassay (LIA)

	Study
HEp-2 IIF pattern compatible with myositis-specific antibodies detected by DIA/LIA	Picard <i>et al</i> ¹⁵ Aggarwal <i>et al</i> ¹⁶ Infantino <i>et al</i> ¹⁷ Infantino <i>et al</i> ¹⁴
High signal intensity of DIA/LIA measured by densitometric quantitation	Cavazzana <i>et al</i> ⁷ Bundell <i>et al</i> ¹⁸ Lecouffe-Desprets <i>et al</i> ¹⁹
No coexisting MSAs (ie, isolate antibody reactivity)	Infantino <i>et al</i> ¹⁴ Lega <i>et al</i> ²⁰
MSA positivity confirmed by another method (immunoenzymatic or fluoroimmunoenzymatic method, chemiluminescence, immunoprecipitation)	Cavazzana <i>et al</i> ⁸ Damoiseaux <i>et al</i> ⁶
DIA, dot immunoassay; LIA, line immunoassay; MSA, myositis-specific autoantibody.	

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