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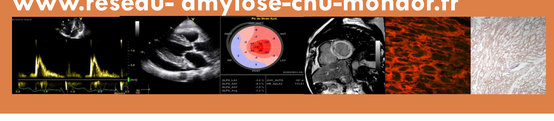

EVALUATION OF THE NEW SEBIA FLC ELISA ASSAY FOR THE QUANTIFICATION OF SERUM FREE LIGHT CHAINS

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BACKGROUND

Since 2001, the introduction of the serum-free light-chain (sFLC) assays has been an important advance for the management of monoclonal light-chain gammopathies: AL amyloidosis, paucisecretory and Light Chain Multiple Myeloma (LCMM). This test is also useful for the diagnosis, prognosis and/or monitoring of other plasma cell dyscrasias, including intact immunoglobulin multiple myeloma (MM) and monoclonal gammopathy of unknown significance (MGUS). In particular, it is now included in international guidelines for the assessment of the response depth in MM.
Several assays are today available based on nephelometry/turbidimetry and lateral flow. The use of different techniques can help resolve analytical and interpretation difficulties.

In this study, we have evaluated the performances and suitability for routine use of a new sFLC assay based on a sandwich ELISA (Sebia FLC Kappa, Sebia FLC Lambda, Sebia, Lisses, France) in comparison with a widely available nephelometry technique (Freelite™, The Binding Site, Birmingham, UK).

METHODS

Free light chain assays

New polyclonal antibody-based ELISA assays for serum FLC κ and λ are now commercially available (Sebia FLC Kappa & Sebia FLC Lambda). We performed the Sebia FLC quantification on the fully automated ELISA processor AP22 ELITE (das, Palombara Sabina, ITALY) and the Freelite™ assays on the BN ProsPec® (Siemens, Marburg, GERMANY), according to manufacturers' instructions.

Patient samples

206 routine serum samples from patients at various stages of several monoclonal gammopathies (MGUS, MM, AL amyloidosis) and control patients (polyclonal increase or decrease of immunoglobins, such as in Common Variable Immune Deficiency [CVID], renal impairment) were analyzed in parallel on both assays. All patients had gave written consent.

Method comparison

The samples were classified according to the κ/λ ratio reference ranges, into low (L), normal (N) and high (H). Concordance analysis was performed.

Clinical performance

True positive (sensitivity) and true negative (specificity) were determined for both assays, using the κ/λ ratio.

Retest Rate

The percentage of additional tests (dilutions) needed to report a result were calculated.

Statistics

All statistics were performed using Analyse-it (Analyse-it R software v2.03, Ltd, Leeds, UK: www.analyse-it.com).

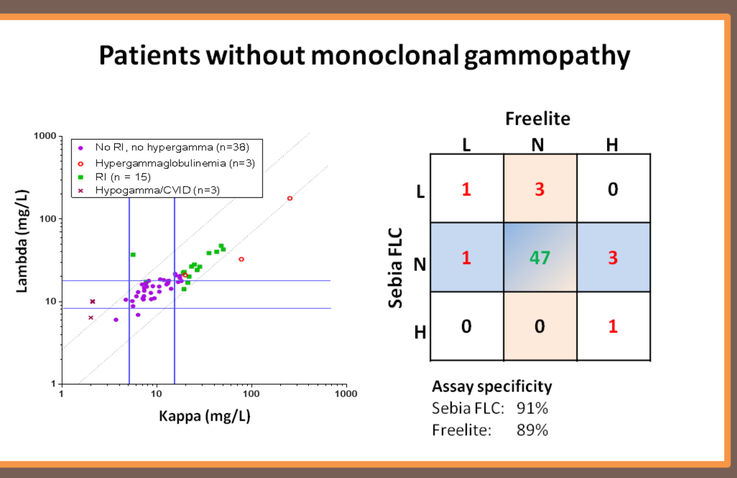
RESULTS

The **reference ranges** of the Sebia FLC assay are comparable to the ranges of the Freelite™ assay:

Sebia FLC κ 5.2 – 15.3 mg/L	Freelite™ κ 3.3 – 19.4 mg/L
Sebia FLC λ 8.2 – 18.1 mg/L	Freelite™ λ 5.7 – 26.3 mg/L
Sebia FLC κ / λ ratio 0.37 – 1.44	Freelite™ κ / λ ratio 0.26 – 1.65 (renal insufficiency [RI]: 0,37-3,1)

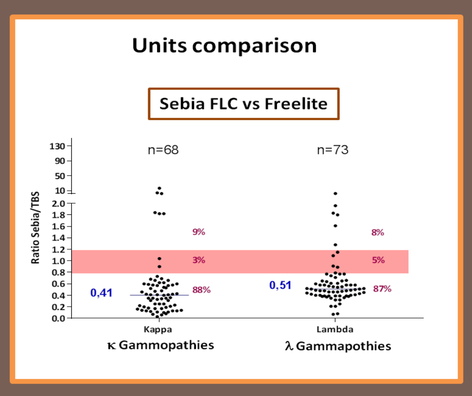
The **percentage of additional tests** needed to report a result was **4 times less** with Sebia FLC than with Freelite™ for both κ and λ quantification.

PATIENTS WITHOUT MONOCLONAL GAMMOPATHIES



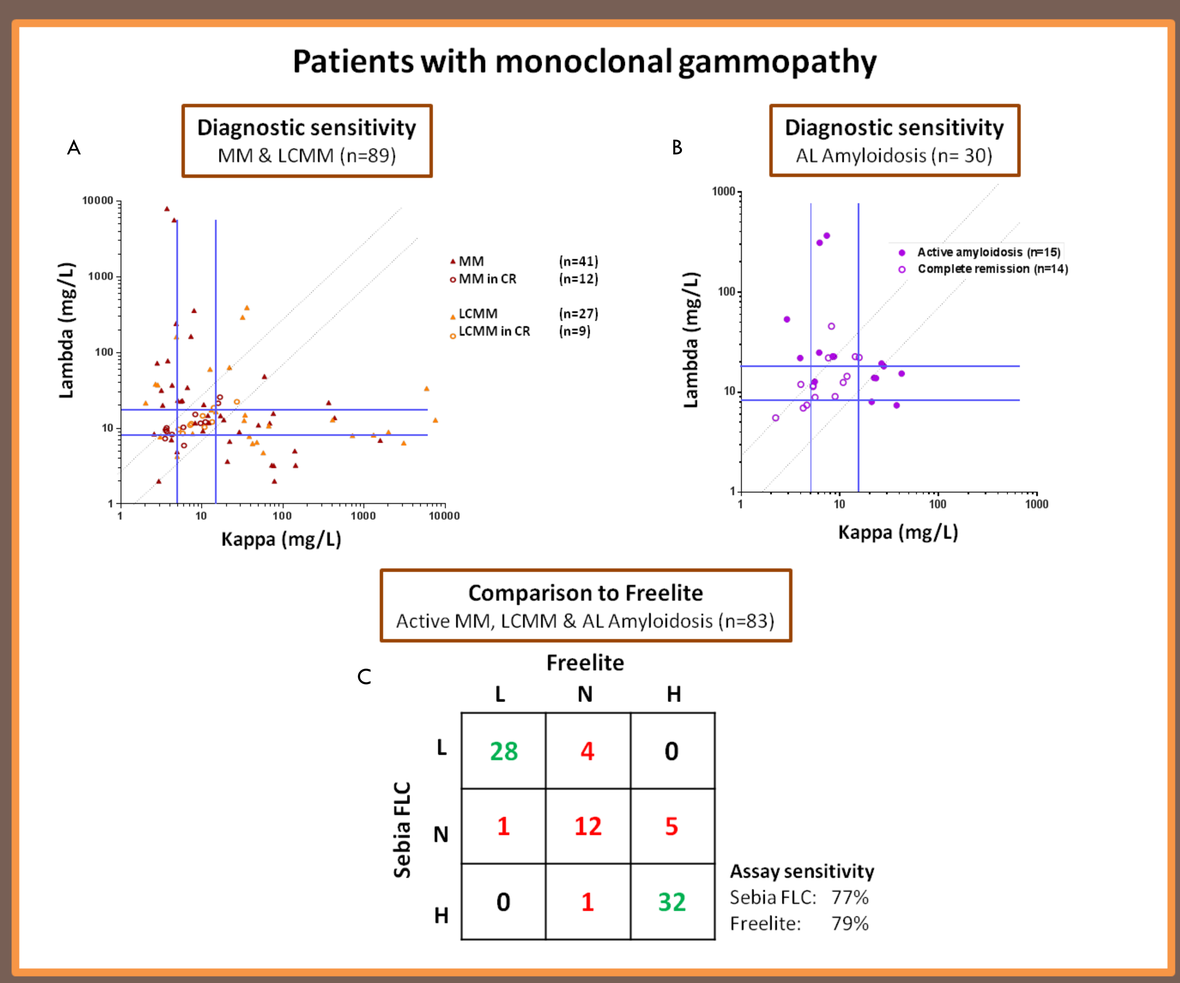
A) κ vs. λ FLC concentrations were plotted for both assays (the Sebia results are shown). The diagonal lines delimit the normal κ/λ ratio. As expected, the Sebia κ and λ concentrations were increased in patients with RI. However, the Sebia ratios did not exceed the standard reference range in contrast to the Freelite ratios (8 patients reclassified as normal with the RI reference range).
(B) The κ/λ ratio concordance between the methods was good at 87 % (discordance: 9/56). The specificities of both assays were similar.

CONCENTRATION COMPARISON



Freelite™ values of the individual monoclonal FLC were significantly higher than the Sebia values. The unit differences between the methods was mainly visible at the high end of the concentration range, especially for κ with Freelite concentrations >10 times higher. Some of these differences may result from the overestimation of monoclonal FLC by Freelite at high serum dilutions due to linearity loss.

PATIENTS WITH MONOCLONAL GAMMOPATHIES



(A) The Sebia FLC assay was able to distinguish the patients with active MM or LCMM from patients in complete response (CR). The same result was observed for patients with AL amyloidosis (B). The results of the 119 patients show a good diagnostic sensitivity of the Sebia FLC, comparable to that of Freelite (C). The concordance between the two assays was 86 % (discordance: 11/83 patients; 5 were picked up by the Sebia assay and 6 by Freelite™). A good concordance was also observed for the 31 MGUS tested (not shown).

CONCLUSION

Sebia FLC satisfactorily distinguished patients with active Multiple Myeloma (MM), Light Chain MM and AL Amyloidosis from those who achieved complete remission and controls, with similar specificity and sensitivity than Freelite™. The use of a specific reference range for the κ/λ ratio of patients with RI was not necessary with the Sebia assay. Freelite™ concentrations were generally higher than the Sebia FLC concentrations. The automation on the das AP22 is suitable with a normal laboratory workflow.
We show here that the new Sebia FLC assay is suitable for FLC quantification in a context of laboratory routine. Its diagnosis performances compare well to Freelite™ and it generates less retests due to a broader measuring rage.