Identification and quantification of urinary monoclonal proteins by capillary electrophoresis in AL amyloidosis

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Introduction

- Identification and quantification of urinary monoclonal proteins (uMPs) is of utmost importance in the diagnosis and monitoring of monoclonal gammopathies.
- Convenient and reliable tools for detecting and measuring uMPs are needed to ensure best sensitivity, and the relatively low concentration can hinder the quantification of uMPs.
- The aim of our study was to assess the performance of the urine peak quantification tool on CAPILLARYS 2 Flex Piercing in patients with AL amyloidosis as a part of a larger study involving patients with plasma cell dyscrasias.

Methods

A total of 75 patients with AL amyloidosis were included. Only patients in whom uMPs were detected by hr-IFE were included in the study.

Samples were tested with:

- homemade high-resolution agarose gel immunofixation electrophoresis (**hr-IFE**) of serum and concentrated (10 times) urine;
- commercial semi-automated agarose gel immunofixation of urine (**Sebia Hydragel** BJ on Hydrasys 2);
- urine protein capillary electrophoresis and immunotyping (**Sebia Capillarys** 2 Flex Piercing Urine);
- quantification of circulating free light chains (FLC) by **Freelite** and **N latex FLC**.
- Urinary MPs were quantified using **Sebia Phoresis** software tools.

Results

Table 1. Patients' characteristics

Variable	N (%) or median (IQR)
Age, years	65 (42-90)
Gender, male	46 (67)
Organ involvement heart / kidney / soft tissue liver / PNS / ANS	 39 (57) / 32 (47) / 12 (17) 5 (7) / 5 (7) / 5 (7)
Proteinuria, g/24h	1.5 (0.2-5.1)
Serum creatinine, mg/dL	1.07 (0.96-1.16)
dFLC N-LATEX FLC, mg/L	111 (63-142)
dFLC Freelite, mg/L	131 (89-210)
eGFR, mL/min per 1.73 m^2	59 (41-160)

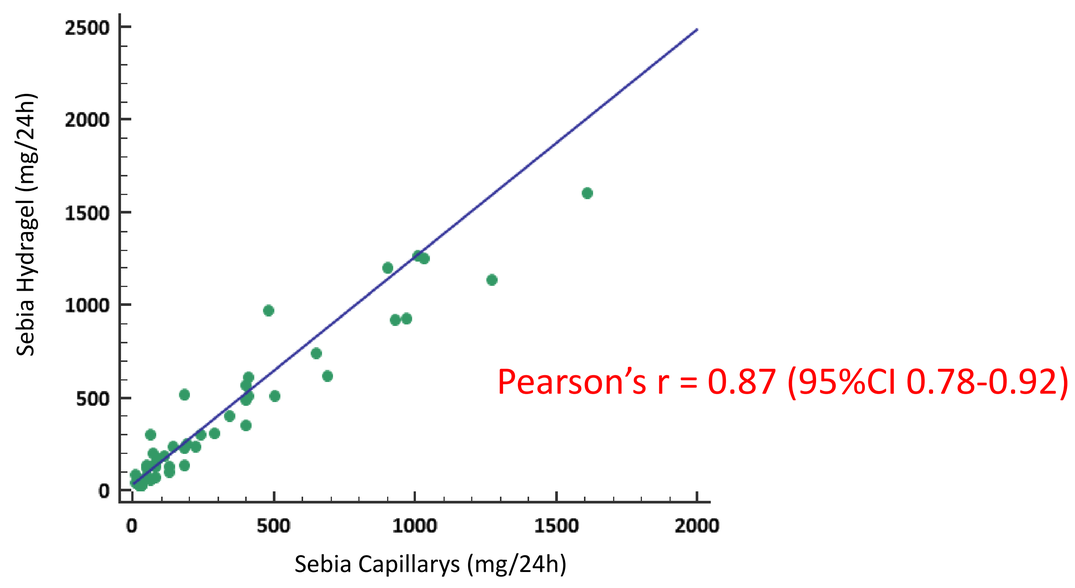
Table 2. Diagnostic sensitivity in 62 patients with a uMP detectable at hr-IFE of concentrated urine

Assay	N (%)	95% CI
hr-IFE of serum	62 (100)	96-100
Freelite FLC ratio	53 (85)	75-92
N latex FLC ratio	52 (84)	73-91
Sebia Hydragel	56 (90)	84-96
Sebia Capillarys	57 (92)	83-96

Results

- The uMP was quantifiable in 55 of 62 cases in whom it was detected by urine protein capillary electrophoresis.
- The median uMP excretion was 130 mg/24h (range 10-1610 mg/24h) as assessed by Sebia Phoresis tool.
- The uMP was also quantifiable on Sebia Hydragel agarose gel in 51 patients (75%).

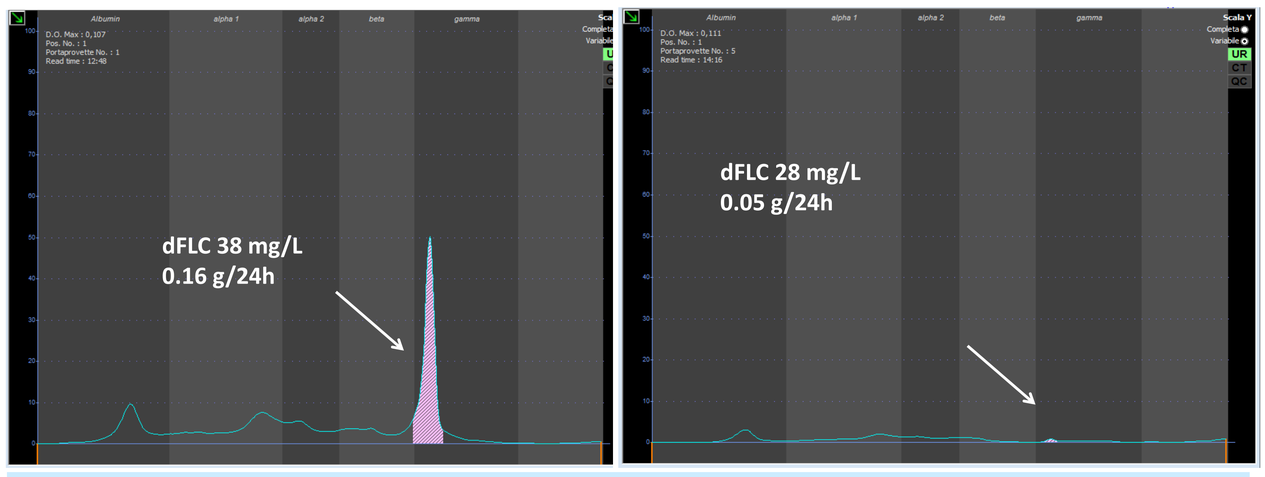
Figure 1. Correlation of uMP quantification between Sebia Capillarys and Sebia Hydragel



FOLLOW UP STUDY

- Sixteen patients with quantifiable uMP and dFLC (*Freelite*) >50 mg/L were treated and had response data at 3 months.
- Five subjects responded (1 partial response, 4 very good partial response) with a median 69% dFLC decrease (range 51-90%). In all of them uMP excretion also decreased (median 100%, range 30-100%).
- Among non-responders, only one patient had a relevant reduction in uMP excretion (from 740 to 250 mg/24h, dFLC from 746 to 619 mg/L) with stable renal function.
- Post-treatment Sebia Capillarys was also available in 5 patients with baseline dFLC (*Freelite*) <50 mg/L. In 2 of them the uMP was still visible but was no longer quantifiable, in 2 it remained stable and in one patient uMP increased from 20 to 40 mg/24h.

Figure 2. Urine protein Sebia Capillarys before and after treatment in a patient without "dFLC measurable" (dFLC <50 mg/L, *Freelite*) disease.



Conclusions

- Sebia Capillarys protein electrophoresis can identify uMPs in patients with AL amyloidosis with a good sensitivity, and can quantify uMP excretion as low as 10 mg/24h.
- Changes in uMP excretion can be monitored during treatment, including some patients without "dFLC-measurable" disease.
- Further studies are warranted to evaluate this tool in response assessment.