

Poster from the Congress

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# Immunonephelometric Determination of Free Light Chains in Urine as Screening for the Detection of Bence Jones Proteinuria

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# IMMUNONEPHELOMETRIC DETERMINATION OF FREE LIGHT CHAINS IN URINE AS SCREENING FOR THE DETECTION OF BENCE JONES PROTEINURIA.

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## INTRODUCTION

The use of immunonephelometric (IN) determination of free light chains (FLC) kappa (FRK) and lambda (FRL) in unconcentrated urine can be a reliable, rapid and economical screening method to detect Bence Jones proteinuria (BJP). Electrophoretic techniques, like immunofixation (IF), are useful in order to determinate the mono- or policlonality of FLC.

## AIM

To evaluate the immunonephelometric determination of free light chains (IN-FLC) in unconcentrated urine samples by comparing these results with those obtained by IF in the same concentrated urines, with the object to integrate IN-FLC in the routine protocol for the BJP study.

## MATERIAL AND METHODS

### Material

100 unconcentrated 24-hour urine samples, which had been selected at random among all the patients with a BJP study requested.

### Methods

- Immunonephelometry (BN II, Dade Behring) determination of FRK and FRL in unconcentrated urine samples using a reagent kit specific for FLC (New Scientific Company).
- Immunofixation (Paragon, Beckman Coulter) in the same urines but x25 concentrated (Minicon, Millipore) with antisera anti: Ig (G, A, M), bound and free (B&F) light chains K and L, and free light chains K and L (New Scientific Company).

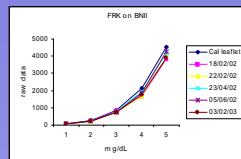
## RESULTS

### ANALYTICAL PERFORMANCE EVALUATION OF IN-FLC

#### 1. Reproducibility of calibration curves:

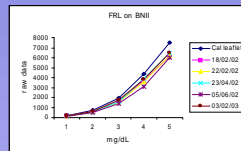
Cal (mg/dL)	Average raw data	CV%
0.5	63	11.3
1	230	7.8
2	753	4.5
4	1794	6.9
8	4031	4.2

FRK



Cal (mg/dL)	Average raw data	CV%
0.5	155	14.2
1	557	7.1
2	1564	8.6
4	3604	8.7
8	6245	3.5

FRL



2. Lower detection limit: FRK = 0.16 mg/dL  
FRL = 0.002 mg/dL

#### 3. Imprecision:

	CV within-day	CV between-day
FRK between 6-8 mg/dL	5 %	11 %
FRK between 1-2 mg/dL	9 %	11 %
FRL between 6-8 mg/dL	8 %	4 %
FRL between 1-2 mg/dL	6 %	8 %

#### 4. Cross-reaction between anti-FLC antiserum and light chains bound to Ig: absent

#### 5. Cross-reaction with free light chains of the opposite type: absent

#### 6. Antigen excess: not demonstrable until over, 3000 mg/dL

#### 7. Matrix effect: results below the lower detection limit

### RESULTS BY IN-FLC

The results obtained by IN-FLC has been interpreted as follows:

CLASSIFICATION (0.5 mg/dL lower calibrator value)	INTERPRETATION	Results
FRK and FRL < 0.5 mg/dL	negative	53
FRK or FRL > 0.5 mg/dL	FRK or FRL positive	34
FRK and FRL > 0.5 mg/dL	FRK and FRL positive	13
		100

### COMPARISON BETWEEN IN-FLC AND IF

I. All the urines without FLC detected by IN (53) resulted also BJP negative by IF.

II. Among the 47 (34+13) urines with FLC detected by IN, 22 showed monoclonal FLC (BJP), 5 policlonal FLC and 20 were negative.

IMMUNONEPHELOMETRY Classification	n°samples	IMMUNOFIXATION		
		with BJP	negative BJP policlonal FLC	negative FLC
FRK or FRL ≥ 0.5 mg/dL	34	16	1	17
FRK and FRL ≥ 0.5 mg/dL	13	6	4	3
<b>TOTAL</b>	<b>47</b>	<b>22</b>	<b>5</b>	<b>20</b>

### EXAMPLES OF URINES WITH BJP

monoclonal FRL		monoclonal, FRL and Ig	
IF	IN-FLC	IF	IN-FLC
	FRK < 0 mg/dL FRL 5.45 mg/dL		FRK < 0 mg/dL FRL 1.9 mg/dL
SPE Ig K <sub>T</sub> L <sub>T</sub> K <sub>F</sub> L <sub>F</sub>		SPE Ig K <sub>T</sub> L <sub>T</sub> K <sub>F</sub> L <sub>F</sub>	

SPE: reference electrophoresis

Antibodies: Ig: anti-Ig (G, A, M); K<sub>T</sub>: anti-K (B&F); L<sub>T</sub>: anti-L (B&F); K<sub>F</sub>: anti-K free; L<sub>F</sub>: anti-L free

## CONCLUSIONS

1. Nephelometric determination of FLC fulfil with all the acceptability criteria.
2. Negative results found by IN-FLC and confirmed by IF would support the use of IN-FLC as screening method for BJP study.
3. Results found by IN-FLC show that this method is more sensitive than IF which would guarantee the absence of BJP false negatives.