Research Article

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Optimizing the Utility of Serum Protein Electrophoresis

Hollie Sheffield[1], Haekyung Jeon-Slaughter[1,6], Nivan Chowattukunnel[2], Waqas Haque[3], Mir Lim[4], Evelyn Shen[5] and Yu-Min Shen[1*]

[1]*Division of Hematology and Oncology, Department of Internal Medicine, University of Texas Southwestern Medical Center, Dallas, Texas, USA*

[2]*Cleveland Clinic Cancer Center, Cleveland, Ohio, USA*

[3]*Department of Medicine, New York University Langone Health, New York, New York, USA*

[4]*Department of Internal Medicine, University of Texas Southwestern Medical Center, Dallas, Texas, USA*

[5]*Department of Medicine, University of Minnesota, Minneapolis, Minnesota, USA*

[6]*VA North Texas Health Care System*

[*]Corresponding Author
Yu-Min Shen, Division of Hematology and Oncology, Department of Internal Medicine, University of Texas Southwestern Medical Center, Dallas, Texas, USA, 2001 Inwood Road Dallas, TX 75390-8872, Tel: (O) 214-648-1939

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Abstract

Introduction: *Serum protein electrophoresis (SPEP) is often obtained to look for monoclonal gammopathy when evaluating nonspecific clinical findings. Currently optimal utilization of SPEP is not well defined. This study was conducted to identify patient characteristics or laboratory data as appropriate indications for ordering SPEP.*

Material and Methods: *A retrospective review of 406 patients referred for abnormal SPEPs from 2012 to 2019 was performed to identify characteristics that predict for development of lymphoplasmacytic malignancy. Indications for ordering the SPEP, serum calcium, serum creatinine, hemoglobin, and presence of bone lesions were recorded. Specific monoclonal (M) component types were also analyzed.*

Results: *Of those patients, 27 were found to have a lymphoplasmacytic malignancy (LPM). The most documented reasons for SPEP testing were renal dysfunction, increased globulin fraction, anemia, and neuropathy. Patients with at least one CRAB criteria (hypercalcemia, renal dysfunction, anemia, or bone lesion) had a significantly increased risk of developing a LPM. In evaluating the M-component subtype, patients with IgA M-components had a significantly higher chance of progression to a LPM when compared to IgG (37% vs 8.2%). An M-component of >1.5 g/dL was associated with significantly increased risk for developing a LPM (Odds Ratio 45.8; 95% Confidence Interval, 10.7-195.8).*

Conclusion: *We conclude that SPEP should be ordered for patients with CRAB criteria or otherwise unexplained neurological or dermatological disorders. Once an SPEP has been ordered and found to be abnormal, patient-specific characteristics such as presence of CRAB criteria and IgA M-component can be used to guide follow-up and need for a hematology referral.*

Keywords: MGUS, SPEP, CRAB, Lymphoplasmacytic Malignancy

1. Introduction

Serum protein electrophoresis (SPEP) is a laboratory assay used to separate serum proteins according to their size, shape, and electric charge, and is frequently used to identify patients with suspected monoclonal gammopathy [1]. Monoclonal gammopathy is the presence of a monoclonal component (M-component) on SPEP, often associated with lymphoplasmacytic malignancies (LPMs), including multiple myeloma (MM), immunoglobulin light-chain (AL) amyloidosis, Waldenstrom macroglobulinemia (WM), or other medical conditions such as cytopenias, hypercalcemia, neuropathy, and renal dysfunction [1]. Monoclonal gammopathy of undetermined significance (MGUS), present in about 3% of the United States population, results from the abnormal proliferation of the lymphoplasmacytic or clonal plasma cells producing an M-component without any resultant clinical disease [2]. Specifically, MGUS does not produce end-organ damage seen with LPMs, including hypercalcemia, renal failure, anemia, or bone lesions, collectively known as CRAB symptoms [3]. However, all cases of MM are preceded by MGUS, which therefore requires monitoring for progression [4].

The risk of progression from MGUS to a LPM is life-long and occurs at a rate of approximately 1% per year overall [2]. Risk stratification of MGUS patients using the size of the M-component (greater than 1.5 g/dL), the type of M-component (non-IgG), and an abnormal free light chain ratio (<0.26 or >1.65) results in risk of progression to LPM ranges from 5% in 20 years of follow up in the low risk group to 58% in 20 years of follow up in the high risk group [5,6].

While it is accepted that MGUS patients should be monitored for the development of LPMs and an SPEP should be ordered for patients with clear signs or symptoms of a LPM, in clinical practice the SPEP is frequently ordered for other reasons [7]. Many patients have isolated clinical findings that can be seen with, but are not diagnostic for LPMs, and the SPEP is ordered in anticipation of an early LPM diagnosis. In our hematology practice, a significant proportion of patients are referred for abnormal SPEP findings, with only a few true LPMs diagnosed. Most patients have MGUS, which necessitates regular monitoring with repeated SPEPs. Currently, in the spectrum of patients with MGUS, asymptomatic MM, and symptomatic MM, evidence-based treatment guidelines only recommend plasma cell- directed therapy for those with symptomatic MM [8]. In addition, those with the highest-risk MGUS only have less than 3% risk per year of developing MM (58% in 20 years) [6]. Contrary to proponents of MGUS screening, we question the wisdom of using SPEPs for early diagnosis of MM as well as the utility of repeated SPEPs for screening purposes [9].

Our study aims to examine the utility of performing an SPEP in routine clinical practice and identify patterns in patient characteristics and laboratory findings that can improve clinicians' understanding of the appropriate and cost-effective indications for ordering an SPEP.

2. Patients and Methods

A retrospective chart review was conducted of patients receiving care at a safety-net, county hospital system in a major metropolitan area from January 01, 2012, to December 31, 2019, who were referred to the hematology clinic for evaluation of an abnormal SPEP. An SPEP is considered abnormal with the presence of an M-component of an intact immunoglobulin and/or a free light chain.

Variables of interest for the study included serum calcium, serum creatinine, hemoglobin levels, presence of bone lytic lesions or pathologic fractures, the amount of the M-component, and the type of M-component (See Table 2). The serum κ to λ free light chain ratio was also collected, if available. Patients were given a CRAB score of 0.

If none of the following four conditions were present: calcium >11 mg/dL, creatinine >2 mg/dL, hemoglobin (hgb) <10 g/dL, and bone lesions present; ≥1 if at least one of the four condition are met [3]. Also recorded were the number of SPEPs done for each patient; for those with subsequent diagnosis of LPM, only the SPEPs done prior to the diagnostic bone marrow biopsy were counted.

Different M-components were categorized into 6 groups: IgG with or without free light chains, IgM with or without free light chains or IgG concurrently, IgA with or without free light chains or IgG concurrently, IgD with or without free light chains, free κ light chains with or without free λ light chains, and free λ light chains only.

A group is defined as those who developed MM or WM later during the observation period and those without (hereafter LPM group).

2.1 Statistical Analysis

Descriptive statistics were reported, means and standard deviations (SD) for continuous variables and frequencies and percent for categorical variables. T-statistics and Chi-square statistics were used to test LPM group differences in continuous and categorical variables, respectively.

The penalized logistic regression model with FIRTH Maximum Likelihood Estimation (MLE) method was used to determine risk factors associated with increased risk of LPM [10]. Odds ratios (OR) and 95% Confidence Intervals (CIs) were reported. Inclusion of covariates in the final model was determined by statistical significance of variables and Akaike Information Criteria (AIC; the smaller values are better). C statistics (Area under the Receiving Operating Characteristics Curve) was used to assess ability of the risk factors included in the model to predict LPM correctly. C statistics >0.7 is considered as a good predictability. A P-value < 0.05 was set as a criterion for statistical significance. All statistical analyses were conducted using SAS 9.4 (SAS Institute, NC).

3. Results

Table 1 lists the reasons for obtaining the 1249 SPEPs from the 406

study participants. The most documented reasons for SPEP testing were renal dysfunction and/or proteinuria (44.8%), increased globulin fraction (11.3%), decreased hemoglobin (11.1%), and neuropathy (8.6%); many patients had multiple reasons for SPEP testing documented. In our cohort, 27 individuals were found to have a LPM; 1 patient had a prior history of mucosa-associated lymphoid tissue (MALT) lymphoma; 25 patients developed MM and 1 patient developed WM in the observation period. All 27 patients diagnosed with a LPM had at least one CRAB criteria.

The mean values for serum calcium, serum creatinine, and hemoglobin among the total samples were 9.09 mg/dL, 1.78 mg/dL, and 11.36 g/dL, respectively. Those with LPM had numerically higher serum calcium and creatinine, and lower hemoglobin, although only serum calcium was significantly different. Meeting at least one CRAB criteria was significantly associated with an increased risk of LPM. As the serum calcium level increases by 1 mg/dL, the risk of LPM increases by 2.7-fold (Table 3). There were 30 patients noted to have lytic bone lesions or pathologic fractures; they were 9 times more likely to develop LPM than those without bone lesions. There were 14 patients who were noted to have an M-component >1.5 g/dL; they were 46 times more likely to develop LPM than those with M component ≤ 1.5 g/dL (Table 3).

Nearly 70% of patients were found to have an IgG M-component. Consistent with the Mayo Clinic risk stratification, a significantly lower proportion of those with an IgG M-component were diagnosed with LPM, while the opposite was observed with the non-IgG M-component groups. Specifically, amongst those with an IgA M-component, a significantly higher proportion developed LPM (37% versus 8.2%; Table 2) with an increased odds ratio of 13.93 (Table 3).

Among the 271 patients with available serum free light chain analysis, 91 can be categorized as low-risk according to the Mayo Clinic MGUS progression risk stratification; 117 were low-intermediate, 61 were high-intermediate, and 2 were high-risk. As predicted by the risk stratification, a progressively higher proportion of the higher-risk groups were found to develop LPM (Table 4).

	Reasons for SPEP	N (%)
CRAB	Hypercalcemia (C)	17 (4.2%)
	Rena dysfunction/Proteinuria (R)	182 (44.8%)
	Anemia (A)	45 (11.1%)
	Lytic bone lesions/Pathologic fractures (B)	17 (4.2%)
Other	Increased globulins	46 (11.3%)
	Neuropathy	35 (8.6%)
	Suspected amyloidosis	8 (2.0%)
		7 (1.7%)
	Osteoporosis/penia	6 (1.5%)
	Others	6 (1.5%)
	Unknown	37 (9.1%)
Total		406*
* 1249 total SPEPs from 406 unique participants done for the cohort in the observation period.		

Table 1: Clinical Indications for Ordering the Serum Protein Electrophoresis (SPEP); Several Patients had more than one Indication for Ordering the SPEP

Characteristics			Total	Myeloma/Lymphoma*		p-value
			(n=406)	Yes (n=27)	No (n=379)	
Calcium (mg/dL)		Mean ± SD	9.09 ± 0.76	9.44 ± 0.83	9.06 ± 0.75	0.0110
Cr (mg/dL)		Mean ± SD	1.78 ± 1.75	2.16 ± 4.0	1.76 ± 6.0	0.2464
Hgb (g/dL)		Mean ± SD	11.36 ± 2.22	10.79 ± 2.11	11.40 ± 2.22	0.1677
Bone Lytic Lesion or Pathologic Fracture	Yes	N (%)	30 (7.4)	10 (37.04)	20 (5.3)	<0.0001
M component >1.5 g/dL	Yes	N (%)	14 (3.5)	8 (29.6)	6 (1.6)	<0.0001
M Component Types						
1. IgG w/wo free light chains		N (%)	283 (69.7)	12 (44.4)	271 (71.5)	<0.0001
2. IgM w/wo free light chains or IgG		N (%)	38 (9.4)	2 (7.4)	36 (9.5)	
3. IgA w/wo free light chains or IgG		N (%)	41 (10.1)	10 (37.0)	31 (8.2)	
4. IgD w/wo free light chains		N (%)	2 (0.5)	1 (3.7)	1 (0.3)	

5. Free κ with or without λ light chains		N (%)	7 (1.7)	2 (7.4)	5 (1.3)	
6. Free λ light chains only		N (%)	35 (8.6)	0 (0.0)	35 (9.2)	
M Component Type (combined)						
IgG w/wo free light chains		N (%)	283 (69.7)	12 (44.4)	271 (71.5)	0.0031
All Others		N (%)	123 (30.3)	15 (55.6)	108 (28.5)	
M Component Types	1	N (%)	283 (69.7)	12 (44.4)	271 (71.5)	<0.0001
	2/4	N (%)	40 (9.85)	3 (11.1)	37 (9.76)	
	3	N (%)	41 (10.10)	10 (37.0)	31 (8.2)	
	5/6	N (%)	42 (10.34)	2 (7.4)	40 (10.6)	
CRAB†	0	N (%)	215 (52.96)	7 (25.93)	208 (54.88)	0.0036
	≥1	N (%)	191 (47.04)	20 (74.07)	171 (45.12)	
* One patient has known history of mucosal associated lymphoid tissue (MALT) lymphoma before M-component was discovered. † CRAB: 0 if none of the following four conditions: Calcium >11 mg/dL, Renal or creatinine >2 mg/dL, Anemia or hemoglobin (hgb) <10 g/dL, and Bone lesions present; ≥1 if at least one of the four condition are met						

Table 2: Baseline and Clinical Characteristics (n=406)

Baseline Predictors		OR (95% CI) †	p-value
Calcium (mg/dL)		2.74 (1.431, 5.260)	<0.01
Cr (mg/dL)		1.11 (0.912, 1.356)	0.29
Hgb (g/dL)		0.79 (0.615, 1.014)	0.06
Bone lytic lesions or pathologic fractures	Yes	8.93 (2.854, 27.928)	<0.01
M component >1.5 g/dL	Yes	45.78 (10.701, 195.824)	<.0001
M component type		13.93 (4.403, 44.056)	
	3 vs. 1		<.0001
	2/4 vs. 1	2.35 (0.472, 11.703)	0.29
	5/6 vs. 1	3.52 (0.725, 17.081)	0.12
C statistics‡	0.88		
* Logistic regressions, † When 95% CI does not include "1" it is statistically significant at α=0.05, ‡ C statistics is Area under the Receiving Operation Characteristics Curve >0.7 is a good fit, §, Akaike Information Criterion (AIC) = 120.11			

Table 3: Estimated odds Ratios (OR) and 95% Confidence Intervals (CI)* of Predictors for Occurrence of Lymphoplasmacytic Malignancies (LPM)

Risk Group	Who developed LPM	
	N	N (% per risk group)
Low	91	2 (2%)
Low intermediate	117	5 (4%)
High intermediate	61	17 (30%)
High	2	2 (100%)
Total	**271**	**26 (9.6%)**

Table 4: Mayo Clinic MGUS Progression Risk of Lymphoplasmacytic Malignancies (LPM)

4. Discussion and Conclusions

The results of our study show an inordinate number of SPEPs (1,249) were performed in patients with low or low-intermediate risk features with only a small number of patients subsequently found to have either MM or WM (26/406). Patients who were found to have either pre-existing lymphoma or subsequently developed MM or WM had significantly higher serum calcium compared to those without a LPM. The presence of bone lesions,

IgA M-component, M-component > 1.5g/dL, or at least one of the CRAB criteria was associated with an increased risk of developing a LPM.

For the clinician who orders an SPEP to look for monoclonal gammopathies, our observations offer guidance to increase the yield of finding a LPM or other diseases associated with monoclonal gammopathy and reduce unnecessary costs. LPM outcomes cannot be altered by early diagnosis and treatment and, as such, the United States Preventive Services Task Force does not recommend screening for LPM [11]. Treatment for multiple myeloma is tailored to patients with symptomatic disease with CRAB criteria; asymptomatic MM and low-grade lymphoma are observed without treatment [8]. Thus finding LPM in patients without CRAB criteria does NOT lead to treatment. In our patient population, a fair number of patients had SPEPs ordered for reasons other than any of the CRAB criteria. Monoclonal gammopathy of clinical significance (renal, neurological, dermatological) has garnered attention in recent years as some of these patients may benefit from treatment of the monoclonal gammopathy [12,13]. Therefore, we advocate for limiting SPEP testing to those with CRAB criteria and otherwise unexplained neurological and dermatological conditions.

The discovery of a monoclonal gammopathy triggers a referral to hematology for additional work-up and follow-up. The overall risk of progression to a LPM is estimated to be 25% in 20 years of follow-up, which means many years of hematology visits with repeated SPEPs that are costly [5]. In addition, since monoclonal gammopathy is statistically more common in the elderly population, a significant proportion of MGUS patients will not develop a LPM or other associated disorders in their lifetime [2]. While the Mayo Clinic MGUS progression risk stratification allows better assessment of the risk of progression from MGUS to LPM and informs the provider of the urgency of a hematology referral, one of the risk factors is an abnormal serum free light chain ratio. However, clinicians outside of hematology are often unaware of the risk stratification and may not order the serum free light chain analysis. Our study shows that only 271 out of 406 patients had the serum free light chain analysis performed, and most of these were ordered after the patients were seen by hematology. Using familiar clinical parameters such as serum calcium, serum creatinine, hemoglobin, and skeletal survey with plain X-rays, our study provides an accessible risk assessment for patients with monoclonal gammopathy. Based on our results, we recommend a referral to hematology if the monoclonal gammopathy is associated with CRAB criteria, the M-component is of IgA isotype or is >1.5 g/dL. The absence of CRAB criteria carries a lower risk of developing a LPM, but it is not zero; parameters including isotype and amount of the monoclonal protein should be used to determine if additional work-up is necessary.

The current recommendation is to repeat SPEP every 2-3 years once MGUS is diagnosed, but it is unclear whether this is high-value practice [5]. Based on our retrospective study, all patients who developed a LPM had one or more of the CRAB symptoms present at the time of diagnosis. Therefore, in a patient without CRAB symptoms and a low-risk MGUS, repeat testing may not be necessary.

If MGUS is present in 3% of the general US population, more than 13,000 patients would have had SPEPs done to have 406 patients with M-component on their SPEPs [2]. At least 3 SPEPs were done per patient in our cohort during the observation period, and many more would have been done if current monitoring guidelines are followed. The cost of an SPEP at our institution is over $300 per test. For our patient population alone, hundreds of thousands of dollars could have potentially been saved by sensibly choosing the appropriate patient to test and considering repeat testing only for patients with at least one of the four CRAB criteria. Targeted testing could also decrease the number of follow-up appointments and patient anxiety regarding the results of their tests.

As typical of retrospective studies, we relied on chart review to obtain the necessary information. Our data could be skewed by inaccurate or incomplete documentation, especially if patients had SPEPs performed at outside institutions not captured in the electronic health record. Some initial SPEPs may have been ordered while the patient was suffering from a systemic illness with associated increased creatinine or decreased hemoglobin, which are commonly seen with many other disease processes. Indiscriminate SPEP testing therefore may have diluted our results. The short-term observation period of 8 years is likely inadequate with the low average risk of progression from MGUS to LPM, which could account for the low number of patients with progression to an LPM.

Based on the observations made in our retrospective review, we recommend ordering SPEP for patients with CRAB criteria or otherwise unexplained neurological or dermatologic disorders to look for monoclonal gammopathy. If discovered, referral to a hematologist should be made in the setting of CRAB symptoms, monoclonal spike of IgA isotype, or a monoclonal spike of >1.5 g/dL. Understanding the indications for ordering an SPEP and tailoring follow-up testing and surveillance to the patient-specific characteristics and symptoms can minimize unnecessary testing and translate into significant cost savings.

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