OPEN ACCESS

GAVIN PUBLISHERS

Journal of Urology and Renal Diseases

Lee SJ, et al. J Urol Ren Dis: JURD-1101.

DOI: 10.29011/2575-7903.000100

Review Article

The Role of Protein Electrophoresis in Differential Diagnosis of Renal Disorders

Shu-Jene Lee[1], I-Hsin Lin[1], Tzung-Hai Yen[2], Fang-Yeh Chu[1,3,4,5*]

[1]Department of Clinical Pathology, Far Eastern Memorial Hospital, New Taipei, Taiwan

[2]Division of Nephrology and Clinical Toxicology, Chang Gung Memorial Hospital, Lin-Kou Medical Center, Taoyuan, Taiwan

[3]School of Medical Laboratory Science and Biotechnology, Taipei Medical University, Taipei, Taiwan

[4]Graduate School of Biotechnology and Bioengineering, Yuan Ze University, Taoyuan, Taiwan

[5]Department of Medical Laboratory Science and Biotechnology, Yuanpei University, Hsinchu, Taiwan

[*]**Corresponding author:** Fang-Yeh Chu, Department of Clinical Pathology, Far Eastern Memorial Hospital, No. 21, Sec. 2, Nanya S. Road, Banqiao, New Taipei City, Taiwan. Tel: +886277282111; Fax: +886277281003; Email: jacpha@mail.femh.org.tw

Citation: Lee SJ, Lin IH, Yen TH, Chu FY (2018) The Role of Protein Electrophoresis in Differential Diagnosis of Renal Disorders. J Urol Ren Dis: JURD-1101. DOI: 10.29011/2575-7903. 001101

Received Date: 26 June, 2018; **Accepted Date:** 13 July, 2018; **Published Date:** 19 July, 2018

Abstract

With the advance of laboratory technologies, protein electrophoresis has been a commonly available screening and monitoring tool for certain diseases in clinical practice. As the pattern of protein electrophoresis varies in different situations involved with renal disorders or complications, it helps the clinical pathologist and laboratory technician for intepretation and differential diagnosis of certain renal disorders. Here, we briefly introduced the specific characteristics of protein electrophoresis in various conditions involved with renal disorders and combined our own results and experiences to outlook the clinical application of protein electrophoresis in differential diagnosis of renal disorders.

Keywords: Protein Electrophoresis, Renal disorders

Introduction

Protein Electrophoresis (PE), developed by Arne Tiselius in the early-mid 20th century, is a laboratory technique for protein separation [1]. Nowadays, PE has been a commonly available screening and monitoring tool for certain diseases in clinical practice [2-6]. The main purpose of PE is to identify whether there is monoclonal gammopathy from other differential diagnosis of serum protein-related disorders [6]. Due to the development of high resolution and capillary electrophoresis [7], the results of PE have been easier to be interpreted by the clinical pathologist and laboratory technician in comparison with the results of agarose gel electrophoresis in conventional. Though PE helps in evaluating a variety of clinical situations at the initial stage, sometimes the electrophoretic results are confusing and needed to be interpreted carefully. The pattern of PE in the serum could be primarily divided into five components, including albumin, alpha-1, alpha-2, beta-1/beta-2, and gamma globulins, on the basis of their electrophoretic mobilities (Figure 1).

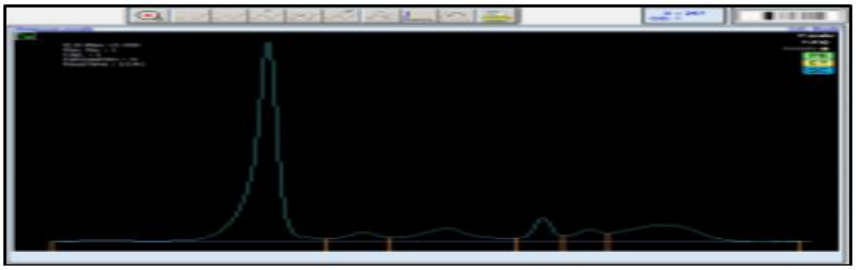
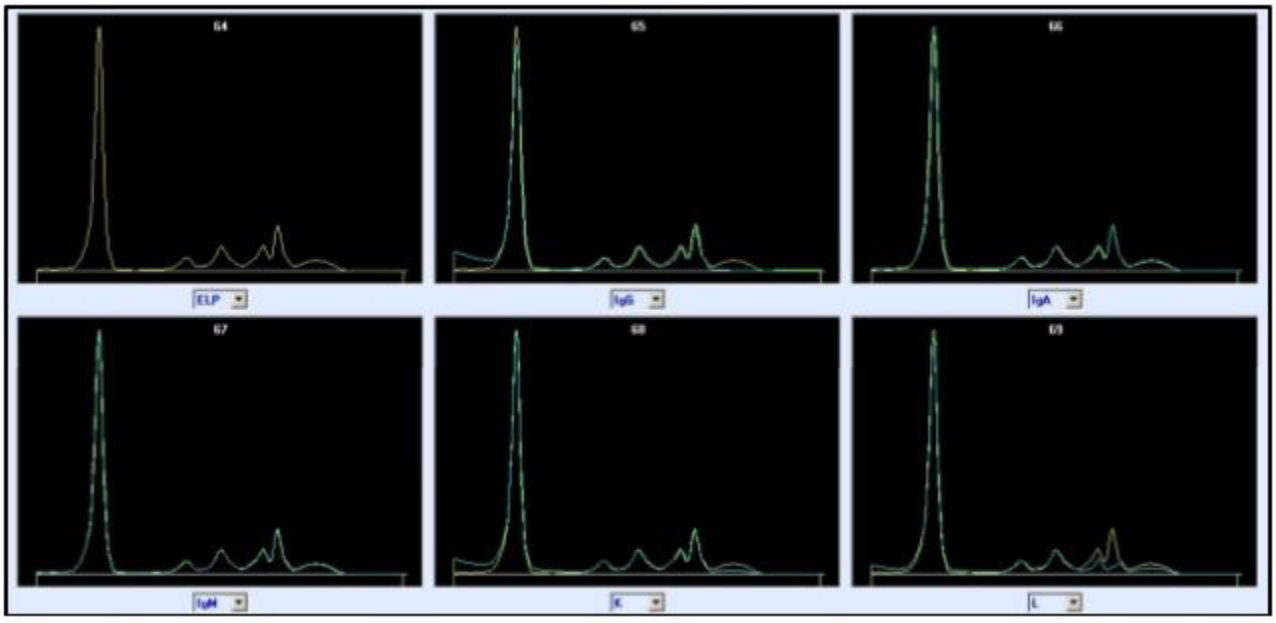


Figure 1: The referenced normal pattern in serum protein electrophoresis, including albumin, alpha-1, alpha-2, beta-1/beta-2, and gamma globulins. The capillary electrophoresis was performed by Sebia Capillarys 2 Flex Piercing systems.

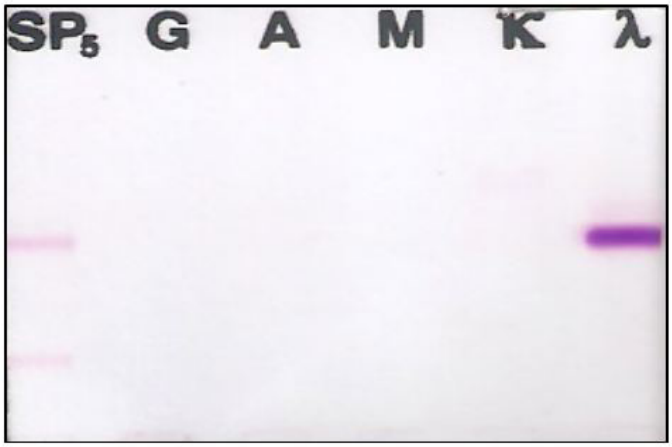
Of these, alpha-1 fraction is mainly composed of antichymotrypsin, antitrypsin and acid glycoprotein; alpha-2-macroglobulin and haptoglobin contribute to alpha-2 fraction; beta fraction is mainly composed of beta-lipoprotein, complement components, transferrin, antithrombin III and fibrinogen; while gamma fraction is manifestly composed of immunoglobulins (Igs; such as IgA, IgD, IgE, IgG and IgM) and C-reactive protein. The pattern of PE varies in different conditions involved with renal disorders. Here, the electrophoretic characteristics of PE in various renal disorders in the serum and urine would be discussed in the following sections.

Protein Electrophoresis in Light Chain Disease and Light Chain Myeloma

Light chain disease was commonly caused by amyloidosis and light chain myeloma. Of which, light chain myeloma belongs to one subtype of myeloma, featured by plasma cell proliferation that produces a monoclonal immunoglobulin in the bone marrow [8,9]. Therefore, serum PE with immunofixation or immunosubtraction is indicated when myeloma was suspected. In light chain disease, only monoclonal light chain of immunoglobulin is overproduced with lack of heavy chain expression. According to the literature review, about 15-20% of myeloma expressed exclusively monoclonal light chain in the serum or urine [9-11]. Patients with light chain disease could present with renal insufficiency, proteinuria and even nephrotic syndrome. It was also shown that light chain disease contributed to approximately 20-60% of renal injury in myeloma, implying that light chain filtration could be nephrotoxic in the kidney [9,11-13]. Free light chain was previously described as Bence-Jones protein in the urine [14]. Thus, detection of Bence-Jones protein used to play an important role in the investigation of light chain disease, especially when monoclonal component was undetectable in serum PE [15, 16]. To date, light chain immunoassay and urine PE with immunofixation were commonly performed for myeloma diagnosis in hematologic laboratories [17,18]. Capillary electrophoresis with urine specimens has not been routinely practiced in clinical yet, as certain organic acids and metablolites in the urine may interfere with the technique and pretreatment of urine samples is needed. Two kinds of light chains have been identified, kappa and lambda. There was no significant difference in distribution of kappa and lambda light chain myeloma [10, 19]. In one study from 2005 to 2012, 96 cases of light chain myeloma were diagnosed from 459 mutiple myeloma patients. Among these 96 patients, 42 were kappa type and 54 were lambda type [10]. In another investigation, 17 kappa and 20 lambda light chain myeloma cases were enrolled [19]. Similar to our experience, a total of 28 cases were diagnosed as light chain disease with the evaluation of urine PE with immunofixation and other evidence of histological pathology from 2014 to 2018 in our institution. Of these, 13 patients were diagnosed as kappa light chain disease and the remaining 15 patients belonged to lambda light chain disease. The serum PE with immunosubtraction and urine PE with immunofixation in case of free lambda light chain myeloma were shown in (Figure 2).



(A)



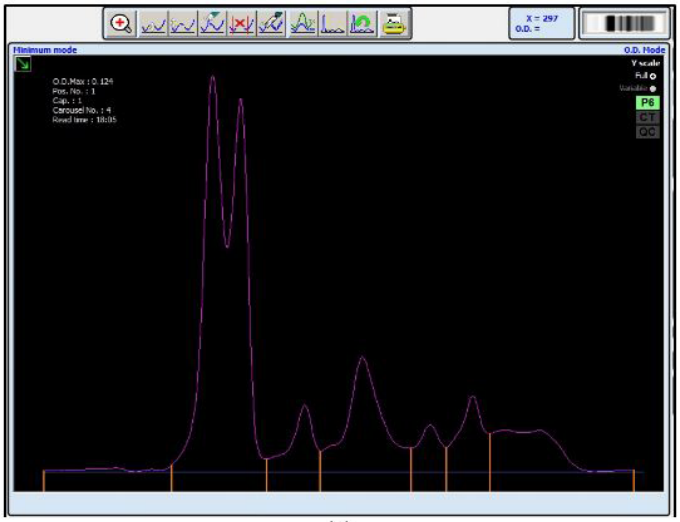
(B)

Figure 2: The serum protein electrophoresis with immunosubtraction **(A)** and urine protein electrophoresis with immunofixation **(B)** in an 81-year-old female who was histopathologically diagnosed as free lambda light chain myeloma. In the serum protein electrophoresis with immunosubtraction, the restricted peak could only be subtracted by anti-lambda light chain antibody. The urine protein electrophoresis with immunofixation also revealed monoclonal gammopathy of free lambda light chain. The serum protein electrophoresis with immunosubtraction was performed by Sebia Capillarys 2 Flex Piercing systems. The urine protein electrophoresis with immunofixation was performed by Helena SPIFE 3000 Electrophoresis Analyzer.

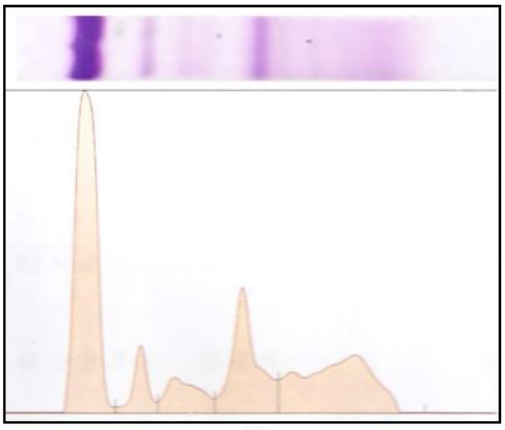
It was noticeable that there were two small peaks could be subtracted by anti-lambda light chain antibody, implying the possibility of polymerization of globular proteins. In such situations, specimens could be treated by beta-mercaptoethanol, a reducing agent that cleaves the disulfide bonds, and therefore prevent the protein polymerization. Actually, pretreatment of beta-mercaptoethanol has been commonly utilized to help identification of monoclonal proteins in serum PE with capillary electrophoresis techniques [20]. Moreover, it was reported that overexpression and deposition of lambda light chain is often associated with light chain amyloidosis [12,21]. Importantly, approximately 10% myeloma cases presented panhypogammaglobulinemia with significant Bence-Jones proteinuria [6,9]. Accordingly, urine PE with immunnnofixation is always recommended in patients who have panhypogammaglobulinemia in serum PE.

Protein Electrophoresis in Protein-Losing Nephropathy and Nephrotic Syndrome

Nephrotic syndrome is presented with heavy proteinuria, hypoalbuminemia, peripheral edema and hyperlipidemia [22]. The urinary protein loss in nephrotic syndrome could be greater than 3 g per day, or more than 3,000 mg per g creatinine in protein-to-creatinine ratio in the spot urine specimen [23]. In serum PE, the pattern of protein-losing nephropathy and nephrotic syndrome is characterized by the increase of alpha-2 fraction and the decrease of albumin and gamma globulins. However, the diagnostic value of PE on a routine basis reamins controversial in nephrotic syndrome, because serological examinations were seldom abnormal without clinical suspicion [24]. Rarely, bisalbuminemia could be observed in serum or urine PE in nephrotic syndrome [25-27] with an estimated incidence of 0.02-0.033% [27]. Bisalbuminemia could be congenital or inherited. And several situations such as pancreatic psuedocyst or beta-lactam use may contribute to acquired bisalbuminemia [27-29], leading to the presence of double peaks in PE. It was reported that there was a better performance of capillary PE in albumin separation than PE with agarose gel, making bisalbuminemia detection easier [30]. However, the underlying mechanism of the association of bisalbuminemia in nephrotic syndrome is still unknown. In our experience, serum PE with capillary electrophoresis technique and urine PE with agarose gel helped to diagnose a case of nephrotic syndrome with bisalbuminemia, which were shown in (Figure 3).



(A)



(B)

Figure 3: Bisalbuminemia in nephrotic syndrome. A 70-year-old male who had stage 4 Chronic Kidney Disease (CKD) presented with progressive leg edema, hypoalbuminemia and heavy proteinuria. Nephrotic syndrome with early stage of focal segmental glomerulosclerosis was diagnosed histopathologically. The serum protein electrophoresis revealed increased

alpha-2 region and decreased of albumin and gamma globulins, compatible with the pattern of protein loss **(A).** Bisalbuminemia was also found in serum and urine protein electrophoresis. It was noticeable that the double peaks of albumin could be identified easier by capillary electrophoresis using serum specimen **(A)** than agarose gel electrophoresis using urine specimen **(B).** The serum protein electrophoresis was performed by Sebia Capillarys 2 Flex Piercing systems. The urine protein electrophoresis was performed by Helena SPIFE 3000 Electrophoresis Analyzer.

Interpretation of Protein Electrophoresis in Some Complicated Conditions

For patients who had multiple comorbidities, the pattern PE could be complicated, leading to difficulties for the clinical pathologist and laboratory technician to intepret. Here we presented some cases with the electrophoretic characteristics of PE that were somewhat complexed. Case 1: A 57-year-old female who was healthy before presented with frothy urine for at least 2 months. Hyperlipidemia and heavy proteinuria were found on health examination. Further investigation accidentally revealed monoclonal gammopathy, IgG lambda, in the serum protein electrophoresis. Increased alpha-2 region and reduced albumin level were also detected, suggestive of protein loss pattern (Figure 4).

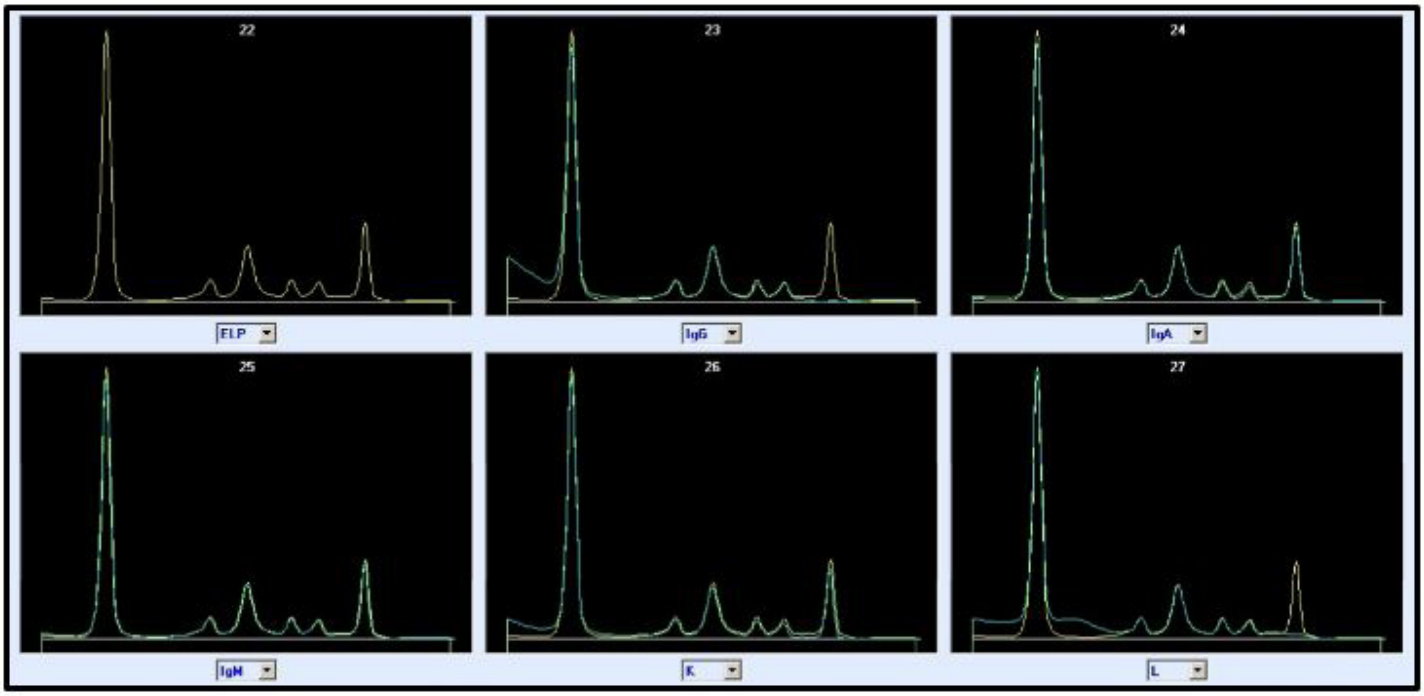


Figure 4: Heavy proteinuria accompanied with monoclonal gammopathy of undetermined significance. A 57-year-old female who was healthy before was found to have hyperlipidemia and heavy proteinuria on health examination. Further investigation accidentally revealed monoclonal gammopathy, IgG lambda, in the serum protein electrophoresis. Increased alpha-2 region and reduced albumin level were also detected, suggestive of protein loss pattern. The serum protein electrophoresis with immunosubtraction was performed by Sebia Capillarys 2 Flex Piercing systems.

Besides, primary long bone imaging studies revealed no osteolytic lesions, and serum creatinine (0.57mg/dL) and IgG (1.15g/dL) levels were within the referenced normal range. Mild anemia with severe microcytosis (hemoglobin: 11.0g/dL; mean corpuscular volume: 62.0 femtoliter) was found on Complete Blood Count (CBC) analysis. The patient was then referred to hematologist for further management. Biopsy of the kidney and bone marrow was suggested but the patient hesitated. Heavy proteinuria with Monoclonal Gammopathy Of Undetermined Significance (MGUS) was impressed and close observation was kept. Case 2: A 97-year-old male patient who was healthy before presented with intermittent fever and generalized malaise for 2 weeks and was thus brought to our emergency room for further assessment. The CBC analysis revealed marked leukocytosis with predominant lymphocytosis, mild normocytic anemia and thrombocytopenia. Pyuria and bacteriuria were also found on urinalysis. Besides, rouleaux formation of red cells was observed on peripheral blood smear and serum albumin and globulin levels were measured to be 2.6 and 7.9 g/dL, respectively. Serum IgG, IgA and IgM concentrations were estimated to be 2.58, 0.28 and 2.59 g/dL, respectively. The serum PE with immunosubtraction revealed a monoclonal peak over beta-gamma region that could be subtracted by anti-IgM and anti-lambda antibodies. Additionaly, one broad peak over gamma region was subtracted by anti-IgG antibody, and partially subtracted by both anti-kappa and anti-lambda antibodies, implying the possiblility of chronic inflammation (Figure 5).

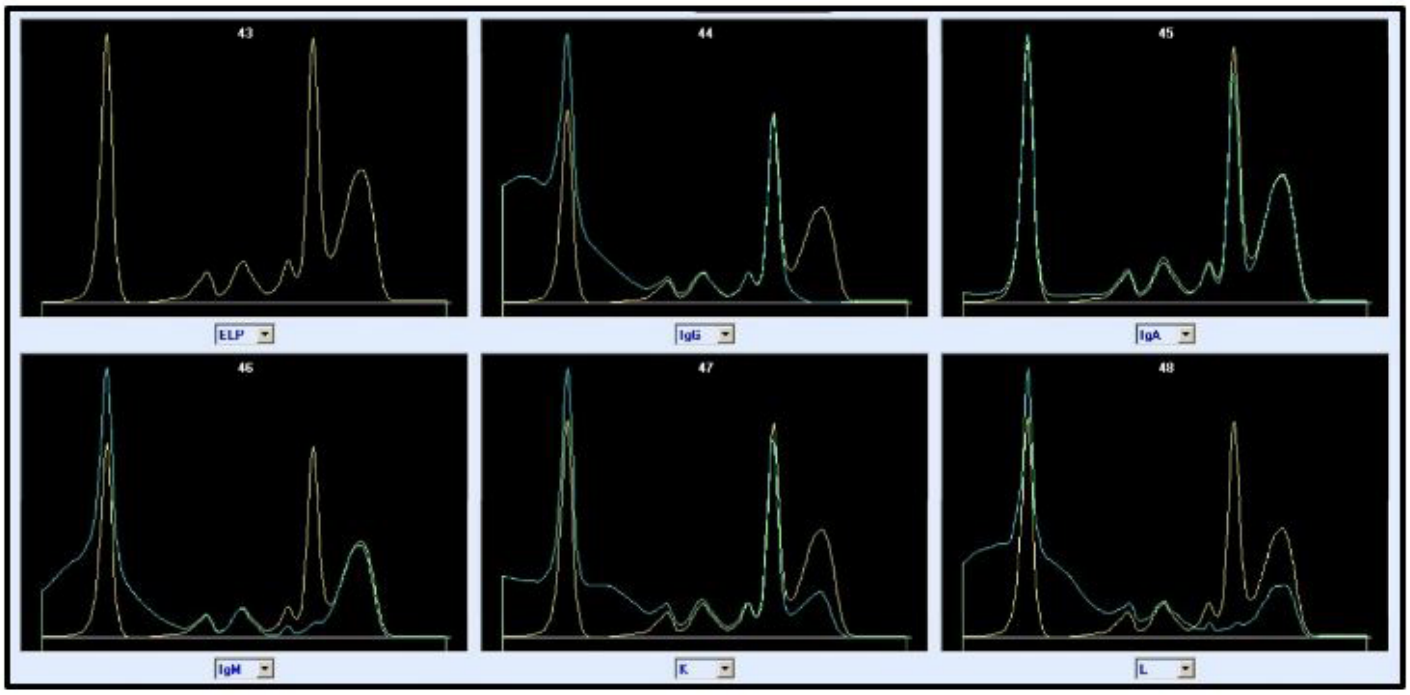


Figure 5: The serum protein electrophoresis with immunosubtraction in a 97-year-old male who was suspected to have Waldenströmmacroglobulinemia with concurrent pyuria and bacteriuria. The monoclonal peak over beta-gamma region could be subtracted by anti-IgM and anti-lambda antibodies. The polyclonal peak over gamma region was subtracted by anti-IgG antibody, and was partially subtracted by both anti-kappa and anti-lambda antibodies. This implied the possibility of chronic inflammation. The serum protein electrophoresis with immunosubtraction was performed by SebiaCapillarys 2 Flex Piercing systems.

The marrow blood smear showed predominant lymphocytes and lymphoplasmacytoid cells. The flow cytometry of bone marrow revealed strong expression of B lymphoid cells (CD20/CD19/surface lambda light chain) with weakly positivity for CD38 and negativity for CD138. The overall picture was compatible with Waldenström macroglobulinemia [31,32]. Considering the extremely old age of this patient, conservative management was given.

Conclusions

As more commonly available with advanced technique in laboratories, PE has been used to help diagnostic establishment in specific disorders as well as monitoring the disease progression. As the pattern of PE varies in different situations involved with renal disorders or complications, the electrophoretic characteristics of PE help the clinical pathologist and laboratory technician for intepretation and differential diagnosis of certain renal disorders.

Conflict of Interest

None

Author Contributions

Shu-Jene Lee and I-Hsin Lin contributed as co-first authors. Fang-Yeh Chu contributed as corresponding author. The manuscript has been read and approved by allauthors.

References

1. Henry's Clinical Diagnosis and Management by Laboratory Methods, 23rd Edition (2016) Chapter 19: Specific Protein.
2. Lahuerta JJ, Martinez-Lopez J, Serna JD, Bladé J, Grande C, et al. (2000) Remission status defined by immunofixation vs. electrophoresis after autologous transplantation has a major impact on the outcome of multiple myeloma patients. Br J Haematol 109: 438-446.
3. Dispenzieri A, Kyle R, Merlini G, Miguel JS, Ludwig H, et al. (2009) International Myeloma Working Group guidelines for serum-free light chain analysis in multiple myeloma and related disorders. Leukemia 23: 215-224.
4. Rajkumar SV, Harousseau JL, Durie B, Anderson KC, Dimopoulos M, et al. (2011) Consensus recommendations for the uniform reporting of clinical trials: report of the International Myeloma Workshop Consensus Panel 1. Blood 117: 4691-4695.
5. Muchtar E, Buadi FK, Dispenzieri A, Gertz MA (2016) Immunoglobulin Light-Chain Amyloidosis: From Basics to New Developments in Diagnosis, Prognosis and Therapy. Acta Haematol 135: 172-190.
6. O'Connell TX, Horita TJ, Kasravi B (2005) Understanding and interpreting serum protein electrophoresis. Am Fam Physician 71: 105-112.

7. Jenkins MA (2009) Serum and urine electrophoresis for detection and identification of monoclonal proteins. Clin Biochem Rev 30: 119-122.

8. Kariyawasan CC, Hughes DA, Jayatillake MM, Mehta AB (2007) Multiple myeloma: causes and consequences of delay in diagnosis. QJM 100: 635-640.

9. Kyle RA, Gertz MA, Witzig TE, Lust JA, Lacy MQ, et al. (2003) Review of 1027 patients with newly diagnosed multiple myeloma. Mayo Clin Proc 78: 21-33.

10. Zhang JJ, Sun WJ, Huang ZX, Chen SL, Zhong YP, et al. (2014) Light chain multiple myeloma, clinic features, responses to therapy and survival in a long-term study. World J Surg Oncol 12: 234.

11. International Myeloma Working Group (2003) Criteria for the classification of monoclonal gammopathies, multiple myeloma and related disorders: a report of the International Myeloma Working Group. Br J Haematol 121: 749-757.

12. Heher EC, Rennke HG, Laubach JP, Richardson PG (2013) Kidney disease and multiple myeloma. Clin J Am Soc Nephrol 8: 2007-2017.

13. Knudsen LM, Hippe E, Hjorth M, Holmberg E, Westin J (1994) Renal function in newly diagnosed multiple myeloma--a demographic study of 1353 patients. The Nordic Myeloma Study Group. Eur J Haematol 53: 207-212.

14. Jones HB (1847) Papers on chemical pathology, lecture III. Lancet 11: 88-92.

15. Perry MC, Kyle RA (1975) The clinical significance of Bence Jones proteinuria. Mayo Clin Proc 50: 234-238.

16. Beetham R (2000) Detection of Bence-Jones protein in practice. Ann Clin Biochem 37: 563-570.

17. Dejoie T, Attal M, Moreau P, Harousseau JL, Avet-Loiseau H (2016) Comparison of serum free light chain and urine electrophoresis for the detection of the light chain component of monoclonal immunoglobulins in light chain and intact immunoglobulin multiple myeloma. Haematologica 101: 356-362.

18. Bradwell AR, Carr-Smith HD, Mead GP, Harvey TC, Drayson MT (2003) Serum test for assessment of patients with Bence Jones myeloma. Lancet 361: 489-491.

19. Song P, An ZM, Zhou XG, Li F, Wang LP, et al. (2015) Test of Serum Free Light Chain and Its Clinical Significance in Light Chain Multiple Myeloma. Zhongguo Shi Yan Xue Ye Xue Za Zhi 23: 1357-1361.

20. Keren DF, Gulbranson R, Carey JL, Krauss JC (2001) 2-Mercaptoethanol treatment improves measurement of an IgMkappa M-protein by capillary electrophoresis. Clin Chem 47: 1326-1327.

21. Preud'homme JL, Ganeval D, Grünfeld JP, Striker L, Brouet JC (1988) Immunoglobulin synthesis in primary and myeloma amyloidosis. Clin Exp Immunol 73: 389-394.

22. Harrison's Principles of Internal Medicine, 19th Edition (2015) Chapter 338: Nephrotic syndrome.

23. Hull RP, Goldsmith DJ (2008) Nephrotic syndrome in adults. BMJ 336: 1185-1189.

24. Howard AD, Moore J Jr, Gouge SF, Lockard JW, Melton KD, et al. (1990) Routine serologic tests in the differential diagnosis of the adult nephrotic syndrome. Am J Kidney Dis 15: 24-30.

25. Ahmad J, Khan AS, Siddiqui MA, Tewari SG, Khan RU (1984) Bisalbuminemia in nephrotic syndrome (a case report). Jpn J Med 23: 45-47.

26. Hoang MP, Baskin LB, Wians FH Jr (1999) Bisalbuminuria in an adult with bisalbuminemia and nephrotic syndrome. Clin Chim Acta 284: 101-107.

27. Akhmouch I, Alayoud A, Bahadi A, Zajjari Y, Montasser D, et al. (2012) Bisalbuminemia during remission of nephrotic syndrome. Saudi J Kidney Dis Transpl 23: 1251-1253.

28. Andre P. Garre M, Youinou P, Saleun JP (1977) Les bisalbuminemies. Trois étiologies: Mutation héréditaire, antibiothérapie, pseudokyste du pancréas. Nouv Presse Med 6: 4123-4124.

29. Bachmeyer C, Grateau G, Dhote R, Dauchy F (1996) Une autre électrophorèse insolite. Revue Médecine Interne 17: 5.

30. Kalambokis G, Kitsanou M, Kalogera C, Kolios G, Seferiadis K, et al. (2002) Inherited bisalbuminemia with benign monoclonal gammopathy detected by capillary but not agarose gel electrophoresis. Clin Chem 48: 2076-2077.

31. Owen RG, Barrans SL, Richards SJ, O'Connor SJ, Child JA, et al. (2001) Waldenström macroglobulinemia. Development of diagnostic criteria and identification of prognostic factors. Am J Clin Pathol 116: 420-428.

32. Sethi B, Butola KS, Kumar Y (2012) A Diagnostic Dilemma: Waldenström's Macroglobulinemia/Plasma Cell Leukemia. Case Rep Pathol 2012: 271407.