Optimization of Serum Protein Separation by Capillary Electrophoresis

To the Editor:

Identification of serum paraproteins is important for diagnosing myeloma and monoclonal gammopathy of undetermined significance (MGUS), which occurs in >3% of the population age 70 years and older. In 1995 we described a prospective comparison of 1000 specimens for protein electrophoresis using high-resolution agarose gel electrophoresis (HRAGE) compared with capillary electrophoresis (CE). We reported two specimens (of a total of 362 paraproteins) in which capillary electrophoresis did not adequately quantify the paraprotein. At the time we noted that the retention time for albumin was markedly increased, and we used that factor to identify these “problem” specimens. Removal of calcium lactate from the borate buffer enabled correct separation of the monoclonal protein.

Since that time a further 5500 specimens have been processed by CE, with identification of a further 6 paraproteins that did not separate correctly on CE.

Of the eight paraproteins that did not separate correctly under the conditions described [1], five were IgM (1.4% of total IgM paraproteins screened) and three were IgG (0.3% of the total IgG paraproteins). We noted on HRAGE that all these proteins migrated in the very slow gamma region.

Isoelectric focusing of the paraproteins revealed that pI of the IgG proteins was higher than 8.5 while that of the IgM paraproteins was between 6.9 and 8.3 (see example in Fig. 1). By modification of the boric acid buffer system from 50 mmol pH 9.7 to 75 mmol pH 10.3, we were able to quantify all of the monoclonal bands.

We believe that by the use of increased ionic strength and increased pH, the modified buffer will correctly separate and quantify all monoclonal paraproteins.

Reference


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