Clinical Chemistry Trainee Council
Pearls of Laboratory Medicine
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TITLE: Monoclonal Gammopathies

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Slide 1:
Hello, my name is Jerry Katzmann. I am a director of the clinical immunology laboratory and an associate professor of laboratory medicine at Mayo Clinic. Welcome to this Pearl of Laboratory Medicine on "Monoclonal Gammopathies."

Slide 2:
Monoclonal gammopathies secrete a monoclonal immunoglobulin, or M-protein, that can be used as a serologic "tumor" marker for a group of disorders that are also known as plasma cell proliferative diseases.

As shown on this slide, this tumor marker is extremely diverse. Monoclonal proteins run the gamut from pentameric IgM at ~900,000 Daltons to monomeric free light chains of 24,000 Daltons. In addition, although some plasma cell proliferative disorders present with M-protein concentrations of grams per liter serum, others have little or virtually no circulating M-protein. The diversity of plasma cell proliferative diseases, the spectrum of their secreted proteins, and the range of concentrations make detection a challenge. No single assay can effectively diagnose and monitor plasma cell proliferative diseases in all patients, and the laboratory needs to define strategies that encompass the spectrum of disease presentations. Protein electrophoresis (PEL) and immunofixation electrophoresis (IFE) of serum and urine have formed the hub of laboratory testing for detecting and monitoring monoclonal gammopathies. These electrophoretic assays analytically separate immunoglobulins based on charge and size. In addition to electrophoresis, quantitative immunoassays of immunoglobulin free light chains may be useful for diagnosis. This presentation will discuss the challenge of detecting plasma cell proliferative diseases and the use of test panels to effectively detect and manage patients.

Slide 3:
In this next slide, I have listed the incidence of 12 different monoclonal gammopathies seen in the Mayo Clinic practice during a span of almost 50 years. This compilation is based on laboratory and clinical presentation. The 3 most common monoclonal gammopathies are highlighted in yellow, and you can see that the most common disorder is Monoclonal Gammopathy of Undetermined Significance (MGUS), followed by multiple myeloma, followed by primary amyloidosis. MGUS is a premalignant disorder, multiple myeloma is a malignancy, and primary amyloidosis is a protein disease.

Slide 4:
The detection of monoclonal gammopathies relies predominantly on the ability to differentiate between monoclonal and polyclonal immunoglobulins, and this has traditionally been done with electrophoretic assays.

On the top left side of this composite slide, you can see the serum protein electrophoresis gel and scan of a normal serum. The gamma fraction has a smooth, Gaussian distribution of all the normal immunoglobulins. Below the PEL is the immunofixation electrophoresis gel showing the distribution of gamma, alpha, and mu heavy chains, as well as the associated kappa and lambda light chains.
On the right side of the slide are the serum PEL and IFE gels from a patient with multiple myeloma. The M-protein is easily seen as a discrete band, and the IFE indicates that it is an IgG kappa protein.

Protein electrophoresis is a relatively simple procedure that can detect monoclonal proteins based on their restricted migration. In addition, the gel scan quantitates the M-protein as a percentage of the serum protein, and when combined with the serum total protein assay, it can quantitate and monitor the monoclonal protein.

Slide 5:
Not all myeloma patients, however, have M-proteins that are so easily detected and quantitated. In this slide, I have listed the distribution of monoclonal proteins detected in a series of 1,027 newly diagnosed myeloma patients. The types of myeloma listed in “red” may be difficult or impossible to detect in serum by electrophoresis. Nonsecretory myeloma represents 3% of all myelomas, and these clones have either lost the ability to synthesize the monoclonal immunoglobulin or are unable to secrete the protein. Free light chain myeloma represents 20% of all myelomas, and these clones have lost the ability to synthesize heavy chain. The clone may secrete large amounts of monoclonal light chain but it is quickly removed from circulation.

Slide 6:
On the left side of this next slide, I am showing you the serum electrophoresis gel of a patient with light chain multiple myeloma. The gamma fraction is suppressed, the beta fraction is asymmetric, but there is no large M-protein. The immunofixation electrophoresis gel contains a discrete lambda band in the beta region but notice that there is no corresponding heavy chain.

On the right side of the slide is the analysis of urine. The monoclonal lambda light chains have been cleared by the kidneys and are easily detected and quantitated. Because of the renal clearance of free light chains, you can see why it previously had been recommended to electrophorese both serum and urine.

Slide 7:
There are some plasma cell proliferative disorders that are difficult to detect even with analysis of serum and urine. On this slide is the electrophoretic analysis of serum and urine from a patient with primary amyloidosis. The monoclonal lambda light chain is a very faint band in the beta/gamma region. Over time, this small amount of light chain has formed amyloid fibrils in the kidneys that have caused the renal damage and proteinuria.

Primary amyloid is caused by a subset of monoclonal free light chains that have the property of forming amyloid fibrils. Although primary amyloid may be a “low tumor burden” disease, the survival is similar to myeloma, and diagnosis and treatment must be done in a timely manner to avoid irreversible organ damage.

Slide 8:
This cartoon illustrates (in red) the antigenic sites that the free light chain (FLC) assay exploits. When a light chain is bound to heavy chain, the interaction is so strong that portions of the light chain are hidden. If the light chain circulates in an unbound form, then the cryptic site is now accessible. The red color in the slide represents the light chain surface that is targeted for generation of antibodies to free light chains. The antisera to kappa and lambda FLC are 10,000-fold more reactive with FLC than light chains contained in intact immunoglobulin. That means that immunoassays can quantitate FLC even in the presence of normal serum immunoglobulin.

Slide 9:
The FLC assay is usually done as a paired assay to quantitate kappa FLC and lambda FLC. The FLC K/L ratio is used to detect excess clonal synthesis of light chains. If the ratio is elevated, it implies an expansion of a clone of kappa plasma cells, and if it is below normal, it suggests an expansion of a clone of lambda producing cells. Because an abnormal ratio implies a monoclonal gammopathy, the reference range for the ratio is generally accepted as a 100% range so that 5% of the population is not defined as having a plasma cell proliferative disorder.

Slide 10:
This slide illustrates free light chain results found in different patient populations. The magenta dots are sera from normal donors and you can see they fall within the red box of the normal kappa and lambda concentrations as well as within the blue rectangle defining the normal FLC ratio. The green dots represent patients with renal failure. They have elevated concentrations but the ratios are normal. The blue triangles and black open squares are sera from patients with light chain myeloma and the ratios are all abnormal.

Slide 11:
Now that we’ve added nephelometric serum FLC assays to serum and urine electrophoretic assays for detection of monoclonal gammopathies, how shall we use them all? This table shows the diagnostic sensitivity for the 3 serum assays when used in isolation. You can see that no single assay identifies all the patients.

Slide 12:
As expected, diagnostic sensitivity improves when we put the assays together into panels. The traditional panel employing serum and urine in the left column is improved by the addition of the serum FLC assay. What is of interest is that the panel of serum assays does almost as well as panels which also contain urine assays (note the darker coral column), and we can eliminate the requirement for a urine sample as part of the diagnostic screen.

In addition, a panel of serum protein electrophoresis and serum FLC quantitation provides a simple screen for the presence of multiple myeloma. The use of a panel of serum tests is now recommended as an efficient screening panel for monoclonal gammopathies, and urine is only required if primary amyloidosis is suspected and to assess renal function.

Slide 13: References

Slide 14: Disclosures

Slide 15: Thank You from www.TraineeCouncil.org

Once again, I am Jerry Katzmann. Thank you for joining me on this Pearl of Laboratory Medicine on "Monoclonal Gammopathies."