Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

(Number 2 of a 9 Module Series)

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TABLE OF CONTENTS

Purpose ................................................................................................................................. 2
Objectives ............................................................................................................................ 2
Advanced Preparation Assignment .................................................................................. 2
Context .............................................................................................................................. 3
Facilitation Schema ......................................................................................................... 3
Readiness Assurance Test (RAT) ..................................................................................... 4
  Item Analysis & Mean Scores ....................................................................................... 5
  RAT Items ..................................................................................................................... 6
Group Activity Exercises ............................................................................................... 11
  Problem 1 ....................................................................................................................... 12
  Problem 2 ....................................................................................................................... 15
Purpose of Module

By the conclusion of this module, learners will have a working knowledge about the function and pathways for activation of complement, effector mechanisms of innate immunity, antibody structure, T cell receptor structure, major histocompatibility complex (MHC) molecule structure, and the molecular basis of antigen recognition by immunoglobulin and T cell receptors. Learner should be able to apply this knowledge in experimental settings.

Objectives

1. Learners will be able to evaluate an immunology experimental design related to innate immunity or the recognition of antigen by B cells and/or T cells, demonstrating the ability to:
   • Identify or provide examples of appropriate controls.
   • Justify the use of specific methods.
   • Assess whether or not a given method provides a desired measure.

2. Learners will be able to interpret data provided for a given experimental protocol, including the ability to:
   • Draw conclusions based on the data.
   • Recognize results that may not fit current understandings.
   • Discuss results in the context of current concepts of immunology.

3. Learners will be able to predict experimental results given an experimental protocol and relevant background information, including the ability to:
   • Decide between alternative experimental outcomes based on current immunology concepts.
   • Justify predicted results in the context of established immunology concepts.

4. Learners will be able to design an experiment to test a hypothesis, including the ability to:
   • Identify required reagents, cell lines, animal strains, etc.
   • Propose appropriate assays to measure experimental outcomes.
   • Organize a sequence of experiments.
   • Justify the proposed design in the context of current immunology concepts.

Advanced Preparation Assignment

1. Reading assignment:
   Chapter 2 “Innate Immunity”
   Chapter 3 “Antigen Recognition by B-cell and T-cell Receptors”

2. Attend relevant lectures.
   (e.g., Complement, Innate Immunity, Antibody Structure & Antigen Recognition, Antigen Recognition by T Cells, TCR & MHC Structure)

3. Review personal notes and lecture handouts.
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Context

This module is the second of nine modules designed to teach experimental immunology to basic scientists and clinician scientists conducting research in immunology and/or using immunological methods. Originally developed for second-year biomedical science MS and PhD students enrolled in a semester-long immunology graduate course, this material is also appropriate for basic science courses for medical and dental students and/or for review of immunology by residents and fellows.

In a graduate-level immunology course entitled “Principles of Immunology”, each TBL module has a specific reading assignment, and is preceded by a block of 3-4 lectures. An exam is administered after each set of three TBL modules. Exams consist of a combination of recall (short answer and essay) and problem-solving questions. The Group Activity Exercise problems in this module were derived from past exam questions.

TBL1: Basic Concepts in Immunology
*TBL2: Innate Immunity & Recognition of Antigen
TBL3: Receptor Diversity & Antigen Presentation
TBL4: Development of Lymphocyte Repertoires
TBL5: T Cell-Mediated Immunity
TBL6: Humoral Immune Response
TBL7: Immunity to Infection & Host Defense Failure
TBL8: Clinical Immunology
TBL9: Manipulation of the Immune Response

Facilitation Schema

For users unfamiliar with team based learning, the following books will be helpful.

Because of scheduling constraints for the graduate course for which this TBL material was developed, we implemented what might be described as a modified form of TBL (described in more detail below), as the activity had to be adapted to a 50-minute classroom session. A manuscript describing this application of TBL in biomedical science graduate education is in preparation, and describes improved student satisfaction and performance after implementation of TBL. The authors acknowledge that this is not the best way to implement TBL, and do not recommend it unless the faculty user has similar scheduling constraints. Regardless of this limitation in our implementation, this material should provide a useful resource that can be adapted to other immunology courses in a more standard TBL session.

Classes were divided into 4-6 teams with 5-7 students per team. Team diversity was maximized by distributing students into different teams based on academic major, gender, and ethnic background.

In our application of TBL, the Individual Readiness Assurance Test (IRAT) was closed book and consisted of only five (5) multiple choice questions answered on a bubble sheet (Scantron). This
number of IRAT questions was sufficient to allow representation from the relevant preceding lectures and elicit student comments in course evaluations about how preparing for the TBL sessions helped them keep up with the material and come to class prepared, thus fulfilling the goals of the readiness assurance process.

The Group Readiness Assurance Test (GRAT) was open book and consisted of the same five multiple choice questions answered on an Immediate Feedback Assessment Technique (IF-AT) card (Epstein Educational Enterprises). GRAT scores were based on awarding 3 points for a correct answer on the first try, 2 points on the second try, etc.

The Group Activity Exercise (GAE) was open book and consisted of a multi-part problem including 2-3 multiple choice questions and 1-3 discussion questions focused on a single problem in experimental immunology. Because of the limited amount of time available, only one problem was included in our application. Some multiple choice questions were answered by the team on the IF-AT card, which allowed students to work out partial solutions to problems in a step-wise fashion and saved some discussion time. GAE scores were based on awarding 3 points for a correct answer on the first try, 2 points on the second try, etc. If time allows, the faculty user of this resource may wish to forego use of the IF-AT cards entirely and use all of the multiple choice questions as discussion questions as described below.

Discussion questions in our application sometimes included multiple choice questions that were not answered on the IF-AT cards. Instead, in order to facilitate inter-team discussion, a set of four answer cards (A-D, printed on cardboard of four different colors), was used for simultaneous reporting of team answers on multiple choice discussion questions. Discussion questions also included open-ended questions that were answered by teams that either volunteered or were called upon by the faculty member leading the class discussion.

If the faculty user has scheduling constraints similar to ours, the following TBL session schedule was found to work well for a 50 minute class period:

- Individual Readiness Assurance Test 5-10 minutes
- Group Readiness Assurance Test 5-10 minutes
- Group Activity Exercise 10-30 minutes
- Faculty-Led Class Discussion 5-10 minutes

In our graduate-level immunology course entitled “Principles of Immunology”, TBL constituted 25% of the students’ final grades, with each of three exams counting 25% each. The relative weights of average scores for each TBL component were determined by the faculty in 2006 and 2007 (10% IRAT, 5% GRAT, and 10% GAE), and by student vote in 2008 (7% IRAT, 9% GRAT, and 9% GAE).

Readiness Assurance Test

A set of 15 multiple choice questions is provided. These questions have been used in sets of five questions (#1-5, #6-10, and #11-15) for the Individual Readiness Assurance Test and the Group Readiness Assurance Test. These questions are provided as a resource, and faculty users are encouraged to select any desired subsets of questions from this and/or other modules to tailor this exercise to their application.

Item analysis data based on student performance on Individual Readiness Assurance Tests is provided below for all questions. Please note that not all items adhere to current guidelines of the National Board of Medical Examiners for multiple choice questions. Faculty users should feel free to modify these questions as needed for their application.
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Item Analysis

The following item analysis based on IRAT performance is provided to help users determine the suitability of each item (Crocker L & Algina J, Introduction to classical and modern test theory, Harcourt Brace Jovanovich College Publishers, Fort Worth, 1986).

Year: Year administered.
N: Number of valid responses (test-takers who completed the form correctly).
Difficulty Index: Percent of correct answers (best if between 40 and 80). Low values indicate a question that is too difficult or an error with the answer key. This item set has 9 items with a difficulty index between 40 and 80.
Point Biserial: Measure of how well questions discriminate between students (best if greater than 0.1). Negative numbers indicate a question that is flawed in some way, e.g., the better examinees are getting it wrong.
Answer Frequency: Percentage of responses for answer choice (best if greater than zero).

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IRAT Possible Points | Mean Score (Range) | Mean Percentage
2006 5 | 3.9 (1-5) | 79%
2007 5 | 4.0 (2-5) | 81%
2008 5 | 3.8 (1-5) | 76%

GRAT Possible Points | Mean Score (Range) | Mean Percentage
2006 15 | 15 | 100%
2007 15 | 15 | 100%
2008 15 | 14.4 (13-15) | 96%
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Readiness Assurance Test Items

IRAT Instructions Provided to Learners:
Please circle your answer for each of the following questions. Then bubble in your response on your answer sheet. For each question, there is one BEST answer.

1. Opsonization is the term used to describe the major activity of which one of the following complement fragments?
   (A) C2b
   (B) C5a
   (C) C1q
   (D) C3b

Correct Answer: (D) C3b binds to pathogen surfaces and promotes phagocytosis (opsonization).
Incorrect Answers: (A) C2b is a precursor to kinin, which results in edema (C2a combines with C4b to form the C3 convertase). (B) C5a is a peptide mediator of inflammation. (C) C1q binds antigen-antibody complexes and pathogen surfaces.

2. Which one of the following characteristics does NOT apply to innate immunity?
   (A) It is ready to react to microbial challenge immediately.
   (B) It only protects the mucosal surfaces.
   (C) It consists of both immediate and induced response mechanisms.
   (D) It is quantitatively and qualitatively the same after a second infection by the same pathogen.

Correct Answer: (B) Innate immunity includes the physical barrier of the skin and effector mechanisms encountered by microbes that get past the skin and mucosal surfaces.
Incorrect Answers: (A, C, D) True statements.

3. The Fab component of antibodies generated by papain digestion contains which one of the following?
   (A) Lambda light chain only
   (B) Gamma heavy chain only
   (C) An antigen-binding site
   (D) The constant region

Correct Answer: (C) Most accurate description.
Incorrect Answers: (A) Fab fragments include a portion of both heavy and light chains. (B) Fab fragments include light chains. (D) Fab fragments include the variable region and a portion of the constant regions (i.e., CH1 domain).
4. The injection of a rabbit with a mouse antibody to human influenza hemagglutinin results in the production of which one of the following responses?
   (A) A polyclonal rabbit anti-mouse Ig response.
   (B) Passive immunity for the rabbit against influenza.
   (C) A monoclonal rabbit anti-human influenza response.
   (D) A polyclonal rabbit anti-human influenza response.

Correct Answer: (A) Best answer.
Incorrect Answers: (B) Mouse antibody would be recognized by the rabbit as foreign and cleared.
(C, D) There is no influenza antigen present to generate a rabbit anti-influenza antibody response.

5. Suppose the following four peptides are eluted from a single MHC allele. Which of the following statements about these results is FALSE?

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(A) These peptides most likely eluted from an MHC class I molecule.
(B) Amino acid residues 1 and 6 appear to be anchor residues.
(C) The MHC molecule that carried these peptides presents these peptides to CD8+ T cells.
(D) The peptides carried by this MHC molecule were of extracellular or intravesicular origin.

Correct Answer: (D) The peptides carried by this MHC molecule were of cytoplasmic origin.
Incorrect Answers: (A, B, C) True statements.

6. The sequence of reaction of the classical complement pathway components is:
   (A) C132456789.
   (B) C142356789.
   (C) C143256789.
   (D) C8675309.

Correct Answer: (B) C1 complex binds to antigen/antibody complex or pathogen surface → cleaves C4 and then C2 to generate C3 convertase → which cleaves C3 → C3b binds to C3 convertase to form the C5 convertase → C5b initiates formation of the membrane attack complex, which includes C6, C7, C8 and C9.
Incorrect Answers: (A, C, D) Incorrect sequences.
7. Which one of the following is NOT considered to be an immediate innate immune agent involved in saliva-mediated defense of the oral cavity?
   (A) mucins
   (B) peroxidase
   (C) defensins
   (D) IgA antibody

Correct Answer: (D) Antibody is part of specific or adaptive immunity, not innate immunity.
Incorrect Answers: (A, B, C) All are components of innate immunity.

8. Which one of the following is a characteristic of a monoclonal antibody produced from the hybridization of a myeloma cell and a secreting B cell?
   (A) not directed towards a specific antigen
   (B) the product of multiple cells in the original spleen tissue
   (C) functional as both the membrane BCR and the secreted product of the hybridoma
   (D) always of the IgG isotype

Correct Answer: (C) True statement.
Incorrect Answers: (A) Directed toward a specific antigen. (B) Usually the product of a single splenic B lymphocyte fusing with a myeloma cell. (D) Monoclonal antibodies may be of any isotype.

9. Which one of the following is a characteristic of the antibody Fc portion?
   (A) binds to receptors on phagocytic cells and aids in opsonization
   (B) responsible for specific antigen recognition properties of Ab
   (C) extremely variable and undergoes somatic hypermutation
   (D) same amino acid sequence for all isotypes

Correct Answer: (A) True statement.
Incorrect Answers: (B) Fab fragments include the antigen-binding sites. (C) Features of the variable region, not included in the Fc portion. (D) Amino acid sequences vary for different isotypes.
10. Suppose the following four peptides are eluted from a single MHC allele. Which of the following statements about these results is TRUE?

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(A) These peptides most likely eluted from an MHC class II molecule.
(B) Amino acid residues 1 and 6 appear to be anchor residues.
(C) The MHC molecule that carried these peptides presents these peptides to CD4+ T cells.
(D) The structure of the MHC molecule resembles Rocky the flying squirrel.

Correct Answer: (B) True statement.
Incorrect Answers: (A) These peptides most likely eluted from an MHC class I molecule. (C) MHC class I molecules present peptides to CD8+ T cells. (D) The structure of the MHC molecule resembles Bullwinkle the Moose.

11. Opsonization is the term used to describe the major activity of which complement fragment?
   (A) C1q
   (B) C2b
   (C) C3b
   (D) C5a

Correct Answer: (C) C3b binds to pathogen surfaces and promotes phagocytosis (opsonization).
Incorrect Answers: (A) C1q binds antigen-antibody complexes and pathogen surfaces. (B) C2b is a precursor to kinin, which results in edema (C2a combines with C4b to form the C3 convertase). (D) C5a is a peptide mediator of inflammation.
(Note: This is a rearranged version of question 1)

12. The major activities of C5a include all of the following EXCEPT:
   (A) chemotaxis.
   (B) erythrocyte clearance of immune complexes.
   (C) increased vascular permeability.
   (D) mast cell degranulation.

Correct Answer: (B) Clearance of immune complexes is mediated by C3b.
Incorrect Answers: (A, C, D) All are major activities of C5a.
Readiness Assurance Test Items (continued)

13. Lysozyme is found in saliva, tears and some of the granules in neutrophils. It is important in host defense due to its antibacterial activity. This is a result of:
   (A) opsonic activity.
   (B) neutralization of toxins.
   (C) disruption of the gram negative bacterial membrane.
   (D) digestion of the bacterial cell wall.

Correct Answer: (D) Lysozyme is an enzyme that functions by attacking peptidoglycans found in the cells walls of bacteria, especially Gram-positive bacteria.
Incorrect Answers: (A-C) These are not activities of lysozyme.

14. By using pattern recognition receptors on their surface, macrophages in the tissues can:
   (A) detect self proteins.
   (B) detect most bacteria.
   (C) detect a virally infected host cell.
   (D) differentiate between two gram-positive bacterial species.

Correct Answer: (B) Membrane-bound pattern recognition receptors recognize molecules identify molecules associated with microbial pathogens, e.g., bacterial carbohydrates [LPS, mannose], bacterial or viral DNA or RNA, peptidoglycans and lipotechoic acids from Gram positive bacteria.
Incorrect Answers: (A, C, D) Not functions of pattern recognition receptors.

15. Suppose the following four peptides are eluted from a single MHC allele. Which one of the following statements about these results is FALSE?

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   (A) The structure of the MHC molecule resembles Bullwinkle the moose.
   (B) These peptides most likely eluted from an MHC class II molecule.
   (C) The MHC molecule that carried these peptides presents these peptides to CD8\(^+\) T cells.
   (D) Amino acid residues 7, 10 and 14 appear to be anchor residues.

Correct Answer: (C) MHC class II molecules present peptides to CD4\(^+\) T cells
Incorrect Answers: (A, B, D) True statements.
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Group Activity Exercises

Two group application exercises are provided, however only one problem set should be used if limited to a single 50-minute TBL session. Each exercise requires learners to understand and evaluate an experimental design, interpret data, predict experimental results, and/or design an experiment to test a hypothesis. Answers to these questions can not be found in the assigned textbook, and can only be answered by a team discussing, debating, and reaching a consensus on a single best answer. Each problem has multiple parts, including 2-3 multiple choice questions that may be answered on an IF-AT card for immediate feedback, plus 4 discussion questions.

Sample Instructions to Learners:

For numbered multiple-choice questions only, please use the IF-AT card for feedback, after your team has discussed the question and arrived at a group consensus. This activity is “open book”, but you should attempt to answer the question first based on what you and your team members know. Simply looking up the answer is often not possible, and may waste time. Keep in mind that part of your grade depends on the success of your team.

Be sure to discuss the pros and cons of each answer, and be prepared to justify your team answers in the class discussion.

Photo Credits

Multiple myeloma photomicrograph (Problem 2)
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 http://commons.wikimedia.org/wiki/Image:Multiple_myeloma_(1)_MG_stain.jpg

Skull X-ray (Problem 2)
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http://orthoinfo.aaos.org/topic.cfm?topic=A00086

Serum Protein Electrophoresis and Immunofixation Test images (Problem 2)
Used with permission by Dr. Edmond S. K. Ma, Dept. of Pathology, University of Hong Kong
Group Activity Exercise - Problem 1

For numbered multiple-choice questions, please use the scratch-off card for feedback, after your team has discussed the question and arrived at a group consensus. This activity is “open book”, but you should attempt to answer the question first based on what you and your team members know. Simply looking up the answer is often not possible, and may waste time. Keep in mind that part of your grade depends on the success of your team.

You have isolated a glycoprotein from the liver of a Florida alligator, which you call FALP (Florida alligator liver protein). You have purified a sample of FALP to homogeneity, and have a total yield of 650 μg. You want to prepare antibodies to FALP so that you can conduct further studies on its distribution and function in the alligator liver.

1. What of the following is the best protocol to use in order to stimulate a rabbit to produce or develop antibodies to FALP?
   (A) Isolate rabbit spleen cells and use in vitro immunization to generate high affinity hybridomas.
   (B) Use a tryptic digest of FALP to immunize the rabbit with FALP peptides.
   (C) Immunize the rabbit with 100 μg of FALP with an adjuvant at intervals of ~2 weeks over a period of ~4-6 weeks.
   (D) Conjugate FALP to a carrier such as bovine serum albumin (BSA) and immunize the rabbit two times, two weeks apart.

Be sure to discuss the pros and cons of each answer. Why is this the best answer?

How would you determine whether the rabbit had produced any antibodies against FALP? When would you make this determination? How would you determine if the rabbit had produced IgG antibodies, IgM antibodies, or both, against FALP?

2. Which of the following is the best description for the kind of anti-FALP antiserum you have isolated from the rabbit?
   (A) homologous, monoclonal
   (B) heterologous, monoclonal
   (C) homologous, polyclonal
   (D) heterologous, polyclonal.
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Group Activity Exercise - Problem 1 (continued)

Discussion Questions

A. Would your anti-FALP antibodies be considered monoclonal if they had been produced in and isolated directly from a mouse instead of a rabbit? Briefly explain.

B. Does the binding of these anti-FALP antibodies to FALP involve covalent bonding between the antibody combining site(s) and FALP? Briefly explain why or why not.

C. How could you use your anti-FALP antibodies to discover the location of FALP in the alligator liver?

D. What experiment could you do in order to determine whether the rabbit anti-FALP antibody you have generated is able to fix complement?
Group Activity Exercise - Problem 1

ANSWER KEY

1. C. Immunizing a rabbit with 100 μg of FALP with an adjuvant at intervals of ~2 weeks over a period of ~4-6 weeks is the most likely approach to producing a good antibody reagent for FALP. In vitro immunization is not likely to produce high affinity antibody (no germinal center reaction). Antipeptide antibodies may not recognize native FALP. Conjugation to a carrier protein is probably not necessary, unless FALP is shown not to be very immunogenic by itself.

How would you determine whether the rabbit had produced any antibodies against FALP? Learners should be able to discuss possible assays for Ab production within their team, e.g., ELISA, radioimmunoassay, immunofluorescence.

When would you make this determination? About two weeks after primary immunization, or one week after secondary immunization (learners should be familiar with kinetics of Ab response).

How would you determine if the rabbit had produced IgG antibodies, IgM antibodies, or both, against FALP? Use isotype-specific Ab reagents to check the isotype of anti-FALP Ab, e.g., goat anti-rabbit IgM, goat anti-rabbit IgG, etc.

2. D. The anti-FALP antiserum would be considered heterologous because it is produced in a different species (rabbit vs. alligator), and polyclonal because it is from serum and represents the Ab response from many rabbit B cells.

This problem was used in 2006 and 2007, with 6 possible points. In 2006 five of six teams had perfect scores and one team had a score of 5 (the average team score was 5.8, or 97%). In 2007 all five teams had perfect scores.

Discussion Questions

A. Would your anti-FALP antibodies be considered monoclonal if they had been produced in and isolated directly from a mouse instead of a rabbit? Not if mouse antiserum was used. Hybridomas must be generated to produce a monoclonal antibody.

B. Does the binding of these anti-FALP antibodies to FALP involve covalent bonding between the antibody combining site(s) and FALP? No. Antigen-antibody interactions are all noncovalent, e.g., electrostatic forces, hydrogen bonds, Van der Waals forces, and hydrophobic forces.

C. How could you use your anti-FALP antibodies to discover the location of FALP in the alligator liver? Immunohistochemical staining of tissue sections, immunofluorescent staining

D. What experiment could you do in order to determine whether the rabbit anti-FALP antibody you have generated is able to fix complement? Determining the Ab isotype will also give you an indication of whether or not it fixes complement. More direct assays would be complement-mediated cell lysis of alligator liver cells or FALP-conjugated sheep red blood cells.

Open-ended questions in this problem were found to be straightforward and teams were able to arrive at complete answers via inter-team discussion. One concept that students sometimes have difficulty with is the use of anti-immunoglobulin reagents made in different species, that is, antibodies recognizing other antibodies, e.g. goat anti-rabbit IgM and goat anti-rabbit IgG.
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

**Group Activity Exercise - Problem 2**

For numbered multiple-choice questions, please use the scratch-off card for feedback, after your team has discussed the question and arrived at a group consensus. This activity is “open book”, but you should attempt to answer the question first based on what you and your team members know. Simply looking up the answer is often not possible, and may waste time. Keep in mind that part of your grade depends on the success of your team.

Be sure to discuss the pros and cons of each answer, and be prepared to justify your team answers.

Mrs. Newberry, 55 years old, has previously been a very healthy person, but is now easily fatigued and suffers from back pain. Her visit to the emergency room is prompted by an upper respiratory infection, which is found to be caused by *Streptococcus pyogenes* and is successfully treated with antibiotics. Blood tests reveal anemia (low red blood cell counts) and neutropenia (low white blood cell count). A blood smear indicates an unusually high numbers of cells that resemble lymphocytes. Her physician diagnoses her with multiple myeloma.

Multiple myeloma results from the malignant transformation of a plasma cell. Clinically this is in part a bone disease, because the plasma cell tumors arise at multiple sites in the bone marrow, where they expand and cause localized bone erosion.

Clinical symptoms include fatigue, anemia, elevated serum immunoglobulin levels, and pain due to bone damage, and complications include increased susceptibility to pyogenic infections.

![Smear preparation of multiple myeloma bone marrow aspirate (May-Grünwald-Giemsa stain).](http://commons.wikimedia.org/wiki/Image:Multiple_myeloma_(1)_MG_stain.jpg)

1. Which of the lanes in the serum protein electrophoresis (SPE) gel shown to the right might be Mrs. Newberry’s serum sample?
   - (A) lane 4
   - (B) lane 3 or 6
   - (C) lane 3 or 7
   - (D) lane 8

2. In this diagnostic assay (immunofixation test), similar to a Western blot, identical patient SPE lanes (#2-8) were probed with antisera specific for immunoglobulin \( \gamma \), \( \alpha \), \( \mu \), \( \kappa \), \( \delta \), \( \varepsilon \), and \( \lambda \) chains, respectively. What is the isotype of Mrs. Newberry’s myeloma protein?
   - (A) IgD\( \lambda \)
   - (B) IgG\( \lambda \)
   - (C) IgD\( \kappa \)
   - (D) IgM\( \kappa \)
Group Activity Exercise - Problem 2 (continued)

3. Which characteristic best describes the heavy chain isotype of Mrs. Newberry’s myeloma protein?
   (A) high concentration in serum and good at complement fixation
   (B) best at complement fixation and present as a pentamer in serum
   (C) expressed on mature B cells but present in only trace quantities in the serum
   (D) sensitizes mast cells and present in only trace quantities in serum

Discussion Questions

A. Multiple myeloma patients often become anemic (low red blood cell count) and neutropenic (low white blood cell count). What causes this?

B. Why do multiple myeloma patients become progressively more susceptible to pyogenic infections, despite high serum immunoglobulin levels? Your team should be able to come up with three reasons.

C. It is usually impossible to identify the antigen specificity for a myeloma protein antibody, but if you could, would you predict that it would have a relatively low or high affinity for its antigen? Why?
   (A) high affinity
   (B) low affinity
   (C) not enough information to tell

D. What would you predict about the affinity of Mrs. Newberry’s myeloma protein antibody?
   (A) high affinity
   (B) low affinity
   (C) not enough information to tell
Immunology Team-Based Learning: Innate Immunity & Recognition of Antigen

Group Activity Exercise - Problem 2

This problem was adapted from the multiple myeloma case study in “Case Studies in Immunology: A Clinical Companion”, R. Geha & F. Rosen, 5th edition, Garland Science, 2008. Learners must be familiar with the characteristics of immunoglobulin expression by myeloma cells, interpretation of serum protein electrophoresis, antibody isotypes and effector functions, and B cell maturation and differentiation (e.g., class switching, affinity maturation).

ANSWER KEY

1. C. Lanes 3 and 7 both show a monoclonal spike in the gamma globulin fraction.

2. A. Positive reactions are seen in the lanes for IgD and \( \lambda \) light chains.

3. C. IgD is expressed on mature B cells but present in only trace quantities in the serum. Answer A, high concentration in serum and good at complement fixation, describes IgG. Answer B, best at complement fixation and present as a pentamer in serum, describes IgM. Answer D, sensitizes mast cells and present in only trace quantities in serum, describes IgE.

This problem was used in 2008 and was scored with 9 possible points. All five teams scored 100%.

Discussion Questions

A. The proliferation of malignant plasma cells in the bone marrow crowds out the RBC and WBC precursors, limiting space in the bone marrow for RBC and neutrophil production.

B. Although serum Ig is elevated, most of the serum Ig is monoclonal. Without the normal polyclonal serum Ig, patients are essentially agammaglobulinemic. Normal lymphocyte production is depressed as in question A. With a low neutrophil count, patients cannot clear bacteria from the bloodstream and lungs effectively.

C. Any answer is correct, if the team can justify their answer.
   (A) high affinity - maybe, if the cell has undergone class switch (IgG, IgA, IgE)
   (B) low affinity - maybe, especially if an IgM or IgD
   (C) not enough information to tell - if you had no info about isotype

D. Based on the reasoning above in question C, one might predict Mrs. Newberry’s myeloma protein antibody to be of low affinity (answer B) based on the IgD isotype.

Open-ended questions in this problem were found to be straightforward and teams were able to arrive at complete answers via inter-team discussion. For question C, at least one team usually picks each answer choice, as revealed using the colored letter cards, prompting good inter-team discussion.