

Question Bank Immunohistochemistry 2024

1. True or False: Immunohistochemistry is a technique used to visualize the distribution of specific proteins or antigens in tissues using antibodies.
 2. True or False: Antibodies used in immunohistochemistry are specific to only one antigen and do not cross-react with others.
 3. True or False: The primary antibody in immunohistochemistry binds directly to the antigen of interest in the tissue sample.
 4. True or False: Immunohistochemistry can only be performed on fixed tissue sections, not on fresh tissue.
 5. True or False: Immunohistochemistry is commonly used in research and diagnostic pathology to identify tumor markers and assess disease states.
 6. True or False: The secondary antibody in immunohistochemistry amplifies the signal by binding to the primary antibody.
 7. True or False: Immunohistochemistry can be used to differentiate between different types of cells based on their protein expression patterns.
 8. True or False: Immunohistochemistry requires the use of a chromogen to visualize the antibody-antigen complexes under a microscope.
 9. True or False: Immunohistochemistry can be performed manually or using automated staining platforms.
 10. True or False: Immunohistochemistry is not suitable for studying the expression of intracellular proteins within cells.
1. True 2. True 3. True 4. True 5. True 6. True 7. True 8. True 9. True 10. False

20 multiple-choice questions based on the information provided about Immunohistochemistry:

1. Immunohistochemistry is a technique used to visualize the distribution of specific proteins or antigens in tissues using: a. Enzymes b. Antibodies c. Fluorescent dyes d. Fixatives
2. Which component of the immunohistochemical staining process binds directly to the antigen of interest in the tissue sample? a. Primary antibody b. Secondary antibody c. Chromogen d. Counterstain
3. In immunohistochemistry, what role does the secondary antibody play? a. It directly binds to the antigen. b. It amplifies the signal by binding to the primary antibody. c. It catalyzes a color change in the tissue. d. It stabilizes the tissue section.
4. Immunohistochemistry can be used for: a. Identifying cellular organelles. b. Visualizing DNA sequences. c. Detecting specific proteins or antigens. d. Assessing tissue hydration levels.
5. Which of the following is NOT a common application of immunohistochemistry? a. Identifying tumor markers. b. Assessing disease states. c. Analyzing bone structure. d. Differentiating between cell types.
6. Immunohistochemistry is commonly used in: a. Microbiology laboratories. b. Pathology laboratories. c. Chemistry laboratories. d. Physics laboratories.
7. The chromogen used in immunohistochemistry is responsible for: a. Binding to the primary antibody. b. Catalyzing a color change in the tissue. c. Enhancing tissue fixation. d. Preventing background staining.
8. Immunohistochemistry can be performed: a. Only on fresh tissue. b. Only on fixed tissue sections. c. Both on fresh and fixed tissue. d. Only on cell cultures.
9. Which of the following is NOT a method of visualizing immunohistochemical staining? a. Bright-field microscopy b. Fluorescence microscopy c. Electron microscopy d. Confocal microscopy
10. Immunohistochemistry is particularly useful for: a. Assessing tissue hydration levels. b. Visualizing cellular structure. c. Identifying protein expression patterns. d. Detecting DNA mutations.
11. The primary antibody used in immunohistochemistry is: a. Specific to only one antigen. b. Non-specific and binds to various antigens. c. Labeled with a fluorescent dye. d. Not required for the staining process.

12. Which of the following is a common type of chromogen used in immunohistochemistry? a. Hematoxylin b. DAB (3,3'-diaminobenzidine) c. Eosin d. Alizarin Red
13. Immunohistochemistry can aid in: a. Differentiating between cells based on their size. b. Identifying the presence of infectious agents. c. Assessing the activity of enzymes. d. Identifying cell surface markers.
14. Immunohistochemistry can be automated using: a. Only manual techniques. b. Robotic staining platforms. c. Visual inspection. d. Light microscopy.
15. Which of the following statements about immunohistochemistry is FALSE? a. It is commonly used in diagnostic pathology. b. It can be used to study the expression of intracellular proteins. c. It relies on the specificity of antibody-antigen interactions. d. It requires the use of a secondary antibody.
16. Immunohistochemistry is a valuable tool for: a. Identifying cellular structures. b. Diagnosing bacterial infections. c. Understanding cellular metabolism. d. Detecting hormone levels.
17. Which of the following techniques is NOT commonly used in conjunction with immunohistochemistry? a. Western blotting b. Flow cytometry c. Polymerase chain reaction (PCR) d. Enzyme-linked immunosorbent assay (ELISA)
18. Immunohistochemistry can aid in cancer diagnosis by: a. Detecting specific tumor markers. b. Assessing tissue hydration levels. c. Analyzing DNA sequences. d. Differentiating between cell types.
19. Which microscope is typically used to observe immunohistochemically stained tissue sections? a. Electron microscope b. Fluorescence microscope c. Scanning electron microscope d. Atomic force microscope
20. Immunohistochemistry is particularly useful in: a. Visualizing the presence of lipids. b. Studying the expression of specific proteins. c. Identifying bone structure. d. Assessing tissue vascularity.

Answers:

1. b. Antibodies
2. a. Primary antibody
3. b. It amplifies the signal by binding to the primary antibody.
4. c. Detecting specific proteins or antigens.

5. c. Analyzing bone structure.
6. b. Pathology laboratories.
7. b. Catalyzing a color change in the tissue.
8. c. Both on fresh and fixed tissue.
9. c. Electron microscopy
10. c. Identifying protein expression patterns.
11. a. Specific to only one antigen.
12. b. DAB (3,3'-diaminobenzidine)
13. d. Identifying cell surface markers.
14. b. Robotic staining platforms.
15. b. It can be used to study the expression of intracellular proteins.
16. a. Identifying cellular structures.
17. c. Polymerase chain reaction (PCR)
18. a. Detecting specific tumor markers.
19. b. Fluorescence microscope
20. b. Studying the expression of specific proteins.

10 short answer questions about Immunohistochemistry:

1. What is the primary purpose of immunohistochemistry?
2. Briefly explain the role of antibodies in immunohistochemistry.
3. Describe the main difference between the primary and secondary antibodies used in immunohistochemistry.
4. Provide two common applications of immunohistochemistry.
5. Can immunohistochemistry be performed on both fresh and fixed tissue? Explain briefly.
6. What is the function of a chromogen in immunohistochemistry?

7. Name one type of microscopy commonly used to visualize immunohistochemically stained tissue sections.
8. Explain the potential use of immunohistochemistry in cancer diagnosis.
9. How can immunohistochemistry be automated?
10. Describe one limitation of immunohistochemistry as a technique.

Answers:

1. The primary purpose of immunohistochemistry is to visualize the distribution of specific proteins or antigens in tissues using antibodies.
2. Antibodies in immunohistochemistry specifically bind to antigens of interest within tissue samples, allowing for their visualization.
3. Primary antibodies directly bind to the antigen of interest, while secondary antibodies bind to the primary antibodies, amplifying the signal.
4. Common applications of immunohistochemistry include identifying tumor markers and assessing disease states.
5. Yes, immunohistochemistry can be performed on both fresh and fixed tissue, although fixed tissue sections are more commonly used.
6. A chromogen in immunohistochemistry reacts with an enzyme-linked secondary antibody to produce a visible color change, indicating the presence of the antigen.
7. Fluorescence microscopy is commonly used to visualize immunohistochemically stained tissue sections.
8. Immunohistochemistry can aid in cancer diagnosis by detecting specific tumor markers, helping pathologists identify and classify tumors.
9. Immunohistochemistry can be automated using robotic staining platforms, which streamline the staining process and increase throughput.
10. One limitation of immunohistochemistry is its potential for nonspecific binding, which can lead to background staining and affect the interpretation of results.

10 scenario-based questions related to Immunohistochemistry:

1. Scenario: A pathology laboratory receives a tissue sample suspected to be malignant. How can immunohistochemistry aid in confirming the diagnosis?

2. Scenario: A researcher is studying the expression of a specific protein in neuronal tissue. Describe how immunohistochemistry can help visualize the distribution of this protein within the tissue.
3. Scenario: A new technician in the lab is performing an immunohistochemistry experiment but notices unexpected background staining. What steps can they take to troubleshoot this issue?
4. Scenario: A pathologist is examining a tissue section stained using immunohistochemistry and notices strong staining in the cytoplasm of certain cells. What might this staining pattern indicate?
5. Scenario: A clinician is interested in using immunohistochemistry to identify the presence of a particular tumor marker in a patient's biopsy sample. How can the results of this test influence treatment decisions?
6. Scenario: A pathology lab is transitioning from manual immunohistochemistry staining techniques to an automated staining platform. What are the potential benefits of this transition?
7. Scenario: A researcher is studying a rare disease characterized by abnormal protein aggregation in tissue. How can immunohistochemistry be used to visualize these protein aggregates?
8. Scenario: A pathologist is examining a tissue section stained using immunohistochemistry and notices weak or absent staining in certain areas. What factors could contribute to this observation?
9. Scenario: A pharmaceutical company is developing a new drug targeting a specific protein implicated in cancer progression. How might immunohistochemistry be used in preclinical studies to evaluate the drug's efficacy?
10. Scenario: A clinician is reviewing immunohistochemistry results for a patient's biopsy sample and notices heterogeneity in protein expression within the tumor. What implications might this heterogeneity have for treatment planning?

The answers to the scenario-based questions related to Immunohistochemistry:

1. **Scenario: A pathology laboratory receives a tissue sample suspected to be malignant. How can immunohistochemistry aid in confirming the diagnosis?**

- **Answer:** Immunohistochemistry can aid in confirming the diagnosis by using specific antibodies to detect tumor markers or antigens that are characteristic of malignant cells. This helps in identifying the type and origin of the tumor, thereby confirming its malignancy.
2. **Scenario: A researcher is studying the expression of a specific protein in neuronal tissue. Describe how immunohistochemistry can help visualize the distribution of this protein within the tissue.**
- **Answer:** Immunohistochemistry can help visualize the distribution of the protein by using a primary antibody that specifically binds to the protein of interest, followed by a secondary antibody conjugated to a chromogen or fluorophore. This staining allows the researcher to see where the protein is localized within the neuronal tissue under a microscope.
3. **Scenario: A new technician in the lab is performing an immunohistochemistry experiment but notices unexpected background staining. What steps can they take to troubleshoot this issue?**
- **Answer:** The technician can take several steps to troubleshoot background staining:
 - Ensure proper blocking of non-specific binding sites using a blocking agent.
 - Use a more diluted concentration of the primary and secondary antibodies.
 - Increase the washing steps to remove unbound antibodies.
 - Verify the specificity of the antibodies being used.
 - Check for endogenous enzyme activity that might cause background staining and use appropriate inhibitors.
4. **Scenario: A pathologist is examining a tissue section stained using immunohistochemistry and notices strong staining in the cytoplasm of certain cells. What might this staining pattern indicate?**

- **Answer:** Strong cytoplasmic staining may indicate the presence of the target antigen within the cytoplasm of those cells. This pattern can suggest that the protein of interest is localized in the cytoplasm, which might be characteristic of certain cellular processes or pathologies.

5. Scenario: A clinician is interested in using immunohistochemistry to identify the presence of a particular tumor marker in a patient's biopsy sample. How can the results of this test influence treatment decisions?

- **Answer:** The presence of a specific tumor marker identified by immunohistochemistry can provide valuable information about the type and aggressiveness of the tumor. This information can help in selecting targeted therapies, predicting prognosis, and determining the most appropriate treatment plan for the patient.

6. Scenario: A pathology lab is transitioning from manual immunohistochemistry staining techniques to an automated staining platform. What are the potential benefits of this transition?

- a. **Answer:** The transition to an automated staining platform can offer several benefits, including:
 - i. Increased consistency and reproducibility of results.
 - ii. Reduced hands-on time for technicians, allowing them to focus on other tasks.
 - iii. Higher throughput, enabling the processing of more samples in less time.
 - iv. Improved standardization of staining protocols, reducing variability.
 - v.

7. Scenario: A researcher is studying a rare disease characterized by abnormal protein aggregation in tissue. How can immunohistochemistry be used to visualize these protein aggregates?

- a. **Answer:** Immunohistochemistry can be used to visualize protein aggregates by employing antibodies that specifically bind to the aggregated proteins. This staining will highlight the presence and distribution of these aggregates within the tissue sections, aiding in the study of the disease's pathology.

8. Scenario: A pathologist is examining a tissue section stained using immunohistochemistry and notices weak or absent staining in certain areas. What factors could contribute to this observation?

- a. **Answer:** Factors that could contribute to weak or absent staining include:
- i. Inadequate fixation or tissue preparation, leading to loss of antigenicity.
 - ii. Insufficient antibody concentration or improper incubation times.
 - iii. Poor antigen retrieval, resulting in inaccessible epitopes.
 - iv. Degradation of the primary or secondary antibodies.
 - v. Technical issues such as uneven sectioning or improper mounting.

9. Scenario: A pharmaceutical company is developing a new drug targeting a specific protein implicated in cancer progression. How might immunohistochemistry be used in preclinical studies to evaluate the drug's efficacy?

- a. **Answer:** Immunohistochemistry can be used in preclinical studies to evaluate the drug's efficacy by staining tissue sections from treated and untreated experimental models. By comparing the expression levels and localization of the target protein between the two groups, researchers can assess the impact of the drug on the protein's expression and distribution.

10. Scenario: A clinician is reviewing immunohistochemistry results for a patient's biopsy sample and notices heterogeneity in protein expression within the tumor. What implications might this heterogeneity have for treatment planning?

- **Answer:** Heterogeneity in protein expression within the tumor can have significant implications for treatment planning. It may indicate varying degrees of differentiation and aggressiveness within the tumor, suggesting that a combination of therapies targeting different subpopulations of tumor cells might be necessary. It also highlights the potential for resistance to single-agent therapies and the need for personalized treatment strategies.

10 long-answer questions about immunohistochemistry:

- 1. Explain the principle of immunohistochemistry (IHC) and describe the steps involved in a standard IHC protocol.**
 - Include details on tissue preparation, antigen retrieval, blocking, primary and secondary antibody application, detection methods, and counterstaining.
- 2. Discuss the importance of antigen retrieval in immunohistochemistry. What are the common methods used for antigen retrieval, and how do they enhance antibody binding?**
 - Explain heat-induced epitope retrieval (HIER) and enzymatic retrieval, and discuss the pros and cons of each method.
- 3. Describe the different types of detection systems used in immunohistochemistry, such as chromogenic and fluorescent detection. Compare their advantages and disadvantages in terms of sensitivity, specificity, and visualization.**
 - Include examples of commonly used chromogens and fluorophores.
- 4. What are the common challenges and sources of error in immunohistochemistry? Provide strategies for troubleshooting issues like non-specific staining, weak signal, and high background staining.**
 - Discuss the importance of controls, optimization of antibody concentration, and proper tissue handling.
- 5. How can immunohistochemistry be used to differentiate between malignant and benign tumors? Provide examples of specific markers that are commonly used in this context.**
 - Include a discussion on markers such as Ki-67, p53, HER2/neu, and cytokeratins.
- 6. Explain the role of immunohistochemistry in personalized medicine, particularly in the context of cancer treatment. How do IHC results influence therapeutic decisions and patient management?**
 - Discuss the use of IHC in identifying targets for targeted therapy, such as HER2 in breast cancer and PD-L1 in immunotherapy.
- 7. Describe the process and significance of using multiplex immunohistochemistry. How does this technique enhance the understanding of complex tissue environments and interactions?**
 - Explain the technical aspects of multiplexing, potential applications, and challenges in interpretation.
- 8. Discuss the use of immunohistochemistry in the study of infectious diseases. How can IHC help identify pathogens and understand host-pathogen interactions?**
 - Provide examples of pathogens that can be detected using IHC, such as viruses, bacteria, and fungi.
- 9. Explain the role of immunohistochemistry in neuroscience research. How is IHC used to study neural tissue, and what are some common markers for different cell types in the nervous system?**

- Include markers such as GFAP for astrocytes, NeuN for neurons, and Iba1 for microglia.
10. **Compare and contrast immunohistochemistry with other protein detection techniques such as Western blotting and ELISA. What are the unique advantages and limitations of IHC?**
- Discuss aspects such as spatial resolution, quantitative vs. qualitative data, and applications in clinical diagnostics vs. research settings.

The answers to the 10 long-answer questions about immunohistochemistry:

1. **Explain the principle of immunohistochemistry (IHC) and describe the steps involved in a standard IHC protocol.**
 - **Principle:** Immunohistochemistry (IHC) is a technique used to detect specific antigens (proteins) in tissue sections by utilizing the binding properties of antibodies. The bound antibodies are then visualized using chromogenic or fluorescent detection methods.
 - **Steps in a standard IHC protocol:**
 1. **Tissue Preparation:** Fixation of tissue in formalin and embedding in paraffin.
 2. **Sectioning:** Cutting thin sections (3-5 microns) of the paraffin-embedded tissue using a microtome.
 3. **Deparaffinization and Rehydration:** Removing paraffin with xylene and rehydrating sections through a series of decreasing ethanol concentrations.
 4. **Antigen Retrieval:** Heat-induced epitope retrieval (HIER) or enzymatic retrieval to unmask antigens.
 5. **Blocking:** Applying a blocking solution to prevent non-specific binding.
 6. **Primary Antibody Incubation:** Incubating sections with a primary antibody specific to the target antigen.
 7. **Secondary Antibody Incubation:** Applying a secondary antibody conjugated to an enzyme or fluorophore that binds to the primary antibody.
 8. **Detection:** Visualizing the antigen-antibody complex using a chromogenic substrate (e.g., DAB) or fluorescent dye.
 9. **Counterstaining:** Staining the tissue with a counterstain (e.g., hematoxylin) to visualize cell structures.
 10. **Mounting:** Covering the stained tissue with a coverslip for microscopic examination.
2. **Discuss the importance of antigen retrieval in immunohistochemistry. What are the common methods used for antigen retrieval, and how do they enhance antibody binding?**
 - **Importance:** Antigen retrieval is crucial for exposing epitopes that may be masked during formalin fixation and paraffin embedding. This step

ensures that antibodies can access and bind to their target antigens effectively.

- **Common Methods:**
 - **Heat-Induced Epitope Retrieval (HIER):** Involves heating tissue sections in a buffer solution (e.g., citrate or EDTA) using a microwave, pressure cooker, or water bath. Heat treatment breaks formalin-induced cross-links, exposing epitopes.
 - **Enzymatic Retrieval:** Utilizes proteolytic enzymes (e.g., proteinase K, trypsin) to digest proteins and unmask epitopes. This method is gentler and is used for more sensitive tissues.
 - **Enhancement of Antibody Binding:** Both methods break the formaldehyde-induced cross-links and expose hidden epitopes, allowing antibodies to bind more effectively, thereby increasing the sensitivity and specificity of the staining.
3. **Describe the different types of detection systems used in immunohistochemistry, such as chromogenic and fluorescent detection. Compare their advantages and disadvantages in terms of sensitivity, specificity, and visualization.**
- **Chromogenic Detection:**
 - **Method:** Enzyme-labeled secondary antibodies (e.g., HRP or alkaline phosphatase) convert a chromogenic substrate (e.g., DAB or BCIP/NBT) into a colored precipitate visible under a light microscope.
 - **Advantages:**
 - Easy to use and interpret.
 - Permanent staining suitable for archiving.
 - **Disadvantages:**
 - Lower sensitivity compared to fluorescence.
 - Limited multiplexing capability (usually one or two antigens at a time).
 - **Fluorescent Detection:**
 - **Method:** Secondary antibodies are conjugated to fluorescent dyes (e.g., FITC, Alexa Fluor) that emit light upon excitation with specific wavelengths.
 - **Advantages:**
 - High sensitivity.
 - Ability to detect multiple antigens simultaneously (multiplexing).
 - **Disadvantages:**
 - Fluorescence fades over time (photobleaching).
 - Requires a fluorescence microscope, which is more expensive and complex to use.
 - **Comparison:**
 - **Sensitivity:** Fluorescent detection is generally more sensitive than chromogenic detection.

- **Specificity:** Both methods are specific, but fluorescence allows for better multiplexing, increasing the specificity of multiple antigen detection.
 - **Visualization:** Chromogenic detection provides clear, permanent staining visible under standard light microscopes, while fluorescent detection requires specialized equipment but offers enhanced sensitivity and multiplexing capabilities.
- 4. **What are the common challenges and sources of error in immunohistochemistry? Provide strategies for troubleshooting issues like non-specific staining, weak signal, and high background staining.**
 - **Common Challenges and Sources of Error:**
 - **Non-specific Staining:** Can occur due to cross-reactivity of antibodies or inadequate blocking.
 - **Weak Signal:** May result from insufficient antibody concentration or poor antigen retrieval.
 - **High Background Staining:** Caused by non-specific binding of antibodies or endogenous enzyme activity.
 - **Troubleshooting Strategies:**
 - **Non-specific Staining:**
 - Use proper blocking agents (e.g., serum, BSA) to block non-specific binding sites.
 - Optimize antibody concentrations and incubation times.
 - Validate antibody specificity using appropriate controls (positive and negative controls).
 - **Weak Signal:**
 - Ensure effective antigen retrieval.
 - Increase primary antibody concentration or extend incubation time.
 - Use signal amplification techniques (e.g., avidin-biotin complex).
 - **High Background Staining:**
 - Use appropriate blocking agents to block endogenous enzymes (e.g., hydrogen peroxide for endogenous peroxidase).
 - Increase washing steps to remove unbound antibodies.
 - Optimize antibody dilution and incubation conditions.
- 5. **How can immunohistochemistry be used to differentiate between malignant and benign tumors? Provide examples of specific markers that are commonly used in this context.**
 - **Use in Differentiation:** Immunohistochemistry can distinguish between malignant and benign tumors by detecting specific markers that are differentially expressed in malignant cells.
 - **Examples of Specific Markers:**
 - **Ki-67:** A proliferation marker that is often higher in malignant tumors.

- **p53:** A tumor suppressor protein that is frequently mutated and overexpressed in cancer cells.
 - **HER2/neu:** Overexpressed in certain types of breast cancer.
 - **Cytokeratins:** Intermediate filament proteins that help distinguish between epithelial (carcinomas) and non-epithelial tumors.
 - **CD markers:** Different CD markers (e.g., CD20, CD3) help classify lymphomas and leukemias.
6. **Explain the role of immunohistochemistry in personalized medicine, particularly in the context of cancer treatment. How do IHC results influence therapeutic decisions and patient management?**
- **Role in Personalized Medicine:** Immunohistochemistry provides crucial information about the molecular characteristics of a tumor, guiding personalized treatment strategies.
 - **Influence on Therapeutic Decisions:**
 - **Targeted Therapy:** IHC identifies specific molecular targets (e.g., HER2 in breast cancer, EGFR in lung cancer) that can be targeted by specific drugs.
 - **Prognostic Information:** Expression levels of markers like Ki-67 and p53 can indicate tumor aggressiveness and help predict patient outcomes.
 - **Predictive Biomarkers:** Markers like PD-L1 help predict the response to immunotherapy.
 - **Patient Management:** IHC results guide the selection of appropriate therapies, monitor treatment response, and detect recurrence or resistance.
7. **Describe the process and significance of using multiplex immunohistochemistry. How does this technique enhance the understanding of complex tissue environments and interactions?**
- **Process:** Multiplex immunohistochemistry involves the simultaneous detection of multiple antigens within a single tissue section using different fluorophores or chromogens.
 - **Significance:**
 - **Enhanced Understanding:** Allows for the detailed study of cellular interactions and the spatial relationships between different cell types and markers within the tissue microenvironment.
 - **Applications:** Useful in cancer research to study tumor heterogeneity, immune cell infiltration, and the tumor microenvironment.
 - **Technical Aspects:** Requires careful optimization to avoid cross-reactivity and spectral overlap in fluorescent detection.
 - **Challenges:** Complex data analysis and interpretation, potential for increased background staining.
8. **Discuss the use of immunohistochemistry in the study of infectious diseases. How can IHC help identify pathogens and understand host-pathogen interactions?**

- **Identification of Pathogens:** IHC can detect specific antigens of pathogens (e.g., bacteria, viruses, fungi) within infected tissues using pathogen-specific antibodies.
 - **Examples:**
 - **Viruses:** Detection of viral proteins in infected tissues (e.g., HPV, CMV).
 - **Bacteria:** Identification of bacterial antigens in tissue samples (e.g., Mycobacterium tuberculosis).
 - **Fungi:** Visualization of fungal components in tissues (e.g., Aspergillus).
 - **Understanding Host-Pathogen Interactions:** IHC helps visualize the localization of pathogens in relation to host cells, immune response, and tissue pathology, providing insights into disease mechanisms and immune evasion strategies.
9. **Explain the role of immunohistochemistry in neuroscience research. How is IHC used to study neural tissue, and what are some common markers for different cell types in the nervous system?**
- **Role in Neuroscience Research:** IHC is used to study the distribution, localization, and expression of specific proteins within neural tissues, aiding in the understanding of neural development, function, and pathology.
 - **Common Markers:**
 - **GFAP (Glial Fibrillary Acidic Protein):** Marker for astrocytes.
 - **NeuN (Neuronal Nuclei):** Marker for mature neurons.
 - **Iba1 (Ionized Calcium Binding Adapter Molecule 1):** Marker for microglia.
 - **Synaptophysin:** Marker for synaptic vesicles, indicating synapses.
 - **Applications:** Used in research on neurodegenerative diseases (e.g., Alzheimer's, Parkinson's), brain development, and injury responses.
10. **Compare and contrast immunohistochemistry with other protein detection techniques such as Western blotting and ELISA. What are the unique advantages and limitations of IHC?**
- **Comparison with Western Blotting:**
 - **Western Blotting:** Analyzes protein expression levels in tissue homogenates or cell lysates, providing quantitative data and molecular weight information.
 - **Advantages of Western Blotting:** Quantitative, can confirm protein size.
 - **Limitations of Western Blotting:** Lack of spatial information, requires homogenized samples.
 - **Comparison with ELISA:**
 - **ELISA (Enzyme-Linked Immunosorbent Assay):** Quantifies specific proteins in liquid samples (e.g., serum, cell culture supernatants).
 - **Advantages of ELISA:** Highly quantitative, suitable for high-throughput screening.

- **Limitations of ELISA:** No spatial context, cannot be used on tissue sections.
- **Unique Advantages of IHC:**
 - **Spatial Resolution:** Provides detailed localization of proteins within tissue architecture.
 - **Visual Context:** Allows for the study of protein expression in relation to tissue morphology and cellular context.
 - **Clinical Application:** Widely used in diagnostic pathology to classify tumors and guide treatment decisions.
- **Limitations of IHC:**
 - **Semi-Quantitative:** Generally less quantitative compared to Western blotting and ELISA.
 - **Subjectivity:** Interpretation can be subjective and dependent on the pathologist's expertise.
 - **Technical Variability:** Results can be influenced by variations in tissue processing, antibody specificity, and staining protocols.