Microscopy Question Bank with Answers

Short Answer Questions

1. Define refraction and explain its role in microscopy. (Remember/Understand)
Answer: Refraction is the bending of light as it passes from one medium to another, such as air to glass. In microscopy, refraction enables lenses to focus light rays and magnify small objects.

2. What is the function of the condenser lens in a bright-field microscope? (Remember)
Answer: The condenser lens focuses light onto the specimen, improving illumination and contrast.

3. Calculate the total magnification when using a 40x objective and a 10x ocular lens. (Apply)
Answer: Total magnification = Objective × Ocular = 40 × 10 = 400x.

4. Why is immersion oil used with the 100x objective lens? (Understand)
Answer: Immersion oil reduces light refraction by matching the refractive index of glass, allowing more light to enter the lens and increasing resolution.

5. State the resolving power formula and identify the variables it depends on. (Remember)
Answer: Resolving power is given by $d = 0.5\lambda / NA$, where λ is the wavelength of light and NA is the numerical aperture.

6. Differentiate between a micrometer and a nanometer in terms of biological scale. (Understand)
Answer: 1 micrometer (µm) = 10■■ m, typical for bacteria. 1 nanometer (nm) = 10■■ m, used for viruses and molecular structures.

7. What is the purpose of the iris diaphragm in a microscope? (Remember/Understand)
Answer: The iris diaphragm regulates the amount of light reaching the specimen, adjusting brightness and contrast.

8. List two advantages of chemical fixation over heat fixation. (Understand)
Answer: Chemical fixation preserves delicate structures and prevents distortion, while heat fixation may damage cells.

9. Mention one limitation of bright-field microscopy. (Understand)
Answer: Bright-field microscopy cannot effectively observe unstained, living cells due to low contrast.

10. Why is resolution more important than magnification in microscopy? (Analyze)
Answer: Magnification enlarges images, but resolution determines clarity. Without resolution, magnification only produces blurry images.

Short Essay Questions

1. Explain how a convex lens produces a magnified virtual image. (Understand)
Answer: A convex lens bends parallel rays of light to converge at a focal point. When an object is placed near the focal point, the lens produces a magnified virtual image that the eye perceives as larger.

2. Describe the sequence of light travel through a bright-field microscope. (Understand)

Answer: Light passes from the illuminator through the condenser, then the specimen, then through the objective lens, and finally through the ocular lens to the eye.

3. Compare and contrast simple, differential, and special staining techniques. (Analyze)
Answer: Simple stains use one dye to highlight cell shape. Differential stains use multiple dyes to distinguish cell types (e.g., Gram stain). Special stains target specific structures like spores or flagella.

4. Discuss the role of numerical aperture (NA) in determining resolution. (Analyze)
Answer: NA indicates a lens's light-gathering ability. Higher NA improves resolution, allowing finer details to be distinguished.

5. Explain why blue light improves resolution compared to red light. (Analyze)
Answer: Blue light has a shorter wavelength than red light, reducing the value of d in the resolution formula and thus providing better clarity.

6. Outline the steps in preparing a bacterial smear for staining. (Apply)
Answer: Steps include: spreading a thin film of bacteria, air-drying, heat or chemical fixation, and applying stain to visualize cells.

7. What are common pitfalls in specimen preparation, and how can they be avoided? (Evaluate)
Answer: Common errors include over-staining, uneven smears, and poor fixation. These can be avoided by careful technique, correct dye use, and proper fixation.

8. Discuss how fixation preserves cellular structures during microscopy. (Understand/Analyze)
Answer: Fixation kills cells while stabilizing proteins and membranes, preventing decay and maintaining morphology for observation.

9. Differentiate between magnification and resolution with appropriate examples. (Analyze)
Answer: Magnification enlarges images (e.g., 400x), while resolution determines the ability to distinguish two points (e.g., bacteria 0.2 µm apart).

10. Explain why mastering light microscopy is essential before using advanced microscopy techniques like fluorescence or electron microscopy. (Evaluate)
Answer: Light microscopy provides fundamental skills in handling specimens, adjusting light, and interpreting images, forming the basis for advanced techniques.

Essay Questions

1. Discuss the essential principles of bright-field microscopy, highlighting the relationship between light, lenses, and image formation. (Analyze/Synthesize)
Answer: Bright-field microscopy uses light transmitted through a specimen. Lenses refract and magnify light, producing images that reveal cell structure. The ocular and objective lenses combine magnification, while resolution and contrast depend on lens quality and illumination.

2. Evaluate the significance of resolution and numerical aperture in studying microorganisms, and explain how immersion oil enhances image clarity. (Evaluate/Apply)
Answer: Resolution determines clarity, not just enlargement. Numerical aperture measures light-gathering ability. Immersion oil reduces light scattering, increases NA, and allows clear visualization of microorganisms like bacteria at 100x magnification.

3. Analyze the process of specimen preparation in light microscopy. How do fixation, staining, and smear preparation contribute to accurate microbial observation?

(Analyze/Evaluate)
Answer: Specimen preparation ensures clarity and stability. Fixation preserves morphology, staining increases contrast, and smears spread cells for visibility. Together, these steps allow accurate identification of cellular features and microbial structures.