FAQ on Scanning Electron Microscope (SEM)

1. Short-Answer Questions (1–2 sentences each)

1. What is the full form of SEM? (Remembering)

   Answer: SEM stands for Scanning Electron Microscope.

2. Who invented the scanning electron microscope? (Remembering)

   Answer: The SEM was developed by Manfred von Ardenne in 1937.

3. What type of beam is used in SEM? (Remembering)

   Answer: A focused beam of high-energy electrons is used in SEM.

4. Mention one main difference between light microscopy and SEM. (Understanding)

   Answer: Light microscopy uses photons (light), whereas SEM uses electrons for imaging.

5. What is the usual voltage range used to accelerate electrons in SEM? (Remembering)

   Answer: Typically between 1–30 kilovolts (kV).

6. Name two types of signals produced when the electron beam strikes the specimen. (Understanding)

   Answer: Secondary electrons and backscattered electrons.

7. What is the main function of electromagnetic lenses in SEM? (Understanding)

   Answer: They focus and control the electron beam on the specimen surface.

8. Why is gold or platinum coating applied to biological samples before SEM observation? (Understanding)

   Answer: To make the specimen conductive and prevent charging under the electron beam.

9. What is meant by “secondary electrons” in SEM imaging? (Understanding)

   Answer: They are low-energy electrons emitted from the specimen’s surface, revealing topographical details.

10. State one advantage and one limitation of SEM. (Understanding)

    Answer: Advantage: Produces high-resolution 3D images. Limitation: Only surface structures can be viewed.

2. Short-Essay Questions (4–6 sentences each)

1. Explain how the interaction between electrons and specimen atoms results in image formation in SEM. (Applying)

   Answer: When high-energy electrons strike the specimen, they interact with its atoms, generating signals such as secondary and backscattered electrons. These signals vary with the surface composition and topography. Detectors collect and convert these signals into electrical outputs, which are then displayed as a high-resolution image representing surface morphology.

2. Describe the purpose of the vacuum system in a scanning electron microscope. (Applying)

Answer: The vacuum system removes air and contaminants from the microscope chamber. This allows electrons to travel freely without scattering, ensuring a clear and focused image. It also prevents oxidation of the filament and damage to sensitive components.

3. Outline the steps involved in biological specimen preparation for SEM analysis. (Applying)

Answer: Biological samples are fixed using glutaraldehyde, dehydrated through graded ethanol or acetone, dried using a critical point dryer, and coated with a conductive layer such as gold or platinum. These steps maintain structural integrity under vacuum and improve imaging.

4. How does SEM provide three-dimensional images compared to optical microscopy? (Analyzing)

Answer: SEM creates 3D-like images because secondary electrons are emitted differently from various surface angles, producing shadow and depth effects. This contrast gives a realistic perception of the specimen's texture and morphology, unlike the flat images from light microscopy.

5. Differentiate between secondary and backscattered electrons in terms of their origin and information provided. (Analyzing)

Answer: Secondary electrons are emitted from the surface layer and give topographical details, while backscattered electrons originate from deeper layers and indicate compositional contrast. Secondary electrons produce high-resolution images, whereas backscattered ones highlight material density differences.

6. Discuss how critical point drying preserves delicate biological structures for SEM observation. (Applying)

Answer: Critical point drying replaces water with a transitional fluid like CO■ and then removes it at its critical point, avoiding surface tension damage. This preserves the sample's natural shape and fine surface features.

7. Explain why SEM is particularly useful in microbiology and botany. (Applying)

Answer: In microbiology, SEM reveals bacterial shapes, colonies, and biofilms. In botany, it provides detailed views of leaf surfaces, stomata, pollen grains, and trichomes, enhancing understanding of structural adaptations.

8. Describe how the scanning system controls the movement of the electron beam across the specimen surface. (Applying)

Answer: The scanning system uses electromagnetic coils to deflect the beam in a raster pattern over the specimen. This systematic scanning ensures each region is imaged, allowing uniform data collection across the surface.

9. Analyze the impact of improper sample preparation on the quality of SEM images. (Analyzing)

Answer: Poor preparation may lead to charging, shrinkage, or collapse of structures, causing blurred or distorted images. Contaminants or moisture can also interfere with vacuum stability and signal clarity.

10. Compare SEM with TEM in terms of the type of information each provides. (Analyzing)

Answer: SEM shows surface morphology and 3D structure, while TEM provides details of internal ultrastructure at higher magnification. SEM is better for topography; TEM excels in studying internal organization.

3. Essay Questions (300–500 words each)

1. Evaluate the role of the scanning electron microscope in advancing modern biological research. Discuss its advantages and limitations in detail with relevant examples. (Evaluating)

Answer: The scanning electron microscope (SEM) has played a transformative role in modern biological research by allowing scientists to visualize minute surface structures with remarkable clarity. SEM's ability to generate three-dimensional images at nanometer resolution has expanded our understanding of cellular and tissue morphology. In cell biology, SEM helps reveal the surface characteristics of membranes, microvilli, and cilia, providing insight into cellular functions. In botany, it allows the study of pollen grain patterns, trichomes, and stomatal complexes, while in zoology, it elucidates adaptations in insect exoskeletons and feathers. The major advantages of SEM include high depth of field, large magnification range, and ability to examine diverse biological materials. However, SEM is not without limitations. Since it requires vacuum conditions, living specimens cannot be studied directly. Moreover, extensive sample preparation steps such as fixation, dehydration, and coating may introduce artifacts. The equipment is also costly and requires trained personnel. Despite these challenges, SEM remains a cornerstone technique in biology, enabling discoveries from microbial morphology to nanostructured biomaterials.

2. Analyze the importance of proper sample preparation in SEM and suggest improvements or modern techniques that minimize specimen distortion in biological imaging. (Analyzing / Evaluating)

Answer: Sample preparation is a critical factor that determines the quality and accuracy of SEM images. Biological specimens are soft, moist, and non-conductive, making them vulnerable to damage in the SEM's high-vacuum and high-energy conditions. Proper fixation with glutaraldehyde stabilizes cellular components, while dehydration through graded solvents removes water. Critical point drying prevents collapse due to surface tension, and metal coating enhances conductivity. Inadequate preparation can cause sample shrinkage, distortion, or charging artifacts, leading to misinterpretation. Recent innovations aim to overcome these limitations. Cryo-SEM allows imaging of frozen-hydrated samples at cryogenic temperatures, preserving natural structures without dehydration. Environmental SEM (ESEM) operates at variable pressures, permitting observation of moist or living specimens without coating. Such advancements enhance image realism and broaden SEM's applications in biological research, offering a more faithful representation of native structures.

3. Design an experimental study where SEM could be used to investigate a specific biological structure (e.g., pollen grains, insect exoskeletons, or bacterial biofilms). Describe the steps, expected observations, and significance of the findings. (Creating)

Answer: An experiment can be designed to study the surface morphology of pollen grains using SEM. First, pollen samples are collected, air-dried, and fixed with glutaraldehyde to preserve their shape. Dehydration is performed using ethanol series, followed by critical point drying. The dried pollen is mounted on aluminum stubs and coated with a thin layer of gold using a sputter coater. The specimen is then examined under SEM at various magnifications. The expected observations include detailed surface ornamentations, apertures, and exine patterns characteristic of each plant species. Comparing different pollen types can reveal evolutionary adaptations and assist in plant taxonomy. This SEM-based investigation demonstrates the utility of surface imaging in biological classification and pollination biology, linking structural features to ecological function and species differentiation.