

## MICROSCOPY SOCIETY OF AMERICA

Affiliate Society of American Institute of Physics - Affiliate Society of AAAS  
MSA Association Management Office, 11130 Sunrise Valley Dr., Suite 350, Reston, VA 20191  
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### GENERAL INFORMATION

The Microscopy Society of America (MSA)<sup>1</sup>, the world's largest professional association of microscopists, provides the only certification of technologists in biological transmission electron microscopy available in the Americas. The program was initiated in 1978 to establish standards of technical skills. In addition to insuring employers that certified technologists are technically proficient, certification can be important in determining job classification, salary level, and potential for advancement or promotion. Many consider certification to be a key benchmark in their professional development.

The program is administered by the Certification Board which is appointed by the Council of the Society. The Board develops regulations, formulates and evaluates examinations, and interprets policies. Individuals with the requisite educational and/or occupational qualifications can attain certification by passing both a written and practical examination. Two examination cycles are offered each year. Complete regulations and an application form appear on the following pages.

The initial period of certification is one year, the calendar year indicated on the certificate furnished to all successful candidates. Certification may be renewed on a 10- year cycle by payment of the appropriate fee (\$75 for MSA members, \$150 for non -members<sup>2</sup>). Certified Technologists who allow their certification to lapse for one year may have it reinstated by paying the appropriate fees; if certification lapses for two or more years, the technologist must submit a new application and take both written and practical examinations again. Communication with the Society regarding certification, should be addressed to:

Microscopy Society of America  
Certification Board  
11130 Sunrise Valley Rd., Suite 350  
Reston, VA 20191  
Phone: ( 703) 234-4115

<sup>1</sup>Before January 1, 1993, the Microscopy Society of America was the Electron Microscopy Society of America (EMSA). "Electron" was dropped from the name to reflect the Society's broadened scope that has come to include all kinds of microscopy and microanalysis. Nevertheless, most of the membership and scientific program still is concerned primarily with electron microscopy.

<sup>2</sup> The Society reserves the right to modify these and other fees from time to time to reflect changes in service, dues, etc.

## APPLICATION AND REQUIREMENTS

An application for certification consists of:

1. A completed application form (included in this package).
2. An application fee of \$75.00 for MSA members<sup>3</sup> or \$150.00 for non-members.<sup>4</sup>
3. Transcripts and/or documentation of ONE of the following:
  - Two years (60 credits) college or equivalent education, including at least 4 semesters of science that include: chemistry, physics, biology, mathematics, and two semesters transmission electron microscopy (TEM). The TEM course must include extensive hands-on experience in sample preparation and microscope operation.

**OR**

- One year (30 credits) college or equivalent education, including at least one semester of laboratory courses each in chemistry and physics AND one year of recent<sup>5</sup> full-time work experience in biological TEM, as a volunteer, internship, or paid employee.

**OR**

- A high school diploma AND two years of recent<sup>5</sup> full-time work experience in biological TEM

**OR**

- Three years of recent<sup>5</sup> full-time work experience in biological TEM

4. Letters of recommendation from two (2) people in supervisory positions having substantial records of research publication, ideally utilizing electron microscopy. Preferably, at least one of them should be a member of MSA. The letters may either be enclosed with the application or sent separately, but the application will not be considered complete until the letters are received by the Society.

All application materials can be sent to the Association Management Office by mail to

Microscopy Society of America  
Certification Board  
11130 Sunrise Valley Rd., Suite 350  
Reston, VA 20191

**OR**

by email to [associationmanagement@microscopy.org](mailto:associationmanagement@microscopy.org).

Applicants are responsible for seeing that all requirements are submitted in time and should confirm with the Association Management Office to make sure their applications are complete. Applications completed after the deadline date will automatically be considered for the next examination cycle.

Completed applications are evaluated by the Certification Board Chair to determine whether the applicant is qualified to take the examinations. Applicants not approved for examination will receive a written explanation and their certification application fee will be refunded (although MSA member dues, if any, will not be refunded). Fees for applicants approved for examination are not refundable.

<sup>3</sup>Application for membership in MSA is separate from application for certification. Candidates for certification may pay the lower member's fee by submitting an application for membership, along with one year's annual MSA dues, at the same time they submit the application and fee for certification.

<sup>4</sup>Payment can be by check (US funds, drawn on a US bank) payable to MSA, or by credit card (Visa or Master Card only). If paying by credit card, supply the complete credit card number and expiration date.

<sup>5</sup>"recent" is interpreted to mean within the five years prior to application.

### EXAMINATIONS: GENERAL

Candidates whose applications are approved must pass both a written and a practical examination in order to be certified. The candidate must pass the written exam before s/he may submit the materials for the practical exam. Both examinations usually are taken during the same cycle in which application was submitted; however, candidates may request deferring either or both examinations until the next cycle. Candidates who fail an examination in the cycle in which they applied may take it again in the next cycle without penalty.

If this occurs, all requirements for certification must be completed by the end of the following cycle. Otherwise, the candidate must submit a new application, including the application fee and letters of recommendation (transcripts need not be re-submitted unless they have become outdated). Examinations taken prior to re-application must be taken again, even if previously passed.

#### Written Examination

The written examination is of the objective type (multiple choice, true-false, etc.); three hours are allotted for completion of the written examination. In most cases the examination is conducted at or near the candidate's home institution. A score of 80% is required to pass. The material covered includes:

- A. Instrumentation including electron optics (approx. 25%)
- B. Tissue processing (fixations, resin chemistry etc.) (approx. 25%)
- C. Sectioning and staining (approx. 15%)
- D. Special techniques (Immuno, shadowing, cryo, etc.) and imaging (approx. 20%)

E. General: chemistry, safety (approx. 15%)

## Written Examination Study Syllabus

### A. Instrumentation

Accessory Equipment: Principles, components, alignment and routine maintenance of:

- Ultramicrotomes,
- Knifemakers
- Light microscopes
- Transmission electron microscope fundamentals:
  - Operation; illumination; imaging systems; alignment; focusing; maintenance; test specimens; astigmatism; resolution, calibration, contamination
- Scanning electron microscopes: general principles; operation
- Vacuum systems:
  - Vacuum evaporator, sputter coaters, mechanical, diffusion, turbomolecular, ion pumps, vacuum gauges

Other Lab Equipment:

- Incubators
- Ovens
- Balances
- pH meters
- Osmometers
- Centrifuges
- Photographic techniques (digital)

### B. Sample/Tissue procurement for TEM processing

Fixation & Processing

- General principles and purpose
- Types, composition, & preparation [glutaraldehyde, paraformaldehyde, OsO<sub>4</sub>, KMnO<sub>4</sub> and others]
- Buffers [eg: phosphate, cacodylate, PIPES, HEPES, s-collidine, veronal- acetate];
- Factors affecting fixation [fixative concentration, time, temperature, pH, osmolarity, buffer, additives, penetration]
- Methods of fixation [immersion, perfusion, vapor]
- Criteria for good fixation
- Washing: general principles and purpose
- En bloc staining
- Dehydration: general principles and purpose
- Dehydrating agents [ethanol, acetone, ethylene glycol, propylene oxide, acetonitrile]
- Factors affecting dehydration [concentration, time, temperature]
- Infiltration: general principles and purpose
- Embedding: general principles and purpose
- Types, composition and preparation of plastics [acrylics, polyesters, epoxies, catalysts, hardeners, plasticizers]
- Methods of embedding [capsules, flat, cell culture, vacuum]
- Polymerization: general principles and purpose

- Safety

### C. Sectioning and Staining

Sectioning: general principles and purpose

- Block Preparation: trimming; facing; re-mounting
- Knife preparation: glass breaking, inspection, troughs (boats)
- Diamond knives: use & handling
- Grid Preparation: types; cleaning; coating [Formvar, Butvar, collodion, carbon]
- “Thick” (semi-thin) sectioning; collection, mounting
- Thin sectioning: orientation, flotation [liquid and meniscus], flattening, collection, thickness [interference colors], problems, factors affecting quality

Staining: General principles and purpose

- Thick Section Staining: Toluidine blue-O, methylene blue, Paragon, azure II, Giemsa
- Thin sections: specific stains [uranyl acetate, lead citrate, phosphotungstic acid, osmium, ruthenium, silver] factors affecting staining quality
- Safety

### D. Digital Imaging/ Power Point

- General principles and purpose.
- Image processing,
- Use of computers
- Illustrations: labeling, magnifications

### E. Special Techniques

- Negative staining
- Shadow casting and replication
- Cytochemistry and immunolocalization
- High Pressure freezing and freeze substitution

### F. General

- Basic cytology, cell morphology, ultrastructure
- Reagents: solvents, solutions, normality, molarity, percentage, acids, bases, salts
- Cleanliness: glassware, distilled and deionized water
- Basic math: metric system, trigonometry, measurements
- Safety: radiation, chemical, biological, fire

## Written Examination

Sample Questions (from past examinations)

Multiple Choice

1. If a cell structure is 60  $\mu\text{m}$  long on a micrograph at 20,000X, its actual length is:

- (a) 6  $\mu\text{m}$
- (b) 3  $\mu\text{m}$
- (c) 2  $\mu\text{m}$
- (d) 0.33  $\mu\text{m}$
- (e) 0.16  $\mu\text{m}$

2. Proper lab attire includes:

- (a) lab coat or jacket
- (b) open toed shoes or sandals

(c) full length slacks

(d) shorts

(e) a and c above

3. Negative staining is often done with:

(a) lead citrate

(b) uranyl acetate

(c) phosphotungstic acid

(d) Toluidine blue-O

(e) b and c above

4. Astigmatism in a TEM can be caused by:

(a) contamination of an aperture

(b) improperly aligned filament

(c) a vacuum leak in the camera chamber

(d) a bent grid

(e) b and c above

5. How much 25% glutaraldehyde is needed to make 50 ml of 3% glutaraldehyde?

(a) 5 ml

(b) 10 ml

(c) 6 ml

(d) 3 ml

(e) 2.5 ml

## Practical Examination

The practical examination consists of preparing blocks, sections, and micrographs from three different samples/tissues and submitting them for evaluation by two (or sometimes three) members of the Certification Board. The examiners base their scoring on the usability of the specimens and grids in everyday practice in a research or clinical setting. Thus, the work submitted should represent the candidate's BEST work. Material submitted should be publication quality and should include image labels that identify key features of the tissues used.

Procedures should be written so that anyone familiar with biological electron microscopy could replicate the work. The relative weight given to various aspects of the submitted material is indicated on the MSA Practical Exam Grading Sheet included in this packet. An average (mean) score of 80 is required to pass.

All work must be done by the candidate alone. However, a supervisor or other qualified individual may assist in obtaining the gross tissue specimens. A signed Pledge of Independent Workmanship (included in this package) must accompany the examination materials Standard (non-pathological and non-human) materials and common processing methods should be used.

Identify all submitted materials with the examination ID number you were assigned when your application was approved. Do not label items with either your name or your lab or institution's name.

The bullet-points outlined in the syllabus above are indicative of the grading points used in evaluating the practical exam materials. ALL aspects of sample preparation and presentation are considered.

Detailed instructions follow:

I. Prepare three different samples for transmission electron microscopy from fixation through sectioning and uranyl acetate-lead staining, Use uncoated 200 or 300 mesh copper grids to mount thin sections. The samples may be plant, animal, cell culture, or microorganism, as the candidate chooses. At least one sample must be from a mammal, cell culture or higher plant. Be aware that the embedding resin you choose will affect the quality of your final images.

II. Submit the following:

a. one trimmed block from each sample

b. four (4) grids with high quality thin sections cut **from each** of the submitted blocks

c. one slide of “thick” (ca. 1  $\mu$ m) sections for light microscopy, appropriately stained and labelled, from each submitted block

d. a detailed description, no longer than one page for each sample, of the preparation methods used. Procedures should be written so anyone else could replicate the work. Be sure to indicate whether a glass or diamond knife was used. This should be modeled after the “Materials and Methods” sections of refereed journals. If the same methods were used for all 3 samples, one copy of the methods may be submitted. It should specify that the methods apply to all submitted samples. The methods may be submitted as printed copy or as a Word document included on the USB drive that contains the images.

e. Submit six (6) images of each sample at 3 magnifications within the microscope magnification range of 2,500x to 30,000. Submit at least one at low magnification (survey), at least one at intermediate magnification featuring a single cell, and at least one at higher magnification showing subcellular/organelle features).

There are three acceptable methods to submit images based on the technology the applicant has available to them in their lab:

### 1. **Digital Image Submission**

- Submit raw images as acquired at the microscope in a folder labeled “raw images”. Create a subfolder for each sample. If images are acquired with proprietary software, submit raw images as acquired and also submit raw images in TIFF format in a file labeled “raw images TIFF”.

- In a file labeled “annotated images” create a subfolder for each sample. Annotated images must be submitted as TIFF files. Annotated images must include labelled structures of interest within the tissue and a scale bar. Figure legends for each submitted image should all be on a separate page. (see “f” below)

- Create a power point presentation with two images from each sample using the annotated images. Include a figure legend. Do not use any “special effects”.

- Image data should be submitted on a USB (flash) drive.

**2. Photographic Film/Print Submission** (for labs without digital acquisition or scanner only)

- Submit original negatives

- Submit an annotated 8x10 photographic print of each micrograph. Include a scale bar on each print and a figure legend.

**3. Hybrid Film and Digital Submission**

- Record images on film and submit all negatives.

- Scan images into digital format as TIFF images. Place in a folder labeled “scanned raw images”.

-Follow 1: submission of digital images.

In the Methods Section:

- Identify the digital camera; manufacturer, model and pixel array (eg: 2K x 2K; 11 megapixel).
- For film/print images, show how you calculated the length of the scale bar for each magnification used. Use three different magnifications.
- Identify the scanner; manufacturer, model pixel array and dpi, if used.
- Create a power point presentation with two images from each sample using the annotated images. Include a figure legend. Do not use any “special effects”.

**f. Complete figure legends for each micrograph should be printed on a separate sheet.**

They should be concise (journal style: e.g., Microscopy and Microanalysis) and should describe any labeled structures and scale bars that appear on the micrographs.

III. Separate the grids into three groups of four and place them in a grid box (slide-type preferred) secured with a rubber band or tape. We recommend packing all materials in a sturdy box or padded shipping envelope. Use packing material so that the contents can't shift during transport.

IV. Send all materials (do not forget the Pledge of Independent Workmanship) to the Chair of the Certification Board to arrive on or before the deadline date. [Sending the practical exam to the Association Management Office delays the grading of the exam and increases shipping costs.] The Certification Board Chairman will provide you with the appropriate shipping address when you are informed of passing the written examination. We recommend using a courier like UPS or Federal Express. If you use the U.S. Postal Service, send the exam by express, certified, or registered mail. MSA is not responsible for damage to examination materials in transit.

VI. Submitted examination materials are held confidential, become the property of MSA, and

are not returned to the applicant. If one or two grids per tissue are damaged or not usable, grades will be based on the remaining grids; if there are more than two damaged grids per sample or unlabeled or missing material, or other deficiencies, the examination may be returned for re-submission in a later cycle.

## Reference Books on Transmission Electron Microscopy

Bozzola, J. J. and Russell, L. D. Electron Microscopy, Jones and Bartlett, Boston, (1999)

Dykstra, Michael J. and Reuss, Laura E. Biological Electron Microscopy: Theory, Techniques, Troubleshooting, Springer- (2003 ).

Griffiths, G. Fine Structure Immunocytochemistry, Springer-Verlag (1993).

Hajibagheri, M. A. Nasser, Electron Microscopy: Methods and Protocol, Springer (1999 ).

Hayat, M. A. Basic Techniques for Transmission Electron Microscopy, Academic Press, New York(1986)

Hayat, M. A. Correlative Microscopy in Biology. Instrumentation and Methods, Academic Press, New York (1987)

Hayat, M. A. Principles and Techniques of Electron Microscopy. Biological Applications, Cambridge University Press (2000)

Kuo, John, Electron Microscopy: Methods and Protocols, Second Edition, Springer (2007)

Maunsbach, Arvid, and Björn Afzelius, Biomedical Electron Microscopy: Illustrated Methods and Interpretations, Academic Press (1998).

Howard, CV and Reed, MG, Unbiased Stereology: Three Dimensional Measurement in Microscopy, Springer (1999)

## Review Articles

Goosmann, C., Abed, U., Brinkmann, V. Infection at the Cellular Level. Methods in Cell Biology, (2008). 88:477-496

Hurbain, I. and Sachse, M. The future is cold: cryo-preparation methods for transmission electron microscopy of cells Biol. Cell (2011) 103, 405–42

Wisse, E. et. al. Fixation methods for electron microscopy of human and other liver. World J Gastroenterol (2010) 16(23): 2851-2866

MICROSCOPY SOCIETY OF AMERICA  
CERTIFICATION BOARD  
Practical Examination  
Pledge of Independent Workmanship

Applicant's Name: \_\_\_\_\_

Applicant's Examination Number: \_\_\_\_\_

I hereby state that all the procedures carried out in the preparation of the enclosed grids, microscope preparations and micrographs were performed exclusively by me and without any assistance.

Applicant's Signature: \_\_\_\_\_

Witness' Name (print): \_\_\_\_\_

Witness' Signature: \_\_\_\_\_

Date: \_\_\_\_\_

Location(s) where work was performed:

**MSA PRACTICAL EXAM GRADING SHEET**

ID# \_\_\_\_\_ Cycle# \_\_\_\_\_ For Certification as Technologist (Biological)

1st Submission\_\_\_\_ Retake\_\_\_\_

**BIOLOGICAL SCIENCE TISSUES (NONPATHOLOGICAL)**

	POINTS		SCORE	<u>TOTAL</u>
1. _____ (Name of Specimen)	BLOCK	3	_____	
	SLIDE	3	_____	
	SECTION	6	_____	
	IMAGES	6	_____	
	POWERPOINT	6	_____	
	LEGEND	3	_____	
		6	_____	
	OVERALL IMPRESSION		_____	
		(33)	_____	
2. _____ (Name of Specimen)	BLOCK	3	_____	
	SLIDE	3	_____	
	SECTION	6	_____	
	IMAGES	6	_____	
	POWERPOINT	6	_____	
	LEGEND	3	_____	
	OVERALL IMPRESSION	6	_____	
		(33)	_____	
3. _____ (Name of Specimen)	BLOCK	3	_____	
	SLIDE	3	_____	
	SECTION	6	_____	
	IMAGES	6	_____	
	POWERPOINT	6	_____	
	LEGEND	3	_____	
	OVERALL IMPRESSION	6	_____	
		(33)	_____	
	<b>TOTAL SCORE</b>		_____	

Date: \_\_\_\_\_

Grader's Signature: \_\_\_\_\_

MICROSCOPY SOCIETY OF AMERICA  
Application for Certification  
Electron Microscopy Technologist  
Biological Transmission Electron Microscopy

Name: \_\_\_\_\_

Mailing Address: \_\_\_\_\_

Is this address your residence? \_\_\_\_\_ Work? \_\_\_\_\_

Company/University (only if not part of above address): \_\_\_\_\_

Daytime Phone: ( \_\_\_\_ ) \_\_\_\_\_ FAX: ( \_\_\_\_ ) \_\_\_\_\_

E-mail address: \_\_\_\_\_

I have read and understand the regulations pertaining to MSA Certification.

Your signature: \_\_\_\_\_ Date: \_\_\_\_\_

EDUCATION (Start with High School)

School/Location/Years Attended Credit Hours Major Field Degree

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

EMPLOYMENT (EM Related)

Current employer (name and address):

\_\_\_\_\_

\_\_\_\_\_

Position/Title: Years employed there: \_\_\_\_\_

Supervisor's name: \_\_\_\_\_

Previous employer (name and address):

\_\_\_\_\_

\_\_\_\_\_

Position/Title: \_\_\_\_\_ Years employed there: \_\_\_\_\_

Supervisor's name: \_\_\_\_\_