Modern Imaging, Spectroscopy and Diffraction Techniques

TIF 030 and FIM 150 January 14th, 2013 Afternoon

Aids: Formula sheets attached to the exam, "Physics Handbook", calculator, and writing tools.

Total marks available from exam: 30 Marks required to pass: 12

1) Basic imaging (3p)

- a) Make schematic drawings of the illumination and imaging paths according to the Köhler design for an inverted optical microscope equipped with an infinity corrected objective. Mark the different conjugate planes and parts of the microscope. (2p)
- b) What is approximately the smallest distance between two point-like objects that one can resolve using the highest resolution classical optical microscope available on the market and using visible light? (1p)

2) Practical microscopy (3p)

- a) Some objectives for inverted microscopes are designed for using immersion oil between the lens surface and the substrate. What is the major advantage of such objectives? (1p)
- b) Wide-field (episcopic) fluorescence microscopes usually contains a filter cube. What are the three primary components of a filter cube and where is the cube placed in the microscope? (1 p)
- c) What is the primary advantage of so-called fluorescent proteins (e.g. GFP) compared to ordinary fluorescent dyes for imaging of biological cells (1p)

3) Fluorescence microscopy (4p)

Name three methods used for improving spatial resolution in fluorescence microscopy. Pick one and explain the basic principle, the basic set up, including schematic drawing (if possible), and explain when the method you have chosen is a better alternative than the other two. (3p)

4) Modern optical microscopy (2p)

In a few sentences and drawings, describe the basic principles and use of **two** of the following techniques/phenomena (2p): FRAP, FRET, NSOM, FLIM, SERS, NIM, FCS, DIC, TIRF.

5) SEM (3p)

- (a) Draw a schematic figure illustrating the depth of focus in the SEM. (1p)
- (b) Calculate the depth of focus for a working distance of 10 mm, an aperture diameter of 30 μm and a spatial resolution of 3 nm. (1p)
- (c) List three of the most common signals used in the SEM order them according to the spatial resolution that can be obtained using each signal. (2p)

6) EDS (2p)

(a) The fluorescence yield is an important factor for magnitude of the X-ray signal. Draw the fluorescence yield as a function of Z number for the K, L, and M lines. (1p)

(b) The acceleration voltage in the SEM affects the spatial resolution of the analysis. Draw a schematic figure illustrating the effect of acceleration voltage on the spatial resolution. (1p)

7) Electron diffraction (3p)

The diffraction pattern in Fig. 1 is obtained for a gold crystal with the electron beam incident along the [110] direction in a TEM operated at 200 kV.

- (a) Draw the Kikuchi lines corresponding to the 6 diffraction spots closest to 000 in Fig 1. (1p)
- (b) Draw the Kikuchi lines for spot A when the crystal is tilted so that the Bragg condition for spot A is fulfilled. (1p)
- (c) How does the intensity of the spot change when going from (a) to (b)? (1p)

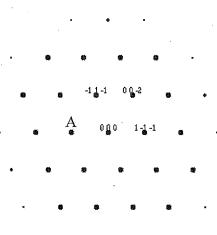


Fig. 1. Diffraction pattern from a gold crystal with the electron beam incident along the [110] direction.

8) TEM (3p)

- (a) Draw a schematic ray diagram that shows how a diffraction pattern and an image are formed in the TEM. Include the specimen and the objective lens in the diagram. All other lenses can be omitted. (1p)
- (b) Describe how Bright Field and Dark Field images can be obtained. Which aperture is used? (0.5p)
- (c) How is an image showing phase contrast obtained in a TEM? (0.5)
- (d) How is an image showing amplitude contrast obtained? (0.5p)
- (e) What is the role of the first condenser lenses? (0.5p)

9) EDS in TEM (4p)

(a) There are three main parts of EELS spectra. Describe them. (1p)

- (b) You are to investigate the presence of light elements like oxygen and nitrogen in a TEM specimen. Would you prefer EDS or EELS? Why? (1p)
- (c) Assume that you would like to investigate if there is segregation of an element to a grain boundary. Explain the effect of specimen thickness and acceleration voltage on the quality of the analysis. (1)
- (d) How should the grain boundary plane in (c) be oriented with respect to the incident electron beam? How is this achieved practically? (1p)

10) SPM: Common denominators and distinguishing characteristics (3p)

There are a large variety of SPM techniques (including STM, AFM, SNOM, etc.). In fact, one often talks about a "family" of SPM techniques. This is related to the fact that most SPM techniques historically have been derived from the mother of all SPM techniques – STM– and that there exist many communalities between the various SPM techniques.

- a) What is the common denominator for most SPM techniques? (0.5p)
- b) What are the major distinguishing characteristics when comparing scanning tunneling microscopy (STM) to atomic force microscopy (AFM)? (1p)
- c) The two most common STM imaging modes are constant-current and constant-height mode. What are the major distinguishing characteristics between these two modes? (1p)
- d) When operated in constant-height mode, which of the two techniques STM or AFM is more susceptible to a small vertical drift and why? (0.5p)

Formula sheet

<u>, Applications</u>	
Element	$k_{ASi}(1)$
(A)	100 kV
- <u>** 76 f. 1841 1945 </u> - ***	GENERAL TEACHERS
Na	5.77
Mg	2.07 ± 0.1
A1	1.42 ± 0.1
Şi	1.0
P	
S	
CI	
K	
Ca	1.0 ± 0.07
Ti	1.08 ± 0.07
V	1.13 ± 0.07
Cr	1.17 ± 0.07
Mn	1.22 ± 0.07
Fe	1.27 ± 0.07
Co	
Ni	1.47 ± 0.07
Cu	1.58 ± 0.07
Zn	1.68 ± 0.07
Ge	1.92
Zr	
Nb Mo	4.3
	8.49
Ag Cd	10.6
In	1,
Sn	10.6
Ba	10.0
	1.

$$\begin{split} \lambda &= h \, / \, [2m_0 eV(1 + eV/2m_0 c^2)]^{1/2} \\ d_p &= (d_g^{\, 2} + d_s^{\, 2} + d_d^{\, 2} + d_c^{\, 2})^{1/2} \\ r_{Sch} &= 0.66 \, \, C_s^{\, 1/4} \, \lambda^{3/4} \\ n &> (5/C)^2 \\ 2 \, d_{hkl} \, \sin\Theta &= n\lambda \\ b &= 7.21 \, x \, 10^5 \, (\rho/A)^{1/2} t^{3/2} \, (Z/E_0) \\ I &\simeq U \rho_s(E,r) e^{-2 \frac{\sqrt{2m_e \phi}}{\hbar} d} \, with \, \phi = \frac{1}{2} \Big(\phi_{sample} + \phi_{tip} \Big) \end{split}$$