

Welcome to the microscopy course

Due to covid-19 the course will be given almost entirely on-line. Course activities will be:

- Lectures on-line. The first half of each lecture will be pre-recorded so that you may view it anytime. The second half will be a real-time zoom session.
- Labs on campus. Will take place in small groups of 2 students and 1 teacher.
- Exam on-line. A combination of oral exam via zoom and written home exam.
- Seminar (only for SK2501/FSK3501) will happen via zoom.

For FSK3500/FSK3501 there will also be a project, communicated individually.

The course literature is two compendia written by Kjell Carlsson. They can be found on the Canvas page once the course starts.

Course responsible and teachers are Anna Burvall and Ilaria Testa.

To pass the course

To pass the course, you should:

- complete all four labs
- complete a lab report on lab 3-4 and reach pass level
- pass the exam
- For SK2501, complete and pass the seminar

The result on the exam determines the grade on the course.

Intended learning outcomes

After completing the course the student should be able to:

- adjust the illumination system to obtain optimal performance in transmission microscopy.
- select a suitable light source and optical filters, and correctly adjust the illumination system for fluorescence microscopy.
- select a suitable objective (correction, immersion etc) for various types of microscopic investigations.
- select a suitable contrast method (phase contrast, DIC, fluorescence, darkfield etc) and correctly use this technique to obtain high-quality images.
- the expected image quality regarding resolution and signal-to-noise ratio for different practical imaging situations.
- understand and be able to describe the physical limitations for microscope performance concerning resolution and signal-to-noise ratio.
- describe performance for different types of microscopes by using (and in some simple cases calculating) optical transfer functions.
- select a suitable sampling density for digital image recording in microscopy.

- do computer processing of microscopic images to visualise three-dimensional structures.
- perform quantitative measurements in microscopic images using a computer.
- extract relevant information from a scientific publication and present this in the form of a seminar.

How to get relevant info

In 2020 the course is given almost completely online due to covid-19. Please make sure you get all the relevant info. You should do two things: first, read the info on the Canvas page. If you still have questions, please contact Anna or Ilaria, by mail or by Canvas message.

Second, there will be new info during the course: definitely reminders and clarifications, but sudden changes due to the covid-19 situation might also happen. This will always be communicated via **Announcements** in Canvas. To make sure you get the info, you should do one of the following:

Option 1. **Adjust your Canvas notification settings** so that you get an e-mail every time we make an announcement. In the gray menu on the left, choose Account and then Notifications. In the list that appears, make sure there is a green check mark by Announcements.

Option 2. Check Canvas announcements every weekday.

Lectures

Due to covid-19, lectures will take place entirely on-line. Each lecture will be split into two 45-min sections.








The first part is pre-recorded. Material for each lecture is found under the module Lectures. The actual viewing time for this material should be around 45 minutes, but might take longer if you follow it thoroughly. An advantage is you can pause to think or take notes, and go through some parts several times if necessary. The pre-recorded part **can be viewed anytime, but must be done before the second part of the lecture starts**. In this part, the basic lecture material is covered.








The second part of the lecture is a real-time session in zoom. Here we use the theory presented in the pre-recorded part to solve problems, do anonymous quizzes and have discussions. We might also go deeper into some subjects covered by the recorded material. This part always starts one hour into the scheduled time. So if the lecture is scheduled 13-15, the zoom session starts at 14.00. The duration is roughly 45 minutes. Link to zoom meeting: <https://kth-se.zoom.us/j/65167287284>.



If you find the second half difficult, go back to the pre-recorded material afterwards and try to make sense of it.

If you prefer, you may read the corresponding material rather than view the videos. In the table below you can find reading instructions plus the slides used for some of the recordings.

Please note this is a new format for all us. If something isn't working or could be done in a better way, please let us know. Preferably early on, when we can still do something about it! E-mail or Canvas message to Anna or Ilaria is the easiest way.

Lecture	Contents	Compendium	Slides
	Basics of light microscopy		
1	Imaging ray-path and illumination ray-path Aberrations, objective types, magnification, numerical aperture (AB)	<i>LM:</i> sect. 1.1 – 1.2, App. I	Lect. 1 
2	Contrast techniques: Absorption, fluorescence, phase contrast, DIC, dark-field (IT)	<i>LM:</i> sect. 1.5	Lect. 2 
3	Fundamentals of radiometry and photometry, microscope photometry, detectors, noise (AB)	<i>LM:</i> sect. 1.6, App. IV <i>IP:</i> p. 5-16	Lect. 3 
4	Resolution, mathematical representation of the imaging process (AB)	<i>IP:</i> p. 17-23 <i>LM:</i> p. 12-14	Lect. 4 
5	The Fourier transform and its interpretation, the optical transfer function OTF (AB)	<i>IP:</i> p. 23-29	Lect. 5 
6	OTF for diffraction-limited optics, 2-dimensional Fourier transforms, 2D OTF, OTF for detectors (AB)	<i>IP:</i> p. 29-47 <i>LM:</i> sect. 1.3	Lect. 6 
7	OTF for an imaging chain, sampling and aliasing, reconstruction calculations, multidimensional sampling (AB)	<i>IP:</i> p. 47-64	Lect. 7 




8	Coherent imaging in microscopy, role of condenser numerical aperture (AB)	LM: sect. 1.4	Lect. 8 
9	Introduction to confocal microscopy, (IT)	LM: sect. 2.1	Lect. 9 
10	Imaging properties of confocal microscopy (IT)	LM: sect. 2.2	Lect. 10 
11	Confocal microscopy: Limitations and errors, multi-channel detection (IT)	LM: sect. 2.3-2.4	Lect. 11 
12	Super resolution fluorescence microscopy, Stimulated emission depletion microscopy (IT)	LM: sect. 3	Lect. 12 
13	Super resolution fluorescence microscopy, Single Molecules based microscopy (IT)	LM: sect. 3	Lect. 13 
14	Problem solving (IT/AB)		Lect. 14 

Literature references: LM = Light Microscopy compendium , IP = Imaging Physics compendium 

Lecturers: AB = Anna Burvall, IT = Ilaria Testa

Labs

Each student will perform four 4-hour laboratory exercises. The contents are given in the table below. A suitable schedule will be worked out at the start of the course.

Lab #	Contents	Instructions
1	Build your own microscope with Koehler illumination	TM 
2	Practical use of research microscopes in different imaging modes	PLM 
3	Confocal microscopy #1	CM 
4	Confocal microscopy #2	CM

Due to covid-19, there will be restrictions on how the labs are carried out. Labs 1 and 2 will be given in the normal fashion, as there are only 2 students attending each lab. Lab 3 will be shortened to 2 hours, to allow for multiple sessions and hence a maximum of 2 students per session. Lab 4 will be given on-line.

Course laboratory: Roslagstullsbacken 21, level 2, see map at the bottom of the page.

Sign-up for labs will start at 13.00 on Friday November 6th.

Please note that the lab sessions start at 8:00 and 13:00, not quarter past like the lectures!

A report on labs 3 and 4 should be handed in (see "Assignments" in the white side bar) by Jan 12th.

Read the introductory parts and literature references (if any) before the lab session. In some cases there are problems that you should look at before the lab session.

Exam - corona special

Due to covid-19 the exam will be done on-line. It will be split into two parts:

- An oral exam via zoom which gives the grades of Pass (E) or Fail (F)
- If you pass the oral, a written home exam to determine your grade A-E.

The oral will be around 30 minutes and concern basic parts of the course. The home exam will be similar to a normal exam so you can still practice using the same material, like old exams.

During the written exam you may use all aids, except any kind of contact with other people.

All of it can't take place during the scheduled hours 14-19 on Jan 11th. Most likely, the home exam will be given within those hours but the orals take place on a different day. You will have the chance to give feedback on suitable dates before the schedule is finalized.

The exam instructions will be on a separate Canvas page which will become available closer to the exam date. Further instructions and material, like examples of questions for the orals, will be on this page.

Exam

You need to sign up for the exam, see info from the science school. Here you can also find general rules.

Written examination (TEN1; 4 credits). 5 hours, 6 problems. Max. 10 points/problem.

Grading:

53-60p = A

45-52p = B

37-44p = C

30-36p = D

25-29p = Fx

< 25p = F.

Students who receive grade Fx can, after completing a supplementary examination, receive grade E (pass).

Times and locations:

- Wed. Jan. 14, 2020, 14-19, FB55

The exam sets the grade on the course. Other tasks such as labs must also be complete before the entire course is reported into ladok.

Old exams are available.

Aids allowed at the written examination:

- Compendium “Imaging Physics,” will be handed out. (Chap. 19, Problems and solutions, and appendices 1-5 and 7 are not included. Appendices 6 and 8 are included.)
- Compendium “Light Microscopy” (will be handed out).
- Extracts from Mathematics Handbook Beta (chapter 13, Fourier series & transforms; will be handed out).
- Pocket calculator.
- Normal aids such as pens/pencils, eraser, ruler, food.

No “pure” math problems will be given at the written examination, but the student may be required to calculate simple Fourier transforms in order to solve problems dealing with imaging physics.

Bringing something to eat and drink is strongly recommended, as the exam lasts for 5 hours.

Seminar on SK2501

Please observe the info on this page applies only to the students taking the extended microscopy course, SK2501 (or FSK3501). The course is identical to SK2500, Physics of Biomedical Microscopy, except for the seminar presentation (1.5 credits. Grade scale P/F).

Time and location of the seminar will be agreed on once the course has started. In 2020 the seminar will be given over zoom.