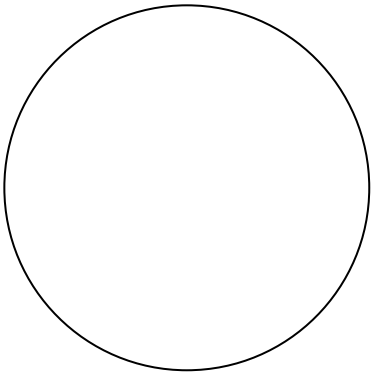
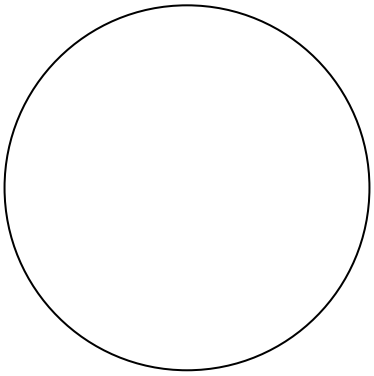
Practical course 1. Microscopy

Name and surname _____

Class _____ Date _____

Exercise 1. Prepare a part of plant tissue, for example a part of the leaf of *Elodea canadensis* by putting it in a drop of water placed on the microscope slide. Cover it with a coverslip.





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objective 10x, ocular 10x objective 40x, ocular 10x

Draw a picture of the leaf part using different (10x and 40x) objective magnification and explain whether you see the same or different part of the leaf.

Exercise 2. Prepare the starch grains isolated from potato in a drop of water placed on microscope slide. On one side of the coverslip gently add a drop of Lugol solution. On the opposite side of the coverslip place a filter paper to enable a Lugol solution to dye the starch grains.

Starch grains in water

Iris a) completely open b) optimum c) completely closed

Starch grains in Lugol solution

Iris a) completely open b) optimum c) completely closed

Explain the differences in starch grains structure in relation to dye solution and their role of Iris diaphragm!

Exercise 3. Draw a picture of onion (*Allium cepa*) cell. Which parts of the plant cell are easily recognized?

Answer to the questions:

HOMEWORK 1. Microscopy

Name and surname _____

Class _____ Date _____

Practical course 2: Cellular organization

Name and surname _____

Class _____ Date _____

Exercise 1. Bacteria

Exercise 2a. Yeast

Exercise 2b. Green algae *Chlorella* sp.

Exercise 2c. Leaf epidermal cell of *Rhoeo discolor*.

Exercise 2d. Epithelial cell from buccal cavity

Answers to the questions:

HOMEWORK 2: Cellular organization

Name and surname _____

Class _____ Date _____

Practical course 3. Biomembranes – indirect visualization

Name and surname _____

Class _____ Date _____

Table1. Preparation of serial dilution of sucrose solution:

Sucrose concentration (M)	0,0	0,2	0,4	0,6	0,8	1,0
Volume of sucrose stock solution (ml)	0					
Volume of water (ml)	5					
Total volume (ml)	5	5	5	5	5	5

Plant material:

Exercise 1.

Plant cell in dH_2O Plant cell in ___ M sucrose solution Plant cell ____ M sucrose solution

Exercise 2.

Solution number	Approx. sucrose concentration

Give a short explanation of how you approximately estimated sucrose concentration!

Answers to the questions:

HOMEWORK 3. Biomembranes – indirect visualization

Name and surname _____

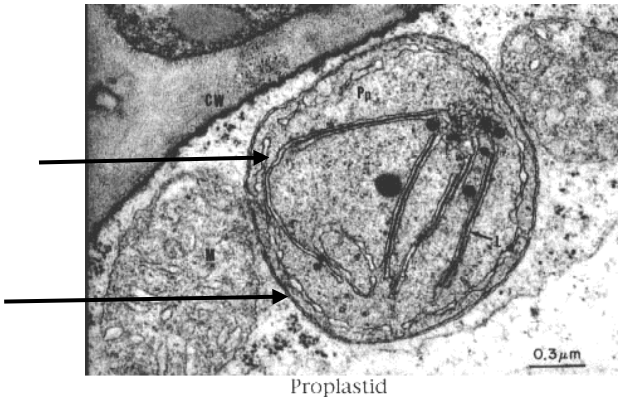
Class _____ Date _____

Practical course 4. Plastids

Name and surname _____

Class _____ Date _____

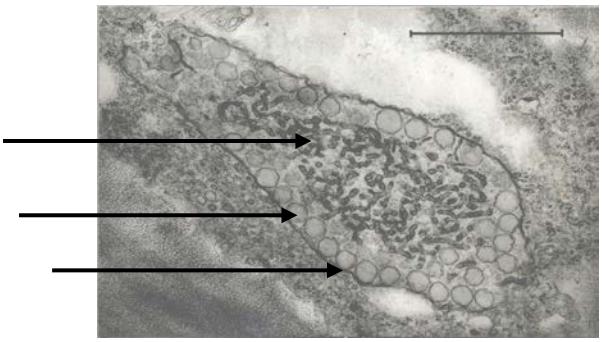
Exercise 1. Draw a picture of vegetation bud and pay attention to the meristematic cells in which PROPLASTIDS can be seen. Compare the structure as seen with light microscope as well as electron microscope. Mark all details of proplastid ultrastructure!



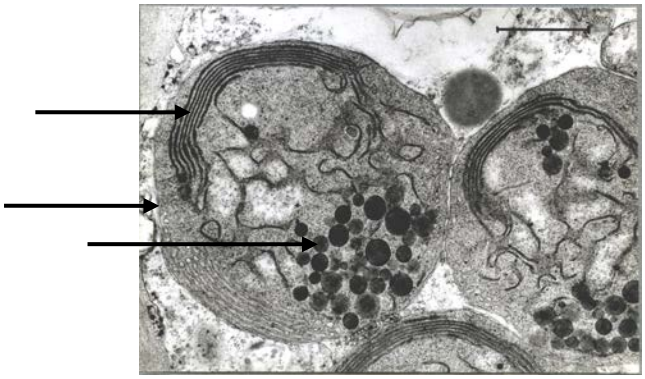
Proplastid

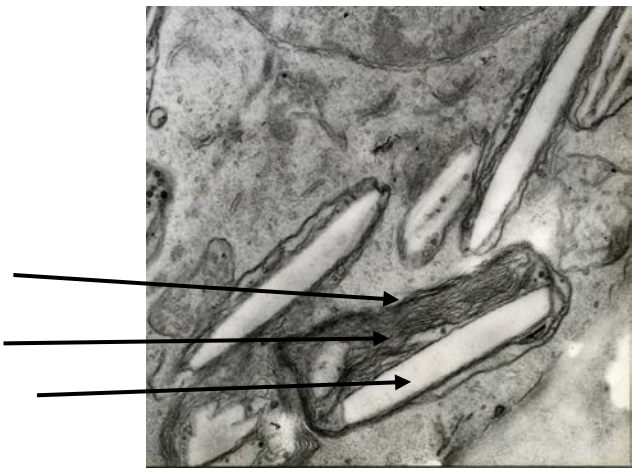
Exercise 2. Draw the epidermal cells which surround the stomata in plant leaf. Define the type of plastids in epidermal cells as well as in guard cell and neighboring cells. What types of plastids are present in parenchyma cells? Draw a part of chloroplast with detailed ultrastructure as seen with electron microscope!

Exercise 3. Define structures in plant cells responsible for different colors of plant petals. Draw plant cells with typical plastids. Mark all details of plastids ultrastructure!



Exercise 4. Chromoplasts are plastids that can be grouped by their shapes in chrysaloid or globular type of plastids, as seen in the pictures bellows (mark all the details of ultrastructure!).
What types of plastids can be found in carrots and pepper cells?





Answer the questions:

1. What type of plastids are present in meristematic cells of vegetation buds?
2. What type of plastids can be found in a) epidermal cells, b)guard cells, c) neighboring cells and d) parenchyma cells of plan leaf?
3. What is the reason that petals can be of different color? What is the reason that the peper tissue is of red colors?
4. What type of plastids are present in carrots tissue?

HOMEWORK 4. Plastids

Name and surname _____

Class _____ Date _____

Practical course 5. Nucleus - Mitosis

Name and surname _____

Class _____ Date _____

Mitosis in root tips of _____.
2n = ____

Exercise 1. Shematic representation of slide preparation.

Exercise 2. Fill in the table!

Phases	**I**	**P**	**PM**	**M**	**A**	**T**	**total number**
Number of cells							

Exercise 3. Calculate the mitotic index.

Mitotic index 1 =

Exercise 4. Draw a cell nucleus in metaphase and anaphase!

Metaphase

Anaphase

Answers to the question:

HOMEWORK 5. Nucleus - Mitosis

Name and surname _____

Class _____ Date _____

Practical course 6. Endomitosis. Polytene chromosomes and C-mitosis

Name and surname _____

Class _____ Date _____

Exercise 1. Schematic representation of polythene chromosome preparation.

Exercise 2. Draw a picture of giant chromosomes isolated from_____

_____.

10x magnification 40x magnification

Exercise 3. Schematic representation of slide preparation for C-mitosis.

Exercise 4. Draw a picture of metaphase plate in *Allium cepa* roots pretreated with colchicine.

HOMEWORK 6. Endomitosis. Polytene chromosomes and C-mitosis

Name and surname _____

Class _____ Date _____

Practical course 7. Meiosis

Name and surname _____

Class _____ Date _____

Exercise 1. Draw specified phases of meiosis as seen on the microscopic image which gives immersion objective. Mark the drawn structures!

Exercise 2. Schematic representation of slide preparation for meiosis.

Exercise 3. Answers to the questions.

HOMEWORK 7. Meiosis

Name and surname _____

Class _____ Date _____

Practical course 8. Isolation of nuclei and DNA fibres

Name and surname _____

Class _____ Date _____

Exercise 1. Schematic representation of the procedure for nuclei isolation.

Exercise 2. Draw the isolated nuclei stained with aceto-carmine! (objective magnification of 40x)

HOMEWORK 8. Isolation of nuclei and DNA fibres

Name and surname _____

Class _____ Date _____

Practical course 9. Isolation of genomic DNA from plant material by using the “home-made ” method

Name and surname _____

Class _____ Date _____

Exercise 1. List all the steps in the isolation of DNA and draw the final result (mark the phases that can be seen in a glass tube after adding alcohol and in which phase the precipitation of DNA occurs).

Exercise 2. Try to explain the role of detergent and table salt in the process of isolation (guidelines for the sub questions: how does table salt dissociate in water, charge of DNA molecule, impact on precipitation).

Exercise 3. Show how you have prepared "extraction solution." Do you think that the experiment would be successful if you used a little more or less detergent and/or salt?

HOMEWORK 9. Isolation of genomic DNA from plant material by using the “home-made ” method

Name and surname _____

Class _____ Date _____

Practical course 10. Isolation of genomic DNA from transgenic plant *Arabidopsis thaliana* and plasmid DNA from *Escherichia coli* laboratory strain

Name and surname _____

Class _____ Date _____

Exercise 1. Draw bacterial cell with incorporated transgene in plasmid DNA. Draw a plant cell with a transgene incorporated in genomic DNA (2n=4).

Exercise 2. Which step in DNA isolation is crucial for lysis of bacterial cell and which one for rupturing the plant cell?

Exercise 3. List the main steps in plasmid isolation! Which step is essential for separating plasmids DNA from genomic DNA!

HOMEWORK 10. Isolation of genomic DNA from transgenic plant *Arabidopsis thaliana* and plasmid DNA from *Escherichia coli* laboratory strain

Name and surname _____

Class _____ Date _____

Practical course 11. PCR, electrophoresis and DNA restriction analysis

Name and surname _____

Class _____ Date _____

Exercise 1. Let's assume that you have two DNA samples. One sample consists of linear DNA and the other one has circular DNA. Both DNA samples have 17 restriction sites recognized by EcoRI. How many DNA fragments will be generated after incubation with enzyme EcoRI and electrophoresis in both cases?

Exercise 2. After 6 cycles of PCR reaction you have 256 DNA molecules. If we assume that PCR reaction is 100% accomplished, how many DNA molecules do you have after 3rd cycle and how many after 9 rounds of PCR?

Exercise 3. Draw a gel in which 3 fragments of different size 150, 250 and 1000 bp (bp=base pairs) are running during electrophoresis. Draw a marker on the left side of the gel which consists of 10 fragments in range from 100 bp to do 1000 bp (difference in size of fragments should be 100 bp)!

HOMEWORK 11. PCR, electrophoresis and DNA restriction analysis

Name and surname _____

Class _____ Date _____