

DNA EXTRACTION

BACKGROUND

The genomes of all living organisms consist of DNA (deoxyribonucleic acid). There are two interconnected strands in the structure of DNA (**double helix**). The strands of a DNA molecule contain nucleotides which have three essential parts: a **sugar**, a **phosphate** and a **base**. There are four different bases: adenine (A), guanine (G), cytosine (C) and thymine (T). The essential information in the genome of the cell is coded by those four bases.

Prokaryotes (such as bacteria) have their DNA in the cytosol (cytoplasm). Eukaryotes (such as animals, plants and fungi) store most of their DNA in a structure called nucleus. There are some DNA in mitochondria and chloroplasts as well.

In the cell, DNA associates with some proteins, and together they form

chromosomes. For instance, there are 46 chromosomes in human cells. A **gene** is a section of DNA that guides the functions of the cell. Most genes contain information about proteins and the information is read and transcribed in a process called **protein synthesis**: The information coded in DNA is transcribed into **messenger RNA**, which delivers the message to the ribosomes, where proteins are produced based on the information of messenger RNA. Proteins are used for e.g. metabolic reactions, transfer, cell movements, cell signalling and reproduction.

In this practical task you will extract DNA from animal cells. You can use liver cells (bovine or porcine) and cells from your cheek epithelium.

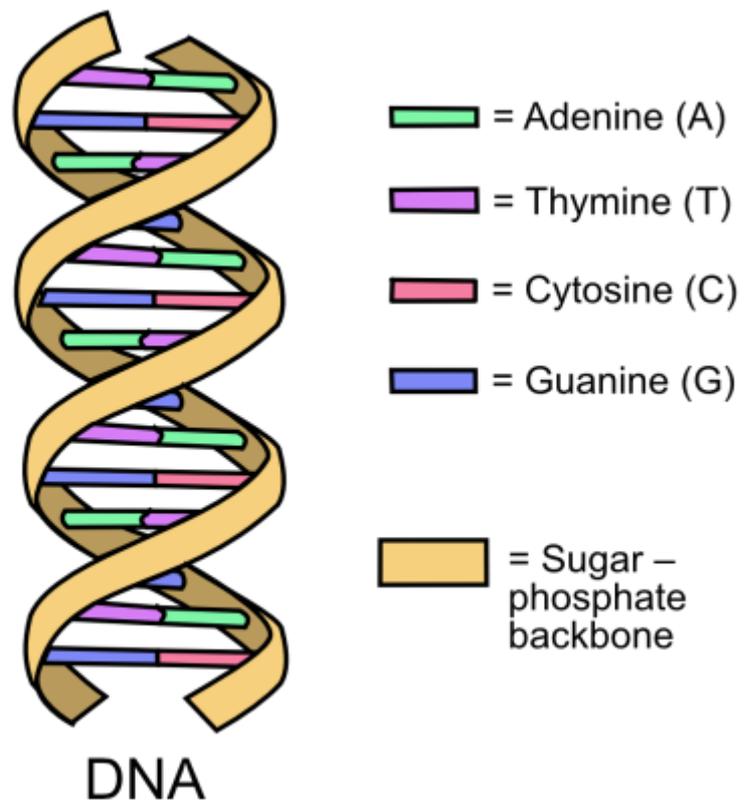


Figure 1. The structure of DNA double helix.

QUESTIONS BEFORE THE TASK

- What parts of a cell contain DNA?
- What structures you should break to release DNA from the cell?
- What is the role of DNA in cells?
- How DNA is packed into chromosomes?

EQUIPMENT

- Pasteur-pipettes
- Filter paper
- Funnels
- Ice cold alcohol (e.g. Sinol)
- Microtubes
- Test tubes
- Soap or other detergent
- Cotton swabs
- Tube holder
- Samples:
 - Liver tissue
 - Your cheek epithelium cells

INSTRUCTIONS

NB! Put the alcohol (ethanol or isopropanol) into freezer at least 15 minutes before starting!

<u>Liver cells</u>	<u>Cheek epithelium cells</u>
1. Take a small piece of tissue and put it into a microtube.	1. Collect the cells from your inner cheek epithelium by rubbing it with a cotton swab. Remember to rub very thoroughly!
2. Add some water and soap. Use e.g. swabs to grind the sample.	2. Put some water and soap in a microtube. Rinse the cotton swab in the microtube to transfer the cells into the solution.

3. Incubate (let stay) the mixture for 5–15 minutes. Meanwhile, prepare the filtering equipment by putting the filter paper in a funnel. Place the funnel in a test tube. You can also answer the questions while waiting.
4. Pour the mixture in the funnel. Let it stay until you have at least 1 cm of filtrate in your test tube. If you don't have enough filtrate, you can add water to the filter.
5. Add ice cold alcohol into the test tube with a Pasteur pipette. Add alcohol by tilting the test tube and pour the liquid slowly by the edge of the tube.

DNA (and proteins that attach to DNA) can be seen in the interface of solution and alcohol. You can try to remove the DNA by using a toothpick.

QUESTIONS AFTER THE TASK

- Why did DNA become visible when you added alcohol?
- Why did you add soap to the mixture?
- There are some other nucleic acids in the cell (such as RNA), which precipitated with DNA in this task. If you would like to remove RNA, what would you do?
- There were also many proteins that attached to DNA and precipitated with it. How could you remove those proteins?

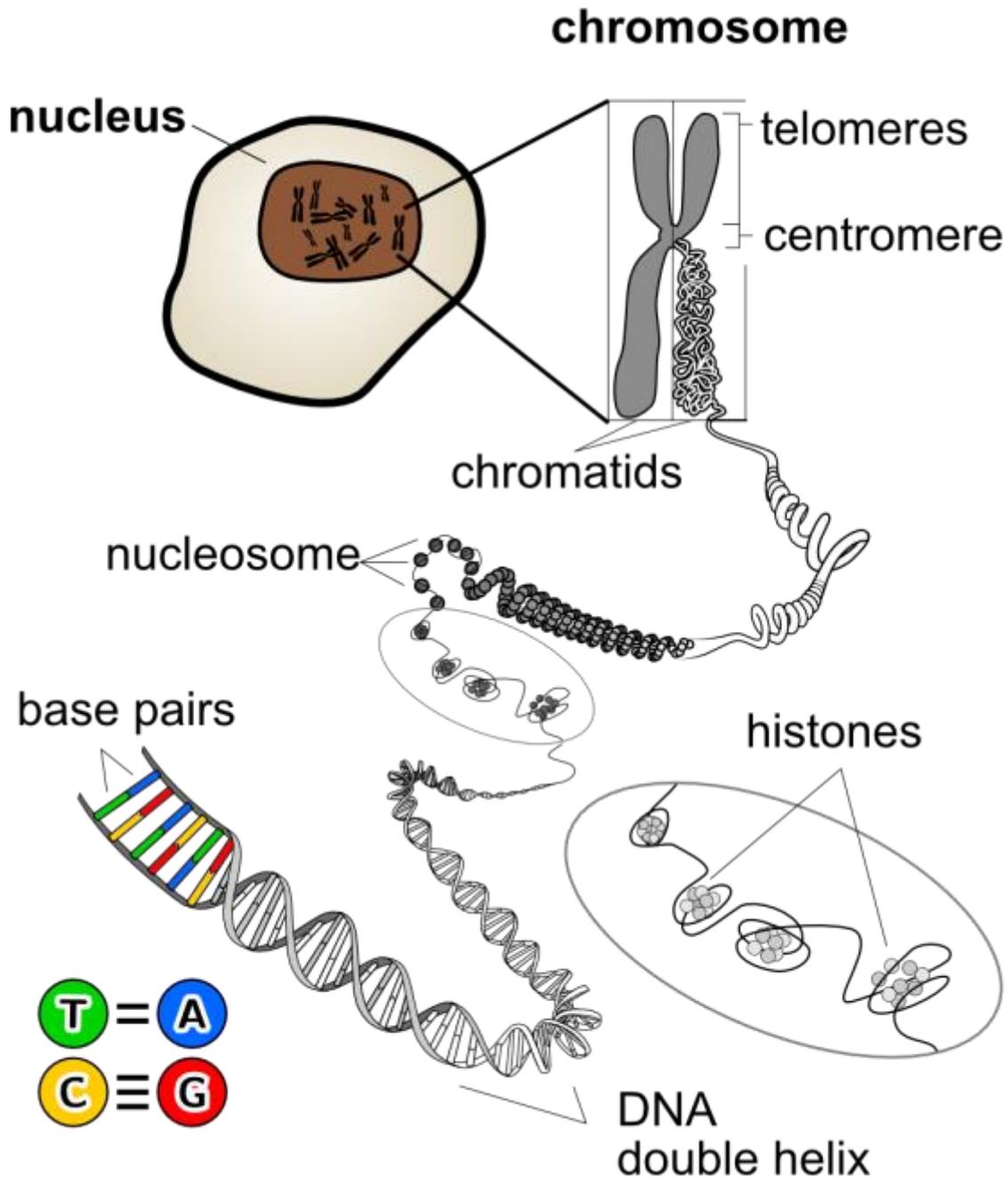


Figure 2. The structure of a chromosome and DNA.