MEDICAL UNIVERSITY – SOFIA

FACULTY OF DENTAL MEDICINE

CURRICULUM

Name of the discipline: BIOPHYSICS

Educational degree: Master

Type of the discipline: Obligatory

Course duration: One semester

Course degree: M degree (Master's)

Evaluation methods: During the practical exercises the students are subjected to **continuous assessment;** at the end of the semester **an exam on the practical exercises** is carried out in form of **a test; the final exam** on the whole course (covering the learning material from the lectures and practicum) contains written answers and discussion on them.

Training methods used during the course: Lectures – 15 hours, **Practical (laboratory) exercises** – 15 hours; before coming to the laboratory exercises the students **have to prepare themselves on a given topic** (list of several keywords) from recommended reference sources (textbook, laboratory guide, new written texts); during the practicum the basic training form is **the discussion**; after finishing the laboratory practice the students have to write **an individual standardized protocol** concerning the experimental work, the obtained results and their discussion – **working out this protocol** is important part of the training.

Final exam at the end of semester: YES

Leading instructor: Associated professor Trayko Traykov

Home department: Department of medical physics and biophysics

ANNOTATION OF COURSE

Subject of biophysics are the physical principles, on which all processes in the living systems are based. It is a precise quantitative interdisciplinary science between physics and biology. It is experimental and theoretical discipline closely related to mathematics, biochemistry and physiology. Biophysics is also connected with many areas of physics, chemistry and physical chemistry. Learning biophysics formats a problem based scientific thinking of the students in medicine, due to the needs of using knowledge of variety of scientific disciplines for clarifying the working mechanisms in biological matter at all levels of organization.

The teaching in BIOPHYSICS is worked out mainly on the parts "Molecular Biophysics" and "Cellular Biophysics" and includes specific knowledge important for the medical education. The theoretical part of the course involves the principles of the biothermodynamics and the quantitative description of transport of substances and energy, as well as energetically and functionally coupling of processes in the biological systems. In the chapter "Biological and artificial membranes" the physical laws concerning the self-organization of biomolecular structures are explained. The interrelationship between biochemical properties of molecules, physical characteristics and biological functions is elucidated. Questions directly related to medical practice and therapy are discussed in the chapters "Transport through biological membranes" and "Passive electrical properties of cells and tissues". In the chapter "Free radical damage of membranes and cells" some specific molecular mechanisms of free radical reactions caused by ionizing radiation are presented. Some formal theories for optimization of radiation therapy are offered. The importance of free radicals in the pathogenesis of several diseases is also discussed. The students learn also about some cellular biotechnological application based on modern biophysical methods – tests for vitality, biophysical basis of electroacupuncture, dielectrophoresis, electroporation, electrofusion of cells, some separation technologies and electromanipulation of cells.

Formation of the overall mark for the course:

- **Final examination** – the mark for written answers on two theoretical questions from the lecture course material and discussion on them; eventually additional questions – 75 %
- **Final mark for the practicum** (25%) – formed on the basis of:
    - **Average grade** from the **continuous assessment**
    - **The mark** from the **final test** on the practicum

Aspects of the student's evaluation:

During the practical exercises:

- Assessment of the students ability to obtain knowledge working individually with the reference sources
- Evaluation of the students participation in the discussion on the keywords given
- Assessment of the students experimental work in the laboratory
- Evaluation of the students skills to process statistically the obtained results (by using computer programs and to represent them in tables, graph and to discuss them in the context of the aim of the study

During the final examination:

- Assessment of the ability of the students to give written answers on the given questions
- Evaluation of the students skill to react on additional questions concerning his written answers and on the eventually formulated additional questions
- Assessment of the students competence to interrelate the theoretical knowledge with the practical problems investigated during the laboratory practice

RESOURCES USED DURING THE COURSE:

Lectures: - computer based multimedia presentation

Laboratory practice: experimental set-ups, chemicals, computers

Course objectives:

The students should gain knowledge and skills for introduction of biophysical methods (theoretical, experimental, and instrumental) in the organization of dental medicine care in accordance with the modern European standards in Health service.

Course goals:

1. Students have to gain the basic knowledge from the Lecture course (see the THEMATIC CALENDAR SCHEDULE of the LECTURES)
2. Students should learn the experimental methods introduced in the laboratory exercises (see THEMATIC CALENDAR SCHEDULE of the LABORATORY EXERCISES)
3. Students have to learn to process statistically (calculate mean values and standard deviations, do linear and non-linear regressions using statistical programs – e.g. Excel, Prism) the obtained experimental results and to represent them in tables, graph and to discuss them in the context of the aim of the study
4. After finishing the laboratory practice the students have to be able to write **an individual standardized protocol** concerning the experimental work, the obtained results and their discussion

Requirements:

Successful finished courses in: chemistry, physics, biology and high school mathematics

Learning outcomes:

From the lecture course the students have to obtain the basic biophysical knowledge

In the practice the students learn:

- to formulate concrete aims and problems for experimental study;
- based on the knowledge offered in these materials, the students must be able to propose and discuss ideas about how the aims of the experiments have to be realized;
- to process statistically the obtained experimental data and represent them in tables and graphs;
- to comment the importance and application of the obtained information about the particular object investigated in biophysical, general biological and medical context;
- to document precisely the discussion, the experimental work and results in an individual protocol, which is close in form and structure to a scientific paper.

The practical skills offered to the students aim to familiarize them with basic knowledge about methods used in research and clinical laboratories. They learn:

- to format a monomolecular layer of amphiphylic substance and determine its molecular dimensions;
- to perform drug ionophoresis through the human skin and estimate the amount of the substance brought into it;
- to acquire a method for investigation of the viscoelastic properties of human skin;
- to estimate the acidic resistance and age distribution of erythrocytes from different blood samples;
- to master the method of cellular electrophoresis and the estimation of the surface electric charge density of cells;
- to use an approach for measuring electric potential differences in model systems;
- to measure the volume fraction of erythrocyte (hematocrit value) and mean cellular volume of those cells;
- to determine the coefficient of permeability for given substance of an artificial membrane used fore hemodialysis.

Literature for preparation:

1. Lecture notes – offered on the web too;
2. Materials for every experimental practice – given as a text
3. Glaser, R., Biophysics, 2001, 361 pp. 162 figs
4. Weiss, T. F., Cellular Biophysics - Two Volume Set, 1996, 1318 pp., 758 illus.

Topics for individual preparation: see the list of the laboratory exercises and the corresponding keywords

Literature for preparation:

1. Lecture notes – offered on the web too;
2. Materials for every experimental practice – given as a printed text
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THEMATIC CALENDAR SCHEDULE

of the LECTURES in BIOPHYSICS

for dental medicine students

during the third (winter) semester of the second academic year

Lectures - 15 hours,
Course Duration: one semester, the third semester (Even weeks - 2 hours, odd weeks - no lectures)

Week No.	Subject	Hours number
2-nd	The subject of biophysics. Divisions and methods of biophysics. Biophysics and Medical Sciences. **I. Biomechanics and biothermodynamics** 1. Subject and importance of biomechanics. Rheology of the solid biomaterials. Viscoelastic properties of bones and muscles. Biomechanics of the chewing apparatus. 2. Passive mechanical properties of blood vessels – Lame's equation, volume-pressure dependencies. Pulse wave. Mechanics of breathing. The role of Laplace pressure and the surfactant. 3. Biothermodynamics: basic concepts. First law of thermodynamics, internal energy. Second law of thermodynamics, entropy.	**2 hours**
4-th	4. Energy sources, types of work and heath in the living systems. Electrochemical potential, free energy and direction of processes. Steady state and thermodynamic equilibrium. **II. Biological and artificial membranes** 5. Subject and importance of the biological membranology, types of membranes. Functions and basic chemical composition of the biomembranes. Membrane phospholipids, glycolipids and cholesterol. 6. Membrane proteins and glycoproteins - functions, localization and binding. Membrane models and methods for investigation of biomembranes. 7. Lipid associates and model artificial membranes - types, preparation methods, properties and significance. Liposomes as drug carriers. 8. Mobility of the membrane components - lateral and transversal diffusion. Liquid-crystalline state of the lipid bilayer. Molecular mechanism of the phase transitions..	**2 hours**
	9. Free radicals - generation in biological systems. Types of free radicals, basic reactions and properties. Reactive oxygen species (ROS). Basic	**2 hours**

6-th	methods for registration and investigation of ROS. 10. Oxidation of the membrane components - consequences. Biophysical mechanisms of the oxidative stress caused by different factors. Antioxidant defense systems of an organism.. **III. Transport of substances through membranes** 11. Subject and importance of the membrane transport. Types of transport (classification). Free diffusion of non-charged particles - first and second Fick's laws. The Stokes-Einstein equation. 12. Free diffusion of ions – the Nernst-Plank equation, dimensionless potential. Non- free diffusion through a pored membrane.	
8-th	13. Osmosis and filtration. Dialysis and hemodialysis. Water exchange through membranes and between blood and tissues 14. Facilitated diffusion. Ionophores i) - mobile carriers (Valinomycin) and ii) - channel-forming carriers (Gramicidin A). Principles of action, molecular structure and selectivity. 15. Active transport. Sodium-potassium pump and calcium pump – importance, role of ATP, molecular structure and model of action.	**2 hours**
10-th	**IV. Electrical properties of cells and tissues** 16. Diffusion, equilibrium (Nernst) and Donnan potentials - conditions and mechanisms of generation, dependencies on concentration and time. 17. Origin of the resting potential - theories. Goldmann equation. The effect of electrogenic pump. Resting potential components.	**2 hours**
12-th	18. Action potential - origin, generation, propagation. The Hodgkin-Huxley's equations. Structure of the ionic channels. The effect of different toxins and drugs on their behavior. 19. Importance and origin of the surface electric charge of the cells – double electric layer and potential distribution. The Poisson-Boltzmann equation. Potential decay across the membrane and near its both sides.	**2 hours**
14-th	20. Electrophoretic mobility – dependence on pH and ionic strength (salt concentration) of the medium. Stability of the biosuspensions. Types of electrophoresis. 21. Electroconductivity of cells and tissues for DC (direct current). Types of polarization of dielectrics and heterogeneous systems. 22. Electroconductivity of cells and tissues for alternating current (AC). Impedance and dispersion of the dielectric permittivity. Physiological and pathological changes in the electrical conductivity of tissues. Biophysical basics of elecroacupuncture diagnostics and therapy.	**3 hours**

COURSE OUTLINE: The course duration is 30 academic hours divided as follows:

Lectures: 15 hours

THEMATIC CALENDAR SCHEDULE of the LABORATORY EXERCISES
for dental medicine students
during the third (winter) semester of the second academic year

LABORATORY PRATICE - 15 hours,
Course Duration: one semester, the third semester
(Even weeks - 2 hours, odd weeks - no practices)

	Topic
1	**Investigation on the mechanical properties of human skin in vivo – 2 hours** Keywords for preparation: passive mechanical properties of biological tissue – mechanical stress, strain, velocity gradient; types of mechanical properties – elastic, Hook's law; viscous – Newton's law or viscoelastic – equation for viscolelastic materials; mechanical models in the rheology – kinetic properties; methods for investigation of the mechanical properties of biological membranes Aim of the study: to determine the type of the mechanical properties of human skin – elastic, viscous or viscoelastic; to estimate the elastic modulus E (Young's modulus) of human skin
2	**The acid hemolysis of red blood cells (RBC) – a dynamic method for investigation of alterations in RBC resistance - 2 hours** Keywords for preparation: hemolysis – types, acidic resistance of RBC – time for 50% hemolysis ($\mathbf{t_{50\%}}$), differential erythrogram – RBC resistance distribution, differential erythrogram – and RBC age distribution, diagnostic importance Aim of the study: to compare the acidic resistance and age distribution of RBC from two different blood samples
3	**The influence of the osmotic pressure on the volume of erythrocytes - 2 hours** Keywords for preparation: osmotic pressure of solutions – equations for ideal and real solutions; osmosis and filtration; osmotic equilibrium at a biological membrane; dependence of red blood cell volume on the osmotic pressure of the suspending medium; apparent osmotically inert volume (V_n). Aim of the study: to determine the dependence between erythrocyte volume and osmotic pressure of the suspending medium; from the obtained relation to estimate the apparent osmotically inert volume (V_n).
4	**Determination of the size of amphipathic molecules by monolayer - 2 hours** Keywords for preparation: chemical composition of the biological membranes; molecular structure and physicochemical properties of the membrane phospholipids; Lipid associates and model artificial membranes – micelles, monolayers, bilayers, liposomes Aim of the study: To determine the length, average area per molecule and the diameter

	of the oleic acid molecule using monolayer.
5	**Assessment of the ionophoretic permeability of human skin - 2 hours** Keywords for preparation: electrophoresis types, drug electrophoresis; coefficient of permeability of human skin – importance, comparison of drug electrophoresis with other methods of drug application (for example – oral and venous); modern methods for drug delivery in the organism – liposomes as a drug carrier. Aim of the study: to determine the coefficient of permeability of human skin for Novocain ions
6	**Transport of substances through artificial membrane - 2 hours** Keywords for preparation: passive transport through membranes – free diffusion and diffusion through a membrane; concentration change with time – the halftime ($t_{0,5}$); dialysis and hemodialysis. Aim of the study: to determine the coefficient of permeability for given substance of an artificial membrane
7	**Estimation of the electrokinetic potential of cells. Dependence on pH and ionic strength - 2 hours** Keywords for preparation: surface electric charge of cells – contributions of the different membrane components; sialic acids; double electric layer – definition, schematic representation, thickness of the electric double layer; quantitative estimation of the surface electric charge density with cell electrophoresis; electrophoretic mobility and electrokinetic potential of cells – relation to surface electric charge; effects of the pH and ionic strength of the suspending medium. Aim of the study: to study the effect of ionic strength of the medium on the electrophoretic mobility and electrokinetic potential of erythrocytes
8	**Equilibrium and diffusion potential in model systems - 2 hours** Keywords for preparation: mechanisms of formation of a potential difference on a membrane – diffusion potential and Nernst equilibrium potential; dependence on the concentration gradient and time; determination of the ratio of ion mobilities in the model system. Aim of the study: to determine the dependence of the diffusion potential in a model system on the concentration gradient. From the obtained relationship to estimate the ratio of the ion mobilities in the model system; to follow up the time dependence of the diffusion potential in the model system
	Exam on the laboratory exercises – final test (60 min, the rest time – checking the results of the test and semester attestation) - the last week in the semester

Practical (laboratory) exercises: 15 hours – every practice lasts **2 hours** and contains seminar part ant experimental work

SYNOPSYS FOR THE FINAL EXAMINATION

1. Subject and importance of biomechanics. Rheology of the solid biomaterials. Viscoelastic properties of bones and muscles. Biomechanics of the chewing apparatus.

2. Passive mechanical properties of blood vessels – Lame's equation, volume-pressure dependencies. Pulse wave. Mechanics of breathing. The role of Laplace pressure and the surfactant.

3. Biothermodynamics: basic concepts. First law of thermodynamics, internal energy. Second law of thermodynamics, entropy.

4. Energy sources, types of work and heath in the living systems. Electrochemical potential, free energy and direction of processes. Steady state and thermodynamic equilibrium.

5. Subject and importance of the biological membranology, types of membranes. Functions and basic chemical composition of the biomembranes. Membrane phospholipids, glycolipids and cholesterol.

6. Membrane proteins and glycoproteins - functions, localization and binding. Membrane models and methods for investigation of biomembranes.

7. Lipid associates and model artificial membranes - types, preparation methods, properties and significance. Liposomes as drug carriers.

8. Mobility of the membrane components - lateral and transversal diffusion. Liquid-crystalline state of the lipid bilayer. Molecular mechanism of the phase transitions.

9. Free radicals - generation in biological systems. Types of free radicals, basic reactions and properties. Reactive oxygen species (ROS). Basic methods for registration and investigation of ROS.

10. Oxidation of the membrane components - consequences. Biophysical mechanisms of the oxidative stress caused by different factors. Antioxidant defense systems of an organism.

11. Subject and importance of the membrane transport. Types of transport (classification). Free diffusion of non-charged particles - first and second Fick's laws. The Stokes-Einstein equation.

12. Free diffusion of ions – the Nernst-Plank equation, dimensionless potential. Non- free diffusion through a pored membrane.

13. Osmosis and filtration. Dialysis and hemodialysis. Water exchange through membranes and between blood and tissues

14. Facilitated diffusion. Ionophores i) - mobile carriers (Valinomycin) and ii) - channel-forming carriers (Gramicidin A). Principles of action, molecular structure and selectivity.

15. Active transport. Sodium-potassium pump and calcium pump – importance, role of ATP, molecular structure and model of action.

16. Diffusion, equilibrium (Nernst) and Donnan potentials - conditions and mechanisms of generation, dependencies on concentration and time.

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19. Importance and origin of the surface electric charge of the cells – double electric layer and potential distribution. The Poisson-Boltzmann equation. Potential decay across the membrane and near its both sides.

20. Electrophoretic mobility – dependence on pH and ionic strength (salt concentration) of the medium. Stability of the biosuspensions. Types of electrophoresis.

21. Electroconductivity of cells and tissues for DC (direct current). Types of polarization of dielectrics and heterogeneous systems.

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Head of Department

Medical Physics and Biophysics:

Assoc.Prof. D. Mihov