Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

# THEMATIC PLAN

2018-2019 study year

N	Торіс	semester	Kinds and hours of auditory works	Control forms
1.	Research Methods in Histology	5	lecture (3) lab work (6)	oral poll (themes 1-4, 7, 11) written work (theme 9) abstract (theme 6) Scientific work report (theme 15)
2.	Research Methods in Genetics	5	lecture (2) lab work (2)	oral poll (theme 12, 14, 18, 26)
3.	Research Methods in Microbilogy	5	lecture (2) lab work (2)	oral poll (theme 16, 20, 27, 28)
4.	Research Methods in Biochemistry	5	lecture (2) lab work (2)	oral poll (theme 8, 10, 24, 25)
	Research Methods in Physiology	5	lecture (2) lab work (3)	oral poll (theme 17, 19, 21-23)
	QUIZ			

# Theme 1. The general representations about methods of scientific research. Use of laboratory animals in an experimental research.

### Laboratory research

Methods of scientific research. Modelling. Experiment. Specificity of experiment as scientific method. Experimental bunches. The kinds of animals used in experimental researches. Features backboneless and backboned (on an example of gnawers) as objects of an experimental research. Kinds of lines of gnawers: inbred, the F1-hybrids, the segregated lines, coisogenic strains, transgene lines, recombinant lines, Heinbred, casually-inbred, outbred lines. The nomenclature inbred and special genetical lines. The basic pure lines of gnawers. Categories of laboratory animals according to demanded conditions of the maintenance and use purposes in biomedical researches. Ethical aspects of use of laboratory animals as objects in biomedical researches. The foreign and domestic legislation regulating use of laboratory animals in biomedical researches. Rules of the maintenance, a food, care of laboratory animals (on an example of gnawers). Bases of surgical interventions on laboratory animals. Anaesthesia, analgesy, aseptics, antiseptics, sterilisation, disinfection. A narcosis, stages. Means and drugs for introduction in a narcosis of laboratory animals. Signs of a penetrating narcosis. A conclusion from a narcosis. Points of the terminal of experiment with use of laboratory animals. Means of a blood sampling at gnawers (from aural, tail veins, ablation of a tip

Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

THEMATIC PLAN

2018-2019 study year

of a tail, from a venous sine of an eye). Means of introduction of materials (per os, rectal, by means of a probe, intramuscularly, intravenously, hypodermically, intradermally, intraperitoneally, retrobulbar).

#### Theme 2. Morphological researches for clinical diagnostics. An obduction.

#### Lecture

The general concept about pathoanatomical service. A subject and problems of pathological anatomy. Methods pathomorphologic researches. A stuff for pathomorphologic researches. An obduction, the carrying out purpose, order and carrying out rules. Technical variants of dissecting. The list of the recommended volume of histological research of a section stuff. Dissecting on G.V.Shor's method.

# Theme 3. Preparation of material for morphological research: bracing, processing and paraffin embedding, decalcification

#### Laboratory research

Deducing of laboratory animals from experiment: the causes, principles, means. Deducing from experiment by a path decapitation, overdoses by a narcosis. Cardiac perfusion: order of carrying out. A cutting of a stuff for histological research in an experimental research. Bracing: the purposes, kinds (thermal, chemical). A choice of a method of bracing. The general rules of bracing of a stuff. The basic fixativing fluids. Bracing in formalin: mechanisms, merits and demerits. Emergency bracing. Conducting of a stuff for paraffin embedding: dehydration, a clarification, inspissation. Paraffin embedding. The most widespread errors at bracing in formalin, conducting, paraffin embedding. Decalcification: acid and acidless. Probes preparation a stuff for electronic microscopy. Criteria of qualitative carrying out decalcification.

### Theme 4. Molecular-genetic methods in clinical practice.

### Lecture

The basic concept of molecular biology. A place of molecular-genetic diagnostics in modern clinical practice. Caryotyping: definition, the purposes, order of procedure, kinds of colouring of chromosomes. In situ hybridization: definition, the purposes, order of procedure, application. Fluorescent and chromogenic in situ hybridization. A polymerase chain reaction: definition, a principle of a method, modification of the PCR-analysis and their application in clinical practice. Sequenation: a method principle, application in clinical practice. Sequenation of new generation, sequenation on Senger. Micro-chiping, a method principle, application in clinical practice, classification of types of a method.

### Theme 5. Microsection

### Laboratory research

The basic phylums (lounger, rotational) and the device of modern section cutters. The device cryotome. Phylums of microlanguid edges. An operating sequence sectioning. Artefacts and the basic errors at sectioning and means of their elimination. Advantages and use lacks cryosections and sections from paraffin blocks.

# Theme 6. Rules of work with biopsy a stuff. Rules of appropriate clinical practice.

### Lecture

Biopsy: definition. Kinds of biopsies. Rules of a cutting of a stuff for histological research in clinical practice. Marking rules biopsy a stuff in clinic. Urgent biopsies. The standard of

Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

# THEMATIC PLAN

2018-2019 study year

Appropriate clinical practice (GCP). Phases and problems of clinical research. The person as object of clinical research. Principles of the Helsinki declaration (1984).

#### Theme 7. Histological and histochemical colourings

#### Laboratory research

Dewaxing, rehydratation paraffin sections: order and characteristic errors. Theoretical bases of histological colouring. Classification of histological dyes. Principles of a constitution of histological dyes. Classification of means of colouring. Colouring hematoxyline and eosine: the order, expected result, characteristic errors. Trihromnye colouring: colouring on Van-Gizon, on Massonu, on Mallori. Histochemistry: a principle, the basic conditions, features probes preparation. The frames revealed with the help histochemical of colouring. The basic histochemical the reactions used in clinical and laboratory practice (colouring by sudan IV, a Feulgen reaction, the PAS-reaction). Ferment histochemistry: a principle, examples. The conclusion under an integumentary glass, order and characteristic errors. Mounting mediums, kinds and their characteristics.

# Theme 8. Research methods in biological chemistry. ELISA. Immunoblotting.

#### Lecture

The immunoenzyme analysis. Immunoblotting (western blot). A chromatography. Chromatography kinds. Spectroscopy and spectrometry. Mass spectrometry. Application in biology and medicine. Microscopy kinds: optical, fluorescent, x-ray, electronic, scanning array.

# Theme 9. Carrying out bases immunohistochemical and immunofluorescent reactions and means detection their products

#### Laboratory research

Immunohistologic reactions: definition, principles. An antigen, types from antigens from the point of view of carrying out immunohistochemical researches. Antibodies: definition, types of antibodies and their frame. Classification of the antibodies used in histological practice: on a reception source, on clonality. Means of reception of antibodies for carrying out immunohistochemical reactions. Advantages and use lacks monoclonal and polyclonal antibodies for carrying out immunohistochemical reactions. Means of a labelling of antibodies. Detection immune complexes, a straight line and indirect methods detection. Modern commercial systems detection. Auxiliary reagents for carrying out immunohistochemical reactions. Blockage of a nonspecific binding of antibodies and endogenous activity of ferments. Demasking an antigen: the purpose, the basic means (thermal. proteoclastic). Double immunohistochemical/immunofluorescent colouring. Positive negative controls and immunohistochemical reactions. Features probes preparation at carrying out immunohistochemical reactions.

# Theme 10. Research methods in biological chemistry. Concept about stem cells.

#### Lecture

Medical-genetic consultation. Stem cells: embriological and somatic. Application of stem cells. Methods of differential and routine colouring of chromosomes. Modern possibilities of auxiliary reproductive technologies.

Theme 11. Morphometry. Bases of a statistical analysis of results morphometric researches.

### Laboratory research

Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

### THEMATIC PLAN

2018-2019 study year

Rules of reception of microphotographs from histological drugs. Features of quantitative analysis of histological drugs. Densitometry. Morphometry. Interpretation of the form and the dimensions of objects on a histological drug. The software for morphometric the analysis. The basic concepts of a statistical analysis: general totality, a sample, normal and abnormal allocation. Descriptive statistics indexes: measures of the central tendency (an average arithmetic, a median, a fashion), dispersion measures (scope, an interval, a standard deflection), their practical value for biomedical research. Comparison of experimental bunches to a studied sign. Check of statistical hypotheses. Methods of an assessment of reliability of distinctions of experimental bunches on a studied index. Statistical criteria (parametrical, nonparametric). Criterion of Student: application conditions, a procedure of payments. Order of carrying out and the report of educational scientific-research work on morphometry.

# Theme 12. Methods of allocation and purification of DNA from cells and tissues.

#### Lecture

Methods of destruction of cells and tissues, reception of cellular lysates. Mechanical, physical and chemical means of destruction of cells and tissues. Separation a fluid phase of the blasted cells from the firm. Purification of DNA by a method of sedimentation from a fluid phase. Sedimentation by means of TCA. Sedimentation of DNA by alcohol. Sedimentation by means of PEG. Dialysis and lyophilizing methods.

## Theme 13. Allocation and purification of DNA from procariotic and eucariotic cells

#### Laboratory research

Allocation and purification genomic DNA from cells of bacteria. Allocation and purification plasmid DNA from bacterial cells. Allocation and purification of DNA from blood leucocytes.

# Theme 14. Qualitative and quantitative methods for determining the parameters of isolated DNA

#### Lecture

The purity of the obtained DNA preparation. Concentration of DNA. The size of DNA. Electrophoresis in agarose and polyacrylamide gel. Hydrolysis of DNA using site-specific restriction enzymes. Electrophoresis of DNA fragments. Spectrophotometric method for determining the amount and purity of DNA.

# Theme 15. Educational research on morphometry and statistical processing of findings of investigation

#### Тема 16. Atomic-power microscopy in biomedical researches Lecture

Basic principles of atomic force microscopy.

Historical review of the development of atomic force microscopy. Introduction to the fundamentals of work on an atomic force microscope. Acquaintance with the device of an atomic force microscope. Explanation of the AFM's work on the example of van der Waals forces. Principle of the probe microscope. Techniques of atomic force microscopy: contact, noncontact and semi-contact. The use of graphical editors for the analysis of images obtained with the help of AFM.

Features of the application of atomic force microscopy for the analysis of biological samples. The main stages of sample preparation before analysis on an atomic force microscope. The principle of choice of substrate, cantilever coating. Study of adhesion and roughness with the help

Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

# THEMATIC PLAN

### 2018-2019 study year

#### of AFM.

# Theme 17. Formation and development of methods of physiological researches. Research of the bioelectric phenomena in an organism.

#### Lecture

Observation as a method of physiological experiment. The concept of "experiment", types of experiment. Vivisection (Magendie, L. Luciani). Methods of studying the nervous system: extirpation and transection of the brain. Study of the localization of functions in the cerebral cortex: from F. Hall's phrenology to K. Brodman's cytoarchitectonic maps. Stereotactic technique.

Investigation of bioelectric phenomena. Electrocardiography. The history of the method and the contribution of scientists to its formation: O. Waller, V. Eithoven (string galvanometer), A. Samoilov.

Electroencephalography. The founders: V.Ya. Danilevsky, V.V. Pravdich-Neminsky, R. Cato, G. Berger. Standard system for the arrangement of electrodes. Background EEG. The main types of electrical activity of the brain at rest and its origin.

Electromyography. Total electrical activity of muscles and individual discharges of motor units with muscle tension. Determination of the dynamics of fatigue by EMG. Diagnosis of movement disorders with EMG. Management of technical devices using EMG.

# Theme 18. Definition of parametres of DNA by electrophoresis and spectrophotometry methods.

#### Laboratory research

Laboratory research. UV a spectrophotometry. UV DNA spectrum. Definition of concentration of DNA by means of a spectrophotometer. The electrophoretic analysis of DNA. DNA restriction. Definition of the dimension of DNA by an electrophoresis method.

# Theme 19. Methods of investigating the functional state of the nervous system. Assessment of the psycho-physiological state of the nervous system

## Lecture

Psychophysiological functional states (PFS).

Levels of regulation of functional states. Methods and criteria for diagnosis of different types of PPS. Typical for professional activity states: monotony, fatigue, tension and stress. Ratio of PFS with motives and goals of activity. Self-regulation of the SPS and its mechanisms. Psychophysiological components of work capacity and its stages. Types of functional states. Methods for diagnosing functional states.

Psychophysiology of the properties of the nervous system. Neurophysiological basis of the human psyche. Principles and laws of higher nervous activity. Typological features of higher nervous activity. Sensation and perception: concept, neurophysiological basis, patterns. Neurophysiological basis of memory. Classification of the phenomena of memory: forms, processes, types and systems of memory. Characteristics of attention. Emotions and feelings: concept, physiological basis, properties, types. Conflict emotional states. Stress.

Expertise as a social phenomenon. Man as an object of expert research. The history of the formation and development of the method of psychophysiological "lie detection". The origin and development of the method as a new field of professional activity of a psychologist. Directions of polygraph checks. Factors determining the intensity of the development of the method of psychophysiological "lie detection" in modern conditions. Expert evaluation of the polygram: reliability of the survey results using a polygraph. Specificity of the formulation of conclusions

Institute of Fundamental Medicine and Biology Programme General Medicine, Dentistry Department of Morphology and General Pathology Course: Research methods in medicine and biology

# THEMATIC PLAN

2018-2019 study year

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### Theme 20. Scanning laser confocal microscopy in biomedical research

#### Lecture

History of the formation of laser confocal microscopy. Principles of confocal microscopy. Optical section. The use of confocal microscopy in biology. Confocal microscope device. Modes of the confocal microscope. Fluorescence correlation spectroscopy.

# Theme 21. Technics of registration EEG. Spectral analysis

#### Laboratory research

Technics of registration EEG. The international system of superposition of electrodes across Jasper, 10:20. Preparation of electrodes, rules superposition of electrodes, connection to computer энцефалографу. Concept functional trials: the occluded and open eyes. Artefacts at registration EEG. The basic rhythms EEG. Reaction desinchronization at opening of eyes. Roughing-out of epoch EEG: spectral analysis of rhythms EEG. The inspection conclusion.

Galvanic skin response registration. Roughing-out of results

# Theme 22. Methods of studying vegetative functions: ECG recording

#### Laboratory research

ECG recording technique: electrodes overlapping, 12 leads. Generation of teeth and intervals of ECG, their characteristics. ECG leads. Some indicators of ECG. Plan for decoding ECG. Self-interpretation of the ECG.

ECG processing and analysis are normal. Writing an opinion.

#### Theme 23. The method of recording the electrical activity of muscles (EMG).

#### Laboratory research

The main classes of methods for studying bioelectric potentials: EMG and its appointment in the clinic. Biopotential extraction system for electromyography (EMG). Electrodes and their classification. Electromyography. Overlapping of electrodes and registration of EMG with muscle contraction.

### Theme 24. Research methods in biological chemistry. ELISA. Immunoblotting.

#### Laboratory research

The immunoenzyme analysis. Immunoblotting (western blot). A chromatography. Chromatography kinds. Spectroscopy and spectrometry. Mass spectrometry. Application in biology and medicine. Microscopy kinds: optical, fluorescent, x-ray, electronic, scanning array.

# Theme 25. Research methods in biological chemistry. Concept about stem cells.

#### Laboratory research

Medical-genetic consultation. Stem cells: embriological and somatic. Application of stem cells. Methods of differential and routine colouring of chromosomes. Modern possibilities of auxiliary reproductive technologies.

### Theme 26. Spectrophotometric analysis of biomolecules

### Laboratory research

Methods of optical analysis of solutions of biomolecules. Principles of spectrophotometry. Qualitative and quantitative analysis of biomolecules. Concentration of substances. The Bouguer-Lambert-Bera law. Extinction coefficient. Construction of a calibration curve. Hypochromic and

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# THEMATIC PLAN

2018-2019 study year

hyperchromic effect of macromolecules.

Head of Morphology and General Pathology Department, Professor\_\_\_\_

\_\_\_\_\_\_A.P.Kiyasov