PREPARATION OF HISTOLOGICAL SPECIMENS FOR EXPERIMENTAL RADIATION NEUROBIOLOGY AND RADIATION THERAPY RESEARCH

1. Introduction

Experimental radiation neurobiology investigates the effects of ionizing radiation on the structure and function of the central nervous system. Understanding radiation-induced damage to brain tissue is essential for improving radiation therapy protocols, reducing adverse side effects, and developing new strategies for radioprotection and tissue recovery.

Modern research in this field combines radiation biology, neuroscience, pathology, and advanced microscopy. Histological analysis remains one of the fundamental methods for evaluating radiation-induced alterations in nervous tissue, including neuronal degeneration, vascular damage, inflammation, demyelination, and changes in cellular morphology. High-quality histological specimens are therefore essential for reliable quantitative and qualitative analysis.

At the Joint Institute for Nuclear Research (JINR), studies in experimental radiation biology support investigations of biological effects of different radiation modalities, optimization of radiotherapy approaches, and development of novel experimental models for preclinical research.

The practical significance of this project lies in providing students with hands-on experience in preparing histological sections suitable for microscopic examination and quantitative image analysis in radiation biology experiments.

2. Main Objectives of the Project

During the internship, the participant will become familiar with the principles of histological sample preparation used in biomedical research related to radiation neurobiology.

The student will learn:

- principles of tissue fixation and preservation;
- tissue sampling and orientation;
- paraffin embedding techniques;
- operation of a rotary microtome;
- preparation of thin histological sections;
- mounting tissue sections onto microscope slides;
- staining procedures (e.g., Hematoxylin and Eosin, Nissl staining, IHC);
- preparation of slides for long-term storage;
- basics of light microscopy;
- evaluation of histological specimen quality;
- documentation and interpretation of obtained results.

The participant will also gain practical laboratory skills including compliance with biosafety regulations, laboratory documentation, and quality control during specimen preparation.

3. Experimental Equipment

The laboratory setup includes:

- Tissue fixation station;
- Paraffin tissue processor;
- Paraffin embedding station;
- Rotary microtome;
- Water bath for section stretching;
- Drying oven;
- Histological staining station;
- Light microscope equipped with a digital camera;
- Computer for image acquisition and analysis.

4. Experimental Procedure

During the project, the student will perform the following steps:

1. Receive fixed experimental brain tissue samples.
2. Perform tissue dehydration and paraffin embedding.
3. Prepare paraffin blocks.
4. Cut tissue sections (4–6 μm thickness) using a rotary microtome.
5. Transfer sections to warm water and mount them onto glass slides.
6. Dry the mounted sections.
7. Deparaffinize and rehydrate the slides.
8. Perform routine histological staining.
9. Mount stained sections using permanent mounting medium.
10. Examine the prepared slides under a light microscope.
11. Select representative microscopic fields.
12. Document observations and assess specimen quality.

3.3 Expected Results

At the end of the project, each participant will provide:

- a complete set of properly prepared histological slides;
- digital micrographs of stained tissue sections;
- a short report describing:
    - specimen preparation protocol;
    - staining procedure;
    - quality assessment of obtained sections;
    - representative microscopy images

4. Prerequisites

The participant should have:

- basic knowledge of cell biology or histology;
- introductory knowledge of anatomy and physiology small laboratory animals;
- understanding of general laboratory safety procedures;
- basic English reading skills sufficient for working with scientific literature and PC.

Previous experience in histology or microscopy is desirable but not mandatory.

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5. Recommended Literature

1. Kiernan J.A. *Histological and Histochemical Methods: Theory and Practice*. 5th Edition. Scion Publishing, 2015.
2. Bancroft J.D., Gamble M. *Bancroft's Theory and Practice of Histological Techniques*. 8th Edition. Elsevier, 2019.
3. Suvarna S.K., Layton C., Bancroft J.D. *Bancroft's Theory and Practice of Histological Techniques*. Elsevier.
4. Mescher A.L. *Junqueira's Basic Histology: Text and Atlas*. 16th Edition. McGraw-Hill, 2021.
5. Hall E.J., Giaccia A.J. *Radiobiology for the Radiologist*. 8th Edition. Wolters Kluwer, 2018.
6. Pawley J.B. *Handbook of Biological Confocal Microscopy*. Springer.

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6. Number of Participants

The project is designed for **1–2 students** working simultaneously.

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7. Project Supervisor (JINR)

Name: *Iurii Sergeevich Severiukhin*

Position: *Head of The Sector*

Academic Degree: *PhD*

Laboratory: *Laboratory of Radiation Biology*

Research Interests:

- Experimental radiation neurobiology
- Histopathology
- Radiation therapy
- Brain radiation injury
- Microscopy and histological analysis

Selected Scientific Achievements:

1. **Severyukhin, Y.S., Lalkovičová, M., Utina, D.M. *et al.*** Comparative Analysis of Behavioral Reactions and Morphological Changes in the Rat Brain After Exposure to Ionizing Radiation with Different Physical Characteristics. *Cell Mol Neurobiol* (2022). https://doi.org/10.1007/s10571-021-01187-z
2. **Yu.S. Severiukhin, T. B. Feldman, M. A. Ostrovsky, A. G. Molokanov** Effects of Cranial Exposure to 170 MeV Proton Radiation at a Dose of 5 Gy on the Visual Behavior and Optomotor Response of Adults Rats // Radiation Biology. Radioecology (Russian), Volume 59, Issue 5. P. 532-537

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