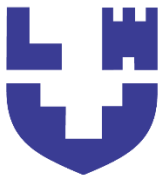


**MINISTRY OF EDUCATION AND SCIENCE OF UKRAINE
LUTSK NATIONAL TECHNICAL UNIVERSITY
FACULTY OF AGRICULTURAL TECHNOLOGIES AND
ENVIRONMENTAL STUDIES
AGRONOMY DEPARTMENT**



**AGRICULTURAL MICROBIOLOGY
(Сільськогосподарська мікробіологія)**

Methodical instructions for performing practical work
for students of the first (bachelor's) level of higher education of
the educational program «Agronomy» branch of knowledge 20 /
H «Agrarian Sciences and Food» specialty 201/ H1
«Agronomy» full-time and extramural form of study

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Методичні вказівки складено відповідно до діючої програми курсу «Agricultural microbiology (Сільськогосподарська мікробіологія)». Видання містить тематику лекційного матеріалу і завдання до виконання практичних робіт. Призначене для здобувачів спеціальності 201 / Н1 Агрономія денної форми навчання, можуть бути використані для здобувачів заочної форми навчання.

У методичних вказівках використано матеріали Гнатуш С.О., Гудзя С.П., Масловської О.Д., Курдиша І.К., Білявської Л.О., Волкогона В.В., Антипчук А.Ф., Бойка П.К. та інших науковців, а також власні напрацювання.

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INTRODUCTION

The training course «Agriculture Microbiology» is designed for students of the specialty 201 – «Agronomy», and can also be useful for training other interested persons according to the specialty.

The student's goal is to acquire competencies, knowledge, abilities and skills for the implementation of professional activities in the specialty in crop planning and crop care, improving soil conditions and increasing/restoring soil fertility, conducting an ecologically safe economy, implementing biotechnological technologies, etc. Discipline «Agricultural microbiology» studies the morphology, physiology and biochemistry of microorganisms as well as their modern taxonomy, ecological features, the participation of microorganisms in the circulation of substances in nature.

The academic course objectives is to form students' basic knowledge about the morpho-physiological features of microorganisms and their functions in the biocenosis, for example: the role of microorganisms in increasing soil fertility both in transformations of N, P and K and the cycle of other chemical elements, their positive and negative impact on people, the environment and the agroecosystem.

The structure of the Manual is as follows:

1. Title and number of practical work.
2. Topic, purpose.
3. A brief description of the equipment and materials needed to carry out the work.
4. Tasks (includes an algorithm of actions for conducting research, performing written tasks, drawing what was seen during a macro-/microscopic examination, performing a calculation, etc.), including recommendations for writing conclusions.

ACADEMIC COURSE PROGRAM

Content module 1. Basics of general microbiology

- Topic 1. Introduction to discipline. History of the development of science.
- Topic 2. The structure of prokaryotic and eukaryotic cells. Movements of microorganisms.
- Topic 3. General characteristics of metabolism. Feeding microorganisms.
- Topic 4. Breathing microorganisms.
- Topic 5. Fermentation (types of fermentation).
- Topic 6. Basics of genetics of microorganisms.
- Topic 7- 8. Basics of systematics of microorganisms according to Bergey.
- Topic 9-10. Microorganisms in production.

Content module 2. The role of microorganisms in the functioning of natural biocenoses

- Topic 11. Microorganisms and soil fertility. Transformation of the main elements of plant nutrition.
- Topic 12. Microorganisms in the conversion of nitrogen compounds.
- Topic 13. Microorganisms in the transformation of phosphorus and potassium compounds.
- Topic 14. Cellulose-degrading microorganisms (aerobic and anaerobic).
- Topic 15. Microorganisms and metals, sulfur. Oxidative and reducing processes (including bioleaching).
- Topic 16. Interaction of microorganisms and plants. Pathogenic forms.
- Topic 17. Useful microorganisms (rhizosphere). Microbial preparations in crop production.
- Topic 18. Pathogenic species for humans and animals.
- Topic 19. Sanitary-indicative microorganisms.
- Topic 20. Bioremediation of the environment.

PRACTICALS

№	Topics of the practicals	Number of hours	
		Full-time Studies	Extramural Form of Study
Content module 1.			
1	Microbiology lab. The structure of the microscope.	4	3
2	Structure of microorganisms. Microscope slide preparation. Movement of microorganisms	4	
3	Methods of bacteria staining (simple and differential).	4	
4	Sterilization	4	
5	Types of microorganisms nutrient / culture media. Preparation.	4	
6	Plating and counting the colonies of microorganisms on nutrient media	6	
For content module 1		26	3
Content module 2.			
7	Aerobic and anaerobic growth of microorganisms	5	3
8	Microorganisms providing NPK compounds conversion in soil	5	
9	Participation of microorganisms in the cycle of other chemical elements (S, Fe)	4	
10	Soil microbiome research	5	
For content module 2		19	3
TOTAL		45	6

FULL-TIME STUDIES

Practice № 1 (4 hours)

Topic: Microbiology lab. The structure of the microscope

The purpose of the work: to familiarize yourself with the rules of operation and safety techniques in the microbiological laboratory, as well as with the structure and principle of operation of the light microscope.

Materials and reagents: laboratory protective clothing, laboratory equipment with usage instructions, guidelines for safety in biological laboratories and a light microscope.

PRACTICAL PART

1. To familiarize yourself with laboratory safety rules and proper handling techniques for chemical reagents.
2. to study the structure of the microscope, create a detailed sketch, and learn the functions of its components.
3. to summarize and draw conclusions based on your observations or findings.

Practice № 2 (4 hours)

Topic: Structure of microorganisms. Microscope slide preparation. Movement of microorganisms

The purpose of the work: to study the structure of a bacterial cell, to get acquainted with the organelles of microorganisms movement and to master the basic methods of making micropreparations.

Materials and reagents: microscopes MBR-1 or Biolam, ready-made micropreparations, cultures of microorganisms, laboratory equipment

PRACTICAL PART

1. Draw the structure of bacterial cell, label its components, be able to characterize them.
2. Examine the prepared micropreparations. Briefly describe what do you see.
3. Make a micropreparation using the “crushed drop” technique.
4. Make a micropreparation using the “hanging drop” technique.
5. Describe and study the character of microorganisms movement.
6. Summarize your observations into conclusions.

Practice № 3 (4 hours)

Topic: Methods of bacteria staining (simple and differential)

The purpose of the work: to study the basic methods of bacterial cells staining.

Materials and reagents: light microscope, slides, cover slides, saline solution, cultures of microorganisms, distilled water, dyes, alcohol, laboratory tools, cultures of microorganisms.

PRACTICAL PART

1. To note and study the types of painting.
2. Perform simple staining of cells using methylene blue or Rebigier dye. Make a microscopic research of micropreparates.
3. Describe the Gram staining algorithm, then you should study it.
4. Stain the culture according to Gram, microscopy, describe what you saw.
5. Draw conclusions

Practice № 4 (4 hours)

Topic: Sterilization

The purpose of the work: Study methods and methods of sterilization of objects, tools, premises.

Materials and reagents: autoclave, dry oven, filters, UV lamp, tools, distillate, illustrative material, disinfection solutions and reagents for their preparation.

PRACTICAL PART

1. Work out theoretical material on the topic. Define key concepts, study.
2. Complete a table «Sterilization methods»
3. Familiarize yourself with the structure of the autoclave, dry fire oven. Schematically depict the diagram of the operation of the autoclave.
4. Produce a 5% hydrogen peroxide solution and a 75% ethyl alcohol solution.
5. In the conclusions to the work, indicate the importance of observing sterility in the laboratory

Practice № 5 (4 hours)

Topic: Types of microorganisms nutrient / culture media. Preparation.

The purpose of the work: learn the concept of «nutrient medium», its role in research, study the basic composition of media, their variations and the cooking algorithm.

Materials and reagents: chemical reagents, electric tiles, 2nd accuracy class scales, spatulas, filter paper, laboratory dishes, autoclave.

PRACTICAL PART

1. Familiarize yourself with the theoretical material. Notice the requirements for nutrient media.
2. Note about the classification of nutrient media.
3. Note the Ingredients and prepare the MPA.

4. Autoclave MPA.

5. Draw conclusions about the dependence of the composition of the nutrient medium on its purpose.

Practice № 6 (6 hours)

Topic: Plating and counting the colonies of microorganisms on nutrient media.

The purpose of the work: learn plating techniques for the nutrient medium.

Materials and reagents: sterile petri dishes, sterile test tubes, sterile and heated nutrient media, alcohol, microbiological loop, sterile pipettes, spatulas, cultures of microorganisms.

PRACTICAL PART

1. Familiarize yourself with the theoretical material on sowing techniques.
2. Pour the agaric nutrient medium into petri dishes, test tubes.
3. Carry out surface (dash, «lawn») and deep plating of the culture of microorganisms on an agaric nutrient medium.
4. Pour the liquid nutrient medium into test tubes
5. Plant in a liquid nutrient medium.
6. Adjust the thermostat and put the cultures on incubation.
7. For 2-3 days, count the colonies of microorganisms with a conversion to their content in 1 g/ml of substrate
8. Draw up conclusions.

Practice № 7 (5 hours)

Topic: Aerobic and anaerobic growth of microorganisms

The purpose of the work: familiarize yourself with the methods of laboratory and industrial cultivation of microorganisms at different levels of aeration.

Materials and reagents: cultures of microorganisms, nutrient media, laboratory equipment, shaker (swing), thermostat, anaerostat, illustrative material.

PRACTICAL PART

1. Familiarize yourself with the theoretical material of the requirements of microorganisms for oxygen. To revise the classification of m.-o. in relation to oxygen (be sure to give examples).
2. Plate aerobic microorganisms on a solid and liquid nutrient medium.
3. Plate anaerobic microorganisms on a solid and liquid nutrient medium.
4. Place aerobes and anaerobes for incubation.
5. Record CFU after incubation.
6. Draw up conclusions.

Practice № 8 (5 hours)

Topic: Microorganisms providing NPK compounds conversion in soil

The purpose of the work: get acquainted with the circulation of N, P and K in nature and microorganisms that implement separate links of cycles.

Materials and reagents: chemical reagents for the preparation of media, distilled water, scales, electric tiles, tools and dishes, autoclave.

PRACTICAL PART

1. Draw a diagram of the cycle of nitrogen-containing compounds in nature, including microorganisms of the corresponding units. Prepare nutrient media according to the census, summarize their composition.
2. Draw a scheme of transformations of phosphorus compounds in nature, including microorganisms of the corresponding units. Prepare nutrient media according to the census, summarize their composition.
3. Record the participation of micro-organisms in the cycle of compounds K. legally inspect the composition of the nutrient medium.
4. Sterilize all media.
5. To draw conclusions about the role of microorganisms in the cycle of NPK compounds in the environment.

Practice № 9 (4 hours)

Topic: Participation of microorganisms in the cycle of other chemical elements (S, Fe)

The purpose of the work: дослідити біогеохімічну активність мікроорганізмів

Materials and reagents: handout material, multimedia equipment.

PRACTICAL PART

1. Work out the theoretical material. Review video materials about the role of microorganisms in the transformation of The purpose of the work: fishing compounds
2. Briefly outline the participation of microorganisms in the transformation of iron.
3. Consider materials on the participation of bacteria in the transformation of sulfur-containing compounds.
4. Briefly outline the participation of micro-organisms in the Sulfur cycle
5. Draw conclusions about the role of microbiota in geochemical transformations.

Practice № 10 (5 hours)

Topic: Soil microbiome research.

The purpose of the work: to investigate the microbiota of the resulting soil sample.

Materials and reagents: nutrient media, sterile dishes and tools, alcohol, soil sample.

PRACTICAL PART

1. Get acquainted with the material about research planning, soil sampling.
2. Select an average soil sample from the selected sample.
3. In boxing, make a series of 10-fold dilutions according to the scheme (write the scheme in a notebook).
4. Prepare and sterilize Hetchenson's medium for cellulose destructors.
5. Sow soil (from 1000 – 100,000 times dilution) on ready-made media. Put the seeded petri dishes in the thermostat for incubation.
6. Keep records of nitrogen fixers (on the Eshby environment), m.-o., which mobilize organic phosphorus (on the village of Menkina), mineral phosphorus (on the village of Muromtseva).
7. Observe the development of destructors on cellulose.
8. Draw conclusions about the stages of microbiological research of substrates.

EXTRAMURAL FORM OF STUDY

Practice № 1 (2 hours)

Topic: setting up a microbiological laboratory. Microscopy. Microscope slide preparation. Structure of microorganisms. Sterilization

The purpose of the work: to familiarize yourself with the rules of operation and safety techniques in the microbiological laboratory, as well as with the structure and principle of operation of the light microscope; with microbe cell structure and the ways of movement.

Materials and reagents: laboratory protective clothing, devices and instructions for working with them, Rules for handling in biological laboratories; light microscope.

PRACTICAL PART

1. To familiarize yourself with laboratory safety rules and proper handling techniques for chemical reagents.
2. to study the structure of the microscope, create a detailed sketch, and learn the functions of its components.
3. Draw the structure of bacterial cell, label its components, be able to characterize them.
4. To note and study the types of painting.
5. Perform simple staining of cells using methylene blue or Rebigier dye. Make a microscopic research of micropreparates
6. Complete a table «Sterilization methods»
7. Familiarize yourself with the structure of the autoclave, dry fire oven. Schematically depict the diagram of the operation of the autoclave.

Practice № 2 (2 hours)

Topic: nutrient media. Microbiological research

The purpose of the work: to study the composition and requirements of nutrient media, to learn how to distinguish, prepare, sterilize them both substrate depending on the purpose of the study.

Materials and reagents: chemical reagents and laboratory equipment.

PRACTICAL PART

1. Familiarize yourself with the theoretical material
2. about the types of nutrient media and their requirements.
3. Familiarize yourself with the theoretical material on sowing techniques.
4. Prepare, sterilize then pour the agaric nutrient medium into petri dishes, test tubes.
5. Sow all nutrient mediums with microbes, put the cultures on incubation

into the thermostat.

6. Familiarize yourself with the theoretical material on the accounting of colonies. Draw the morphology (shape, profile, edge and structure) of colonies of m.-o. on agar mediums.
7. Calculate CFU per each nutrient medium.

Practice № 3 (2 hours)

Topic: microorganisms in the circulation of substances in nature

The purpose of the work: to get acquainted with the needs of microorganisms in various substances and their role in the transformation of compounds N, P, K, S, C, etc.

Materials and reagents: cultures of microorganisms, nutrient media, alcohol, sterile tools and dishes.

PRACTICAL PART

1. Work out the theoretical material, briefly review.
2. To revise the concept of «donor and electron acceptor» for m.-o.
3. Prepare different nutrient media according to the census. Autoclave them.
4. Sow cultures of N-, P-, K-mobilising microorganisms on appropriate nutrient media. Incubate and count the results.
5. Draw a scheme of transformations of N, P, K and S compounds as well in nature, including microorganisms of the corresponding units.
6. To sum up about the role of microorganisms in geochemical processes.

LITERATURE

Main literature resources:

1. Diakiv Svitlana. Soil microbiota state as a highly informative soil characteristic. Digital Transformation: Challenges and Strategies: Proceedings of the International Scientific and Practical Conference, February 25, 2025, Lutsk: LNTU, 2025. p. 137-139.
2. Diakiv S., Ilkevych A., Kuzmishyna I., Komovych L. Biological activity of urban soils microbiota, formed by the introduction of construction waste. Молодь і поступ біології / Youth and Progress of Biology: збірник тез доповідей XXII Міжнародної наукової конференції студентів і аспірантів (м. Львів, 15–17 квітня 2026 р.). Львів : ПАІС, 2026. С. 203. ISBN 978-617-7694-89-1
3. Diakiv S.V., Zinchuk M.I., Boyko P.K., Kuzmishyna I.I., Merlenko I.M. Catalase activity of lutsk urban soils / Microbiology and Immunology – the Development Outlook in the 21st century. Abstracts book of the V International Scientific Conference, April 29-30, 2025, Kyiv. Kyiv, 2025. p. 49.
4. Hnatush S. O., Maslovska O. D., Komplikevych S. Ya., Segin T. B., Diakiv S. V. Relationships between prooxidant and antioxidant parameters of bacteria of sulfur cycle under the influence of heavy metal compounds. The current state of fundamental and applied natural sciences research: Scientific monograph. Riga, Latvia : “Baltija Publishing”, 2022. 382 pp. 100-125. <https://doi.org/10.30525/978-9934-26-212-8-5>.
5. Бойко П.К., Дяків С.В., Зінчук М.І., Аджиева Л.Г., Коробейко Л.С. Ґрунтова мікробіологія: методичні рекомендації до лабораторних занять для бакалаврів і магістрів медико-біологічного факультету. Луцьк: ПП Іванюк В.П., 2020. 76 с.
6. Бойко П.К., Дяків С.В., Титюк О.В., Бойко О.П., Мандигра Ю.М. Загальна мікробіологія. Лабораторні роботи: навчальний посібник для спеціальності 091 Біологія, освітньо-професійних програм «Біологія» і «Лабораторна діагностика» факультету біології та лісового господарства. Луцьк, 2022. 140 с.
7. Дяків С.В., Кузьмішина І.І., Бойко П.К., Комович Л.В., Бурачук Х.В. Целюлазна активність ґрунтів м. Луцька та приміських територій. Тези доповідей XVI з'їзду Товариства мікробіологів України ім. С.М. Виноградського, Тернопіль, ТНМУ ім. І. Я. Горбачевського, 2-6 червня 2025 р., 302 с., С. 30
8. Дяків С. В., Кузьмішина І. І., Комович Л. В., Галас В. А., Головій А. С.. Каталазна активність ґрунту за впливу сполук важких The purpose

of the work: лів. Збірник наукових праць. Охорона ґрунтів. Спеціальний випуск: Моніторинг ґрунтів. Реалії, виклики, перспективи. З нагоди 60-річчя агрохімічної служби України: матеріали міжнародної науково-практичної конференції. Київ: Охорона ґрунтів, 2024. с. 134-135.

9. Дяків С.В., Максимчук А.В. Уреазна активність ґрунту для індикації забезпечення рослин доступними формами нітрогену. Матеріали Всеукраїнської науково-практичної конференції студентів, аспірантів та молодих вчених «Сучасна освіта і наука Волині» (22 травня 2025 р.). Луцьк: ФОП Мажула Ю. М., 2025. С.31–32.

10. Дяків С.В., Мерленко І.М., Зінчук М.І. Ідентифікація ферментативної активності для діагностики стану ґрунту. Талановита організатор, вчена-практик, педагог: присвячено 85-річчю від дня народження докторки біологічних наук, професорки Любові Калинівни Тараненко: матеріали круглого столу; збірник праць:, Київ, 16 квітня 2024 р. / НААН, ННСГБ, Ін-т історії аграр. науки, освіти та техніки, МОН України, ЗВО «ПДУ»; наук.ред. В.А. Вергунов. – Вінниця: ТВОРИ, 2024. С. 80-84.

11. Технічна мікробіологія: практикум для здобувачів вищої освіти / В.В. Т 38 Євлаш, Л.В. Газзаві-Рогозіна, А.С. Бикова, О.В. Циганков – Х. : НТУ «ХПІ», ХДУХТ, 2020. 180 с.

Additional literature resources:

1. Komplikevych S., Maslovska O., Peretyatko T., Moroz O., Diakiv S., Zaritska Ye., Parnikoza I., Hnatush S. Culturable microorganisms of substrates of terrestrial plant communities of the maritime Antarctic (Galindez Island, Booth Island). Polar Biology, 46, 1–19 (2023). <https://doi.org/10.1007/s00300-022-03103-7>.

2. Бурачук Х., Дяків С., Кузьмішина І., Комович Л. Загальна біологічна активність ґрунту та урбанозему на прикладі м. Луцька і його околиць: тези IV студентської науково-технічної конференції «Сучасні технології у агровиробництві та природокористуванні». Луцьк: Факультет аграрних технологій та екології, Луцький НТУ. 2024. с. 4-7.

3. Доведено ефективну дію триходерми в ґрунтах, інфікованих фузаріозними інфекціями. Бойко П., Зінчук М., Дяків С., Комович Л., Матрунчик В. The Ukrainian Farmer, 2024, Вип. 3. URL: <https://agrotimes.ua/article/gryby-proty-grybiv/>

4. Зінчук М.І., Дяків С.В., Комович Л.В., Довгаюк-Семенюк М.В. Вплив часових експозицій на виживання мікробних тіл в бакових сумішах препарату «AZOTER» та КАС. Інноваційні технології в АПК: збірник тез доповідей X Міжнародної науково-практичної конференції, 3-4 квітня 2025 р., м. Луцьк [Електронний ресурс] – Луцьк: Луцький НТУ, 2025. с. 94-95.

Useful Internet resources:

1. LPSN - List of Prokaryotic names with Standing in Nomenclature/ Режим доступу: <https://www.bacterio.net/>
2. Інститут мікробіології і вірусології ім. Д.К.Заболотного. НАН України. Режим доступу: <https://imv.org.ua/>
3. Microbiologyinpictures.com. Режим доступу: <https://www.microbiologyinpictures.com/> index.php
4. MicrobeWiki. Режим доступу: <https://microbewiki.kenyon.edu/index.php/MicrobeWiki>
5. Microbe Notes. Classification of Bacteria: Methods and Criteria Explained Режим доступу: <https://microbenotes.com/classification-of-bacteria/>