### MINISTRY OF HEALTH OF UKRAINE BOGOMOLETS NATIONAL MEDICAL UNIVERSITY

# **HANDBOOK**

# on manufacturing practice on standardization of herbal medicines for auditory and independent students work

# Laboratory handbook

**Direction:** second (master's) level of higher education

Specialty: 226 "Pharmacy, industrial pharmacy"

**Department:** Pharmacognosy and botany

| Name   |  |  |
|--------|--|--|
| Course |  |  |
| Group  |  |  |

### **UDC 615.322(076.5)**

Handbook on on manufacturing practice on standardization of herbal medicines for auditory and independent students work. Laboratory handbook. / Minarchenko V. M., Karpiuk U. V., Cholak I. S., , Makhynia L. M., Pidchenko V.T., Kovalska N. P., Dvirna T. S., – Kyiv., 2024. – 62 p.

Approved at the meeting of the department on August 28, 2024, protocol No. 1 Considered and approved: CMC on specialty 226 "Pharmacy, industrial pharmacy" dated August 30, 2024, protocol No. 1

#### **Reviewers:**

Zhuravel I.O., professor, doctor of pharmaceutical sciences, professor of Pharmacognosy and Nitriciology Department of National Pharmaceutucal University Sholoiko N.V., Associate Professor, Candidate of Pharmaceutical Sciences, Associate Professor of Department of Organization and Economy of Pharmacy, of Bogomolets National Medical University

The diary is intended for recording the results of research on industrial practice for the standardization of herbal medicines in accordance with the working curriculum and calendar and thematic plan.

The publication is intended to be used for studying the discipline - industrial practice on standardization of herbal medicinal products and studying individual sections of professionally oriented topics in the discipline in order for students to master knowledge on standardization of herbal medicinal products and medicinal products based on it.

The diary is supplemented with topics for extracurricular study for self-study of students

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#### SAFETY PRECAUTIONS

Execution of educational and scientific experimental work at the Department of Pharmacognosy and Botany in educational and research laboratories associated with various chemicals (organic solvents, acids, alkalis), plant materials using, different chemical utensils, equipment and devices. Therefore, in the laboratory spaces there are continuously possible dangerous and harmful factors which can effect on students. These factors can lead to work-related injury and fire hazards.

Students are admitted to practical training in chemical lab only after a detailed briefing on safety and fire precautions.

Each student works in the laboratory must know the location of fire fighting and be able to know where the first aid kit and know how to provide first aid for various injuries.

Experimental part begins only after a thorough acquaintance with chemical dishes, technique experiments, properties, purpose reagents and solvents used, and the rules of work with devices. The workplace should only has necessary reagents, instruments and a notebook to record results.

Before using glass and porcelain tableware check its purity and integrity. Do not work with the dishes that have chips, cracks, deep scratches.

All transactions with flammable liquids, concentrated acids and alkalis, experiments with the formation of gases and work with metallic sodium should be performed only in a fume hood, if necessary, should use personal protective equipment (masks, goggles, mask, gloves, etc.). Smell of substance in a test tube or flask determine carefully directing couples to himself flick of the wrist. Mixing and dilution of chemicals, accompanied by heat, spend with heatproof bowl and porcelain.

Do not allow heat flasks with inflammable liquids over an open fire, avoid getting water on the heated external surface of glass vessels, gently and carefully to treat laboratory glassware and equipment.

Acids and bases to dial in using only the dropper rubber pear, forbidden to absorb acid and alkali liquid in the pipette mouth, because it can cause burns and poisoning.

Heating of substances in hermetically sealed vessels (safety explosion!) is strictly forbidden. To prevent the release of liquid from the reaction vessel should be carried out uniformly heating the to put to the bottom of the vessel 2-3 boiling stones (pieces of porous inorganic material).

Heating tubes of substances should be carried out at periodic shaking, vent tubes should be directed away from yourself and others that work.

Take and carry glass with substances should, covering them with hand from side, not the neck. Do not leave without supervision laboratory installation, operating and equipment included.

It is strictly forbidden to drink water from the chemical dishes, eat, smoke at the laboratory.

After work should be thoroughly washed and put to dry dishes, cups and place shtanhlasy on their location, wipe the work surface of the table, close the gas and water valves, turn off appliances and exhaust ventilation.

*In case of pouring concentrated acid*, it must first fill with sand so that it is absorbed acid. Sand collect in the container and make out of room to place waste collection. Polluted place pouring rinse with water and wipe dry.

In case *of pouring concentrated and ammonia* - they can fill as sand and sawdust. Pouring place by weak solution of acetic acid after collecting sand or sawdust.

In case of fire in a laboratory situation should turn off gas appliances, exhaust ventilation and remove all flammable material from the fire area. Shouting loudly advertise on fire people working together and in neighboring areas.

It is necessary to take urgent measures to eliminate the fire using fire extinguishers or sand. Do not fill the flames with water that in many cases this leads to expansion of the fire. Only water-soluble substances (alcohol, acetone, etc.) quenched with water. In case of fire clothes should not run, you need to throw the victim's robe located in a prominent and accessible place.

**Provision for first aid** is the duty of everyone! In providing assistance priority should be to eliminate the cause of the injury, turn off the power grid, extinguish the flame, remove from the

wound pieces of glass or substance that causes burns, etc.; the victim must create conditions for the most comfortable position and provide first aid.

If *cuts* must be removed with tweezers glass pieces of glass and wash the wound 3% solution of hydrogen peroxide. The skin around the wound grease 5% solution of iodine and apply a sterile bandage. In severe bleeding tourniquet and attach a note with precisely specified time imposing and send poterpitsloho doctor.

When I degree *thermal burns* (redness) burnt areas should be cool running water, while more severe burns to the provision of skilled care - apply dry aseptic bandage. Do not remove the skin from the burnt remains of clothes that burned.

Burns by concentrated acids affected skin should be washed with plenty of water for 10-15 minutes and then process the 2% solution of sodium bicarbonate and again rinsed with water.

Burns by concentrated alkalis affected area should be washed with plenty of water, and then - a 1% solution of acetic acid.

When hit acids or alkalis to the eyes, they should immediately wash with water for 10-15 minutes, then, if getting acid - 2% solution of sodium bicarbonate, and when it enters the meadow - isotonic sodium chloride solution for 30-60 minutes. After thorough rinsing eyes should consult a doctor.

After burns by phenol rub the affected area till restore the natural skin colour.

If poisoning by gas substances bring the victim to fresh air and create him complete rest and call a doctor.

When *electric shock* turn off power and, using a wooden or plastic objects poterpioho release from contact with electric wire. It is necessary to ensure the victim calm and bring it to life.

If *breathing or heartbeat stops* it's necessary to carry out artificial respiration and chest compressions and do not stop these operations to full functional recovery or the arrival of the medical workers.

# Safety precautions at work, harvesting, drying, processing and storage of plant material that contains toxic and potent substances (alkadoides, cardiac glycosides, etc.):

- 1. Teenagers, students are allowed to collect only under the supervision of the responsible team leader or instructor. By collecting MP, which contain these substances, it is better to include the adult population to collect datura, henbane, hellebore teenagers do not allow!
- 2. During the assembly should not touch your eyes, the face, not to eat. After collecting wash hands thoroughly with soap and water.
- 3. During the processing, drying, sorting, packaging protecting mouth and nose with a respirator, wet gauze, eye protective glasses. Do not take food or smoke.
- 4. After thoroughly shake out of clothes, wash clothes, wash the face with soap and water, wipe with a dust mask, goggles, gauze.
  - 5. When the need to have a first aid kit.
  - 6. To work with the potent and poisonous MP not allowed zhinkm pregnant and lactating.

**TOPIC 1:** tandardization of the living plant material and medicinal products based on it. Problems of standardization of herbal medicines and medicinal products. General provisions and articles of the State Pharmacopoeia on the study of the quality of the medicinal products and herbal medicines.

**Task 1.** Analyze and compare the SPhU monographs on medicinal products and drugs on the example of the SPhU monograph "Echinacea Roots" and "Echinacea Tincture"

| «Echinacea Roots» | «Echinacea Tincture» |
|-------------------|----------------------|
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**Task 2.** Провести стандартизацію ЛРС та ЛЗ за монографією ДФУ (наприклад: «Нагідок квітки<sup>N</sup>» та «Нагідок настойка<sup>N</sup>») To standardize the MPM and medicine according to SPhU monographs for medicinal products and drugs (e.g., "Calendula flower<sup>N</sup>" and "Calendula tincture<sup>N</sup>")

| «Calendula flower <sup>N</sup> » |             |  |  |
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| Identification A                 |             |  |  |
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# Identification C

Add a scheme of the chromatogram. Compare the results with the monograph on this type of MPM.

| Mobile phase        | Detection           |
|---------------------|---------------------|
|                     |                     |
| The tested solution | Comparison solution |
|                     |                     |

|          | Chromatogram scheme |              |
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| Quantita                   | ative determination, %          |   |
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| Descrip                    |                                 | a tincture <sup>N</sup> »                   |
| Identific Add a stype of l | cheme of the chromatogram. Comp | pare the results with the monograph on this |
|                            | Mobile phase                    | Detection                                   |
|                            | The tested solution             | Comparison solution                         |
|                            | Chromatog                       | ram scheme                                  |
|                            |                                 |   |
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|                            |                                 |   |

# Identification B

Add a scheme of the chromatogram. Compare the results with the monograph on this type of MPM.

| Mobile phase        | Detection           |
|---------------------|---------------------|
|                     |                     |
| The tested solution | Comparison solution |
|                     |                     |

|         | Chromatogram scheme    |  |
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- 1. Name the main documents that regulate the quality standards of medicines.
- 2. What factors determine the suitability of the drug for use?
- 3. Name national and international organizations that introduce standards and guidelines necessary to ensure the quality of pharmaceutical products.
- 4. What is the modern international concept of quality assurance of drugs?
- 5. What are the functions of the State Pharmacopoeia?
- 6. What issues are within the competence of the State Enterprise "Ukrainian Scientific Pharmacopoeial Center for Quality of Medicines"?
- 7. List the characteristics of medicinal products that are included in the main sections of monographs on medicinal products.
- 8. When did Ukraine start publishing its own pharmacopoeia and how many supplements have been published?

| 9. Provide a list of names that are classified as medicinal products according to the Law of Ukraine "On Medicinal Products". |  |  |
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**TOPIC 2:** Pharmacopoeial methods of research to confirm the quality of the analyzed herbal medicines. Rapid methods for the study of the main groups of BAS.

**TOPIC 2.1:** Pharmacopoeial methods of research to confirm the quality of the analyzed herbal medicines.

**Task 1.** To analyze pharmacopoeial methods for the study of various groups of BAS in herbal medicines.

| a) chemical reactions;                  |
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| в) physical and chem | ical research n | nethods; |   |
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- 1. Name the main documents that regulate the quality standards of medicines.
- 2. What factors determine the suitability of the drug for use?
- 3. Name national and international organizations that introduce standards and guidelines necessary to ensure the quality of pharmaceutical products.
- 4. What is the modern international concept of quality assurance of drugs?
- 5. What are the functions of the State Pharmacopoeia?
- 6. List the characteristics of drugs that are introduced in the main sections of monographs on the drug.
- 7. Name the qualitative reactions to the main groups of BAS.
- 8. What methods of chromatographic analysis are used to determine the main groups of BAS.

| 9. Name the physicochemical methods for the quantitative determination of the main groups of BAS. |
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# **TOPIC 2.2:** Express methods of studying the main groups of BAS.

**Task 1.** Carry out histochemical and microchemical reactions to confirm the presence of different groups of bioactive substances:

| The name of the reagent                      | Conditions   | The result of the reaction |
|--|--|----------------------------|
| Reaction on cellulose                        | (fiher)  | TCACCION                   |
| with chlorine-zinc-                          | Chlorine-zinc-iodine is applied to the cut of the root         |                            |
| iodine                                       | and the color is observed                                      |                            |
| with iodine and                              | Iodine with sulfuric acid is applied to the root               |                            |
| sulfuric acid                                | section and the color is observed                              |                            |
| with an ammonia                              | An ammonia solution of copper oxide is applied to              |                            |
| solution of copper                           | the cut of the root and the color is observed                  |                            |
| oxide  |  |                            |
| with Lugol's solution                        | Lugol's solution is applied to the root section and            |                            |
|  | the color is observed  |                            |
| Reaction on mucilage                         |  |                            |
| with methylene blue                          | A section of the altea root is placed for a few                |                            |
| solution                                     | minutes in a solution of methylene blue in alcohol             |                            |
|  | (1:5000), then transferred to glycerin                         |                            |
| with copper sulfate                          | A section of the altea root is placed for 5-10                 |                            |
| and alkali                                   | minutes in a saturated solution of copper sulfate,             |                            |
|  | washed with water and transferred to a 50%                     |                            |
|  | solution of potassium hydroxide                                |                            |
| with mascara solution                        | Flax seed powder is placed on a glass slide in a               |                            |
| (1:10)                                       | drop of freshly prepared carcass solution (1:10) and           |                            |
|  | mixed with a needle, covered with a cover glass and            |                            |
| 1.1.2.20/                                    | observed under a microscope                                    |                            |
| with 3-5% sodium                             | Flaxseed powder is placed in 1-2 drops of 3-5%                 |                            |
| hydroxide solution                           | sodium hydroxide solution, covered with a cover                |                            |
| - · · · · · · · · · · · · · · · · · · ·      | glass and examined under a microscope.                         |                            |
| Reaction on inulin                           |  |                            |
| Molish's reaction                            | A cross-section of the root of a dandelion or                  |                            |
|  | elecampane is placed in 1-2 drops of an alcoholic              |                            |
|  | solution of α-naphthol (or thymol) and a drop of               |                            |
| D 4! 4 1.                                    | concentrated sulfuric acid is added                            |                            |
| Reaction on starch                           | 121 CT 11 12 11 14   |                            |
| with Lugol's solution                        | 1-2 drops of Lugol's solution are applied to a                 |                            |
|  | section of the altea root, covered with a cover glass          |                            |
| Danetian an liquidad                         | and observed under a microscope                                |                            |
| <i>Reaction on lignified</i> with 1% alcohol |  |                            |
|  | A section of althea root is placed on a slide in a 1%          |                            |
| solution of                                  | alcohol solution of phloroglucin and a drop of                 |                            |
| phloroglucin                                 | concentrated hydrochloric acid is applied to the               |                            |
|  | section and a drop of glycerin is added after 1-2              |                            |
|  | minutes; cover with a cover glass and study under a microscope |                            |
| Reaction on essential                        |  |                            |

| with a solution of<br>Sudan III | A section of the root is placed for several minutes in<br>a solution of Sudan III and a drop of water or<br>glycerin is added. Cover with a cover glass and |  |
|---------------------------------|---|--|
|                                 | observe under a microscope  |  |
| Reaction on fatty oils          |   |  |
| with a solution of              | A slice of castor seed is placed for a few minutes in   |  |
| Sudan III                       | Sudan III solution and washed with 50% alcohol  |  |
|                                 | and glycerol is added, covered with a cover glass   |  |
|                                 | and observed under a microscope   |  |
| Reaction on anthraces           | ne derivatives  |  |
| with a 5% solution of           | A slice of castor seed is placed for a few minutes in   |  |
| sodium hydroxide or             | Sudan III solution and washed with 50% alcohol  |  |
| ammonium hydroxide              | and glycerol is added, covered with a cover glass   |  |
|                                 | and observed under a microscope   |  |
| Reaction on tannins             |   |  |
| with 1% ferric                  | A slice of oak bark is placed in a drop of a 1%   |  |
| chloride or 1%                  | solution of ferric chloride or a 1% solution of iron-   |  |
| aqueous solution of             | ammonium alum, covered with a cover glass, and  |  |
| iron-ammonium                   | the staining of the drug is observed under a  |  |
| alums                           | microscope.   |  |

**Task 2.** Carry out chemical reactions to confirm the presence of different groups of BAS:

| The name of the           | Conditions                                     | The result of the |  |  |
|---------------------------|--|-------------------|--|--|
| reagent                   |  | reaction          |  |  |
| CARBOHYDRATES. GLYCOSIDES |  |                   |  |  |
| Reactions on starch       |  |                   |  |  |
| preparation of paste      | Place 1 g of starch in a 100 ml flask and add  |                   |  |  |
|                           | 50 ml of water. The mixture is heated for 5    |                   |  |  |
|                           | minutes constantly stirring                    |                   |  |  |
| with iodine solution      | 1 drop of Lugol's solution is added to 2 ml of |                   |  |  |
|                           | cooled starch paste                            |                   |  |  |
| with Fehling's reagent    | To 2 ml of starch paste, add 2 drops of an     |                   |  |  |
|                           | aqueous solution of CuSO4 (solution A) and 2   |                   |  |  |
|                           | drops of an alkaline solution of ferric salt   |                   |  |  |
|                           | (solution B) and heat in a water bath          |                   |  |  |
| Reactions on cellulose    |  |                   |  |  |
| with iodine solution      | A drop of iodine solution is added to the      |                   |  |  |
|                           | cellulose powder                               |                   |  |  |
| with iodine in a          | Appropriate reagents are added to the          |                   |  |  |
| solution of zinc          | cellulose powder                               |                   |  |  |
| chloride and potassium    |  |                   |  |  |
| iodide                    |  |                   |  |  |
| Reaction on inulin        |  |                   |  |  |
| with α-naphthol           | A drop of α-naphthol and a drop of             |                   |  |  |
| (Molish reaction)         | concentrated sulfuric acid are applied to a    |                   |  |  |
|                           | cross-section of the raw material (chicory     |                   |  |  |
|                           | root, dandelion, echinacea, amaranth)          |                   |  |  |
| Reactions on mucus        |  |                   |  |  |

| with a solution of alkali            | 2 drops of sodium hydroxide solution are  |   |
|--------------------------------------|---|---|
|                                      | applied to a cross section of the altea root  |   |
| with concentrated                    | 1 ml of 10% infusion of althea root and 2   |   |
| hydrochloric acid                    | drops of concentrated hydrochloric acid are   |   |
|                                      | added to the test tube  |   |
| with a solution of lead              | Add 2 ml of a solution of lead acetate to 2 ml  |   |
| acetate                              | of a 10% infusion of althea root  |   |
|                                      | FATS AND FATTY SUBSTANCES   |   |
| Reaction on seed oils (B             |   | T |
| with nitric acid and                 | 2 ml of the tested oil is poured into the test  |   |
| 0.15% resorcinol                     | tube, 1 ml of nitric acid and 0.15% solution of   |   |
| solution                             | resorcinol in benzene are carefully layered.  |   |
| D (1 1 11                            | The contents are vigorously stirred   |   |
| Reaction on seed oils                |   |   |
| with water and                       | Place 2.5 ml of oil in a test tube, carefully   |   |
| concentrated sulfuric                | add 1 ml of a cooled mixture of equal   |   |
| and nitric acids                     | volumes of water and concentrated   |   |
| Donation on figh oil                 | sulfuric and nitric acids   |   |
| Reaction on fish oil with chloroform | 0.1 a of fat is dissalyad in 1 ml of ablamafarm and   |   |
| with chlorotorin                     | 0.1 g of fat is dissolved in 1 ml of chloroform and 5 ml of a solution of stibium (III) chloride is added |   |
| Reaction on lanolin                  | 3 IIII of a solution of sublum (III) chloride is added  |   |
| with concentrated                    | 0.1 g of fat is dissolved in 5 ml of chloroform and   |   |
| sulfuric acid                        | carefully layered in a test tube with 5 ml of   |   |
| Sulfulic acid                        | concentrated sulfuric acid  |   |
|                                      | TERPENOIDS. IRIDOIDS. BITTERS   |   |
| with Stahl's reagent                 | 0.5 ml of Stahl's reagent is added to 1 ml of   |   |
| with Stain's reagent                 | the extract, the mixture is heated in a water   |   |
|                                      | bath for 2 minutes.   |   |
| with Trim-Hill's                     | 0.5 ml of Trim-Hill's reagent is added to 1 ml  |   |
| reagent                              | of the extract, the mixture is heated in a water  |   |
|                                      | bath for 2 minutes.   |   |
|                                      | ESSENTIAL OILS  |   |
| Reactions on aldehydes               |   |   |
| obtaining oximes                     | 3 drops of an alcoholic solution of   |   |
|                                      | hydroxylamine chloride (15 g of   |   |
|                                      | hydroxylamine chloride in 100 ml of 80%   |   |
|                                      | alcohol) and a few drops of methylene orange  |   |
|                                      | are added to 2 drops of essential oil   |   |
| nitroprusside reaction               | 5-10 drops of essential oil are mixed with the  |   |
|                                      | same number of drops of sodium  |   |
|                                      | nitroprusside solution and 3 drops of 5%  |   |
|                                      | alkali solution   |   |
| Reactions on phenols                 |   |   |
| reaction with iron III               | 3-4 drops of iron III chloride solution are   |   |
| chloride                             | added to 1 ml of concentrated alcohol solution  |   |
|                                      | of essential oil  |   |
| reaction of the                      | 3-4 ml of 25% sodium hydroxide solution and   |   |
| formation of azo dyes                | 1-2 drops of diazotized sulfanilic acid are   |   |
|                                      |   |   |
| Reactions on azulenoge               | added to 1 ml of essential oil  |   |

| Ehrlich-Muller reaction  | 5 drops of essential oil are mixed in a test tube   |          |
|--------------------------|---|----------|
|                          | with 1 ml of reagent and heated in a water  |          |
|                          | bath  |          |
| Sabetay's reaction       | Dissolve 5-10 drops of essential oil in 1-2 ml  |          |
|                          | of chloroform and add 0.1-1 ml of a 5%  |          |
|                          | solution of bromine in chloroform drop by   |          |
|                          | drop.   |          |
|                          | TRITERPENOIDS. STEROIDS. SAPONINS   |          |
| foaming test             | 2-3 ml of the aqueous extract of the raw  |          |
|                          | material is vigorously shaken for 1 minute.   |          |
| Precipitation Reactions  | ,   |          |
| with barite water        | 3-4 drops of barite water are added to 1 ml of  |          |
| William Saries Water     | aqueous extract in a test tube  |          |
| with lead acetate        | 3-4 drops of 10% lead acetate solution are  |          |
| With four decide         | added to 1 ml of aqueous extract in a test tube   |          |
| with cholesterol         | 1 ml of 1% alcohol solution of cholesterol is   |          |
| solution                 | added to 1 ml of alcohol-water extract in a test  |          |
| Solution                 | tube  |          |
| Color reactions          | tuoc  |          |
| the Lafon reaction       | 1 drop of a 10% solution of copper sulfate, 1 ml  |          |
| the Laton reaction       | of concentrated sulfuric acid is added to 2 ml of   |          |
|                          | the alcohol-water extract in a test tube and  |          |
|                          | heated carefully  |          |
| Salkovsky's reaction     | 1 ml of chloroform and 5-6 drops of   |          |
| Salkovsky s Icaction     | concentrated sulfuric acid are added to 2 ml of   |          |
|                          | the alcohol-water extract in a test tube  |          |
| reaction with a solution | 0.5 ml of a saturated solution of stibium (V)   |          |
| of stibium (V) chloride  | chloride in chloroform is added to 1 ml of the  |          |
| of stibium (v) chioride  | alcohol-water extract in a test tube  |          |
| Vanillin-sulfuric acid   |   |          |
|                          | 1 ml of 0.5% alcohol solution of vanillin, 3-4 drops of concentrated sulfuric acid are added to |          |
| assay                    | 2 ml of alcohol-water extract in a test tube and  |          |
|                          |   |          |
|                          | heated in a water bath at a temperature of 60 °C.   |          |
| Determination on the cl  | nemical nature of saponins  |          |
| foaming reaction         | Take 2 measuring tubes of the same diameter   |          |
| roaming reaction         | with ground stoppers. 5 ml of 0.1 M   |          |
|                          | hydrochloric acid is poured into one of them,   |          |
|                          | and 5 ml of 0.1 M sodium hydroxide solution   |          |
|                          | is poured into the other. Add 0.5 ml of   |          |
|                          | aqueous extract to both test tubes and shake  |          |
|                          | both test tubes with the same intensity for 1   |          |
|                          | min.  |          |
|                          | CARDIOGLYCOSIDES  | <u> </u> |
| Reactions on the steroid | part of cardioglycosides  |          |
| Lieberman-Burchard       | The dry residue is dissolved in 1 ml of acetic  |          |
| reaction                 | anhydride, transferred to a dry test tube, and 2  |          |
| 1 Caction                | drops of concentrated sulfuric acid are   |          |
|                          | -   |          |
| Rosenheim reaction       | carefully added along the wall  1 ml of trichloroacetic acid in ethanol is added                |          |
| Roseinienn reaction      | to 1 ml of chloroform extract   |          |
|                          | to 1 mi of chiofololili extract   | <u> </u> |

| Reactions on the lacton | e ring  |  |
|-------------------------|---|--|
| the Kedde reaction      | The dry residue is dissolved in 2 ml of a 3%  |  |
| the fredde federion     | solution of 3,5-dinitrobenzoic acid and 1 ml of                                       |  |
|                         | a 1M sodium hydroxide solution is added   |  |
| Raymond's reaction      | The dry residue is dissolved in 1 ml of a 3%  |  |
| Raymond's reaction      | solution of m-dinitrobenzene in benzene and 2   |  |
|                         |   |  |
|                         | drops of an alcoholic solution of potassium   |  |
| ~                       | hydroxide are added   |  |
| Legal's reaction        | The dry residue is dissolved in 1 ml of 5%  |  |
|                         | sodium nitroprusside solution and 2 drops of  |  |
|                         | 10% sodium hydroxide solution are added   |  |
|                         | ydrate part of the molecule   |  |
| the Keller-Kiliani      | The dry residue is dissolved in 1 ml of acetic  |  |
| reaction                | acid with traces of ferric sulfate (III), and 1 ml                                    |  |
|                         | of concentrated sulfuric acid is poured over the                                      |  |
|                         | walls of the test tube. Do not shake the  |  |
|                         | contents of the test tube!  |  |
| with Fehling's reagent  | 0.5 ml of a 1% solution of hydrochloric acid is                                       |  |
| 2 2                     | added to 2 ml of the obtained extract and   |  |
|                         | heated on a water heater for 1 hour. After that,                                      |  |
|                         | a few drops of a 10% solution of sodium   |  |
|                         | hydrogen chloride are added to the test tube,   |  |
|                         | and then 1 ml of Fehling's reagent and heated   |  |
|                         | in a boiling water bath   |  |
|                         | PHENOLIC COMPOUNDS  |  |
| Reactions on arbutin    | THENOEIC COM OUNDS  |  |
| with iron (II) sulfate  | A few crystals of iron (II) sulfate are added to                                      |  |
| ( )                     | 1 ml of extract   |  |
| with sodium             | 4 ml of ammonia solution and 1 ml of a 10%  |  |
| phosphoric              | solution of sodium phosphoric-molybdic acid   |  |
| molybdenum solution     | in hydrochloric acid are added to 1 ml of the   |  |
|                         | extract   |  |
| Reaction on salidroside | CALLOT  |  |
| with 10% lead acetate   | 2-3 drops of a 10% solution of lead acetate are                                       |  |
| solution                | added to 1 ml of aqueous infusion of raw  |  |
| Solution                | materials, the precipitate is filtered, 2 drops of                                    |  |
|                         | a 1% alcoholic solution of 1-nitroso-2-   |  |
|                         |   |  |
|                         | naphthol and 3 drops of concentrated nitric   |  |
|                         | acid are added to the filtrate  COUMARINS AND CHROMONES                               |  |
| lactone test            | 1 ml of alkalized extract is diluted with four  |  |
| lactoric test           |   |  |
|                         | times the amount of water, the mixture is neutralized with a 20% solution of sulfuric |  |
|                         |   |  |
| ana diva fameration     | acid.   |  |
| azo dye formation       | 3-5 drops of freshly prepared solution of   |  |
| reaction                | diazotized sulfanilic acid are added to 1 ml of                                       |  |
|                         | alkalized extract   |  |
| reaction with potassium | Add 15 ml of purified water to 1.0 g of   |  |
| hydroxide (on           | crushed raw material and boil in a water bath   |  |
| chromones)              | for 15 minutes. The resulting extract is  |  |
|                         | filtered through cotton wool into a porcelain   |  |

|                          | 1  |  |
|--------------------------|--|--|
|                          | cup and evaporated. A crystal of potassium       |  |
|                          | hydroxide is added to the dry residue            |  |
|                          | FLAVONOIDS                                       |  |
| cyanidine reaction       | 2-3 drops of concentrated hydrochloric acid      |  |
|                          | and 1-2 shavings of magnesium metal are          |  |
|                          | added to 1 ml of the extract                     |  |
| with alkali              | 1-2 drops of 10% alcohol-water solution of       |  |
|                          | potassium or sodium hydroxide are added to 1     |  |
|                          | ml of the extract                                |  |
| with iron (III) chloride | 2-3 drops of 10% iron (III) chloride solution    |  |
|                          | are added to 1 ml of extract                     |  |
| with lead acetate        | 3-5 drops of a 10% solution of basic lead        |  |
|                          | acetate are added to 1 ml of the extract         |  |
|                          | QUINONES   |  |
| Borntraeger reaction     | Place 1.0 g of crushed raw material in a flask,  |  |
| $\mathcal{E}$            | pour 10 ml of 10% alcoholic NaOH solution,       |  |
|                          | boil for several minutes and filter. After       |  |
|                          | cooling, the filtrate is acidified with 10% HCl  |  |
|                          | to a slightly acidic reaction (according to the  |  |
|                          | universal indicator), transferred to a           |  |
|                          | separatory funnel and extracted with 10 ml of    |  |
|                          | chloroform. After settling, the chloroform       |  |
|                          | layer turns yellow (anthraquinone                |  |
|                          | derivatives). 5 ml of chloroform extract is      |  |
|                          | shaken in a test tube with 5 ml of 5% alcohol    |  |
|                          | solution of NH4OH. Note the color of the         |  |
|                          | ammonia layer.                                   |  |
|                          | TANNINS  |  |
| with gelatin solution    | A 1% gelatin solution is added dropwise to 2     |  |
| ··· 8                    | ml of the extract under study, avoiding its      |  |
|                          | excess   |  |
| with alkaloid solution   | 1% alkaloid solution (quinine hydrochloride,     |  |
| Williamarora Boration    | cytisine) is added drop by drop to 2 ml of the   |  |
|                          | tested extract                                   |  |
| with iron-ammonium       | 4-5 drops of iron-ammonium alum solution are     |  |
| alums                    | added to 2 ml of the tested hood                 |  |
| with lead acetate        | 4 ml of a 10% solution of acetic acid and 2 ml   |  |
| solution                 | of a 10% solution of lead acetate are added to   |  |
| Solution                 | 2 ml of the tested hood. The precipitate         |  |
|                          | formed is filtered. Add a few drops of 1%        |  |
|                          | iron-ammonium alum solution to the filtrate      |  |
| with bromine water       | The reaction is carried out under a hood! A      |  |
| with bioinine water      | 2% solution of bromine water is added            |  |
|                          | dropwise to 5 ml of the tested extract until the |  |
|                          | smell of bromine appears.                        |  |
|                          | ALKALOIDS  |  |
| Cananal presinitation    |  |  |
| General-precipitation r  |  |  |
| with the Wagner-         | a solution of iodine in a solution of potassium  |  |
| Bouschard reagent        | iodide   |  |
| with Mayer's reagent     | a mixture of solutions of mercury dichloride     |  |
|                          | and potassium iodide                             |  |

| with Dragendorff's      | a solution of basic bismuth nitrate, potassium |  |
|-------------------------|--|--|
| reagent                 | iodide and acetic acid                         |  |
| with Bertrand's reagent | 1% aqueous solution of silicon-tungstenic acid |  |
| with Sonnenstein's      | 1% aqueous solution of phosphoric-molybdic     |  |
| reagent                 | acid   |  |
| with picric acid        | 1% aqueous solution of picric acid             |  |
| with tannins            | 0.1% aqueous solution of tannin                |  |

**Task 3.** Carry out the TLC of rutine according to the requirements of the SPhU. Draw a diagram of the chromatogram. Compare the results with the SFC monograph.

| Mobile phase        | Detection           |
|---------------------|---------------------|
|                     |                     |
| The tested solution | Comparison solution |
|                     |                     |

| Chromatogram schen | ne |
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- 1. Name the qualitative reactions to the main groups of BAS.
- 2. What methods of chromatographic analysis are used to determine the main groups of BAS.
- 3. What types of analysis are used in pharmacognostic practice?
- 4. What are the histochemical and microchemical reactions for the detection of inulin, essential and fatty oils, tannins, mucus, alkaloids, anthracene derivatives, starch?

| star |  |
|------|--|
| 5.   | Explain the essence of the chromatographic method of research. |
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herbs, briquettes, tinctures, extracts, essential oils, teas). Task 1. Define and give examples of different technological forms of herbal medicines. Teacher's signature

TOPIC 3: Classification and prescription of herbal remedies (medicinal products,

- List the characteristics of medicines that are included in the main sections of monographs on medicines.
- Provide a list of names that belong to medicinal products according to the Law 2. of Ukraine "On Medicinal Products".

  Provide a classification of herbal medicines according to their purpose.

| <i>3</i> . 4. | What is the difference between tinctures and extracts?                    |  |  |
|---------------|---|--|--|
| 5.            | Features of the preparation of herbal medicines (teas, briquettes, etc.). |  |  |
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**TOPIC 4:** Development of quality control methods for herbal medicines for various purposes. Additional quality indicators for the plant-based medicinal products and drugs.

**TOPIC 4.1:** Development of quality control methods for herbal medicines for various purposes.

| ask 1. De                             | fine the concept of "quality control methods" (QCM)                 |
|---------------------------------------|---|
|                                       |   |
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|                                       |   |
| ask 2. De                             | scribe the algorithm and structure of the QCM for herbal medicines. |
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- 1. Name the main documents that regulate the quality standards of medicines.
- 2. What factors determine the suitability of the drug for use?
- 3. Name national and international organizations that introduce standards and guidelines necessary to ensure the quality of pharmaceutical products.

| 4.          | guidelines necessary to ensure the quality of pharmaceutical products.  What is the modern international concept of quality assurance of drugs? |  |  |
|-------------|---|--|--|
| 5.          | What are the functions of the State Pharmacopoeia?  |  |  |
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| <b>TOPIC 4.2:</b> Additional quality indicators for LRS and medicines based on it.  |
|---|
| <b>Task 1.</b> Provide examples of additional quality indicators for the drug substance and medicinal products based on it  |
|   |
|   |
|   |
|   |
| <b>Task 2.</b> Define the "Swelling Index" and select monographs of the State Financial Institution in which the standardization of the drug or drug products is carried out according to this index. Determine the "Swelling Index" according to the SPhU in mucus-containing MPM. |
| Definition:   |
|   |
| Monographs of SPhU:   |
| Observations:   |
|   |
| <b>Task 3.</b> Define the "Foam Number" and select monographs of the SPhU in which the standardization of the drug or drug products is carried out according to this indicator. Determine the "Foam Number" according to the SPhU for a saponin-containing drug product.            |
| Definition:   |
| Monographs of SPhU:   |
|   |

| Observations:   |
|---|
| Task 4. Define the "Bitter Index" and select monographs of the SPhU in which the standardization of the MPM or drugs by this index is carried out. Determine the "Bitter Index" according to the SPhU in the MPM containing iridoids. |
| Definition:   |
| Monographs of SPhU:   |
| Observations:   |
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- 1. Name the main documents that regulate the quality standards of medicines.
- 2. Define additional quality indicators such as "swelling index", "foam number", "bitter index", etc.
- 3. What factors determine the suitability of a drug for use?
- 4. Name national and international organizations that introduce standards and guidelines necessary to ensure the quality of pharmaceutical products.
- What is the modern international concept of quality assurance of drugs? 5. What are the functions of the State Pharmacopoeia? 6.

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**TOPIC 5:** Development of choleretic and diuretic herbal preparations and standardization of their components.

Task 1. Offer a composition of choleretic or diuretic herbs

| MPM, MP, Family | The main BAS | Pharmacological activity | Daily doses<br>(ratio) |
|-----------------|--------------|--------------------------|------------------------|
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
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| Eng.            |              |                          |                        |
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| Eng.            |              |                          |                        |
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| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |
| Lat             |              |                          |                        |
| Eng.            |              |                          |                        |

| Lat                   |          |                         |                       |
|-----------------------|----------|-------------------------|-----------------------|
| Eng.                  |          |                         |                       |
| Lat                   |          |                         |                       |
| Eng.                  |          |                         |                       |
| Lat                   |          |                         |                       |
| Eng.                  |          |                         |                       |
| Lat                   |          |                         |                       |
| Lai                   | <u> </u> |                         |                       |
| requirements of the S |          | for the developed fee i | n accordance with the |
| Identification A      |          |                         |                       |
|                       |          |                         |                       |
|                       |          |                         |                       |
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| r.1                   |          |                         |                       |
| Identification B      |          |                         |                       |
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| Standardization of the qualitative composition and quantitative content of the developed collection of BAS |
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| 1. Name the MPM with choleretic effect |
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| 2. Name the MPM with diuretic effect   |
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**TOPIC 6:** Development of collections of antiseptic and anti-inflammatory action based on raw materials containing essential oils and standardization of its components.

Task 1. Offer the composition of the collection of antiseptic and anti-inflammatory

action based on raw materials containing essential oils

| MPM, MP, Family | The main BAS | Pharmacological activity | Daily doses (ratio) |
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| <b>Task 2.</b> Propose qu with the requiremen Identification A | ods for the dev | veloped collectio | n in accordance |
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| tandardization of the qualitative composition and quantitative content of BAS of the eveloped collection | he |
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- 1. What are essential oils? Their localization in plants.
- 2. Justify the classification of LRS containing essential oils.
- 3. Specify ways to use LRS containing essential oil.
- 4. Name the possible impurities and their distinguishing features with the following types of medicinal plants: chamomile, mountain arnica, bitter wormwood.
- 5. Name the methods of quantitative determination of essential oils depending on their physical properties.
- 6. What groups of chemical compounds can be detected in essential oils using qualitative reactions?
- 7. What are the pharmacopoeial methods of establishing the authenticity and good quality of essential oils?
- 8. For what purpose is the ether number determined in essential oils before and after acetylation?

| 9. Name the medicinal products prepared from essential oil raw materials, their pharmacological action and application in medicine. |
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**TOPIC 7:** Development of collections of hypoglycemic action and tea of sedative action and standardization of their components.

Task 1. Offer the composition of the collection of hypoglycemic and sedative action

| MPM, MP, Family | The main BAS | Pharmacological activity | Daily doses<br>(ratio) |
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| <b>Task 2.</b> Propose qual with the requirements Identification A | for the developed col | llection in accordance |
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| Standardization of the qualitative composition and quantitative content of BAS of the developed collection |
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| 1. Na | ame MPM wit | th hypoglyc                           | emic action           | • |   |
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| 2. N  | ame MPM wit | th sedative a                         | action.               |   |   |
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# **TOPIC 8:** Development of vitamin tea and standardization of its components.

Task 1. Offer the composition of tea with vitamin effect

| MPM, MP, Family | The main BAS | Pharmacological activity | Daily doses<br>(ratio) |
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| <b>Task 2.</b> Propose quarequirements of the S Identification A | for the developed tea i | n accordance with the                 |
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| Standardization of the qualitative composition and quantitative BAS content of the developed tea |
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# Questions for student self-preparation for the practical lesson: Technology of making teas from MPM

| . Name MP  | y of making to<br>M with vitami | n effect.   |                                       |                                       |                                       |                                       |
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**TOPIC 9:** Development of laxative briquettes and tinctures to improve digestion and standardization of their components.

**Task 1.** Offer a composition of briquettes with a relaxing effect or a tincture to improve digestion

| MPM, MP, Family | The main BAS | Pharmacological activity | Daily doses (ratio) |
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|      | lity control methods cording to the require      |                                    | quettes or tinctures to |
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|      | e qualitative composit<br>or tincture to improve | tion and quantitative le digestion | BAS content of the      |
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- 1. Technology of making MPM briquettes.
- 2. Name the MPM with a relaxing effect.
- 3. Name MPM containing bitterness.

| 4. Name MPM with antacid ef | fect. |      |  |
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| <b>TOPIC 10:</b> Analysis of the thick extract according to the monograph of the SPhU "Extracts".   |
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| <b>Task 1.</b> Define "Thick extract" in accordance with the SPhU and specify the requirements of the SPhU for obtaining and standardizing such extracts. |
| Definition:   |
| Requirements of the SPhU for obtaining and standardization:   |
| Task 2. Standardize the thick extract in accordance with the requirements of the SPhU   |
| Observation:  |
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| 1. Technology of preparation of extracts from MPM. |  |                                       |  |  |  |
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## **Рекомендована література**

#### Базова

- 1. Фармакогнозія: базовий підручник для студентів вищих фармац. навч. закл. (фармац. ф-тів) ІVрівня акредитації / В.С. Кисличенко, І.О. Журавель, С.М. Марчишин та ін.; за ред. В.С. Кисличенко. Харків: НфаУ: Золоті сторінки, 2015. 736 с. (Національний підручник). <a href="https://gnosy.nuph.edu.ua/wp-content/uploads/2020/10/Фармакогнозія 2015.pdf">https://gnosy.nuph.edu.ua/wp-content/uploads/2020/10/Фармакогнозія 2015.pdf</a>
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#### <mark>Допоміжна</mark>

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