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DETERMINATION OF THE AMOUNT OF WATER-SOLUBLE VITAMINS IN THE ROOT OF RUBIA TINCTORUM L.

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ABSTRACT
In this article, the amount of water-soluble vitamins in the root of the medicinal plant Rubia tinctorum was determined by the high-performance liquid chromatography method, and information on its physicochemical properties and spectral characteristics and its use in medicine is highlighted.

Keywords: *Rubia tinctoru L*, anthracene, alizarin, ruberythric acid, galiosin, purpurin, xanthopurpurin, pseudopurpurin, rubiadin-glucoside, ibericin, urinary tract stone, kidney stone.

INTRODUCTION
Today, comprehensive measures are being taken to organize scientific research at a high level and to supply the national pharmaceutical market with high-quality drugs in the direction of developing the creation of effective drugs based on local raw materials. On the basis of the measures implemented in this direction, a number of important practical results are being achieved in terms of organizing the development of competitive preparations based on natural plant raw materials. It is of urgent importance to study the biology of *Rubia tinctorum* plant species and to create a base of primary raw materials for the preparation of cheap and high-quality drugs that can replace imports by separating natural medicines from local raw materials. Such preparations can be isolated from the medicinal plant *Rubia tinctorum* and used in medical practice and folk economy.

THEORETICAL PART
Rubia tinctorum L. belongs to the rubicaceae family. The plant is native to Mediterranean countries. It is found in Ukraine, Moldova, southeast of the European part of Russia, Azerbaijan, Georgia, Armenia, Dagestan and Central Asia. In Uzbekistan, it mainly grows along streams, among bushes, along canals, in fields and

gardens[1-4].

Rubia tinctorum is a perennial herb with a height of 30-150 cm. The rhizome is long, creeping, branched, cylindrical, thick, jointed, many-headed. The stem is several, four-lobed, jointed, coarse and looped. covered with feathers. The leaf is oval-ovate, shiny, the veins on the lower side are covered with rough hairs with loops, they are arranged in bundles of 4-6 on the stem with a very short band. The flowers are small, greenish-yellow, gathered in a semi-umbrella growing from the axils of the leaves, forming a flower cluster. The calyx is not clearly known, the corolla is 5, united, funnel-shaped, the paternity is 5, the maternal node is 2-digit, located below. The fruit is a 1-2-seeded, globular, first red, then black wet fruit. It blooms in June-August, the fruit ripens in August-September, the above-ground part of *Rubia tinctorum L.* Fig. 1. [1-4]

Figure 1. Surface and root part of Rubia tinctorum plant

The finished product of the *Rubia tinctorum* plant consists of rhizomes and root pieces. The thickness of rhizome pieces is 2-18 mm, the upper side is painted in reddish-brown color. When it is cut crosswise, the bark layer is red-brown, and the wood part is red. The product has a characteristic weak smell, sweeter at first, and then a slightly sour and bitter taste. The rhizome turns the water brownish-red, the moisture content of the harvested Rubia tinctorum plant is 13%, total ash is 10%, other parts of the plant (stem, leaves, etc.) are 1.5%, organic impurities more than 1% and mineral impurities more than 1%, the amount of anthraglycosides (those combined in the form of glycosides) in the product should not be less than 3%. [1-4]

The rhizome of *Rubia tinctorum* contains 5-6% of anthracene compounds (alizarin, ruberythric acid, galiosin, purpurin, xanthopurpurin, pseudopurpurin, rubiadin-glucoside, munistin, lucidin, ibericin, etc.).

Rubia tinctorum plant has antispasmodic and diuretic effects in medicine and softens kidney stones (phosphates). Therefore, medicinal preparations are used in ureteral stones, kidney stones, gallstones and gout.[1-4]

Vitamins were first discovered in 1880 by N.I. Lunin. He came to the conclusion that in addition to proteins, carbohydrates, fats and mineral substances that provide normal life, there are also organic substances necessary for life. Vitamins are called substances necessary for life (vita means life, vitamin means life amines). Vitamins are small molecular organic compounds that are extremely important in the life and activity, growth and reproduction of organisms. Vitamins have the following properties: - they are not synthesized in the human body; - does not participate in the formation of structures; - when they are not enough in the body, the metabolism is disturbed and causes specific diseases; - vitamins consumed with food affect biochemical processes in the body as coenzymes.

A change in the amount of vitamins in the body leads to the following conditions: 1. Avitaminosis - diseases caused by the lack of some vitamin in the body. 2. Hypovitaminosis - diseases caused by vitamin deficiency. 3. Hypervitaminosis - diseases caused by an excess of vitamins.

So far, more than thirty vitamins have been identified, and they are divided into three groups: water-soluble vitamins, fat-soluble vitamins, and vitamin-like substances. Water-soluble vitamins include: Vitamin B1, Vitamin B2, Vitamin B6, Vitamin B12, Vitamin PP, Biotin, Vitamin N, Vitamin C, Vitamin P. Fat-soluble vitamins include: vitamin A, vitamin D, vitamin E, vitamin K. Water-soluble vitamins: Vitamin B1 - thiamine, is the first vitamin to be isolated in pure form. Lack of vitamin B1 - avitaminosis occurs in beriberi or polyneuritis. Vitamin B1 causes a violation of carbohydrate metabolism. Vitamin B1 is a coenzyme of pyruvate decarboxylase. This vitamin is found in large quantities in eggs, meat, and peas. The daily requirement of the body is 1-3 mg. Vitamin B2 is called riboflavin and it has a yellow color. Deficiency of this vitamin leads to avitaminosis, colds of the mucous membrane of the oral cavity, impaired vision, and anemia. Riboflavin is a coenzyme of flavin enzymes. A person receives 65-70% of this vitamin through dairy, meat and bread products, 30-35% through vegetables and fruit products. Daily requirement - 2 mg. Vitamin B6 (pyridoxine). Vitamin B6 deficiency causes a disturbance in the metabolism of amino acids and leads to a skin disease called dermatitis. It also causes anemia and growth retardation. This vitamin is a coenzyme of enzymes that catalyze the reamination reaction of amino acids. Vitamin compounds: pyridoxine, pyridoxal and pyridoxamine. Vitamin B6 is mainly found in meat, fish, and grain products. Adults need 2 mg of this vitamin. is Vitamin PP (nicotinic acid). Nicotinic acid is important in metabolic processes in living organisms. It is a coenzyme of dehydrogenase enzymes that catalyze oxidation-reduction reactions by becoming part of NAD and NADF. It is a derivative of pyridine: nicotinic acid and nicotinamide. Vitamin PP

deficiency causes pellagra disease. The nervous system and digestive system are disturbed. Vitamin PP is found in cereals and vegetables. The daily requirement for an adult is 7 mg. Vitamin C (Ascorbic acid). Ascorbic acid is not synthesized in the body of humans, monkeys and guinea pigs, so they consume vitamin C with ready-made food. If there is a lack of products rich in vitamin C in the food, people and some animals develop ringworm. Milks cause bleeding, blood accumulation under the skin. If vitamin C is not taken into the body, it leads to death. Vitamin C increases the body's antioxidant capacity. Ascorbic acid serves as an intermediate that transfers hydrogen in oxidation-reduction reactions in living organisms. Vitamin C is abundant in plums, oranges, lemons, dill and other plants. The daily requirement of an adult is 0.2-1 g. [1-6].

Water-soluble vitamins contained in the root of Rubia tinctorum plant were determined using the high-performance liquid chromatography (HPLC) method. 5-10 g of Rubia tinctorum roots are taken on an analytical scale and placed in a 300 ml flat flask. 50 ml of 40% ethanol solution is added to it. The mixture was heated under vigorous stirring for 1 h, equipped with a magnetic stirrer, reflux condenser, and then stirred at room temperature for 2 h. The mixture is cooled and filtered. 25 ml of 40 percent ethanol was added to the remaining part and re-extracted 2 times. The filtrates were combined and filled to the mark with 40% ethanol (5-10%) in a 100 ml volumetric flask. The resulting solution is spun in a centrifuge at a speed of 7000 rpm for 10 minutes. The resulting solution was taken from the upper part for analysis. Working solutions of water-soluble vitamins with a concentration of 1 mg/ml were prepared. For this purpose, 50.0 mg of each vitamin standard is taken on an analytical balance and dissolved in 40% ethanol in a 50 ml volumetric flask and filled to the mark.

Acetate buffer system and acetonitrile were used as an eluent for the determination of water-soluble vitamins in the roots of the Rubia tinctorum plant with YuSSX. Chromatographic conditions:-Chromatograph Agilent-1200 (equipped with an autodoser);-Column Exlipse XDB C 18 (obraschenno-faznyy), 5 µm, 4.6 x250mm; - Diode matrix detector (DAD), 250 nm identified; - Flow rate 0.8 ml/min; - Eluent acetate buffer: acetate buffer: acetonitrile: 0-5 min 96:4, 6-8 min 90:10, 9-15 min 80:20, 15-17 min 96:4, thermostat temperature 25 0C, -5 µl amount entered. First, a working standard solution was prepared in the chromatograph, then a solution prepared from the root of the Rubia tinctorum plant was introduced, and the amount of vitamins was determined by comparison.(Figure 2)

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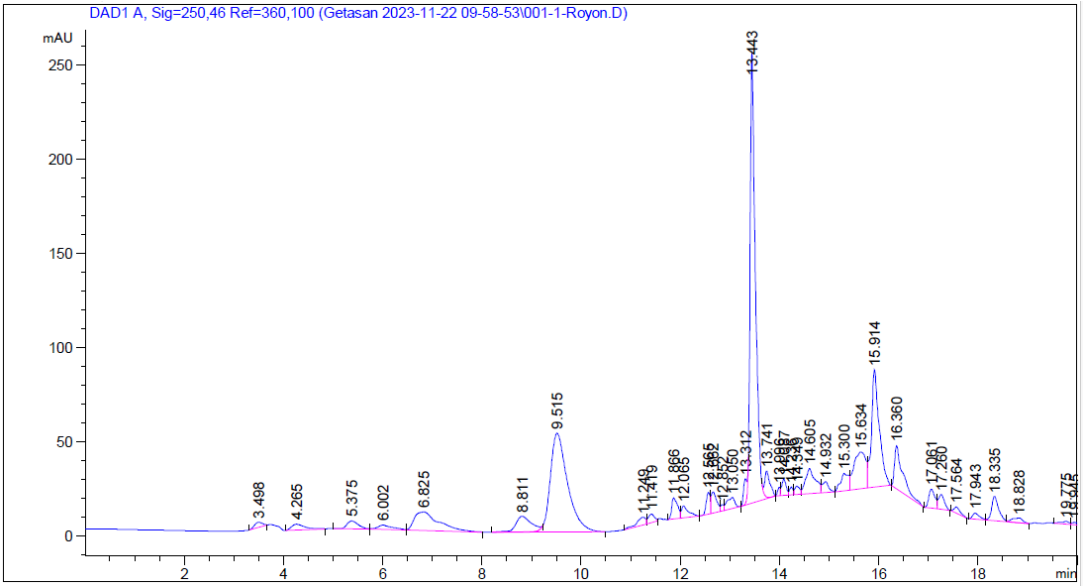

Figure 2. Solution chromatography of *Rubia tinctorum* root solution to determine the vitamin content of Rubia tinctorum root

Chromatography shows that in the roots of *Rubia tinctorum* plant B1=1.566249 mg., B2=0.914181 mg., B6= 2.615811 mg., B9= 2.901639 mg., (PP) B3= 0.112042 mg. Vitamin C was found to be in the amount of 0.177824 mg.

CONCLUSION

Rubia tinctorum root contains B1=1.566249 mg., B2=0.914181 mg., B6= 2.615811 mg., B9= 2.901639 mg., (PP) B3= 0.112042 mg. Vitamin C was found to be in the amount of 0.177824 mg.

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