# СУЧАСНІ ДОСЯГНЕННЯ ФАРМАЦЕВТИЧНОЇ НАУКИ В СТВОРЕННІ ТА СТАНДАРТИЗАЦІЇ ЛІКАРСЬКИХ ЗАСОБІВ І ДІЄТИЧНИХ ДОБАВОК, ЩО МІСТЯТЬ КОМПОНЕНТИ ПРИРОДНОГО ПОХОДЖЕННЯ

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**Conclusions:** Based on the analysis of the world's leading pharmacopoeias, a draft specification for a dietary supplement containing magnesium citrate was created, which includes methods for identification and quantification using both pharmacopoeial and non-pharmacopoeial physicochemical methods for use in laboratory conditions.

Pharmaco-technological tests for this form in the form of capsules were proposed for further use and testing in laboratory conditions. Criteria for controlling microbiological purity according to the requirements of the Eur.Ph. for the presented form in the form of capsules were determined.

**References**

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## METHOD FOR QUANTITATIVE DETERMINATION OF HEMOGLOBIN IN URINE

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**Introduction.** It is known that in glomerulonephritis, kidney and bladder tumors, pyelitis, cystitis, as well as during menstruation, a blood dye may appear in the urine - hemoglobin, which enters the urine from the blood flowing in the kidneys and urinary tract, or separately (hemoglobinuria). Along with hemoglobin, blood formed elements, often altered – hematuria, can also enter the urine. Hematuria occurs in many pathological conditions, so it is always necessary to find out the cause of a positive test result. To definitively determine the cause of this pathology, it is necessary to have a method for quantitative determination of hemoglobin in the urine.

The basis of the known chemical methods for determining hemoglobin in urine is the peroxidase properties of hemoglobin or myoglobin or acetic acid-ether extract (hematin) to change the color of the chromogen as a result of its catalytic oxidation by hydrogen peroxide - amidopyrine or benzidine, as well as ozonated turpentine - guaiac resin (Weber reaction) [1]. Hemoglobin in the urine is also determined by special chemical diagnostic test strips, for example, «Hemophan». The indication zone contains a stabilized organic hydroperoxide, an acid buffer and a chromogen, which in the presence of hemoglobin is oxidized by hydroperoxide to form colored products in an intense blue color. The strips are highly sensitive and allow for the reliable detection of 5 erythrocytes/μl of urine. The test complements microscopic examination of urine [2].

**Research objective**: Development of a new highly sensitive method for quantitative determination of hemoglobin in urine by the chemiluminescence method based on the catalytic oxidation of luminol with hydrogen peroxide.

**Materials and methods.** LLC «RPE Chemstandard» (Kharkiv, Ukraine) Luminol sodium salt (NaHL). Molecular Formula $C_8H_6N_3NaO_2$. Molecular Weight 199.14 g/mol. Melting Point 319-320°C. $w$=98%. Human hemoglobin produced by «Simko Ltd.» (Lviv, Ukraine). All reagents were analytical reagent grade and double

distilled water (DDW) was used throughout these experiments. The 0.01 $mol \cdot L^{-1}$ NaHL stock solution was prepared by dissolving 0.20320 g NaHL with 1 mL 1 $mol \cdot L^{-1}$ NaOH solution and diluted with double distilled water to 100 ml. Working standard solutions of NaHL were freshly prepared from the stock solution by appropriate dilutions before use and adjusted its pH with 0.1 $mol \cdot L^{-1}$ NaOH. A stock solution of hemoglobin (Hb) 0.1 mg/ml was prepared by dissolving 10 mg of Hb in 75 ml of 0.5% $K_2HPO_4$ by heating to 323 K. The volume was adjusted to 100 ml with DDW. A working solution with a hemoglobin concentration of 1.0 μg/ml was prepared by diluting the stock solution with DDW (or child's urine). Hydrogen peroxide ($H_2O_2$) 58 % (wt.) high pure preparation (produced by LTD «Inter-Syntes», Boryslav, Ukraine). Working solution of $H_2O_2$ 0.058 % (wt.) ($1.7 \cdot 10^{-2}$ $mol \cdot L^{-1}$) was obtained by the appropriate dilution of the stock solution exactly 1000 times. The working solution can be used throughout the day. The intensity of chemiluminescence ($I_{Chl}$) was measured in discrete mode in relative units (rel. units) on a device with a photomultiplier FEU-84-A, a small current meter IMT-0.5 and a high-speed (time constant 0.1s) potentiometer-recorder. In a quartz cuvette 0.50 ml of hemoglobin solution was added to the mixture of alkaline NaHL solution and hydrogen peroxide using a P-1 pipette dispenser and the kinetic curve of chemiluminescence intensity ($I_{Chl}$) - time (min) was recorded. The dispenser is arranged in a removable holder that isolates the photocathode of the photomultiplier from extraneous light, thus allowing operation in normal lighting. All experiments were performed at a temperature of 293 K.

**Results and their discussion.** In the course of studies, it was found that under optimum conditions: ($c$(NaOH) = 0.05 $mol \cdot L^{-1}$, $c(H_2O_2)$ =0,015 $mol \cdot L^{-1}$, $c$(NaHL)= $1 \cdot 10^{-4}$ $mol \cdot L^{-1}$, $C$(Hb) = 0.05-0.5 $\mu g \cdot mL^{-1}$) Hb exhibits an enhance effect on intensity of CL in NaHL – $H_2O_2$ system. A sensitive, rapid and relatively simple chemiluminescence (CL) method for the assay of hemoglobin (Hb) in urine has been proposed. The scheme of chemical transformations that form the basis of the proposed analysis method is shown in Fig. The determination of Hb was achieved by monitoring CL intensity from the luminol-hydrogen peroxide reaction in the presence of Hb. Under optimal conditions, the maximum CL intensity was found to be proportional to concentration in the range from 0.05 to 0.5 μg/ml. The regression equation: $I_{CL}$ = (74.24±6.67) C, r=0.999; LOQ (10S) = 0.084 μg/ml. The accuracy and reliability of the proposed method were validated by conducting recoveries experiments.

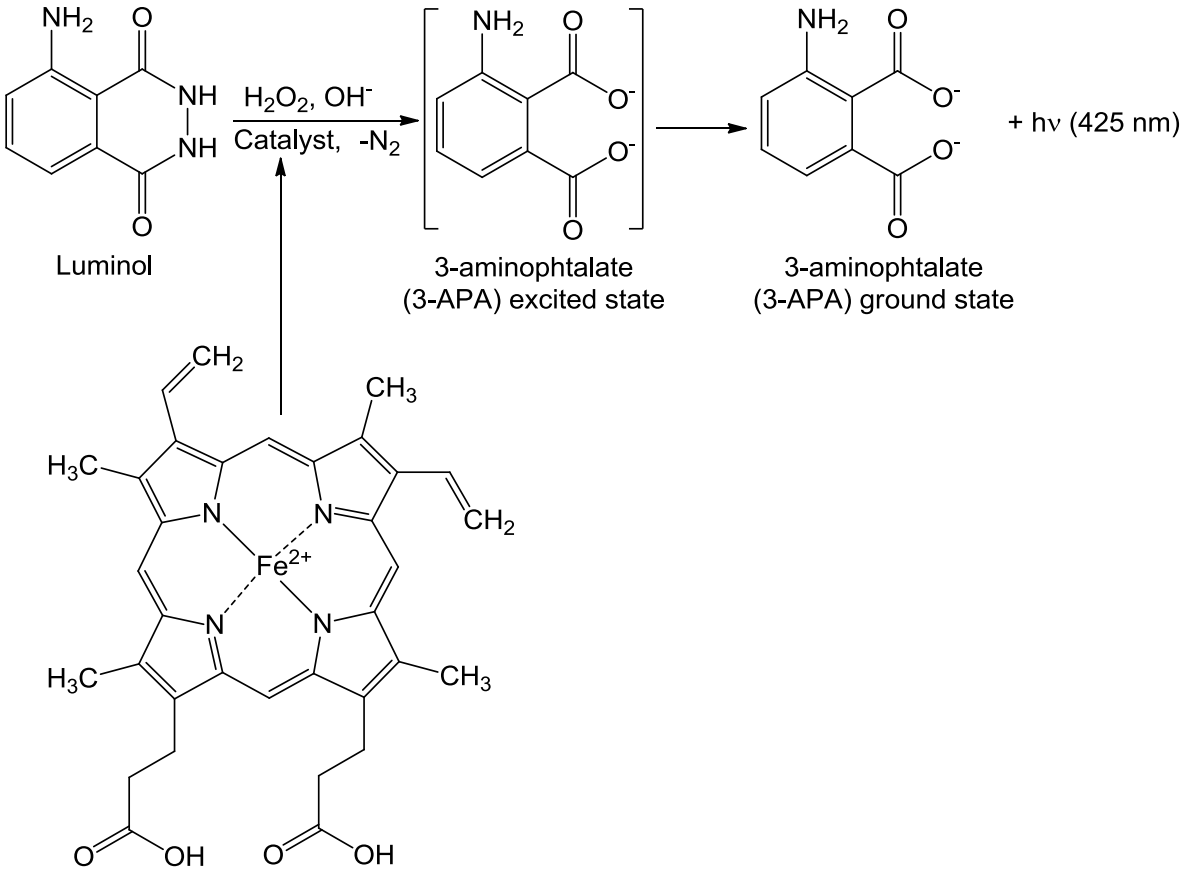


Fig. Scheme of the process of occurrence of chemiluminescence of luminol in the presence of hemoglobin

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## PHENOLIC COMPOUNDS IN FRUITS OF *SAMBUCUS NIGRA*

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**Introduction.** Elderberry's (*Sambucus nigra* L.) fruits are widely used as ingredients in food supplements, food products, beverages, and personal care products. This species is naturally distributed in Europe and has been introduced into different countries as a source of fruit and flowers. This species can adapt to various soil types, including sand, loam, and clay. However, bushes' yield is significantly better in well-drained, fertile soils with a pH of 6.0–6.5 [1]. So, the resources of this species are significant, even in natural habitats.

**Materials and methods.** Three elderberry cultivars –'Haschberg', 'Dana', and 'Sambo 5' and two clones SN1 and SN2 were selected from the genetic resource collection of the Botanical Garden at Vytautas Magnus University. Three replicates of 0.2 kg of fresh fruit were prepared for each genotype. All samples were stored in plastic bags in a freezer at −28 °C until analysis. Using appropriate methods, the amounts of phenolic compounds, flavonoids, proanthocyanidins, and phenolic acids