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MINISTRY OF HEALTH OF UKRAINE
NATIONAL UNIVERSITY OF PHARMACY
faculty for foreign citizens' education
department chemistry of natural compounds and nutriciology

QUALIFICATION WORK

on the topic: « PHYTOCHEMICAL STUDY OF CARAMBOLA FRUITS »

Prepared by: higher education graduate of group
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Kharkiv – 2023 year

ANNOTATION

The qualification work presents the results of the study of the qualitative composition and quantitative content of biologically active substances (Polysaccharides, Organic acids, Flavonoids, Fatty acids, Pigments), quality indicators, technological parameters of Carambola.

Qualification work consists of an introduction, literature review, experimental part, general conclusions, list of used literature sources and is set out on 41 pages, includes 11 tables, 13 figures, 45 literature sources.

Key words: Carambola, qualitative analysis, quantitative analysis.

АНОТАЦІЯ

В кваліфікаційній роботі представлено результати дослідження якісного складу і кількісного вмісту біологічно активних речовин (полісахаридів, органічних кислот, флавоноїдів, жирних кислот, пігментів), показники якості, технологічні параметри карамболи.

Кваліфікаційна робота складається зі вступу, огляду літератури, експериментальної частини, загальних висновків, переліку використаних літературних джерел та викладена на 41 сторінці, включає 11 таблиць, 13 рисунків, 45 джерел літератури.

Ключові слова: карамбола, якісний аналіз, кількісний аналіз.

TABLE OF CONTENTS

INTRODUCTION

Rationale. In recent years, exotic fruits have gained considerable popularity and firmly entered our everyday life. The variety of exotic fruits is simply amazing, but still a large part of them is little known to us and not studied enough. Fresh fruits are important in human nutrition. They are the main sources of vitamins, macro- and microelements, easily digestible sugars, organic acids, enzymes, etc. Fruits have many medicinal properties, they are used to treat colds, to improve the work of the gastrointestinal tract [31, 39, 42]. Therefore, it is urgent to carry out more detailed pharmacognostic studies of carambola fruits.

Purpose. The purpose of the Master's thesis was the phytochemical study of Carambola fruits.

Tasks of the research. The following tasks were set:

- analyze the modern literature regarding the botanical characteristic, chemical composition, practical use, application in medicine carambola;
- determine the quality indicators of Carambola fruits;
- define the presence of biologically active compounds;
- determine the quantitative content of biologically active compounds.

The subject of the research: definition of quality indicators, the study of the qualitative composition and the quantitative content of biologically active compounds in Carambola fruits.

The object of the research: the phytochemical study of Carambola fruits.

Methods of the research. The paper chromatography method and the well-known chemical reactions were used to identify biologically active compounds in Carambola fruits. Quantitative determination was carried out by gravimetric, titrimetric, spectrophotometric, and gas chromatography methods.

The practical significance and scientific novelty of the results. The results of the phytochemical study of Carambola fruits are represented in scientific work.

The quality indicators of Carambola fruits were determined: the weight loss on drying, the total ash, and the extractable matter.

Polysaccharides, organic acids, flavonoids, chlorophyll A, chlorophyll B, and carotenoids were identified via qualitative reactions and paper chromatography method in Carambola fruits.

The quantitative content of the following biologically active compounds was determined: polysaccharides, organic acids, flavonoids, fatty acids, chlorophyll A, chlorophyll B, and carotenoids.

The experiment results were processed by statistical methods according to the requirements of the State Pharmacopoeia of Ukraine.

Approbation of the research results.

The results to Identification and quantitative determination of organic acids in carambola fruits. *Current approaches of pharmaceutical science in development and standardization of medicines and dietary supplements that contain components of natural origin :* The Proceedings of the V International Scientific and Practical Internet-Conference (April 14, 2023, Kharkiv).

The structure and scope of the qualification work – consists of an introduction, a literature review, an experimental part, general conclusions, a list of used literary sources, laid out on 41 pages, including 11 tables, 13 figures, 45 literature sources.

CHAPTER 1. BOTANICAL CHARACTERISTICS, CHEMICAL COMPOSITION, PRACTICAL USE, APPLICATION IN MEDICINE CARAMBOLA

1.1. Botanical characteristics, distribution, distinguishing features of *Averrhoa bilambi* L. from *Averrhoa carambola* L.

Averrhoa carambola (*Averrhoa carambola* L.) is a family of *Oxalidaceae*. An evergreen tree whose family is considered to be Indonesia, this tree also grows in Sri Lanka. Cultivated varieties of carambola are grown and bred in Indonesia, India, Vietnam, the Philippines, South China, the USA (the state of Florida). Common sorrel (*Oxalis acetosella* L.), which grows in Asia and the coniferous forests of Europe, is the closest relative of Carambola [13, 31, 32, 39, 42].

Classification of *Averrhoa carambola*:

Scientific Name: Averrhoa carambola

Kingdom *Plantae* – Plants

Subkingdom *Tracheobionta* – Vascular plants

Superdivision *Spermatophyta* – Seed plants

Division Magnoliophyta – Flowering plants

Class *Magnoliopsida* – Dicotyledons

Subclass Rosidae

Order Geraniales

Family *Oxalidaceae* – Wood-Sorrel family

Genus *Averrhoa* Adans. – averrhoa

Species *Averrhoa carambola* L. – carambola

Carambola is traditionally considered to originate from Malaysia, although it has also been speculated to be a tropical American species introduced to Asia by the Spanish galleons. Carambola has a wider climate range and can grow within the latitudinal range from 32°N to 30°S and withstand growing in both the hot

humid tropics and subtropical countries including Egypt and Israel, and can tolerate short periods of freezing temperatures as low as −3°C. It prefers well-drained soils ideally between pH 5.5–6.5 but can tolerate pH between 5 and 8.5. In recent years, it has been extensively distributed and widely cultivated in most parts of the world (Figure 1.1), e.g., Asia countries including China and India, Africa countries including Madagascar and Tanzania, North America countries including Mexico and Honduras, Oceania countries including Australia and French Polynesia, South America countries including Brazil and Bolivia, etc [14, 27, 31, 39, 40, 42].

Fig. 1.1. Geographical distribution of Carambola throughout the world

Carambola is also called: Carambola – the Spanish folk name of the tree; Sarambola, starfruit – in English; carambolier – in French; carambolero – in Spanish; kalamarak, kamaranga – in India; related to Averrhoa bilimbi, carambola is also called kamias in Tagalog.

Carambola is a small, evergreen shrub or tree, 5-12 m high, with a short trunk and a thick and rounded crown with many branches that hang down. The wood is white, thin-layered, of medium hardness, and acquires a red color over the years. The leaves are pinnately compound, from dark green to pale green in color,

soft, 15-30 cm long, arranged alternately around the branches; consists of 5-11 oppositely located oval or ovate-oblong leaves 3-9 cm long and up to 5 cm wide. The upper side of the leaves is smooth, and the lower side is white and has fine pubescence. The leaves react to light and can curl up at night, they are also sensitive to a sharp blow, and when the carambola is shaken it can curl up.

The flowers are fragrant, fluffy, mauve, purple or purple with red veins, located in small clusters, growing in the axils of the leaves at the end of the branches or sometimes on large stems and trunks; the width of the flower is about 6-10 mm, with 5 petals and sepals that have curved ends; blooms continuously, but most actively - from spring to autumn. Each tassel is attached to the tree with red stems [1, 7, 9, 13, 32, 31, 39, 40, 42].

Fruits are oblong in shape, 5-15 cm long and up to 9 cm in diameter; has 5 longitudinal ribs, the depth of which is approximately 2 cm. The average weight of the fruit is 75-120 g. Carambola fruits are covered with a thin waxy film and have a yellow or orange-yellow color. Juicy fruits are yellow inside, do not contain fibers, have a clear and almost transparent texture when fully ripe, and a star shape when cross-sectioned. The flesh of the fruit has a sweet-sour or sweet taste, most reminiscent of a harmonious combination of green gooseberry, apple, grape and cucumber (Figure 1.2.) [1, 7, 9, 13, 13, 31, 32, 39, 40, 42].

Inside the fruit there are up to 12 flat seeds, brown in color, which are 0.6 cm to 1.5 cm long, but some growers cultivate carambola seedless. Fruits ripen 60-70 days after flowering. The sweetest fruits come out when they ripen naturally on the tree. In industrial cultivation, the fruits are harvested when they become pale green and begin to turn yellow. Unripe fruits can be stored in the refrigerator for up to 3 weeks, they ripen at room temperature, but contain less sugar. Carambola bears fruit twice a year: from April to May and from September to October, at other times the plant can produce single flowers and fruits [1, 7, 9, 13, 31, 32, 39, 40, 42].

Fig. 1.2. Carambola: A – whole plants;
B – fruits;
C – flowers and woods;
D – leaves

Distinguishing features of Bilimbi (*Averrohaa bilimbi* L.) from Carambola (*Averrhoa carambola* L.)

The genus Averroa consists of two species: Bilimbi (*Averrohaa bilimbi* L.) and Carambola (*Averrhoa carambola* L.). Carambola fruits are also known as star fruits and this is considered to be the more important difference between the two species.

Averrhoa bilimbi (*Averrhoa bilimbi* L.), or Cucumber tree, native to Malaysia. The plant grows very quickly, has a tree-like shape with evergreen leaves. The height of bilimbi can reach 6 meters, but in areas with a tropical

climate there are specimens up to 10 meters. The trunk is single, short, branches into several vertical branches.

The petioles of the leaves are long, densely pubescent at the base. The leaves are complex, 30-60 cm long, consist of 11-40 leaflets, which are placed oppositely and directed downwards. The length of each leaf plate reaches 2-15 cm, and the width is from 1,2 to 5 cm, the shape is elongated-ovoid or oval-oblong, asymmetrical, rounded at the base, and pointed at the top, green above and pale green below, covered with down. As soon as it starts to get dark, the leaves start to curl up in pairs into a tube.

The flowers are small, fragrant, five-petalled, yellowish-green or purple in color, the surface of which has dark purple spots. Loose inflorescences are collected from them, which include up to 15-20 buds. They grow directly on the trunk or old thick branches, practically devoid of leaves, flowers grow in the axils of the leaves. After flowering, the fruits ripen, which resemble small cucumbers, they grow in bunches and have an elliptical shape with weakly expressed five ribs; the surface of the fruit is thin and shiny, the length ranges from 4 to 10 cm long. At the base, the fruits are covered with a small star-shaped cup, and the remains of flowers are visible at the ends (Figure 1.3.).

Fig. 1.3. Bilimbi fruits

Unripe fruits have a bright green color with very crispy flesh, with ripening the color changes to yellow, ivory or almost white. One adult tree can ripen up to 100 fruits per season [42].

1.2. Chemical composition

Carambola fruits contain proteins, fats, water, polysaccharides, pectin substances (14%), alkaloids, flavonoids (apigenin-6-C-β-glucofuranoazide and apigenin-6-C-β-1-fucopyranoside, also called carambolaflavone) [4, 14], proanthocyanidins, tannins (epicatechin, gallic acid) [8, 19, 33, 37, 39]; saponins, in particular phytosterols (β-sitosterin, campesterol, lupeol, isofurosterol) [14, 33].

Fresh carambola fruits contain fatty acids: unsaturated (omega-3 and omega-6, palmitic, stearic) and saturated fatty acids (oleic, linoleic, linoleic) [14, 33].

Simultaneously, GC-MS analysis has demonstrated that the abundant fatty acids existing in Carambola leaves were α-linolenic acid (62.04%) and oleic acid (55.44%) in fruits. Moreover, the proportion of total unsaturated fatty acids existing both in the fruits and leaves of Carambola comprise more than 77% of total fatty acid.

The fruits also contain macroelements: potassium (167.13–168.0 mg/100 g), calcium (6.37–6.40 mg/100 g), manganese (0.04–0.52 mg/100 g), sodium (3.8–3.85 mg/100 g), phosphorous (17.87–17.88 mg/100 g) and microelements: iron (0.34–0.45 mg/100 g), copper (0.19–0.45 mg/100 g), selenium, zinc (0.29–0.51 mg/100 g), manganese (0.04–0.52 mg/100 g) [13, 14, 18, 20, 32, 40].

The formulas of some compounds are shown in Figure 1.4 - 1.7.

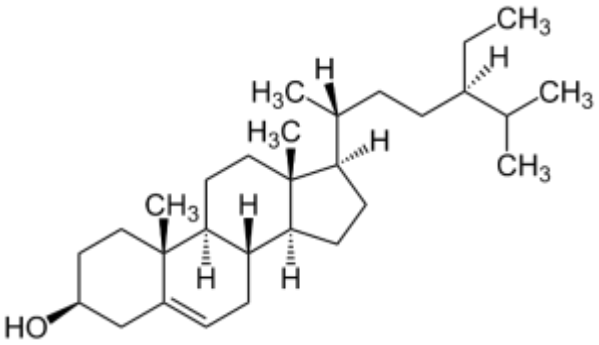


Fig. 1.4. β-sitosterin

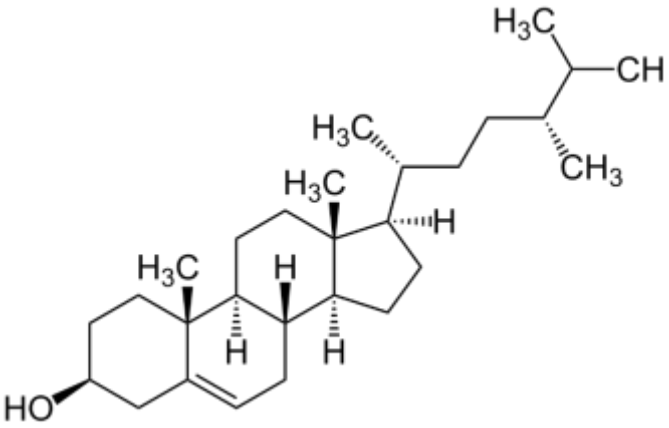


Fig. 1.5. Campesterol

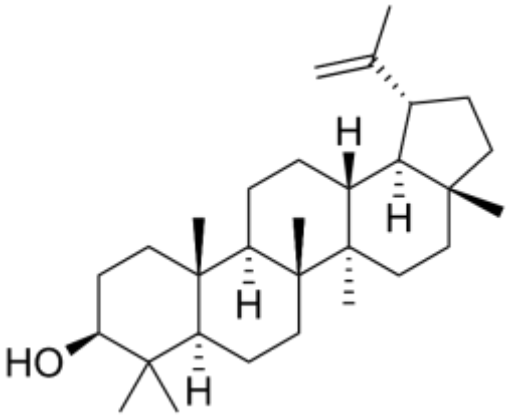


Fig. 1.6. Lupeol

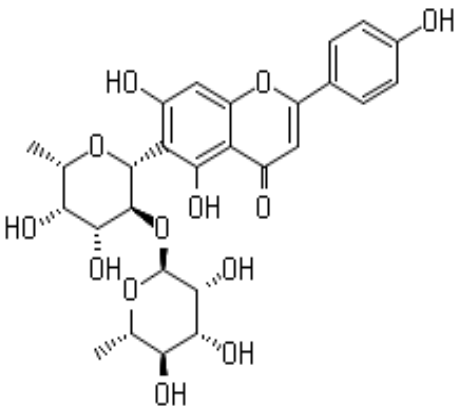


Fig. 1.7. Carambolaflavone

Amino acids were found in fruits: essential (arginine, valine, histidine, leucine, isoleucine, lysine, methionine, threonine, tryptophan, phenylalanine) and replaceable (alanine, glycine, proline, serine, tyrosine, aspartic and glutamic acids) [13, 14, 33, 40].

51 flavonoids have been separated and characterized by nuclear magnetic resonance (NMR) and mass spectrometer (MS) technologies from the leaves, fruits, and roots of Carambola. Information of these isolated flavonoids is listed in Figure 1.8. Their chemical structures were drawn by ChemBioDraw Ultra 14.0 and are described in Figure 1.9.

No.	Chemical constituents	Molecular formula	Extracts	Parts	References
Flavonoids					
1	Carambolaside R1	$C_{32}H_{36}O_{14}$	EtOH	Leaves	Yang et al. (2020a)
2	Carambolaside R2	$C_{32}H_{36}O_{14}$	EtOH	Leaves	Yang et al. (2020a)
3	Carambolaside R3	$C_{32}H_{36}O_{15}$	EtOH	Leaves	Yang et al. (2020a)
4	Carambolaside S1	$C_{41}H_{48}O_{18}$	EtOH	Leaves	Yang et al. (2020a)
5	Carambolaside S2	$C_{41}H_{48}O_{18}$	EtOH	Leaves	Yang et al. (2020a)
6	Carambolaside T1	$C_{41}H_{48}O_{18}$	EtOH	Leaves	Yang et al. (2020a)
7	Carambolaside T2	$C_{41}H_{48}O_{18}$	EtOH	Leaves	Yang et al. (2020a)
8	Carambolaside T3	$C_{41}H_{48}O_{19}$	EtOH	Leaves	Yang et al. (2020a)
9	3-Hydroxycarambolaside T1	$C_{41}H_{48}O_{19}$	EtOH	Leaves	Yang et al. (2020a)
10	3-Hydroxycarambolaside P	$C_{47}H_{58}O_{24}$	EtOH	Leaves	Yang et al. (2020a)
11	Carambolaside M	$C_{32}H_{42}O_{18}$	EtOH	Fruits	Jia et al. (2018)
12	Carambolaside N	$C_{38}H_{52}O_{22}$	EtOH	Fruits	Jia et al. (2018)
13	Carambolaside O	$C_{47}H_{56}O_{23}$	EtOH	Fruits	Jia et al. (2018)
14	Carambolaside P	$C_{47}H_{58}O_{23}$	EtOH	Fruits	Jia et al. (2018)
15	Carambolaside Q	$C_{41}H_{48}O_{19}$	EtOH	Fruits	Jia et al. (2018)
16	Carambolaside A	$C_{21}H_{24}O_9$	MeOH	Fruits	Yang et al. (2015)
17	Carambolaside B	$C_{30}H_{32}O_{10}$	MeOH	Fruits	Yang et al. (2015)
18	Carambolaside C	$C_{27}H_{34}O_{14}$	MeOH	Fruits	Yang et al. (2015)
19	Carambolaside D	$C_{36}H_{40}O_{15}$	MeOH	Fruits	Yang et al. (2015)
20	Carambolaside E	$C_{27}H_{34}O_{13}$	MeOH	Fruits	Yang et al. (2016)
21	Carambolaside F	$C_{36}H_{40}O_{14}$	MeOH	Fruits	Yang et al. (2016)
22	Carambolaside G	$C_{36}H_{40}O_{14}$	MeOH	Fruits	Yang et al. (2016)
23	Carambolaside H	$C_{36}H_{40}O_{15}$	MeOH	Fruits	Yang et al. (2016)
24	Carambolaside I	$C_{41}H_{48}O_{18}$	MeOH	Fruits	Yang et al. (2016)
25	Carambolaside Ia	$C_{41}H_{48}O_{18}$	MeOH	Fruits	Yang et al. (2016)
26	Carambolaside J	$C_{47}H_{58}O_{22}$	MeOH	Fruits	Yang et al. (2016)
27	Carambolaside Ja	$C_{47}H_{58}O_{22}$	MeOH	Fruits	Yang et al. (2016)
28	8-carboxymethyl-(+)-epicatechin methyl ester	$C_{18}H_{18}O_8$	EtOH	Fruits	Jia et al. (2018)
29	(+)-Epicatechin	$C_{15}H_{14}O_6$	EtOH	Fruits	Jia et al. (2018)
30	Epicatechin-(5,6-bc)-4β-(p-hydroxyphenyl)-dihydro-2(3H)-pyranone	$C_{24}H_{20}O_8$	EtOH	Leaves	Yang et al. (2020b)
31	Epicatechin-(7,8-bc)-4α-(p-hydroxyphenyl)-dihydro-2(3H)-pyranone	$C_{24}H_{20}O_8$	EtOH	Leaves	Yang et al. (2020b)
32	Epicatechin-(7,8-bc)-4β-(p-hydroxyphenyl)-dihydro-2(3H)-pyranone	$C_{24}H_{20}O_8$	EtOH	Leaves	Yang et al. (2020b)
33	6-(S-2-Pyrrolidinone-5-yl)-epicatechin	$C_{19}H_{19}NO_7$	EtOH	Leaves	Yang et al. (2020b)
34	6-(R-2-pyrrolidinone-5-yl)-epicatechin	$C_{19}H_{19}NO_7$	EtOH	Leaves	Yang et al. (2020b)
35	(−)-Epicatechin	$C_{15}H_{14}O_6$	EtOAc	Fruits	Gunawardena et al. (2015)
36	Pinobanksin 3-O-β-D-glucoside	$C_{21}H_{22}O_{10}$	EtOH	Fruits	Jia et al. (2018)
37	Aromadendrin 3-O-β-D-glucoside	$C_{21}H_{22}O_{11}$	EtOH	Fruits	Jia et al. (2018)
38	Helicioside A	$C_{21}H_{22}O_{12}$	EtOH	Fruits	Jia et al. (2018)
39	Taxifolin 3′-O-β-D-glucoside	$C_{21}H_{22}O_{12}$	EtOH	Fruits	Jia et al. (2018)
40	Norathyriol	$C_{13}H_8O_6$	EtOH	Fruits	Jia et al. (2018)
41	Isorhamnetin 3-O-rutinoside	$C_{28}H_{32}O_{16}$	EtOH	Fruits	Jia et al. (2018)
42	Hovertichoside C	$C_{36}H_{40}O_{14}$	MeOH	Fruits	Yang et al. (2015)
43	Isovitexin 2″-O-α-L-rhamnopyranoside	$C_{27}H_{30}O_{14}$	MeOH	Fruits	Yang et al. (2015)
44	Carambolaflavone	$C_{27}H_{30}O_{13}$	MeOH	Fruits	Yang et al. (2015)
45	(+)-Catechin	$C_{15}H_{14}O_6$	Aqueous	Roots	Liao et al. (2019a)
46	Isovitexin	$C_{21}H_{22}O_{10}$	EtOH	Leaves	Araho et al. (2005)
47	Carambolaflavone A	$C_{27}H_{30}O_{13}$	EtOH	Leaves	Araho et al. (2005), Moresco et al. (2012), Wang et al. (2018)
48	Carambolaflavone B	$C_{27}H_{30}O_{13}$	EtOH	Leaves	Araho et al. (2005), Moresco et al. (2012), Wang et al. (2018)
49	Apigenin 6-C-(2″-O-α-L-rhamnopyranosyl)-β-D-glucopyranoside	$C_{36}H_{37}O_{19}$	EtOH	Leaves	Moresco et al. (2012)
50	Cyanidin-3-O-β-D-glucoside	$C_{21}H_{21}O_{11}Cl$	EtOH	Leaves	Gunasegaran (1992)
51	Cyanidin-3, 5-O-β-D-diglucoside	$C_{27}H_{31}O_{16}Cl$	EtOH	Leaves	Gunasegaran (1992)

Fig. 1.8. Chemical components isolated and structurally identified from Carambola.

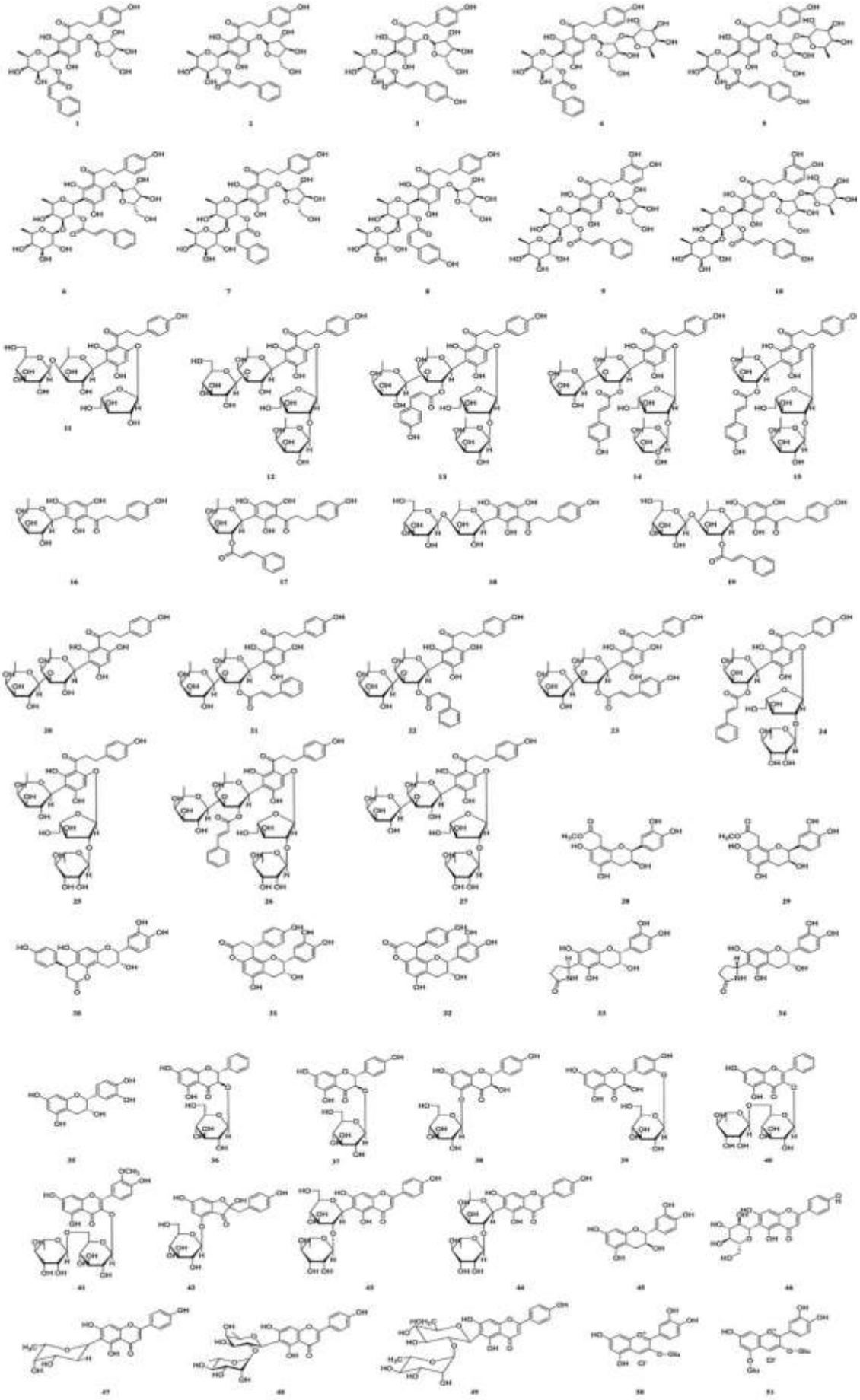


Fig. 1.9. Chemical structures of compounds isolated from Carambola

Carambola fruits are rich in vitamins, the largest amount contains ascorbic acid (38.4%), carotenoids (α-carotene and β-carotene), thiamin, riboflavin, choline, pantothenic acid, pyridoxine, α-tocopherol, nicotinic and folic acids [14, 32, 33, 36] and carboxylic acids, such as: oxalic, tartaric, citric, α-ketoglutaric acid [14, 33].

Caramboxin (CBX) is a neurotoxin whose chemical structure is similar to the amino acid phenylalanine and is a non-proteinogenic amino acid that stimulates glutamate receptors in neurons. Caramboxin is an agonist of glutamatergic ionotropic receptors NMDA and AMPA with powerful excitatory, convulsive and neurodegenerative properties (Figure 1.10) [16].

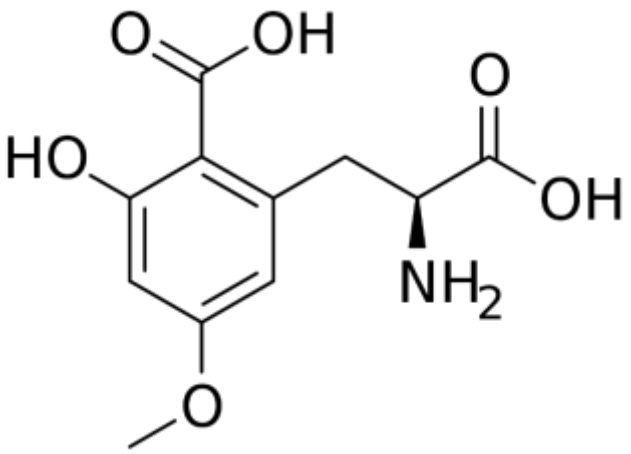


Fig. 1.10. Caramboxin

1.3. Pharmacological properties and use of carambola fruits

In Indian medicine, dried and fresh fruits are used as a hemostatic agent, flowers are used as spices, and leaves are used as vegetables because they taste like spinach. For the treatment of skin diseases, such as psoriasis, atopic dermatitis, an alcoholic extract of leaves and fruits is used [2]. Juice and dried fruits are used in fever as an antipyretic. The fruits are also considered a diuretic. An infusion of carambola flowers is a natural anthelmintic [15].

In Brazil, crushed leaves and young shoots are used in the treatment of ringworm and chicken pox. Teas and infusions from carambola leaves are used to treat headaches, coughs, and insomnia [26]. Seed powder lowers blood pressure,

relieves colic and coughing attacks. Ground roots with sugar are taken as an antidote for poisoning. In Chinese medicine, for several centuries, the fruits have been used as a natural diuretic and choleretic agent, as well as to quench thirst and increase the secretion of saliva.

Carambola fruits are used to increase the body's protective properties, remove excess fluid from tissues, lower blood pressure, quench thirst, and are a hepatoprotective agent. They are very useful for hypovitaminosis, increase lactation during breastfeeding, reduce headache and toothache. Such they are used as an aphrodisiac for men and women. Fresh carambola fruits are effective as an antiallergic and hemostatic agent [13, 25, 32, 33, 38. 40].

A decoction of fruits and leaves can be used as an antiemetic. The seeds are ground into powder and used as a sedative, hypotensive agent. Candied canned fruits are used as a choleretic and astringent for diarrhea. Antimicrobial and antioxidant properties of carambola fruits are known [12, 59, 34, 35]. Carambola leaves and fruits are widely used in traditional medicine as an antihyperglycemic agent in the treatment of diabetes [5, 6, 20, 36, 41]. Anti-inflammatory, analgesic [11, 37], antitumor [4] and hepatoprotective [8] activity of carambola fruits is known.

But the use of carambola fruits, in addition to the positive effect, has a number of contraindications. For example, in acidic varieties of carambola, due to the large amount of oxalic acid, with kidney diseases, in particular, kidney failure, after consuming fresh juice or fruits, such symptoms as sudden numbness of the limbs, muscle weakness, constant hiccups, psychomotor excitement, insomnia, paresthesias and convulsions, varying degrees of unconsciousness, neurological disorders. With such symptoms, a person should immediately undergo dialysis, because fatal cases are known [17, 23].

Carambola fruits should be consumed with caution by people with such diseases as enterocolitis, gastritis, gastric and duodenal ulcers in the acute stage, in some cases, salt metabolism disorders can be observed after consumption [5, 20, 36].

Carambola fruits are also used in cosmetology, its nutrients, in particular vitamin C, make hair strong, accelerate its growth, strengthen the roots, and moisturize the skin of the face and maintain its elasticity. That is why BAS fruits are part of shower gels, soaps, scrubs, creams. Due to the content of zinc and anti-inflammatory components, carambola is considered an effective cosmetic for oily skin, it removes sebaceous shine, treats acne, inflammation and other skin infections [24, 31, 39, 40, 42].

In addition, carambola fruits are used in perfumery, the specific aroma of this fruit is used as the top notes of refreshing men's perfumes or in bright fruity women's perfumes. Due to the high content of oxalic acid, carambola fruits are able to remove stains from clothes, and the pulp can be used to polish copper and brass products, and dentures can be whitened with the juice. The carambola derivative is used for the manufacture of furniture and wooden structures [13, 31, 39, 40, 42].

Conclusions

The analysis of literary data shows that Carambola fruits are widely used in traditional medicine for the treatment of various diseases, they show anti-inflammatory, antioxidant, hemostatic, antibacterial activity. In addition, they are widely used in cooking and cosmetology.

However, the chemical composition of carambola has not been studied enough. This became the basis for conducting pharmacognostic studies of Carambola fruits.

CHAPTER 2. DEFINITION OF QUALITY INDICATORS IN CARAMBOLA FRUITS

2.1. Weight Loss on Drying

The moisture content of the raw material is understood as the loss in mass due to hygroscopic moisture and volatile substances, which is determined in the raw material during drying to a constant mass.

Determination of the weight loss on drying in the plant raw material studied was carried out according to requirements from the State Pharmacopoeia of Ukraine [44]. 2.0 g of raw material was taken, placed in a box and dried to a constant mass.

Loss in mass during drying of raw materials (X, %) was calculated according to the formula:

$$X = \frac{(m - m_1) \cdot 100}{m}, \qquad (2.1)$$

Where: m – mass of raw material before drying in grams;

m1 – mass of raw material after drying in grams.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 2.1.

Table 2.1

Results of the weight loss on drying in Carambola fruits

m	n	X i	X mean	S2	S mean	P	t(P, n)	Confidence interval	ε, %
5	4	9,24	9,37	0,07393	0,07393	0,95	2,78	9,37 ± 0,34	3,60
		8,97							
		9,56							
		9,64							
		9,47							

As can be seen in Table 2.2, the weight loss on drying in the studied raw materials is 9,37 ± 0,34 %.

2.1 Total Ash

Ash is the non-combustible residue of inorganic compounds obtained after burning and calcination of raw materials. Total ash consists of the sum of mineral compounds inherent in this raw material and extraneous mineral impurities (soil, sand) that fall into it during collection.

Determination of the total ash in the plant raw material studied was carried out according to requirements from the State Pharmacopoeia of Ukraine (2.4.16) [44]. About 3.0 g of raw material was taken, placed in a porcelain crucible and burned in a muffle furnace. Next, the crucible with ash was placed in a desiccator, cooled and weighed.

The content of total ash (X, %) in absolutely dry raw materials was calculated by the formula:

$$X = \frac{m \cdot m_1 \cdot 100}{m \cdot (100 - W)}, \qquad (2.2)$$

Where: m1 – mass of ash, g;

m – mass of raw material, g.

W – raw material moisture, %.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 2.2.

As can be seen in Table 2.2, the total ash in the studied raw materials is 3,13 ± 0,06 %.

Table 2.2

Results of the total ash determination in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	3,12 3,07 3,12 3,15 3,21	3,13	0,00263	0,02293	0,95	2,78	3,13 ± 0,06	2,03

2.3 Extractable Matter

Determination of the extractable matter in the plant raw material studied was carried out according to requirements from the State Pharmacopoeia of Ukraine [45].

1.0 g of raw material was poured with an extractant (water, 40%, 50 %, and 96 % ethanol), insisted at room temperature for 1 hour, and then for 2 hours in a water bath. Then 25 ml of the extract was cooled, filtered, placed in a porcelain cup and evaporated. The dry residue in the cup was dried to a constant mass [7].

The content of extractive substances (X, %) in terms of absolutely dry raw materials was calculated by the formula:

$$X = \frac{m \cdot 200 \cdot 100}{m_1 \cdot (100 - W)}, \quad (2.3)$$

Where: m – mass of dry residue in grams;

m_1 – mass of raw materials in grams;

W – weight loss during drying of raw materials in percent.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 2.3.

Table 2.3

Results of the extractable matter determination in Carambola fruits

m	n	Xi	X mean	S2	S mean	P	t(P, n)	Confidence interval	ε, %
solvent – water									
5	4	8,41 8,53 8,67 8,82 9,04	8,69	0,06093	0,11039	0,95	2,78	8,69 ± 0,31	3,83
solvent – 40 % ethyl alcohol									
5	4	2,50 2,47 2,44 2,32 2,36	2,42	0,00572	0,03382	0,95	2,78	2,42 ± 0,09	3,899
solvent – 50 % ethyl alcohol									
5	4	7,02 7,18 7,27 7,35 7,49	7,26	0,03127	0,07908	0,95	2,78	7,26 ± 0,22	3,03
solvent – 96 % ethyl alcohol									
5	4	2,56 2,60 2,64 2,68 2,73	2,64	0,00442	0,02973	0,95	2,78	2,64 ± 0,08	3,12

As can be seen in Table 2.3, the best solvent for the studied raw materials is water and 50 % ethyl alcohol extracting the maximum amount of biologically active compounds (8,69 ± 0,31 % and 7,26 ± 0,22 % respectively).

The generalized results of the conducted experiment to study the yield of extractive substances in Carambola fruits are shown in Figure 2.1.

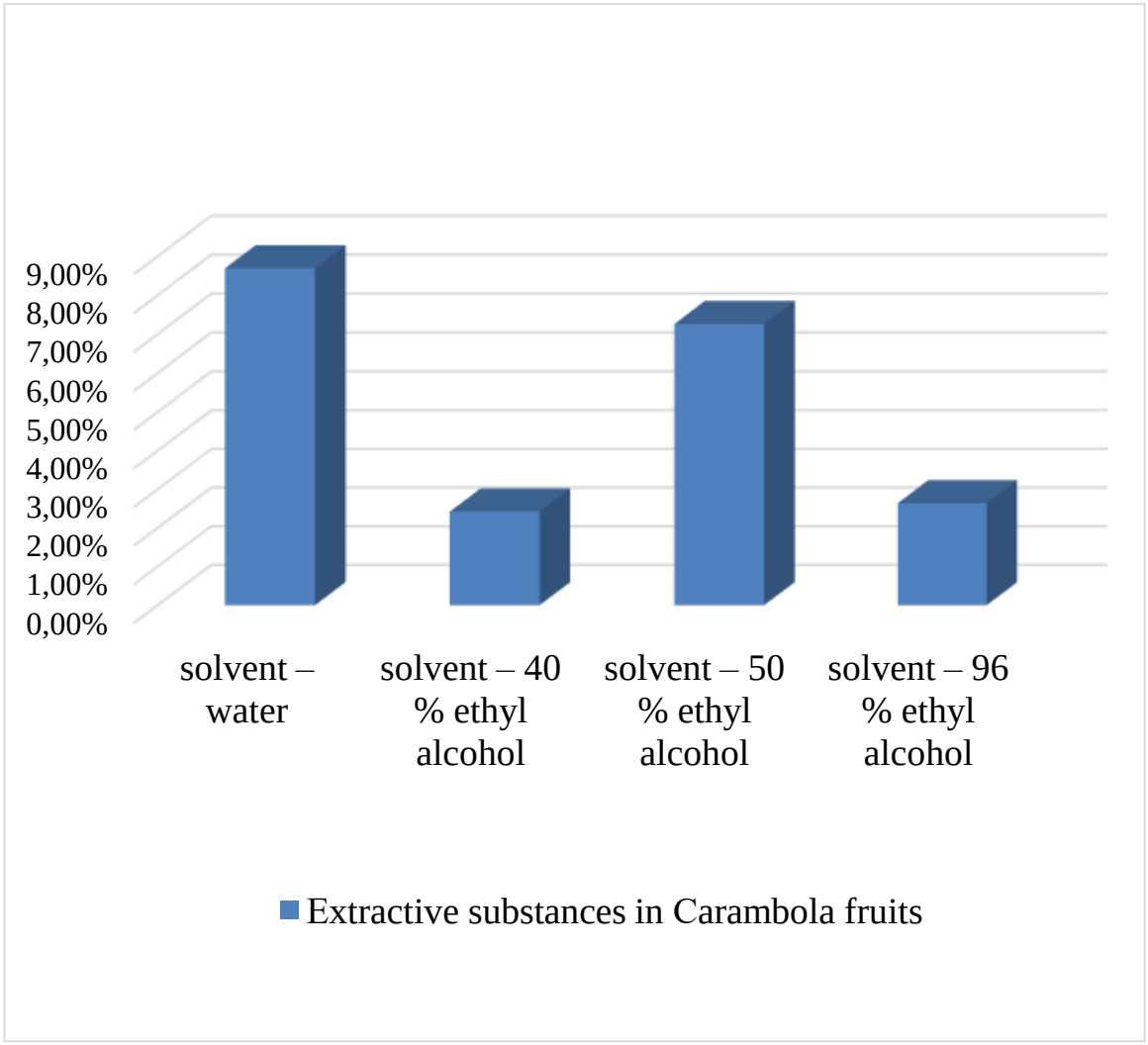


Fig. 2.1. Diagram of the determination of the content of extractive substances in Carambola fruits

Conclusions

For Carambola fruits, the quality indicators were determined according to the requirements of the SPU:

- weight loss during drying (9,37 ± 0,34 %),
- total ash content (3,13 ± 0,06 %),
- content of extractive substances during extraction with water, 40 % 50%, and 96 % ethanol. The maximal content of extractable matter in Carambola fruits is 8,69 ± 0,31 and 7,26 ± 0,22 % when used water and 50% ethanol as solvent.

CHAPTER 3. STUDY OF THE QUALITATIVE COMPOSITION AND QUANTITATIVE CONTENT OF BIOLOGICALLY ACTIVE SUBSTANCES IN CARAMBOLA FRUITS

3.1. Study of Polysaccharides

Polysaccharides exhibit various types of pharmacological activity, in particular, expectorant, laxative, anti-inflammatory, antibacterial, immunomodulatory, antioxidant, antidiabetic, antitumor activity [30]. The polysaccharides in the raw materials of Carambola have not been studied enough, so their study was relevant.

Reaction with 96% ethanol. 96% ethanol was added to the studied aqueous extract [29, 30].

An amorphous white precipitate was formed, which indicated the presence of polysaccharides in the studied extract.

Determination of the quantitative content of polysaccharides. The quantitative content of polysaccharides was determined by the gravimetric method according to the method described in the monograph State Pharmacopoeia of Ukraine 2.0.3 "Marshmallow herb ^{N}" [45].

The content of the amount of water-soluble polysaccharides (X, %) in terms of absolutely dry raw materials was calculated according to the formula:

$$X = \frac{(m_1 - m_2) \cdot 500 \cdot 100}{m \cdot (100 - W)}, \quad (3.1)$$

Where: m – weight of the raw material, g;

m_1 – mass of the filter, g;

m_2 – mass of filter with sediment, g;

W – mass loss during drying of raw materials, %.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 3.1.

Table 3.1

Results of the quantitative determination of polysaccharides in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	6,52 6,37 6,52 6,85 6,51	6,55	0,03143	0,07928	0,95	2,78	6,55 ± 0,22	3,36

As can be seen in Table 3.1, the quantitative determination of polysaccharides in the studied raw materials is 6,55 ± 0,22 %.

3.2. Study of Organic acids

Organic acids are promising compounds of plant origin with anti-inflammatory, choleretic, antibacterial properties [28, 30]. To expand knowledge on the chemical composition of carambola fruits, this class of compounds was studied.

Organic acids were studied by paper chromatography in the solvent system ethanol-chloroform-ammonia-water (70:40:20:2) with reliable samples of organic acids. After passage, the chromatogram was dried in air, treated with a sodium solution of 2,6-dichlorophenolindophenolate, followed by heating in an oven at a temperature of 100-105ºC. Organic acids appeared as yellow zones, ascorbic acid – as a pink zone on a blue background, which disappeared with time [28-30].

The chromatogram for the detection of free organic acids is shown in Figure 3.1.

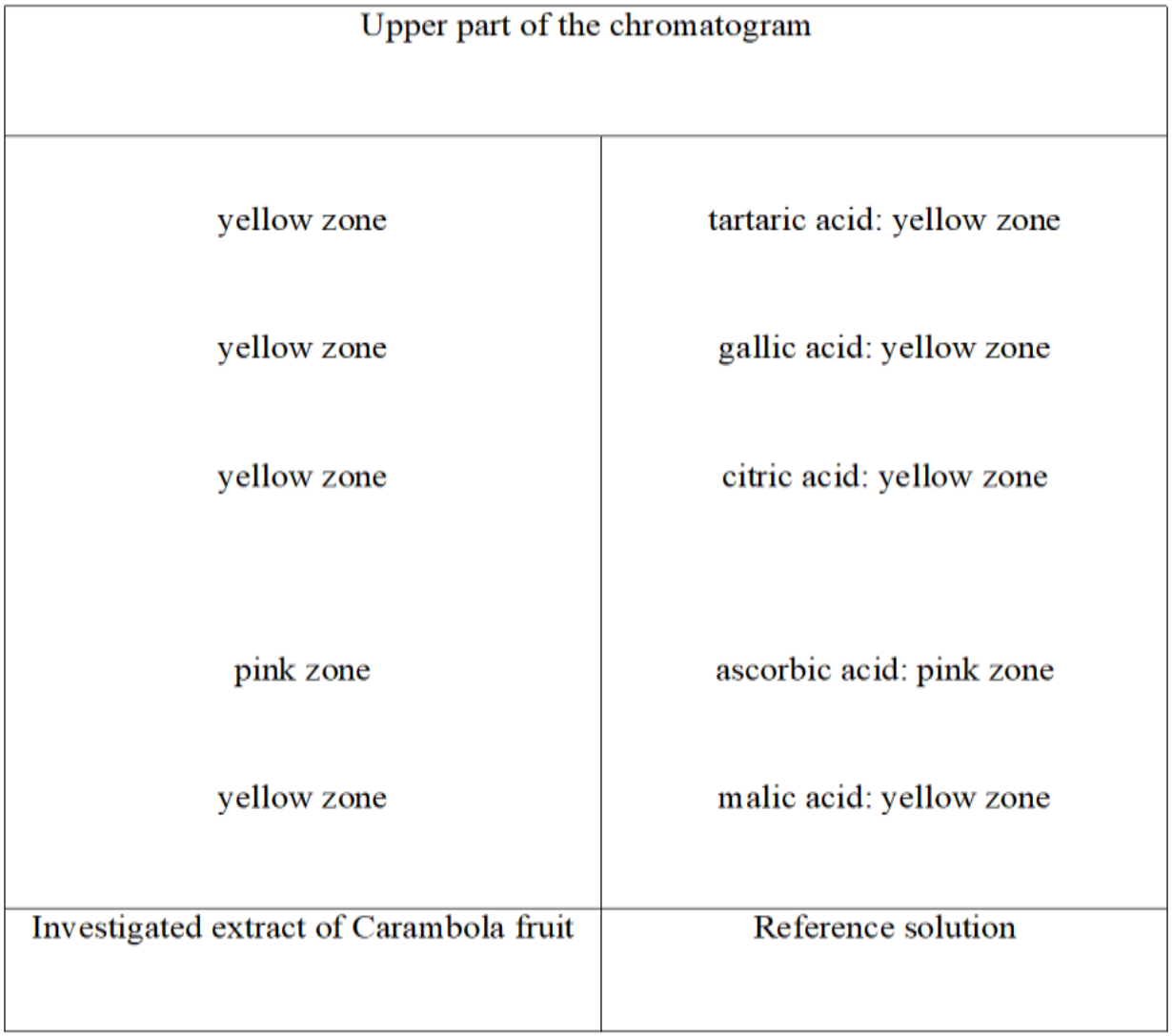


Fig. 3.1. Chromatogram for the detection of free organic acids in Carambola fruit.

As a result of the chromatographic analysis in comparison with standard samples, the following organic acids in Carambola fruit were identified: malic, citric, tartaric, gallic, and ascorbic acids.

Determination of the quantitative content of free organic acids. For the quantitative determination of the amount of free organic acids, the method of alkalimetry was used according to State Pharmacopoeia of Ukraine 2.0, appendix 1, the monograph "Rosehip fruit ^{N}" [43].

The content of the total organic acids (X, %) in terms of malic acid and absolutely dry raw material was calculated by the formula:

$$X = \frac{V \times 0{,}0067 \times 2500 \times 100}{m \times (100 - W)}, \quad (3.2)$$

Where: 0.0067 – amount of malic acid, which corresponds to 1 ml of 0.1 M sodium hydroxide solution, g;

V – volume of 0.1 M sodium hydroxide solution used for titration, ml;

m – sample weight, g;

W – weight loss during drying of raw materials, %.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 3.2.

Table 3.2

Results of the quantitative determination of free organic acids in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	4,77 4,79 4,85 4,77 4,92	4,82	0,00420	0,02898	0,95	2,78	4,82 ± 0,19	1,67

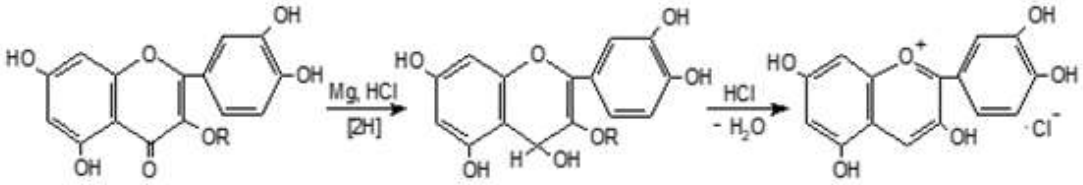
As can be seen in Table 3.2, the quantitative determination of free organic acids in the studied raw materials is 4,82±0,19 %.

3.3. Study of Flavonoids

Flavonoids are a class of biologically active substances widely distributed in the plant world, which are important active pharmaceutical ingredients with anti-

inflammatory, antioxidant, antiviral, hepatoprotective, hypotensive, immunomodulatory, antiallergic, antimicrobial activities [21, 22, 29, 30]. Therefore, given the high pharmacological activity of flavonoids, it was advisable to conduct their study of Carambola fruits.

Cyanidin reaction. Concentrated hydrochloric acid and metallic magnesium were added to ethanol extracts of Carambola fruits [29, 30].

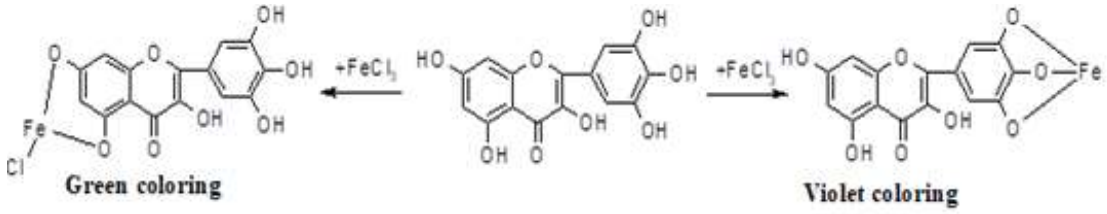


The contents in the test tube turned pink-orange.

Briant's modification of Cyanidin reaction. Butanol was added to the test colored solution, after which the contents in the test tube were diluted with water until the layers separated [29, 30].

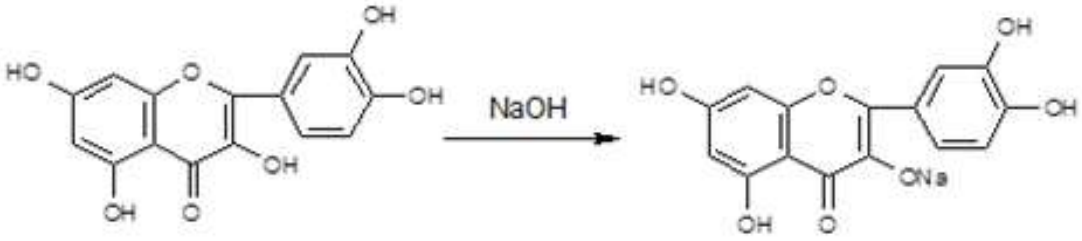
A pink-orange coloration was observed, which was the same in both layers, indicating an equal ratio of aglycones and flavonoid glycosides in the studied extracts.

Reaction with iron (III) chloride. A 10% solution of iron (III) chloride was added to the extracts [29, 30].



The extracts acquired a black-green color, which indicated the presence of phenolic compounds, including flavonoids.

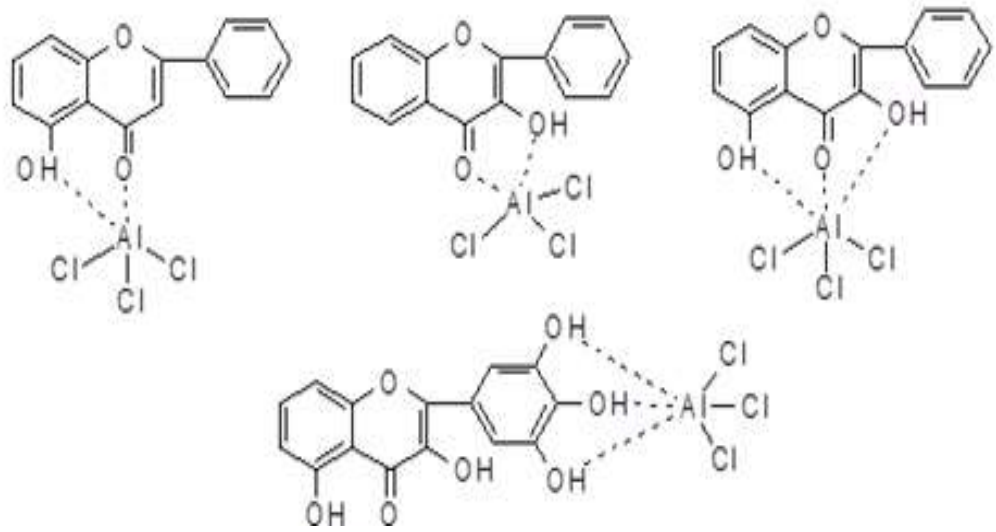
Reaction with 10% alcohol solution of alkali. A 10% ethanol solution of potassium hydroxide was added to the ethanol extracts [29, 30].



During the reaction, the extracts acquired a yellow-green color.

Reaction with 2% alcohol solution of aluminum (III) chloride. A 2% ethanolic solution of aluminum (III) chloride was added to ethanol extracts from the studied raw material [29, 30].

A green-yellow color was observed in the extracts.



Quantitative determination of the content of the sum of flavonoids. The spectrophotometric method was used to determine the amount of flavonoids. Quantitative determination of the content of the sum of flavonoids was carried out according to the State Pharmacopoeia of Ukraine 2.0 method, appendix 1, monograph "Pagoda tree flowers" [43].

The content of the sum of flavonoids (X, %) in terms of rutin was calculated by the formula:

$$X = \frac{A \times 1000}{m \times 37}, \qquad (3.3)$$

Where: A – optical density of the test solution at a wavelength of 425 nm;

m – weight of the sample of raw materials, g.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 3.3.

Table 3.3

Results of the quantitative determination of the sum of flavonoids in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	4,54	4,33	0,02643	0,07727	0,95	2,78	4,33 ± 0,20	4,66
		4,19							
		4,15							
		4,37							
		4,42							

As can be seen in Table 3.3, the quantitative determination of the sum of flavonoids in the studied raw materials is 4,33 ± 0,20 %.

3.4. Study of Fatty acids

Fatty acids are part of the structural components of cells, tissues and organs; they participate in the synthesis of biologically active compounds. Fatty acids have anticoagulant, antihypertensive, hypocholesterolemic, antioxidant, cardioprotective, anti-inflammatory effects, regulate lipid metabolism, improve the conduction of nerve impulses, and contribute to more efficient brain cells. Therefore, the study of the qualitative and quantitative composition of fatty acids in Carambola fruits is relevant [30].

Method of fatty acids' composition study is based on transformation of triglycerides of fatty acids into their methyl esters and gas chromatographic analysis of the last-mentioned. Analysis of fatty acid composition of studied extracts was carried out by gas chromatography of fatty acids' methyl esters

using gas chromatograph “Selmichrom-1” (Ukraine) with flame ionization detector. The compounds were separated in gas chromatography column of stainless steel 2,5 m in length and with inner diameter 4 mm. The column was filled in by stationary phase – inerton, having been processed by 10% diethylene glycol succinate (DEGS). The following operating parameters were set on the chromatograph: temperature of the column heating oven – 180°C; vaporizer’s temperature – 230°C; detector’s temperature – 220°C; carrier-gas flow rate (nitrogen) 30 cm3/min; sample volume – from 2 mm3 solution of acids’ methyl ethers in hexane.

Fatty acids’ methyl ethers were identified by peaks’ retention time in comparison with this index of the standard mixture. Calculation of methyl ethers’ composition was performed by internal normalization method according to the generally accepted procedure.

The fatty acids’ methyl esters were obtained by the modified Peisker’s method. The mixture of chloroform, methanol and sulfuric acid in correlation 100:100:1.

30-50 μl of lipophilic fractions were dispensed into glass ampoules with addition of 2,5 ml of methylating mixture, the ampoules bulbs were sealed and then placed into a thermostat with temperature 105°C for 3 hours. After methylation the ampoules were opened, their content was transferred to a test tube with adding zinc sulfate powder on the tip of a scalpel, 2 ml of distilled water and 2 ml of hexane for methyl ethers’ extraction. After thorough stirring and decantation, hexane extract was filtered and used for further chromatographic analysis [10].

Analysis results are shown in Table 3.5. Gas chromatogram of the fatty acid composition of lipophilic fractions from Carambola fruits in Figure 3.2.

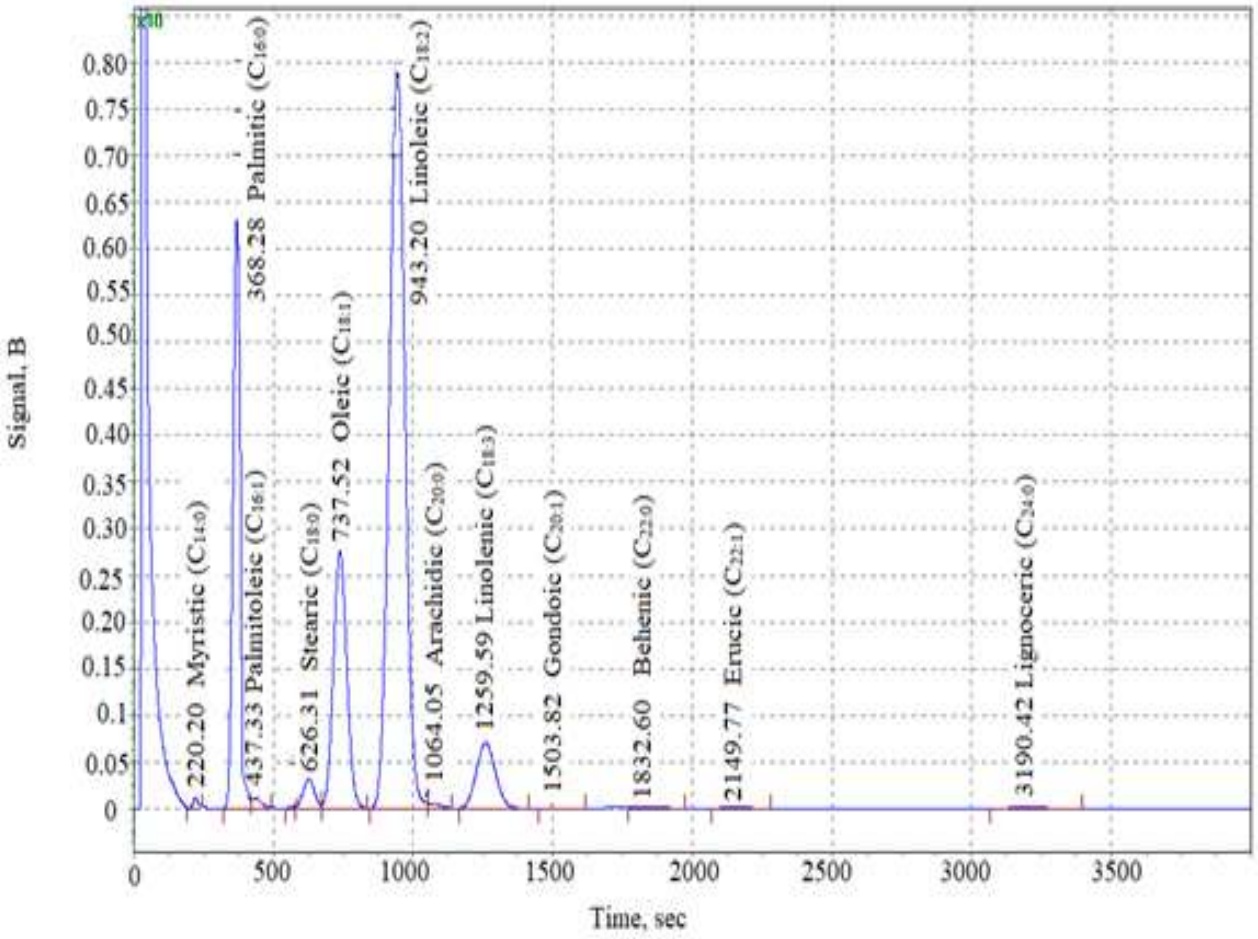


Fig. 3.2. Gas chromatogram of the fatty acid composition of lipophilic fractions from Carambola fruits

15 fatty acids were found in Carambola fruits, of which 13 were identified. In the studied raw material, the maximum content was linolenic acid (53,94 %). In addition to linoleic acid, oleic acid and linoleic acid were accumulated in significant amounts in carambola fruits among unsaturated acids, their content was 15,88 % and 5,81 %. Among saturated fatty acids, palmitic acid dominated – 21,11 %.

Table 3.5

Results of the fatty acids composition in Carambola fruits

No.	Fatty Acid	Content in lipophilic fraction, % of the total
Saturated fatty acids		
1.	Lauric (C12:0)	–
2.	Myristic (C14:0)	0,26
3.	Palmitic (C16:0)	21,11
4.	Stearic (C18:0)	1,56
5.	Arachidic (C20:0)	0,31
6.	Behenic (C22:0)	0,26
7.	Lignoceric (C24:0)	0,17
Monounsaturated acids (ω-9)		
9.	Myristoleic (C14:1)	–
10.	Palmitoleic (C16:1)	0,47
11.	Oleic (C18:1)	15,88
12.	Gondoic (C20:1)	0,07
13.	Erucic (C22:1)	0,13
Polyunsaturated acids (ω-3 and ω-6)		
14.	Linoleic (C18:2)	53,94
15.	Linolenic (C18:3)	5,81
Total amount of saturated fatty acids		23,67
Total amount of unsaturated fatty acids		76,30
Total		100,00

3.5. Study of Pigments

According to the literature, chlorophylls exhibit antimicrobial, antioxidant, antitumor, and anti-inflammatory activity [30].

Reaction with a 10% solution of copper sulfate. A 10% solution of copper sulfate was added to the ethanol extract. In the test tube with the tested hood, the benzene layer was colored bright green, which confirmed the presence of chlorophyll in Carambola fruits.

Reaction with concentrated sulfuric acid. Concentrated sulfuric acid was added to 70% ethanol extract from the fruits of Carambola. During the reaction, a dark blue color appeared, which indicated the presence of carotenoids in the studied raw material of Carambola.

Identification of chlorophylls and carotenoids was carried out by the method of thin-layer chromatography, using the solvent system hexane - acetone (8:2). Chromatograms were dried at room temperature.

On the chromatograms, chlorophylls were identified by the green coloring of the zones in daylight and red fluorescence in UV light. Carotenoids in daylight appeared as zones with a yellow or yellow-hot color, which changed color to pink after treatment with a 2% ethanolic solution of n-dimethylaminobenzaldehyde [30]. The chromatogram for the detection of chlorophylls and carotenoids is shown in Figure 3.3.

Chromatography revealed at least 5 substances, 3 of which were attributed to chlorophylls based on their green color in daylight and red fluorescence in UV light; 2 zones in daylight had yellow and color, and after processing the chromatogram with a solution of *n*-dimethylaminobenzaldehyde changed its color to pink.

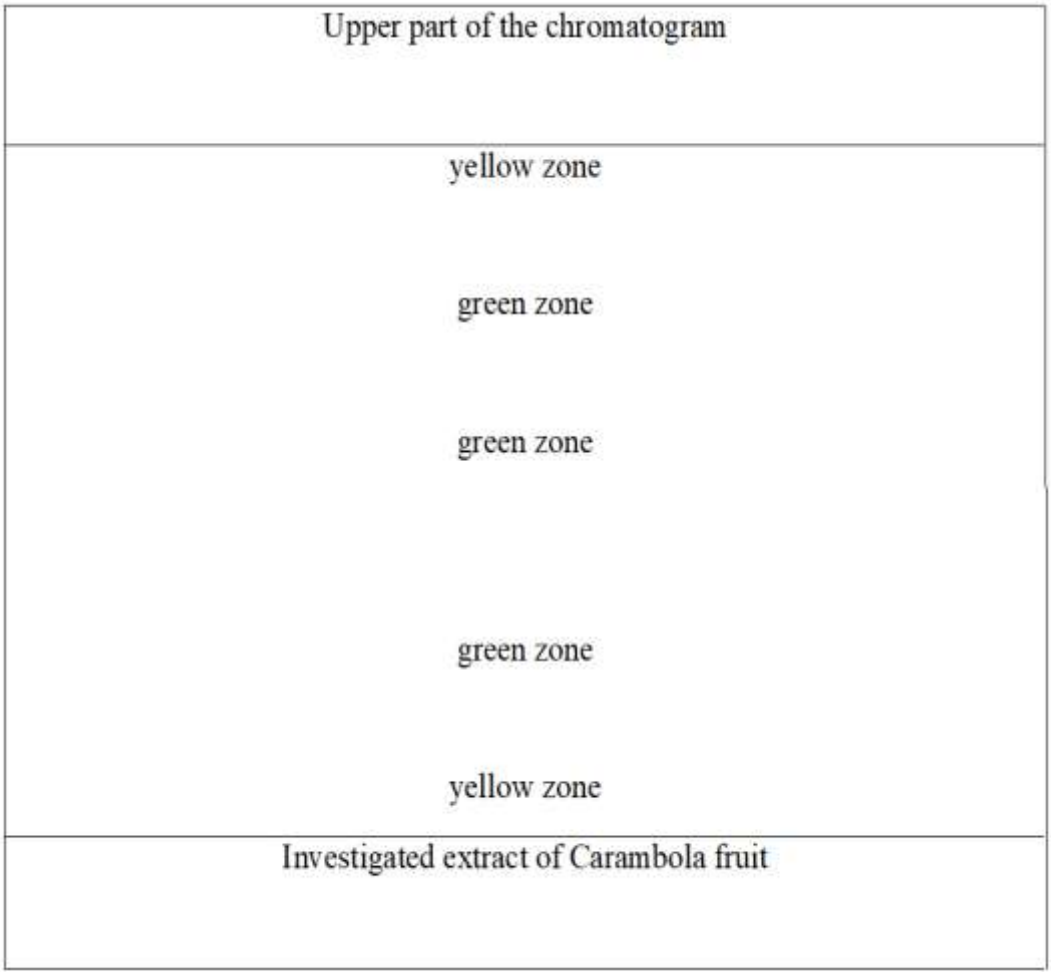


Figure 3.3. Chromatogram for the detection of chlorophylls and carotenoids in Carambola fruit

The pigments (chlorophyll A, chlorophyll B, carotenoids) in the studied raw materials were determined quantitatively using the spectrophotometric method [30].

The medicinal raw material (0,25) was placed in a mortar, a small amount of magnesium carbonate or calcium carbonate and 5 ml of cooled 96% ethanol were added to it. The obtained composite was ground thoroughly for 5 minutes. The resulting extract was carefully poured on a glass filter using a glass rod and filtered. This operation was performed several times until the solvent stopped

staining. The filtrate was placed in a volumetric flask with 25,0 ml capacity, then the solution volume was led to the mark with 96% ethanol.

The absorbance was measured with the Mecasys Optizen POP spectrophotometer in a cuvette with a layer thickness of 10 mm. Maximal absorbance for chlorophyll A was at 665 nm wavelength, and for chlorophyll B – at 649 nm wavelength. Carotenoids were determined at 441 nm wavelength. The comparison solution (the reference solution) was 96 % ethanol.

The concentrations of chlorophyll A (C_{chlA}, mg/l) and chlorophyll B (C_{chlB}, mg/l) were calculated by the formulas:

$$C_{chlA} = 13{,}70 \times A_{665} - 5{,}76 \times A_{649}, \quad (3.4)$$

$$C_{chlB} = 25{,}80 \times A_{649} - 7{,}60 \times A_{665}, \quad (3.5)$$

where: A_{665} – absorbance of the extract at 665 nm wavelength;

A_{649} – absorbance of the extract at 649 nm wavelength.

The concentration of carotenoids A (C_{car}, mg/l) was calculated by the formula:

$$C_{car} = 4{,}695 \times A_{441} - 0{,}268 \times (C_{chlA} + C_{chlB}), \quad (3.6)$$

where: A_{441} – absorbance of the extract at 441 nm wavelength;

(C_{chlA} + C_{chlB}) – total content of chlorophyll A and chlorophyll B in solution, mg/l.

After determination of pigment concentration in the extract, their quantitative content was calculated by the formula:

$$X = \frac{V \cdot C \cdot 100}{m \cdot 1000 \cdot (100 - W)}, \quad (3.7)$$

where: V – volume of alcohol extract, ml;

C – concentration of pigment in alcohol solution, mg/l;

m – mass of plant material, g;

W – weight loss on drying, %.

The results of the quantitative determination of pigments in the studied raw materials are shown in Table 4.3.

5 series of experiments were performed to obtain reliable results that adequate for statistical probability. Analysis results are shown in Table 3.6 – 3.8.

Table 3.6

Results of the quantitative determination of chlorophyll A in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	0,063 0,062 0,064 0,059 0,062	0,062	0,00001	0,00083	0,95	2,78	0,062 ± 0,002	3,75

As can be seen in Table 3.6, the quantitative determination of chlorophyll A in the studied raw materials is 0,062 ± 0,002 %.

Table 3.7

Results of the quantitative determination of chlorophyll B in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	0,091 0,089 0,088 0,092 0,090	0,09	0,00001	0,00070	0,95	2,78	0,09 ± 0,002	2,18

As can be seen in Table 3.7, the quantitative determination of chlorophyll B in the studied raw materials is 0,09 ± 0,002 %.

Table 3.8

Results of the quantitative determination of carotenoids in Carambola fruits

m	n	X_i	X_{mean}	S^2	S_{mean}	P	t(P, n)	Confidence interval	ε, %
5	4	0,042 0,041 0,044 0,042 0,041	0,042	0,00001	0,00054	0,95	2,78	0,042 ± 0,002	3,62

As can be seen in Table 3.8, the quantitative determination of carotenoids in the studied raw materials is 0,042 ± 0,002 %.

Conclusions

1. The content of biologically active compounds was determined in Solanum muricatum fruits using different methods:

- polysaccharides (gravimetric method) amounted to 6,55 ± 0,22 %;
- organic acids (titrimetric method) amounted to 4,82±0,19 %;
- flavonoids (spectrophotometric method) amounted to 4,33 ± 0,20 %;
- carotenoids (spectrophotometric method) amounted to 0,042 ± 0,002 %;
- chlorophyll A (spectrophotometric method) amounted to 0,062 ± 0,002 %;
- chlorophyll B (spectrophotometric method) amounted to 0,09 ± 0,002 %.

2. Quantitative content of fatty acids in Carambola fruits was identified and established by gas chromatography. The results of the research indicate a rich fatty acid composition of the studied raw materials and can be used to create drugs based on them.

GENERAL CONCLUSIONS

1. The literature data regarding the botanical characteristic, history of origin and distribution, chemical composition, application in medicine and culinary of Carambola fruits were analyzed. The detailed study of biologically active compounds of Carambola fruits is a relevant and modern direction of pharmacognosy.

2. The quality indicators of Carambola fruits were determined: the total ash, the weight loss on drying, and the extractable matter. The maximal content of extractable matter was when used water as solvent.

3. The presence of polysaccharides, flavonoids, and organic acids in Carambola fruits was confirmed by the performed qualitative chemical reactions.

4. Five organic acids (malic, gallic, citric, tartaric, and ascorbic acids) were identified in Carambola fruits as the result of chromatographic analysis.

5. The content of biologically active compounds in Solanum muricatum fruits was determined: polysaccharides flavonoids, organic acids, carotenoids, chlorophyll A, chlorophyll B.

6. Quantitative content of fatty acids in Carambola fruits was identified and established by gas chromatography.

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APPENDIX

Appendix A


СУЧАСНІ ДОСЯГНЕННЯ ФАРМАЦЕВТИЧНОЇ НАУКИ В СТВОРЕННІ ТА СТАНДАРТИЗАЦІЇ ЛІКАРСЬКИХ ЗАСОБІВ І ДІЄТИЧНИХ ДОБАВОК, ЩО МІСТЯТЬ КОМПОНЕНТИ ПРИРОДНОГО ПОХОДЖЕННЯ
Матеріали V Міжнародної науково-практичної інтернет-конференції
14 КВІТНЯ 2023
м. Харків

Continuation of Appendix A

МІНІСТЕРСТВО ОХОРОНИ ЗДОРОВ'Я УКРАЇНИ
МІНІСТЕРСТВО ОСВІТИ І НАУКИ УКРАЇНИ
НАЦІОНАЛЬНА АКАДЕМІЯ НАУК ВИЩОЇ ОСВІТИ УКРАЇНИ
НАЦІОНАЛЬНИЙ ФАРМАЦЕВТИЧНИЙ УНІВЕРСИТЕТ
КАФЕДРА ХІМІЇ ПРИРОДНИХ СПОЛУК І НУТРИЦІОЛОГІЇ

MINISTRY OF HEALTH OF UKRAINE
MINISTRY OF EDUCATION AND SCIENCE OF UKRAINE
NATIONAL ACADEMY OF HIGHER EDUCATION OF SCIENCES OF UKRAINE
NATIONAL UNIVERSITY OF PHARMACY
DEPARTMENT OF CHEMISTRY OF NATURAL COMPOUNDS AND NUTRICIOLOGY

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

CURRENT APPROACHES OF PHARMACEUTICAL SCIENCE IN DEVELOPMENT AND STANDARDIZATION OF MEDICINES AND DIETARY SUPPLEMENTS THAT CONTAIN COMPONENTS OF NATURAL ORIGIN

Матеріали V Міжнародної науково-практичної інтернет-конференції

The Proceedings of the V International Scientific and Practical Internet-Conference

ХАРКІВ
KHARKIV
2023

Continuation of Appendix A

МІНІСТЕРСТВО ОХОРОНИ ЗДОРОВ'Я УКРАЇНИ
МІНІСТЕРСТВО ОСВІТИ І НАУКИ УКРАЇНИ
НАЦІОНАЛЬНА АКАДЕМІЯ НАУК ВИЩОЇ ОСВІТИ УКРАЇНИ
НАЦІОНАЛЬНИЙ ФАРМАЦЕВТИЧНИЙ УНІВЕРСИТЕТ
КАФЕДРА ХІМІЇ ПРИРОДНИХ СПОЛУК І НУТРИЦІОЛОГІЇ

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

Матеріали V Міжнародної науково-практичної інтернет-конференції

14 квітня 2023 року
м. Харків

Харків
2023

Continuation of Appendix A

УДК 615.1 : 615.32 : 615.07
С 89

Електронне видання мережне

Редакційна колегія: проф. А. А. Котвіцька, проф. А. І. Федосов, проф. І. М. Владимирова, проф. В. С. Кисличенко, асист. В. В. Процька, ст. лаб. О. О. Іосипенко

Конференція зареєстрована в Українському інституті науково-технічної і економічної інформації (УкрІНТЕІ), посвідчення № 546 від 19.12.2022 року

С 89 *Сучасні досягнення фармацевтичної науки в створенні та стандартизації лікарських засобів і дієтичних добавок, що містять компоненти природного походження* : матеріали V Міжнародної науково-практичної інтернет-конференції (м. Харків, 14 квітня 2023 р.). – Електрон. дані. – Х. : НФаУ, 2023. – 186 с. – Назва з тит. екрана.

У збірнику розглянуто теоретичні та практичні аспекти розробки, виробництва лікарських засобів рослинного походження і дієтичних добавок, контролю якості, стандартизації лікарських засобів рослинного походження та визначення безпечності дієтичних добавок, а також їх реалізації в умовах сучасного фармацевтичного ринку.

Для широкого кола науковців, магістрантів, аспірантів, докторантів, викладачів вищих фармацевтичних та медичних навчальних закладів, співробітників фармацевтичних підприємств, фармацевтичних фірм.

Друкується в авторській редакції. Автори опублікованих матеріалів несуть повну відповідальність за підбір, точність наведених фактів, цитат, економіко-статистичних даних, власних імен та інших відомостей. Матеріали подаються мовою оригіналу. Матеріали пройшли антиплагіатну перевірку за допомогою програмного забезпечення StrikePlagiarism.

УДК 615.1 : 615.32 : 615.07

Continuation of Appendix A

IDENTIFICATION AND QUANTITATIVE DETERMINATION OF ORGANIC ACIDS IN CARAMBOLA FRUITS

Imane Malhi, Tartynska G. S.
National University of Pharmacy, Kharkiv, Ukraine

Introduction. In recent years, exotic fruits have gained considerable popularity and firmly entered our everyday life. The variety of exotic fruits is simply amazing, but still a large part of them is little known to us and not studied enough. Fresh fruits are important in human nutrition. They are the main sources of vitamins, macro- and microelements, easily digestible sugars, organic acids, enzymes, etc. Fruits have many medicinal properties, they are used to treat colds, to improve the work of the gastrointestinal tract. The analysis of literary data shows that carambola fruits are widely used in traditional medicine for the treatment of various diseases, they show anti-inflammatory, antioxidant, hemostatic, antibacterial activity. In addition, they are widely used in cooking and cosmetology. Therefore, it is urgent to carry out more detailed pharmacognostic studies of carambola fruits [1].

Materials and methods of research. The paper chromatography method was used for the qualitative identification of organic acids. The chromatography study was carried out in the solvent system: ethanol – chloroform – ammonia – water (70:40:20:2) in parallel with reliable samples of organic acids. After drying, the chromatogram was processed with the reagent (solution of sodium 2,6-dichlorophenolindophenolate) and heated in the drying oven at 105 °C temperature. The organic acids on the chromatogram appeared as yellow stains on a blue background, but ascorbic acid appeared as a pink stain, which disappeared with time.

The quantitative determination of organic acids content in carambola fruits was carried out by the alkalimetry method described in the State Pharmacopoeia of Ukraine (article "Rosehip fruits") [2, 3].

Results and their discussion. Malic, citric, tartaric, gallic, and ascorbic acids were identified among organic acids in carambola fruits as a result of the chromatographic analysis. The quantitative content of organic acids in carambola fruits amounted to 4.82±0.19%.

The new promising source of plant raw material – the fruits of carambola (*Averrhoa carambola*) – contains various groups of biologically active compounds. The detailed study of these BAC is a relevant and modern direction of pharmacognosy.

Referencers:

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3. Тулуб І. О., Процька В. В., Бурда Н. Є. Ідентифікація та визначення кількісного вмісту органічних кислот у сировині цинії елегантної. Управління якістю в фармації : матер. XVI наук.-практ. internet-конференції з міжнар. участю, (м. Харків, 20 травня 2022 р.). Харків. 2022. С. 90.

Continuation of Appendix A

ЗМІСТ **CONTENT**

Continuation of Appendix A

Наукове електронне видання мережне

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

Матеріали V Міжнародної науково-практичної інтернет-конференції

14 квітня 2023 року
м. Харків

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Процька Вікторія Василівна
Іосипенко Олена Олександрівна

Комп'ютерний набір та верстка
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Свідоцтво суб'єкта видавничої справи серії ДК № 3420 від 11.03.2009

Appendix B

MINISTRY OF HEALTH OF UKRAINE
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NATIONAL ACADEMY OF HIGHER EDUCATION OF SCIENCES OF UKRAINE
NATIONAL UNIVERSITY OF PHARMACY
DEPARTMENT OF CHEMISTRY OF NATURAL COMPOUNDS AND NUTRICIOLOGY

CERTIFICATE

№ 23

This is to certify that

Imane Malhi

has participated in the V International Scientific and Practical Internet-Conference

"CURRENT APPROACHES OF PHARMACEUTICAL SCIENCE IN DEVELOPMENT AND STANDARDIZATION OF MEDICINES AND DIETARY SUPPLEMENTS THAT CONTAIN COMPONENTS OF NATURAL ORIGIN"

(Duration - 6 hours)
April, 14, 2023, Kharkiv, Ukraine

МІНІСТЕРСТВО ОХОРОНИ ЗДОРОВ'Я УКРАЇНИ Національний фармацевтичний університет №02010936

Rector of the NUPh, prof. — **Alla KOTVITSKA**

Vice-Rector for scientific and pedagogical work of the NUPh, prof. — **Inna VLADIMIROVA**

Head of the department of chemistry of natural compounds and nutriciology of the NUPh, prof. — **Viktoriia KYSLYCHENKO**

Ф А 2.2.1–32-366

National University of Pharmacy

Faculty for foreign citizens' education
Department chemistry of natural compounds and nutritiology
Level of higher education master
Specialty 226 Pharmacy, industrial pharmacy
Educational program Pharmacy

APPROVED
The Head of Department
of chemistry of natural
compounds and nutritiology

_____
Prof. Viktoria KYSLYCHENKO
"28" September 2022

ASSIGNMENT

FOR QUALIFICATION WORK

OF AN APPLICANT FOR HIGHER EDUCATION

Imane MALHI

1. Topic of qualification work: « Phytochemical study of carambola fruits », supervisor of qualification work: Ganna TARTYNSKA, PhD,

approved by order of NUPh from "6th" of February 2023 № 35

2. Deadline for submission of qualification work by the applicant for higher education: April, 2023

3. Outgoing data for qualification work: Phytochemical study of carambola fruits

4. Contents of the settlement and explanatory note (list of questions that need to be developed): Review of the literature on botanical description, chemical composition, use of carambola fruits; determination of the qualitative composition and quantitative content of the main biologically active substances

5. List of graphic material (with exact indication of the required drawings):
Tables – 11, pictures – 13.

6. Consultants of chapters of qualification work

Chapters	Name, SURNAME, position of consultant	Signature, date	
		assignment was issued	assignment was received
1.	Ganna TARTYNSKA, assistant of higher education institution of department of chemistry of natural compounds and nutriciology, PhD	03.10.2022	03.10.2022
2.	Ganna TARTYNSKA, assistant of higher education institution of department of chemistry of natural compounds and nutriciology, PhD	07.11.2022	07.11.2022
3.	Ganna TARTYNSKA, assistant of higher education institution of department of chemistry of natural compounds and nutriciology, PhD	05.12.2022	05.12.2022

7. Date of issue of the assignment: "28" _September_ 2022

CALENDAR PLAN

№ з/п	Name of stages of qualification work	Deadline for the stages of qualification work	Notes
1.	BOTANICAL CHARACTERISTICS, CHEMICAL COMPOSITION, PRACTICAL USE, APPLICATION IN MEDICINE CARAMBOLA	03.10.2022-21.10.2022	**done**
2.	DEFINITION OF QUALITY INDICATORS IN CARAMBOLA FRUITS	07.11.2022-28.11.2022	**done**
3.	STUDY OF THE QUALITATIVE COMPOSITION AND QUANTITATIVE CONTENT OF BIOLOGICALLY ACTIVE SUBSTANCES IN CARAMBOLA FRUITS	05.12.2022-07.02.2023	**done**

An applicant of higher education _____ Imane MALHI

Supervisor of qualification work _____ Ganna TARTYNSKA

ВИТЯГ З НАКАЗУ № 35
По Національному фармацевтичному університету
від 06 лютого 2023 року

нижченаведеним студентам 5-го курсу 2022-2023 навчального року, навчання за освітнім ступенем «магістр», галузь знань 22 охорона здоров'я, спеціальності 226 – фармація, промислова фармація, освітня програма – фармація, денна форма здобуття освіти (термін навчання 4 роки 10 місяців та 3 роки 10 місяців), які навчаються за контрактом, затвердити теми кваліфікаційних робіт:

Прізвище студента	Тема кваліфікаційної роботи		Посада, прізвище та ініціали керівника	Рецензент кваліфікаційної роботи
• **по кафедрі хімії природних сполук**				
Малхі Іман	Фітохімічне вивчення плодів карамболи	Phytochemical study of Carambola fruits	ас. Тартинська Г.С.	проф. Георгіянц В.О.

Підстава: подання декана, згода ректора

Ректор

Вірно. Секретар громадян

Ф А2.8-47-110

ВИСНОВОК

Комісії з академічної доброчесності про проведену експертизу

щодо академічного плагіату у кваліфікаційній роботі

здобувача вищої освіти

№ 113140 від « 9» травня 2023 р.

Проаналізувавши випускну кваліфікаційну роботу за магістерським рівнем здобувача вищої освіти денної форми навчання Малхі Іман, 5 курсу, _____ групи, спеціальності 226 Фармація, промислова фармація, на тему: «Фітохімічне вивчення плодів карамболи / Phytochemical study of Carambola fruits», Комісія з академічної доброчесності дійшла висновку, що робота, представлена до Екзаменаційної комісії для захисту, виконана самостійно і не містить елементів академічного плагіату (компіляції).

Голова комісії,
професор **Інна ВЛАДИМИРОВА**

7%

31%

Ф А 2.2.1-32-353

REVIEW

of scientific supervisor for the qualification work of the level of higher education master of the specialty 226 Pharmacy, industrial pharmacy

Imane MALHI

on the topic: «Phytochemical study of Carambola fruits»

Relevance of the topic. A new promising source of plant materials – the fruits of *Averrhoa carambola* (L.) – contains various groups of biologically active compounds. A detailed study of these compounds is a relevant and modern direction of pharmacognosy.

Practical value of conclusions, recommendations and their validity. Analyzed and summarized the literature data on the carambola, botanical characteristics of *Averrhoa carambola* (L.) and *Averrohaa bilimbi* (L.), chemical composition and pharmacological activity of carambola are analyzed and summarized. The qualification work presents the results of the study of the qualitative composition and quantitative content of biologically active substances, quality indicators, technological parameters of carambola fruits.

In the process of performing the qualification work, Imane MALHI mastered the methods of phytochemical analysis of medicinal plant raw materials.

Assessment of work. Imane MALHI qualification work was performed at a high scientific level. When conducting phytochemical analysis on the topic of the applicant's work, various methods of analysis were used.

Statistical processing of the results of quantitative determination of biological active compounds and quality indicators in accordance with the requirements of the State Pharmacopoeia of Ukraine

General conclusion and recommendations on admission to defend. Qualification work of Imane MALHI on the topic: “Phytochemical study of Carambola fruits” can be submitted for defense to the State Examination Commission.

Scientific supervisor _____ Ganna TARTYNSKA

«05» of April 2023

Ф А 2.2.1-32-356

REVIEW

for qualification work of the level of higher education master, specialty 226 Pharmacy, industrial pharmacy

Imane MALHI

on the topic: «PHYTOCHEMICAL STUDY OF CARAMBOLA FRUITS»

Relevance of the topic. Medicinal plants are safe, they are widely used for a long time as a source of biologically active substances in the food, pharmaceutical and other industries. The appearance and taste of Carambola fruits make them a very popular gastronomic and decorative element in many countries around the world. They are used in folk medicine as an antioxidant, anti-inflammatory agent. Carambola fruits are rich in vitamins and macro- and microelements, pectins. In addition, the study of the chemical composition and pharmacological activity of carambola fruit is increasingly attracting the attention of scientists around the world.

Theoretical level of work. The author of the qualification work analyzed the literature on botanical characteristics, distribution area, chemical composition and use of plant of the *Averrhoa carambola* L. and make analysis of pharmacological activities and methods of determination of the quality of herbal drugs in many countries.

Author's suggestions on the research topic. In the qualification work, Imane MALHI presents the results of a phytochemical study of Carambola fruits: polysaccharides, flavonoids, organic acids, and pigments were discovered and identified. The quantitative content of the main classes of biologically active compounds was determined: polysaccharides, organic acids, flavonoids, fatty acids, pigments – carotenoids, chlorophyll A, and chlorophyll B. The main numerical indicators for the requirements of State Pharmacopoeia of Ukraine for the studied raw materials were established. According to the research results, the work was published in the proceedings of the conference.

Practical value of conclusions, recommendations and their validity. The author studied the qualitative composition of biologically active compounds in the studied herbal drugs. The content of the main groups of BAC and quality indicators of the studied raw materials.

Disadvantages of work. The work contains spelling errors and unsuccessful statements.

General conclusion and assessment of the work. The proposed work is of practical importance and meets the requirements for qualification work. Qualification work of Imane MALHI on the topic: “Phytochemical study of Carambola fruits” can be submitted for defense to the Examination Commission

Reviewer _____ prof. Victoriya GEORGIYANTS

«11» of April 2023

Витяг
з протоколу засідання кафедри хімії природних сполук і нутриціології
Національного фармацевтичного університету
№ 4 від 18 квітня 2023 року

ПРИСУТНІ: Бурда Н.Є., Журавель І.О., Кисличенко В.С., Комісаренко А.М., Король В.В., Новосел О.М., Попик А.І., Попова Н.В., Процька В.В., Скребцова К.С., Тартинська Г.С., Хворост О.П.

Порядок денний:

1. Щодо допуску здобувачів вищої освіти до захисту кваліфікаційних робіт у Екзаменаційній комісії.

СЛУХАЛИ: про представлення до захисту в Екзаменаційній комісії кваліфікаційної роботи на тему «Фітохімічне вивчення плодів карамболи» здобувач вищої освіти випускного курсу Фм18(5,0д)англ-06 групи Іман МАЛХІ.
Науковий керівник: асистент Ганна ТАРТИНСЬКА
Рецензент: професор Вікторія ГЕОРГІЯНЦ

УХВАЛИЛИ: рекомендувати до захисту в Екзаменаційній комісії кваліфікаційну роботу здобувача вищої освіти Фм18(5,0д)англ-06 групи Іман МАЛХІ на тему «Фітохімічне вивчення плодів карамболи».

Завідувачка кафедри хімії природних
сполук і нутриціології Вікторія КИСЛИЧЕНКО

Секретар кафедри ХПСіН Надія БУРДА

Ф А2.2.1-32-042

НАЦІОНАЛЬНИЙ ФАРМАЦЕВТИЧНИЙ УНІВЕРСИТЕТ

ПОДАННЯ
ГОЛОВІ ЕКЗАМЕНАЦІЙНОЇ КОМІСІЇ
ЩОДО ЗАХИСТУ КВАЛІФІКАЦІЙНОЇ РОБОТИ

Направляється здобувач вищої освіти Іман МАЛХІ до захисту кваліфікаційної роботи
за галуззю знань 22 Охорона здоров'я
спеціальністю 226 Фармація, промислова фармація
освітньою програмою Фармація
на тему: «Фітохімічне вивчення плодів карамболи».

Кваліфікаційна робота і рецензія додаються.

Декан факультету _____ / Світлана КАЛАЙЧЕВА /

Висновок керівника кваліфікаційної роботи

Здобувач вищої освіти Іман МАЛХІ засвоїла основні методи фітохімічного аналізу, дана кваліфікаційна робота має практичне значення та відповідає вимогам, що висуваються до роботи певного рівня

Керівник кваліфікаційної роботи

_____ Ганна ТАРТИНСЬКА

«05» квітня 2023 р.

Висновок кафедри про кваліфікаційну роботу

Кваліфікаційну роботу розглянуто. Здобувач вищої освіти Іман МАЛХІ допускається до захисту даної кваліфікаційної роботи в Екзаменаційній комісії.

Завідувач(ка) кафедри хімії природних сполук і нутриціології

_____ Вікторія КИСЛИЧЕНКО

«18» квітня 2023 року

Qualification work was defended

of Examination commission on

« ___ » _____ 2023

With the grade _____

Head of the State Examination commission,

DPharmSc, Professor

_____ / Oleh SHPYCHAK /