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PLANTS OF *NEPETA CATARIA* VAR. *CITRIODORA* BECK. AND ESSENTIAL OILS FROM THEM FOR FOOD INDUSTRY

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ABSTRACT

Nepeta cataria var. *citriodora* Beck. (catmints) is a source industrial production of citral and attractive raw material for food industry and cooking. Aerial part of *Nepeta* are characterized by high antimicrobial activity and fungicidal action against mold fungi, used in folk medicine, as ingredient in recipes for sausages, liqueurs and soft drinks, vegetable and fruit canned food, in the manufacture of vermouth. Ukrainian variety 'Melody' was created specifically for growing in the Forest-Steppe zone, and variety 'Peremozhets' – in the Steppe zone. Data on the yield aerial part and essential oil *Nepeta* was determined. The dry aerial part of plants *N. cataria* we used to create a dry spicy mixture for sweet dessert dishes. Quantitative content and qualitative composition of essential oil of plants by organs and phases of vegetation are presented in the article. In our research we used essential oils obtained by hydro distillation procedure for 2 h using Clevendger-type apparatus from the flowering parts of plants *N. cataria* 'Peremozhets' and 'Melody'. Investigate of components was carried out by high effective gas chromatography with HP 6890 chromatograph coupled with HP 5972 mass selective detector. The most abundant components of *Nepeta* essential oil was citral, geraniol, as well as nerol, citronellol, citronellal, carvacrol, camphor, eugenol. We proposed fractional distillation of essential oils to obtain a line of flavors with stable sensory and physicochemical indicators for food industry. The separation of essential oils into fractions was carried out on a pilot installation of fractional distillation DFD (Device of Fractional Distillation). Calculations of parameters controlled dispersal of essential oils (residual pressure, temperature regimes, number of theoretical plates, reflux number) were carried out. During fractionation of essential oil of *N. cataria* four fractions were obtained with a content of 96 ±0.5% to the total mass of samples. Sensory and physicochemical analysis of aromatic fractions announced them as promising flavours for food industry.

Keywords: *Nepeta cataria*; yield; essential oil; fraction; flavour

INTRODUCTION

The total number of aromatic and spicy-aromatic plants of the world flora is estimated at 2500 – 3000 species. The main families, which include a large number of aromatic plants, are *Lamiaceae, Apiaceae* and *Asteraceae* **(Tkachenko, 2011)**. *Nepeta cataria* var. *citriodora* Beck. (common name – catnip, catmints) is a source of industrial production citral and attractive raw material for the food industry and cooking, it is widely used in the world due to the very pleasant aroma of essential oil **(Burt, 2004; Bhattacharya, 2016)**. The plant originates from the Mediterranean and the Western Asia. It is distinguished by its high ecological plasticity. *N. cataria* is characterized by a significant intravivid variability, which results to selection of high-yielding samples – source forms for breeding. One of the ways to intensify production raw materials of aromatic plants in Ukraine for the production of essential oils is the creation of new high-yielding varieties **(Korablyova, 2001)**. Variety *Nepeta cataria* 'Melody' was created specifically for growing in the Forest–Steppe zone of Ukraine and the variety 'Peremozhets' – for growing in zone Steppe.

Essential oil of *N. cataria* is a colourless, mobile liquid with a pleasant herb-citrus aroma with tones of geraniums. The main components of essential oil *Nepeta cataria* are citral – 12%, geraniol – 25%, as well as nerol, citronellol, citronellal, carvacrol, camphor, eugenol. The composition of the oil includes the main classes of compounds: monoterpene alcohols – 71.2%, monoterpene aldehydes – 19.3%, sesquiterpenes – 2.5%. Nepetalactone accumulates in this essential oil 0.8 – 1.9% **(Ríos, 2016)**. It should be noted that in literary sources data are given on the composition of essential oils *N. cataria* with significant differences, even with the content of key components. These are citronellol and geraniol, the content of which varies for citronellol from 8 to 70%, citral from 0 to

18.33%, and geraniol – from 0 to 28%. World production of essential oils is more than 5000 tons per year **(Franz, 2006)**. Most often, essential oils are used in the production of perfumery and cosmetic products, hygiene products, in the form of drugs. The average annual outputs of such products in the world are fixed at 7.5 million units **(Khan and Abourashed, 2011)**.

Plants of *N. cataria* are used in folk medicine **(Naghibi et al., 2005; Adiguzel et al., 2009)**. The broth from them has immunomodulatory properties, normalizes the work of the cardiovascular, nervous and respiratory system, and stimulates appetite. It is used for malignancy, cough, liver disease, jaundice, intestinal atony, hysteria, headache, gynecological diseases, as well as anti-helminthes agent **(Gilani et al., 2009)**. At a fairly low concentration of essential oil as aerosols in the air, blood pressure is rapidly normalized.

Essential oil of *N. cataria* is characterized by high antimicrobial activity and fungicidal action in relation to mold fungi: *Mucor*, *Penicillium*, *Aspergillus* **(Zomorodian et al., 2012; Foltinová, Tančinová and Císarová 2017; Tančinová et al., 2018; Stević et al., 2014)**, like as potent source of nematicidal compounds for use in phytopathology and entomology **(Pandey et al., 2000; Peterson and Ems-Wilson, 2003; Tworkoski, 2002; Juglal, Govinden and Odhav, 2002; Amer and Mehlhorn, 2006)**.

As a flavouring *Nepeta* is an ingredient in recipes for sausages, liqueurs and soft drinks, vegetable and fruit canned food, dry spicy mixtures. It is used fresh and dry in the confectionery industry, in the manufacture of vermouth, tea and different cheeses, as tea and drinks additives **(Korablova and Rakhmetov, 2012; Shanaida et al., 2018)**. Natural flavours – this is primarily the essential oils, as well as individual aromatic components that are derived from essential oils by physical methods. GC-FID is the traditional method for essential oils quantification while GC-MS is the most common analytical method for qualitative analysis **(Smelcerovic et al., 2013; Baranauskiene et al., 2003; Frolova et al., 2005; Ukrainets and Frolova, 2009)**.

In food technologies, with considerable demand for natural aromatic substances, using of essential oils are quite limited **(Utami et al., 2011)**. Objective obstacle is a rather narrow range of commercial essential oils, which is associated with the difficulties of their qualitative selection and storage **(Tkachenko, 2011)**. The component composition of the essential oils varies depending on the climatic conditions of cultivation, and the terpene components of the oil when in contact with air during storage are rapidly oxidized **(Méndez-Tovar et al., 2016)**.

This leads to a change in the colour and organoleptic characteristics of the oil. Hardly controlled changes in the quality of essential oils make producers find alternatives to flavouring products, use synthesized substitutes **(Surburg and Panten, 2006)**. Distribution of the release of synthetic sources of aroma is considered the main subjective reason for the narrow use of essential oils in food technologies.

Essential oils in their natural form practically do not use. A significant amount of them is processed by different technologies. The most effective technologies are the following: the receipt of monofractions with subsequent chemical transformation **(Utami et al., 2011)**, deterpening, which releases essential oils from the group of terpene components **(Fantin et al., 2010)**, aims to separate the fractions by distillation, extraction, membrane technology **(Brose et al., 1995)**. Studies of recent years extend to the study of the possibilities of fractionation of essential oils by supercritical fluids **(Chiyoda et al. 2011)**.

We considered the experience of medicine, the perfume and cosmetics industry regarding the mechanisms and practices of fractional disintegration of organic mixtures and proposed fractional distillation of essential oils to obtain a line of flavours of stable sensory and physicochemical indicators for the food industry.

Essential oils are not technologically processed in Ukraine. A common practice is the combination of solutions of essential oils in ethyl alcohol, propylene glycol, as well as in so-called "heavy" ethers. Such mixtures are the basis for the receipt of perfumery and cosmetic products **(Peshuk, Bavkina and Demidov, 2007)**.

Getting natural flavours of improved stability from essential oil *Nepeta cataria* with the development of parameters for its processing into separate fractions (flavouring) of various flavours and their use in formulations of composite flavours for the food industry has urgency, social and industrial request.

Scientific hypothesis

Growing conditions and varietal characteristics of *N. cataria* can affect the productivity of green mass and the composition of the essential oil. Separating different oil fractions will allow us to create flavours with a planned smell.

MATERIAL AND METHODOLOGY

The work was carried out at the National University of Food Technologies and M. M. Gryshko National Botanical Gardens of NASU, located on the border of the Forest-Steppe zone and the Polissya of Ukraine**.** The object of the study was plants of Ukrainian varieties *N. cataria* 'Peremozhets' and 'Melody' **(Korablova and Rabotyagov, 2007)**. The crop accounting was carried out during the period of mass flowering of plants in 2011 – 2015 by the method of field experiments **(Dospekhov, 1986)**. The raw material was cut by hand and immediately weighed. The yield of the above-ground mass was calculated by weighing the raw material from the whole plot.

In our researches essential oils were obtained by water distillation procedure for 2 h using Clevendger apparatus, according to the method described by **El-Seedi et al. (2008)** from the flowering parts of plants *N. cataria*.

Investigate of components was carried out by high effective GC-MS-Analysis with HP 6890 chromatograph coupled with HP 5972 mass selective detector. Injection volume – 1 µL. Inlet: temp – 250 °C, split ratio 10:1. Column – JW DB-5MS, Kat.Nr. 122-5532, length – 30.0 m, nominal diameter – 250 µm, nominal film thickness – 0.25 µm. Carrier gas – He. Mode was constant flow. Initial flow – 1.3 mL.min^{-1}. Nominal init pressure – 11.06 psi. Average velocity – 42 cm.sec^{-1}.Temperature of the thermostat was linearly programmed from 50 °C (5 min) up to 280 °C (2 min)with speed 3 ºC.min^{-1}. MSD Transfer Line – 280 °C. Solvent Delay – 6 min, scan range: 40 – 550 AMU, threshold – 20. Sample – 3

(0.91 Scan.sec^{-1}). The chromatograms represent the total ion current. Identification of the components according their mass spectra carried out using the data bases NIST and Wiley 275 and their retention indices **(Adams, 2001; McLafferty, 1989; NIST, 1994)**.

Fractioning of essential oil *Nepeta cataria* was performed on a pilot universal automatic facility – DFD (Device of Fractional Distillation). This chromatographic method of investigation was developed in the research laboratory National University of Food Technologies **(Frolova et al., 2005; Frolova and Korablova 2016)**. The type of column – three-section; number of real plates, pcs – 20; number of side - bars, pcs – 3; diameter of refractive part 30 mm; head type – full condensation; regulation of the reflux ratio and temperature in a cube from the control unit; control of temperature – automatic. Facility elements are made of inert material – heat-resistant glass produced by Simex (Kavalierglass, USA).

Statistic analysis

The mathematical processing of results the experimental studies was carried out using dispersion statistical methods under the program Microsoft Excel-2010 and the package of programs of statistical analysis in crop production "AGROS" **(AGROS, 2000)**. All experiments determinations were performed in triplicate and the values were expressed as mean ±*SD*.

RESULTS AND DISCUSSION

Nepeta cataria var. *citriodora Beck.* – a perennial grassy plant of a pale green colour, strongly short-cut. Leaves are a pubescent silvery-gray (Figure 1). The root is rod-shaped, long. Stems are quadrilateral, pubescent, from the base branched, up to 80 cm high, with plenty of leaves. Each lateral branch ends with a dense spike-shaped inflorescence from the strongly converged unreal, multi-threaded crests. Flowers are small five-membered, double-haired, corolla white, coupled with the tiny twists of tubular flowers. Fruit – dark brown nuts. The quality of aromatic plants is primarily determined by the content of essential oils (Table 1).

The research has established that the amplitude of the variability the mass fraction of essential oil *N. cataria* varies not only over the years. The mass fraction of essential oil increases in the process of development starting from the vegetative growth phase, reaching the maximum to period of full flowering, and the ending of flowering is accompanied by its reduction (Table 2).

So, the maximum of essential oil of the *N. cataria* during flowering is associated with its high content during this period in the flowers. We have found that the essential oil of flower-lemon flavour in the plants is contained throughout the growing season and is accumulated both in the generative and in the vegetative organs (Table 3).

The maximum amount of essential oils contains fresh inflorescences in the phase of mass flowering. In dried raw materials it contains 3.3 times less essential oils than in fresh inflorescences, which proves the inexpediency of drying the grass before processing. We recommend adhering to the optimum cut height of the upper part of the plant stem at the level up to lower fresh leaves (25 – 35 cm). The dynamics of essential oil in the aboveground mass of the whole plant is determined by the aggregate of its content in separate parts at a definite phase of development. It has been established that essential oil is contained in all organs of *N. cataria*, but is distributed unevenly, and its amount varies in the process of ontogenetic development.

In the structure of the *N. cataria* yield dominates the fate of stems, in which the content of essential oils is very small compared with leaves and inflorescences. This fact greatly affects the overall collection of essential oils.

The component composition of essential oil *N. cataria* was investigated according to the developed by us method of gas chromatographic analysis oxygen-containing components of sources aromatic substances. Chromatogram of essential oil *N. cataria* is shown in Figure 2.

Sample of essential oil was injected into a chromatograph in amount 100 µL oil + 500µL CH_2Cl_2. The main components that determine the qualitative indices of essential oil from the *N. cataria* are citral (neral and geranial), nerol, geraniol and citronellol, as well as geranylacetate (Table 4). Depending on the predominance of these components, several chemotypes of the *N. cataria* are can be distinguished. Quality indicators of the samples essential oil of *N. cataria* are shown in Table 5.

It was established that the amplitude of the variability the mass fraction of essential oil of *Nepeta cataria* under the conditions of Forest-Steppe zone of Ukraine varies by cultivars and organs ranging from 0.795% to 2.863% of the absolutely dry mass of plant material.

The quantitative relations of the *Nepeta cataria* essential oil components are represented with Figure 3, and it is allowed estimate typicalness of the essential oil.

As a result of biochemical research, we have recommended aromatic plants *Nepeta cataria* var. *citriodora* as component of spicy seasonings that are suitable for use in the food industry. Spicy mixtures were prepared by mixing dried and crushed parts of spicy plants in different combinations and quantities.

According to the results of the organoleptic evaluation of the various mixtures and the tasting of dishes with the *Nepeta cataria*, dry plants was included to seasoning 'Citrina' as the most abundant component. Seasoning 'Citrina' is a loose spicy powder without lumps, with lemon tones, beige-greenish colour. It was proposed to use the mixture 'Citrina' for prepare sweet and dessert dishes.

The most abundant component of *N. cataria* was geraniol (Figure 3). His content for cultivar 'Melody' was 23.26% of the total oil, as well as nerol – 22.37%, citronellol – 11.36%, geranial – 9.43%, neral – 7.41%, nepetalactone – 6.08%. Content of geraniol for cultivar 'Peremozhets' was 24.41% of the total oil, as well as nerol – 22.04%, citronellol – 12.32%, nepetalactone – 10.62%, geranial – 9.57%, neral – 7.72%. It was established that the second among the main components of essential oil of the *N. cataria*is the citral (the sum of its cis- and trans isomers – neral and geranial). A group of components of flowery note aroma is citral, citronellol, geraniol, geranyl acetate.

The key components have different contents and their own original aroma. It was affected on the sensory characteristics of the fractions. Due to that each from four fractions has their own unique aroma and can be an independent flavour.

Table 1 Yield of the herb and essential oil of *N. cataria* under condition of Forest-Steppe zone of Ukraine.

Sample	Direction of use	Yield green mass. t.ha^{-1}	Essential oil. kg.ha^{-1}
Nepeta cataria 'Peremozhets'	Perfumery	26.5 ±0.05	89.6 ±0.1
Nepeta cataria 'Melody'	Food	28.5 ±0.05	146.2 ±0.1

Table 2 Essential oil content depending on plant organs and their humidity in of *N. Cataria* 'Peremozhets'.

Samples	Amount of sample	Contents essential oil	
	g	g	%
Fresh inflorescences	200	2.57 – 2.67	1.06 ±0.01
Fresh inflorescences with stems	200	0.72 – 0.92	0.46 ±0.01
Dried inflorescences	200	1.78 – 2.05	1.03 ±0.01
Dried inflorescences with stems	200	0.52 – 0.64	0.31 ±0.01

Table 3 The content of essential oil in the plants *N. cataria* by phases of vegetation. %.

Samples	Regrowth phase	Flowering phase	
	leaves	leaves	inflorescence
Nepeta cataria 'Peremozhets'	0.811 ±0.01	1.374 ±0.01	2.572 ±0.02
Nepeta cataria 'Melody'	0.795 ±0.01	1.485 ±0.01	2.863 ±0.02

Table 4 Composition of essential oil of *N. cataria* cultivars 'Peremozhets' and 'Melody'. % of total oil

Component	Content	
	'Melody'	'Peremozhets'
Neral	7.41 ±0.03	7.72 ±0.03
Geranial	9.43 ±0.04	9.57 ±0.04
Nerol	22.37 ±0.05	22.04 ±0.05
Citronellol	11.36 ±0.04	12.32 ±0.04
Geraniol	23.26 ±0.05	24.41 ±0.05
Nepetalactone	6.08 ±0.03	10.62 ±0.04
6-Methyl-5-heptene-2-one	1.2 ±0.02	1.5 ±0.02
Citronellyformate	0.5 ±0.01	0.4 ±0.01
Geranylformate	0.5 ±0.01	0.4 ±0.01
Geranylacetate	1.6 ±0.02	1.5 ±0.02
Ethylgeranate	1.1 ±0.02	0.7 ±0.01
Caryophyllene oxide	1.4 ±0.02	1.1 ±0.02
Total	86.21	92.28

Table 5 Quality indicators of *N. cataria* essential oil

Quality indicator	Norms of quality indicators	Analysisresults
Raw material for production	Dried plants	Fresh plants
Method of production	Rejection by pair	Hydrodistilation
Appearance	Liquid yellow color	Liquid of amber color
Scent	Pleasant, appropriate for this plant	Grassy-Citrus with floral tint
Density at 20 ºC, g.cm^3(d_4^{20})	0.900 – 0.9980	0.962 ±0.015
Refraction index at 20 ºC, (n_D^{20})	1.4700 – 1.500	1.4861 ±0.012
Acid index, mg KOH.g^{-1}	not more than 30.0	19.44 ±2.45
Solubility 1 volume EO in 70% ethyl alcohol	in three volumes	corresponds
Availability of water	is not allowed	corresponds

Table 6 Parameters of separation binary systems of *Nepeta cataria* essential oil.

System	Dispersion temperature, ˚C	Pressure, kPa	Relative volatility, α	The degree of separation, $\mathbf{n_{min}}$	Reflux number, $\mathbf{V_{work.}}$
mircen – cineol	70.5 – 74	1.32	3.85	2.6	1:5
cineol – linalool	76 – 79	0.96	3.45	2.8	1:4
linalool – citral	94 – 104	0.33	3.3	3.1	1:7
citral + nerol-citronellol	115 – 120	0.33	1.36	7	1:7
citronellol + geraniol + geranylacetate (cubic balance)	142 – 147.5	0.33	1.32	7.5	–

Figure 1 Plants of *Nepeta cataria* var. *citriodora* (1) and their inflorescences (2).

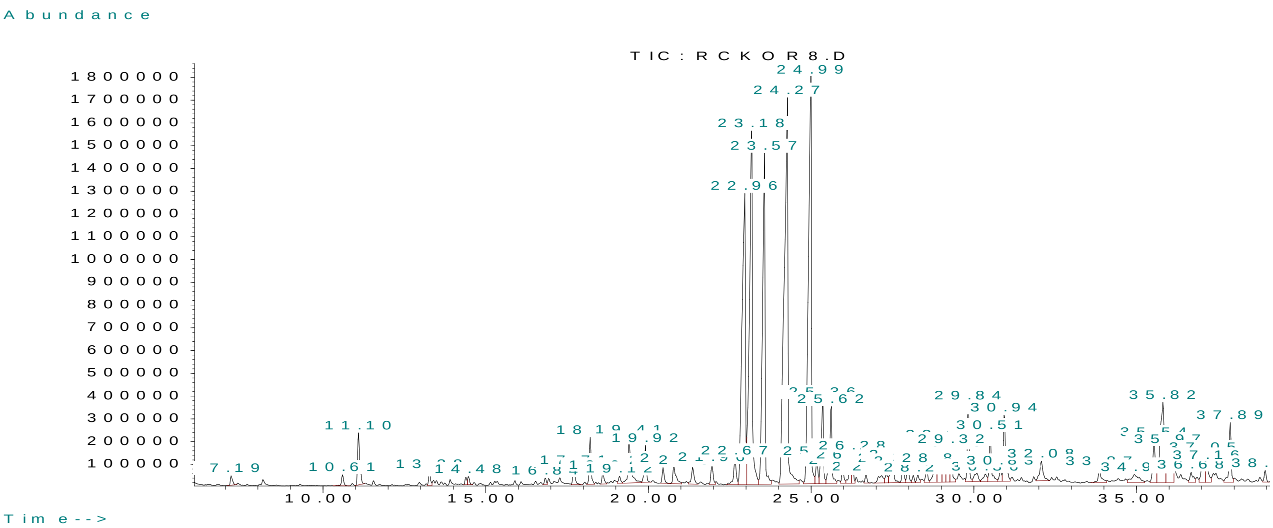

Figure 2 Chromatogram of essential oil *N. cataria* 'Melody'.

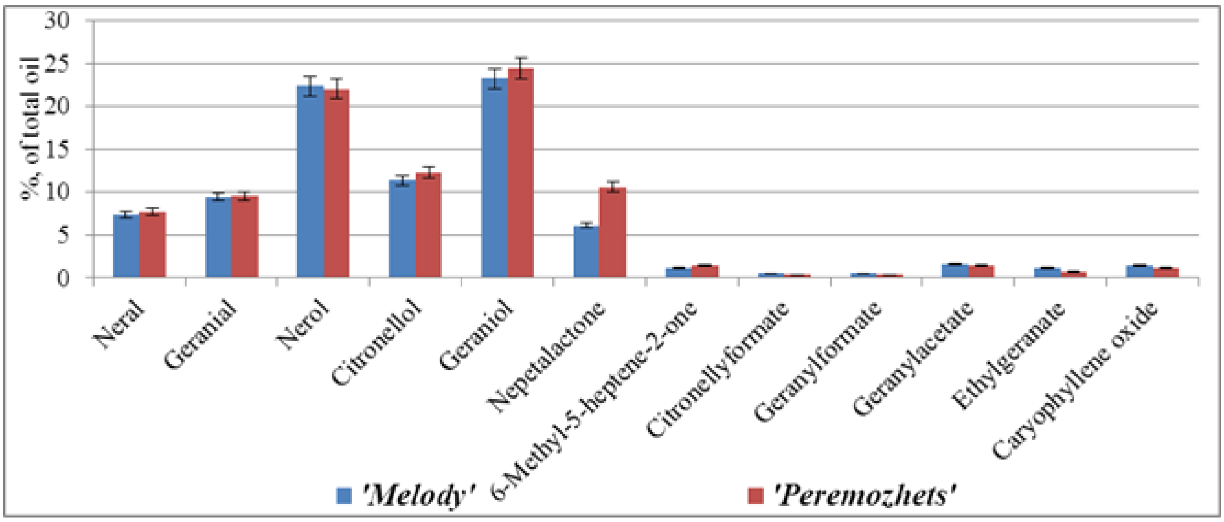

Figure 3 The quantitative relations of the *Nepeta cataria* essential oil components.

Group of components α-pinene, β-pinene, camphene, α-phellandrene, cineol – add a pine note to the general aroma; group β-myrcene, d-limonene, β-phellandrene, n-cymene – have intensive citric note; and group d-camphor and l-borneol – have camphor aroma.

The development parameters of fractional dispersal essential oil of *Nepeta cataria* was aimed to obtaining fractions with the above-mentioned concrete tonality of flavours. It was used the concept of 'key components' – the boundaries, between which fractionation is carried out. By the key components method, *Nepeta cataria* essential oil was considered as the sum of five binary systems – 4 fractions and distillation residue. The modes of distillation essential oil are presented in Table 6.

Calculations of parameters controlled dispersal of essential oils (residual pressure, temperature regimes, number of theoretical plates, reflux number) were carried out according to the rules of distillation and data of quantitative composition.

The process of obtaining a fraction occurs with the enrichment of the most volatile key component, as well as components with similar boiling temperatures. Such components greatly affect the aroma tone of the fractions.

After the discontinuation of the distillate selection at the initial temperature, the temperature of the cube was raised, thereby achieving new equilibrium conditions and receiving a new fraction. Similarly, the following fractions were collected. The fractions were selected from the top of the column. During the fractionation of essential oil *Nepeta cataria* were obtained 4 fractions with a total content of 96 ±0.5% of the total mass essential oil.

Loss due to incomplete capture of low boiling components was 3 ±0.5%.

The fractioning process allows concentrating the key aromatic components and receiving highly concentrated flavours of original pure notes. Results of the study **(Frolova and Korablova, 2016)** indicate the possibility to combine the components of the fractions for use in the product to provide the desired flavour notes.

The flavours not only give products a special scent, but also are characterized by the orientation of the physiological action, saturation, and improved stability. Application of flavour enables to expand assortment of developed products to improve them tasting properties.

CONCLUSION

The formation and accumulation of essential oils occurs in all overground organs of plants *Nepeta cataria* var. *citriodora*. The analysis of the component composition of essential oil *Nepeta cataria* var. *citriodora*, grown in the Kyiv region, have had made it possible to identify several basic components, the most valuable of which is citral. Dedicated fractions of *Nepeta cataria* essential oil had been offered to the food industry as natural flavours for functional and dietary dishes. Seasoning "Citrina", the main component of which is *Nepeta cataria*, is included in the Ukrainian standard "Dry seasoning with spicy aromatic plants". Our research confirms that *Nepeta cataria* var. *citriodora* and their essential oil are promising aromatic raw materials for use in the food industry as flavours.

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