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FEATURES OF ACCUMULATION OF FLAVONOIDS BY QUINQUELOBATE MOTHERWORT (*LEONURUS QUINQUELOBATUS*) HERB COLLECTED IN VARIOUS URBAN BIOCENOSSES AND AGROBIOCENOSSES OF THE VORONEZH REGION

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An important and little-studied aspect of the impact of human economic activity on medicinal plants is that in response to increased anthropogenic load, additional synthesis of secondary metabolites especially flavonoids, is induced, which play an important role in the adaptation of plants to changing conditions. As part of the study, the content of flavonoids in terms of rutin was determined in 51 samples of quinquelobate motherwort herb collected in various urban and agrobiogeocenoses of the Voronezh region which differ in anthropogenic impact. It has been revealed that in quinquelobate motherwort herb collected in agrobiogeocenoses, the content of flavonoids is on average 1.5 times higher than in samples collected in natural biocenoses of protected areas, and in raw materials collected in a number of urbobiogeocenoses that is 2–3.5 times higher than in samples from the controlled protected areas. This fact can be explained by the biochemical adaptation of the plant to significant oxidative stresses, in response

to which the synthesis of polyphenol substances, the main representatives of which are flavonoids, is induced. At the same time, near some production enterprises and along roads with high traffic intensity, we selected samples with reduced content of flavonoids in terms of rutin in comparison with other samples of raw materials. This suggests that in case of excessive toxic influence of pollutants it is also possible to suppress the antioxidant system of plants.

Keywords: Voronezh region, quinquelobate motherwort (*Leonurus quinquelobatus*), flavonoids, rutin

Currently, more than 6.5 thousand medicinal products prepared from medicinal plant raw materials are used in medical practice in Russia. The growing interest in herbal medicines is due to the fact that in the case of rational use, they combine a good therapeutic effect with relative harmlessness. The main part of preparations

of medicinal plant raw materials is traditionally concentrated in the European part of Russia, and in its most populated and industrially developed regions, in particular in the Central Chernozem region. At the same time, most of the exploited resources of wild medicinal plants are located in the zone of active human economic activity, on transport-accessible territories [1,2].

However, due to the growth of cities, sharp increase in the number of vehicles, and the expansion of production areas, the probability of collecting medicinal plant raw materials by the population near sources of pollutants emission increases significantly. In this regard, from a practical point of view, it is necessary to find out the influence of anthropogenic pollution on the chemical composition of medicinal plants, determine the possibility of their use for medical purposes, and identify the most environmentally unfavorable harvesting areas [3].

One of the synanthropic species, the raw material of which is harvested from wild specimens, is the quinquelobate motherwort (*Leonurus quinquelobatus* Gilib.) that is a perennial herb which is widely used in medicine and pharmacy as a sedative, hypotensive, antispasmodic, hemostatic, diuretic. This widespread use is due to the rich chemical composition of the quinquelobate motherwort herb, which is based on flavonoids, iridoids, alkaloids (up to 0.4%), essential oil (up to 0.9%), tannins (up to 2.5%), bitterness, vitamin C, carotene, macro – and microelements [4].

An important and little-studied aspect of the impact of human economic activity on medicinal plants is that in response to the increased anthropogenic load, the additional synthesis of secondary metabolites is induced, which play an important role in adaptation of plants to changing conditions. It is well known that amino acids, organic acids and peptides can serve as ligands for chelating toxic substances, but it has been shown that some secondary metabolites,

especially phenolic compounds, can also serve as chelators and participate in detoxification of pollutants by plants. The main group of low-molecular-weight phenolic compounds consists of flavonoids. Increasing of their content is one of the non-specific reactions to environmental stress [5–7].

Study purpose – study of the accumulation of flavonoids in the quinquelobate motherwort herb collected in various urban and agrobiogeocenoses of the Voronezh region from the point of view of anthropogenic impact.

MATERIALS AND METHODS

To conduct the study in the Voronezh region, points of sampling of soil and medicinal plant raw materials were selected. The choice of the studied areas is determined by the nature of the specific anthropogenic impact on it (Fig. 1, Table 1): chemical enterprises of Voronezh Giprokauchuk LLC, Minudobreniya JSC, Bormash LLC, VOGRES thermal power plant (TPP), Novovoronezh nuclear power plant (NPP), airport, city street (Leningradskaya street), high-voltage power transmission lines (HV PTL), Voronezh reservoir, cities with developed light industry (Kalach, Borisoglebsk), areas of active agricultural activity with the introduction of large amounts of fertilizers (Liskinsky, Olkhovatsky, Podgorensky, Petropavlovsky, Gribanovsky, Khokholsky, Novokhopersky, Repyevsky, Vorobyevsky, Paninsky, Verkhnekhavsky, Rossoshansky districts), zones that were exposed to radionuclide contamination as a result of the Chernobyl NPP accident (Nizhnedevitsky, Ostrogzhsky, Semiluksky districts) and as a comparison – a protected zone (Voronezh biosphere reserve, Khopersk state nature reserve). In addition, we have paid great attention to the issue of collecting medicinal plant raw materials near roads and railways. Actual reference books and manuals on medicinal plants are not unanimous:

somewhere the prohibition zone is indicated as 100 m, somewhere 200 m, and somewhere 300 m from the road. There are no instructions about railway transport at all. Therefore, we decided to analyze this problem in order to find out the proper prohibition zone for collecting medicinal plants near highways. For this study, the Voronezh region was also suitable as well as possible: it presents different natural zones – the forest zone (Ramonsky district), forest – steppe (Anninsky district), steppe (Pavlovsky district), there are major traffic interchanges – M4 “Don” highway, A144 Kursk-Saratov, as well as the non-high-speed road (Bogucharsky district) and the railway (Ramonsky district) were considered.

Natural biogeocenoses were selected for collecting samples. Each analyzed point is subject to the determining influence of one object of economic use, and cross-influence on other studied areas is practically excluded, since

all territories are located at a significant distance from each other.

Determination of the amount of flavonoids in terms of rutin in selected samples of quinquelobate motherwort herb was carried out according to the standard Pharmacopoeia method [8] on the SF-2000 spectrophotometer. Each determination was performed three times. Data obtained in the course of studies of the upper layers of soils and medicinal plant raw materials for determination of the content of radionuclides were statistically processed using Microsoft Excel.

RESULTS AND DISCUSSION

The determined parameters of the content of biologically active substances in quinquelobate motherwort herb are shown in the table.

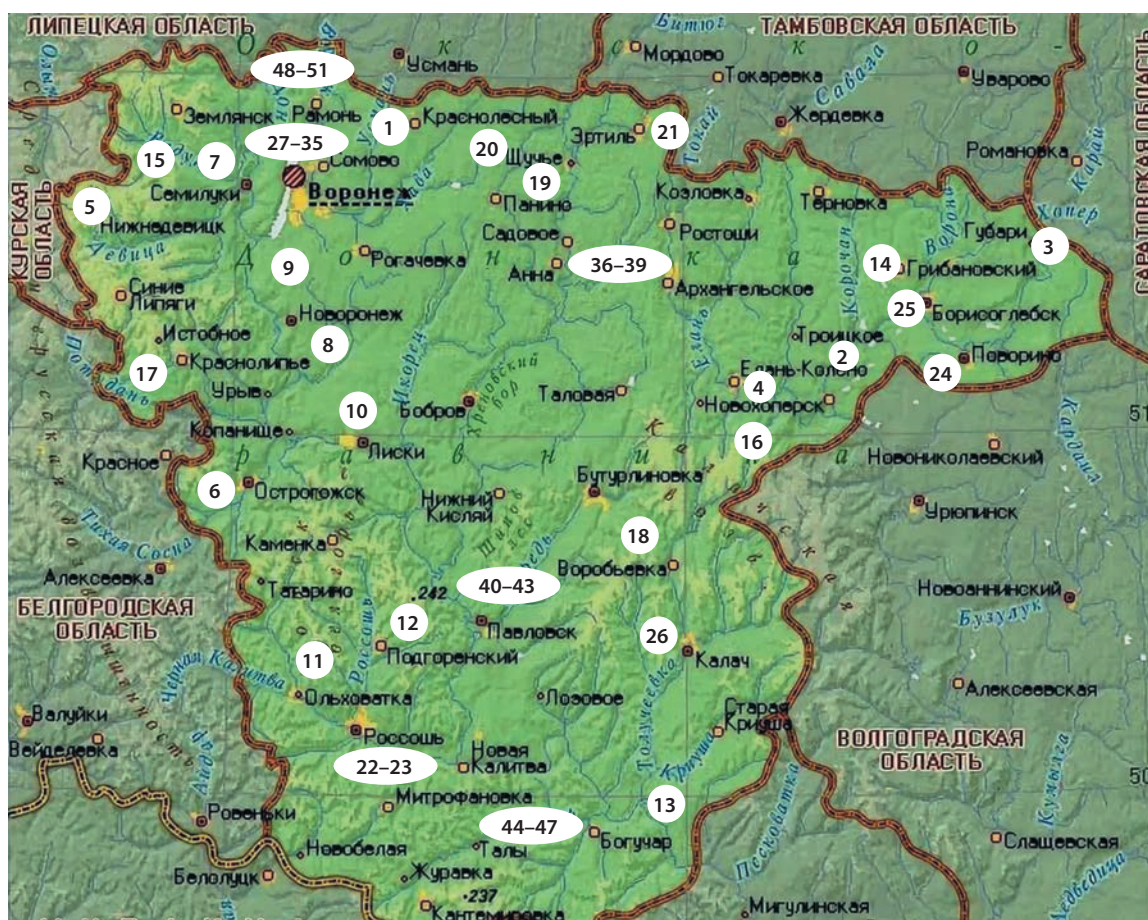


FIG. Map of sampling of medicinal plant raw materials (interpretation – in the table)

**CONTENT OF BIOLOGICALLY ACTIVE SUBSTANCES IN SAMPLES OF QUINQUELOBATE
MOTHERWORT HERB (*LEONURUS QUINQUELOBATUS* GILIB.)**

№	Territory for collecting medicinal plant raw materials	Content of the sum of flavonoids in terms of rutin, %
1.	Voronezh state natural biosphere reserve	0,35±0,02
2.	Khoper nature reserve	0,37±0,03
3.	Borisoglebsk district	0,48±0,05
4.	the settlement of Elan-Koleno	0,41±0,04
5.	the settlement of Nizhnedevitsk	0,89±0,04
6.	the town of Ostrogozhsk	0,95±0,05
7.	the town of Semiluky	0,78±0,03
8.	the town of Novovoronezh	0,85±0,05
9.	High-voltage power transmission line (Novovoronezh municipal district)	0,84±0,03
10.	Lisky District	0,44±0,06
11.	Olkhovatskiy District	0,98±0,04
12.	Podgorensk District	0,42±0,06
13.	Petropavlovsk District	0,53±0,04
14.	Gribanovsk District	0,65±0,05
15.	Khokholsk District	0,50±0,06
16.	Novokhopersk District	0,77±0,03
17.	Repjevsk District	0,44±0,03
18.	Vorobievsk District	0,42±0,05
19.	Paninsk District	0,68±0,04
20.	Verkhnekhavsk District	0,74±0,02
21.	Ertilsk District	0,55±0,06
22.	Rossosh District	0,51±0,04
23.	Near Minudobreniya JSC (Rossosh)	1,11±0,03
24.	Near Bormash LLC (the town of Povorino)	0,30±0,03
25.	the town of Borisoglebsk	0,90±0,04

Table end

№	Territory for collecting medicinal plant raw materials	Content of the sum of flavonoids in terms of rutin, %
26.	the town of Kalach	1,08±0,05
27.	Near VOGRES TPP (Voronezh)	0,75±0,02
28.	Near SIBUR LLC (Voronezh)	0,93±0,06
29.	Along the reservoir (Voronezh)	0,67±0,05
30.	Near the Peter I airport	0,67±0,02
31.	Street in Voronezh (Dimitrov street)	1,04±0,03
32.	Along the M4 highway (Ramonsky District)	0,57±0,06
33.	100 m from the M4 (Ramonsky District)	0,66±0,05
34.	200 m from the M4 (Ramonsky District)	0,87±0,05
35.	300 m from the M4 (Ramonsky District)	1,17±0,02
36.	Along the A144 road (Anninsk District)	0,38±0,03
37.	100 m from A144 (Anninsk District)	0,64±0,04
38.	200 m from A144 (Anninsk District)	0,67±0,05
39.	300 m from A144 (Anninsk District)	0,80±0,03
40.	Along the M4 highway (Pavlovsk District)	0,28±0,02
41.	100 m from M4 (Pavlovsk District)	0,51±0,04
42.	200 m from M4 (Pavlovsk District)	0,76±0,02
43.	300 m from M4 (Pavlovsk District)	0,84±0,04
44.	Along the low-speed road (Bogucharsky District)	1,38±0,03
45.	100 m from the low-speed road (Bogucharsky District)	0,83±0,02
46.	200 m from the low-speed road (Bogucharsky District)	0,84±0,05
47.	300 m from the low-speed road (Bogucharsky District)	0,57±0,04
48.	Along the railway	1,28±0,02
49.	100 m from the railway	0,83±0,05
50.	200 m from the railway	0,82±0,03
51.	300 m from the railway	0,64±0,04
Value for API [8]		At least 0,2

All selected medicinal plant raw materials of quinquelobate motherwort herb according to the results of our studies are recognized as benign in relation to flavonoid content in terms of rutin.

Samples collected in control territories contain this group of biologically active substances on average 2 times more than the lower numerical value specified in the Pharmacopoeial monograph.

In agrocenoses of the Voronezh region the content of flavonoids in terms of rutin in the quinquelobate motherwort herb varies in the range of 0.42% to 0.98% (in the Lisky, Olkhovatskiy, Podgorensk, Petropavlovsk, Gribanovsk, the Khokholsk, Novokhopersk, Repjevsk, Vorobievs, Paninsk, Verkhnekhavsk, Rossosh Districts), but the average is 0.59%, which is almost 3 values specified by the statutory regulatory documentation and 1.5 times higher than the content of flavonoids in the samples from the control conservation areas. Collection of raw materials in these areas was carried out in places of active agricultural activity. They are characterized by the introduction of a large amount of fertilizers, under the influence of which, apparently, phenylalanine ammonia-lyase is activated, which is a key enzyme in the phenylpropanoid pathway of flavonoid biosynthesis [5,9,10].

Samples of quinquelobate motherwort herb collected in the conditions of urban-biocenoses of the Voronezh region are characterized by highly varying results. Thus, in samples collected near Minudobreniya JSC in the Rossosh District, near SIBUR LLC in the city of Voronezh, on the streets of Borisoglebsk, Kalach, Voronezh, along the low-speed highway and along the railway, as well as at distance of 300 m from highways with high traffic intensity (M4 in Ramonsk and Pavlovsk Districts and A144 in Anninsk District), there are very high concentrations of flavonoids in terms of rutin (from 0.80% to 1.38%), which is 2–3.5 times higher than the

content of flavonoids in samples of control protected areas. This fact can be explained by the biochemical adaptation of the plant to significant oxidative stress, in response to which there is an induction of synthesis of polyphenolic substances, the main representatives of which are flavonoids. Flavonoids make a corresponding contribution to the mechanisms of higher plants' response to a variety of abiotic stresses. In addition to the long-known function of shielding from damage by short-wave solar radiation, flavonoids in stressed plants play a key antioxidant function by inhibiting the generation and reduction of reactive oxygen intermediates as soon as they are formed. These properties are mainly inherent in flavones and flavonols, especially quercetin derivatives, in particular rutin. At the biochemical level, the results can be explained by the fact that the key enzyme of flavonoid synthesis – phenylalanine-ammonia-lyase – has a pronounced stress-inducibility [5,7,9,10]. Therefore, the synthesis of phenolic compounds is always enhanced in conditions of anthropogenic impact and pollution of the habitat of the species with toxic substances which are stress for the plant organism.

However, it is possible to distinguish samples also collected under conditions of significant anthropogenic impact, characterized by significantly lower content of flavonoid compounds, for example, near the machine-building enterprise Bormash LLC in the Povorinsk District, along the M4 highway in the Pavlovsk District, along the A144 highway in the Anninsk district, where the content of flavonoids in terms of rutin is noted at the level of 0.28–0.38%. At the same time, at distance of 100 m from the highway, there is sharp, almost twofold increase in the content of flavonoids. Thus, this example implements the ecological law of interaction of factors, which states that the optimal zone and tolerance limits of organisms can be shifted in relation to any environmental factor

depending on the strength and combination of other factors acting simultaneously [1]. That is, for example, near major roads and industrial enterprises, the habitat conditions of the species differ not only in increased concentrations of toxic substances in the soil, but also in high gas content, dustiness of territories, which also affects both the physiological characteristics of the plant organism and the strength of the toxic effects of pollutants. In this case, it seems that instead of stimulation, the enzyme system of flavonol biosynthesis is suppressed. Thus, it is likely that the excessive toxic effect of pollutants may also inhibit the antioxidant system of plants, which is manifested by reduced content of flavonoids.

CONCLUSION

1. More than 50 samples of quinquelobate motherwort herb collected in various urban and agrobiogeocenoses of the Voronezh region were analyzed, where the content of flavonoids in terms of rutin was determined. All selected medicinal plant raw materials according to the results of our study are considered to be of good quality for this factor.

2. It was found that the content of flavonoids in quinquelobate motherwort herb collected in agrobiogeocenoses is 1.5 times higher on average than in samples collected in natural biocenoses of protected areas. The raw material of quinquelobate motherwort herb collected in a number of urban-biogeocenoses of the Voronezh region also has a significant content of flavonoids in terms of rutin, which is 2–3,5 times higher than the content of this group of biologically active substances in samples of control protected areas. This can be explained by the fact that the key enzyme of flavonoid synthesis – phenylalanine-ammonia-lyase – has a pronounced stress-inducibility. Therefore, the synthesis of antioxidant substances, among

which flavonoids play a crucial role, is always enhanced under conditions of toxic stress.

3. The study has revealed that the anthropogenic impact, characterized by a significant release of various toxic substances into the environment, is not always accompanied by the induction of flavonoid synthesis in the plant body. Thus, near some manufacturing plants and along highways with high traffic intensity, we selected samples with reduced content of flavonoids in terms of rutin in comparison with other raw materials. This suggests that the excessive toxic effect of pollutants may also inhibit the antioxidant system of plants.

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