



Synergism and antagonism of the beneficial properties of the fungus *Inonotus obliquus* and plants in tea drinks

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ABSTRACT: Five combinations of *Inonotus obliquus* with plant materials and individual tea ingredients were studied in vitro for the content of biologically active substances (BAS). Crushed samples of raw materials from the temperate climate, *I. obliquus*, *Chamaenerion angustifolium*, *Melissa officinalis*, leaves of *Hippophaë rhamnoides*, fruits of *Pyrus malus*, and from the subtropical zone, *Zingiber officinale*, *Cymbopogon nardus*, fruits of *Citrus limon*, *Cinnamomum verum*, *Hibiscus sabdariffa*, were extracted separately and in ternary mixtures with drinking water (100 °C, 20 minutes). The extracts were evaluated by the total content of polyphenols (TPC), tannins (TN), and antioxidant activity (AOA). Among subtropical plants, *C. limon* (TPC $0.77 \pm 0.08 \text{ mg cm}^{-3}$) and *C. verum* (TN $0.64 \pm 0.06 \text{ } \mu\text{g cm}^{-3}$; AOA $0.25 \pm 0.03 \text{ mg cm}^{-3}$) had the highest BAS content. From the raw materials of the temperate climate, *H. rhamnoides* was maximally enriched with BAS (TPC $0.68 \pm 0.07 \text{ mg cm}^{-3}$; TN $0.67 \pm 0.07 \text{ } \mu\text{g cm}^{-3}$; AOA $0.24 \pm 0.01 \text{ mg cm}^{-3}$). The antagonism of all biologically active substances was established by combining *I. obliquus*, *C. limon*, and *H. rhamnoides*. The maximum synergism was observed in the compositions of *I. obliquus*, *C. nardus*, and *M. officinalis* (to TPC, TN). Therefore, these combinations can reasonably be attributed to a superfood.

Keywords: antioxidant activity; medicinal plants; polyphenolic compounds; tannins.

Sinergismo e antagonismo das propriedades úteis do fungo *Inonotus obliquus* e de plantas em chás

RESUMO: Cinco combinações de *Inonotus obliquus* com materiais vegetais, bem como ingredientes individuais de chá, foram estudadas in vitro quanto ao teor de substâncias biologicamente activas (BAS). Amostras trituradas de matérias-primas do clima temperado *I. obliquus*, *Chamaenerion angustifolium*, *Melissa officinalis*, folhas de *Hippophaë Rhamnoides*, frutos de *Pyrus malus* e da zona subtropical *Zingiber officinale*, *Cymbopogon nardus*, frutos de *Citrus limon*, *Cinnamomum verum*, *Hibiscus sabdariffa* foram extraídas separadamente e em misturas ternárias de água potável (100 °C, 20 minutos). Os extratos foram avaliados pelo teor total de polifenóis (TPC), taninos (TN) e atividade antioxidante (AOA). Entre as plantas subtropicais, *C. limon* (TPC $0,77 \pm 0,08 \text{ mg cm}^{-3}$) e *C. verum* (TN $0,64 \pm 0,06 \text{ } \mu\text{g cm}^{-3}$; AOA $0,25 \pm 0,03 \text{ mg cm}^{-3}$) apresentaram o maior teor de BAS. A partir das matérias-primas do clima temperado, o *H. rhamnoides* foi enriquecido ao máximo com BAS (TPC $0,68 \pm 0,07 \text{ mg cm}^{-3}$; TN $0,67 \pm 0,07 \text{ } \mu\text{g cm}^{-3}$; AOA $0,24 \pm 0,01 \text{ mg cm}^{-3}$). O antagonismo de todas as substâncias biologicamente ativas foi estabelecido na combinação de *I. obliquus*, *C. limon*, *H. rhamnoides*. O sinergismo máximo foi observado nas composições de *I. obliquus*, *C. nardus* e *M. officinalis* (para TPC, TN). Portanto, essas combinações podem ser razoavelmente atribuídas ao superalimento.

Palavras-chave: atividade antioxidante; plantas medicinais; compostos polifenólicos; taninos.

1. INTRODUCTION

The effects of the combined action of substances initially gained fame and explanation of mechanisms in the field of environmental chemistry, as well as pharmacology. In traditional medicine, information about synergism and antagonism in combination with cancer treatment has also been disseminated (CAESAR; CECH, 2019). Modern physicochemical research methods allow us to quantitatively show the strengthening or weakening of the beneficial properties of compositions compared to the original ingredients. So, before the study, the antioxidant potential of the extracts was measured using a modified method using 2,2-diphenyl-1-picrylhydrazyl (DPPH) using an antagonistic

extract of seeds of *Nigella sativa* (Kalonji), *Trachyspermum ammi* (Ajwain), and *Trigonella foenum graecum* (Fenugreek) (QURESHI; BARABDE, 2021). The analytical method established that the combination of Yupingfeng san and Flos Sophorae Immaturus had a synergistic antioxidant effect in vitro (HUANG et al., 2020). However, the classification of combination effects within complex mixtures and the identification of contributing compounds remains a difficult task (VAOU et al., 2021).

Modern stress loads on people, chemical intoxication due to environmental pollution, and viral attacks together create the development of a free radical process and oxidative stress

of the body at the cellular and intracellular levels. People objectively need functional products that have a positive, biologically active effect.

Water extracts from the birch mushroom *I. obliquus* (chaga - fungus) have been proven to have therapeutic and preventive properties. Phenolic, hydroxylated fatty acid, and terpenoid compounds produced by sclerotia of chaga mushrooms are bioactive constituents (PENG; SHAHIDI, 2022). Scientists' attention to the chemical composition of chaga is still great. The work (Stanislaw et al., 2004) published data on previously unknown representatives of isocoumarins (neolignan, cyclic diarylheptanoid) with neuroprotective properties in *I. obliquus*. The anticancer effect of betulin and betulinic acid, part of *I. obliquus*, was found on isolated human colon adenocarcinoma cells (HT 29-MTS) (MITRAKOVA, 2018). To date, antiproliferative, antimicrobial, and other properties have been established for alcoholic extracts from chaga, in addition to a powerful antioxidant effect (HINOJOSA et al., 2004). Less studied in this respect are the aqueous extracts from the fungus.

Recently, phyto-compositions for the production of hot and cold drinks rich in polyphenols have attracted manufacturers and consumers, which is due to their positive role in the prevention and treatment of various diseases, including those caused by SARS-CoV-2 (KICKER et al., 2022). It is known that fungus polyphenols actively absorb free radicals and provide antioxidant properties to prepared beverages (EREMCHENKO et al., 2016; OLKOVA; ASHIKHMINA, 2021). Natural additives to chaga can improve the consumer composition properties (taste, aroma) and enrich the finished drink with additional biologically active substances. At the same time, the combination of some types of raw plant materials leads to an interaction between substances that pass into the extract, which can manifest itself in the manifestation of properties both positively (synergy) and negatively (antagonism) (KUMAR et al., 2013).

This article aimed to study the antioxidant activity, polyphenol content, and tannins in aqueous extracts (tea) of fungus *I. obliquus*, raw plant materials from subtropical, tropical, and temperate climatic regions, and compositions from their mixtures.

2. MATERIAL AND METHODS

2.1. Plant materials and preparation of extracts

Five formulations for the preparation of herbal tea have been developed. The basis of each composition is the birch mushroom *I. obliquus*, to which two ingredients have been added. The first ingredient added to each composition is traditionally used in the food culture of countries located in subtropical and tropical climates: Ginger (*Zingiber officinale*), Lemongrass (*Cymbopogon nardus*), Lemon (*Citrus limon*), Cinnamon (*Cinnamomum verum*), and Karkade (*Hibiscus sabdariffa*). The second ingredient refers to vegetable raw materials harvested in temperate climates: leaves of *Chamaenerion angustifolium*, Melissa (*Melissa officinalis*), Sea buckthorn leaves (*Hippophaë rhamnoides*), Apple, fruit (*Pyrus malus*). The choice of ingredients for the preparation of drinks was due to their use in the folk medicine of various countries and their valuable taste properties.

Fungus *I. obliquus* and plant raw materials from temperate climates were purchased from Russian phytoproduct manufacturers. Raw materials from tropical and subtropical areas were purchased from producers in other countries. All raw materials had quality and safety certificates. A botanist

cross-checked plants and their parts. The raw materials of chaga and vegetables were air-dried, with no more than 10% humidity. The chaga was crushed to a particle size of 0.1-2.5 mm; the rest of the raw materials were no more than 5 mm, except cinnamon, which was powder.

Water extracts from phytocomposition were prepared. In the phyto composition, the basis was fungus - 70% by weight, ingredient No. 1 - 15% by weight, and ingredient No. 2 - 15% by weight. The ratio of the finished mixture to water was 1:20 by weight. Water extracts were also prepared from individual ingredients used in the compositions. At the same time, we considered each ingredient's contribution to the finished product. So, for 200 mL of water, 7 g of the fungus or 1.5 g of other raw vegetable materials were required.

For extraction, we used artesian water, which is monitored monthly according to the regulations of the Eurasian Economic Union (TR EAEU 044/2017 Technical Regulations of the Eurasian Economic Union "On the safety of packaged drinking water, including natural mineral water"). The water temperature was 100 °C, and the infusion was 20 minutes. The infusion was filtered through filter paper (Whatman 42, 125 mm). The cooled extract (22 ± 2.0 °C) was analyzed for 2 hours.

2.2. Determination of total polyphenolic content

The finished aqueous extracts were analyzed. The concentration of polyphenolic compounds was determined by the spectrophotometric method using the Folin-Chokaltau reagent on a PE5300VI spectrophotometer, Ekroschem LLC, Russia (glass cuvettes with a layer thickness of 1 cm). Gallic acid was used as an internal standard.

2.3. Determination of tannin content

Determination of the total content of tannins in terms of tannin in aqueous extracts from plant raw materials was carried out by the Leventhal-Neubauer titrimetric method. This method is based on the ability of tannins containing phenolic groups to be easily oxidized by potassium permanganate in an acidic environment in the presence of an indicator of indigo sulfonic acid, which also acts as an indicator. At the equivalence point, the indigo sulfonic acid is oxidized to isatin, which can be seen in the change in the color of the solution from blue to golden-yellow.

2.4. Evaluation of antioxidant activity

Potentiometry determined the antioxidant content, and quercetin standardized the potassium permanganate solution.

2.5. Statistical analysis

All measurements in this study were obtained by analysis in triplicate ($n=3$). The results were presented as mean \pm standard deviation (SD). ANOVA evaluated differences in the data sets at a significance level of 0.05. Pearson's linear correlation analysis at the 5% probability level evaluated correlations between the total antioxidant activity and the number of tannins and polyphenols.

3. RESULTS

3.1. Total polyphenolic content (TPC)

Compared to other raw materials, the polyphenol content in the extract from Chaga *I. obliquus* was low – 0.101 ± 0.010 mg cm^{-3} . Extracts from plant products of tropical and subtropical regions differed in the maximum

variability of the values of the studied indicator. Thus, ginger *Z. officinale* and lemongrass *C. nardus* were characterized by minimum values of TPC – 0.0340 ± 0.0034 and 0.052 ± 0.005 mg cm⁻³, respectively, and lemon *C. limon* and cinnamon *C. verum* maximum – 0.77 ± 0.08 and

0.64 ± 0.06 mg cm⁻³, respectively. In extracts from temperate plants, TPC decreased in the row: sea buckthorn leaves *H. rhamnoides* (0.68 ± 0.07) > melissa *M. officinalis* (0.46 ± 0.05) > leaves *Ch. angustifolium* (0.43 ± 0.04) > apples, fruits *P. malus* (0.141 ± 0.014).

Table 1. TPC in aqueous extracts from phytocompositions and individual ingredients¹.

Tabela 1. A solução de polifenização na extração de água de uma combinação de fitoterápicos e ingredientes adicionais¹.

Composition No.	TPC (mg cm ⁻³)		Arithmetic sum of TPC (mg cm ⁻³)	TPC phytocompositions (mg cm ⁻³)
	<i>I. obliquus</i>			
1		0.0340 ± 0.0034	0.43 ± 0.04	0.57 ± 0.05 ^a
2		0.052 ± 0.005	0.46 ± 0.05	0.61 ± 0.1 ^a
3	0.101 ± 0.010	0.767 ± 0.077	0.68 ± 0.07	1.56 ± 0.1 ^b
4		0.64 ± 0.06	0.141 ± 0.014	0.88 ± 0.09 ^b
5		0.123 ± 0.012	0.46 ± 0.05	0.68 ± 0.07 ^a
				0.87 ± 0.09 ^{ab}

1 - Each value in the table represents the mean ± standard deviation. Means within each column sharing different superscripts differ significantly ($\alpha=0.05$) at 95% confidence.

1 - Cada valor na tabela representa a média ± desvio padrão. As médias dentro de cada coluna que compartilham sobrescritos diferentes diferem significativamente ($\alpha = 0,05$) com 95% de confiança.

Figure 1 shows the values of TPC in aqueous extracts from *I. obliquus* and from individual ingredients of the developed compositions, as well as TPC in the extract from the finished composition.

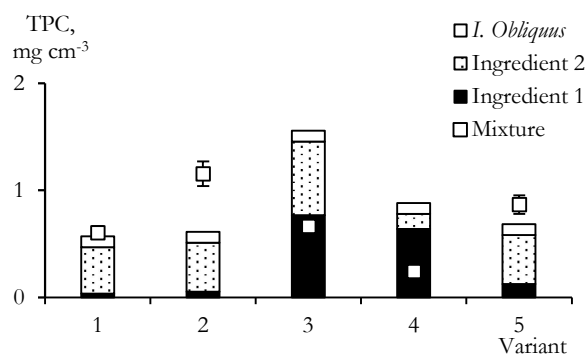


Figure 1. Total polyphenolic content in vegetable ingredients and their mixtures with fungi.

Figura 1. Conteúdo polifenólico Total em ingredientes vegetais e suas misturas com fungo.

The results of the analysis of the extract from composition No. 1, including *I. obliquus*, ginger *Z. officinale* and leaves Chamaenerion, showed the effect of a simple

summation of TPC: the arithmetic sum of the values of the indicator for individual ingredients and TPC mixtures in direct measurement are equal and amount to 0.6 mg cm⁻³. The combination of chaga with lemon fruits and sea buckthorn leaves (No. 3), as well as chaga with cinnamon and apples (No. 4) led to antagonism in terms of TPC: the value of TPC in extracts from mixtures decreased compared to the arithmetic sum of TPC of individual ingredients by 23 and 3.6 times, respectively.

The analysis of extracts from mixture No. 2 and No. 5 showed an increase in the amount of TPC compared to the sum of the ingredients by 1.9 and 1.3 times, respectively. This result is the best characteristic of the developed compositions.

3.2. Tannins content (TN)

Tannins are a component of polyphenolic compounds, so their content in aqueous and other extracts is usually three orders of magnitude less than the total content of polyphenols in the sample. This is typical for the samples under study. Tannins are valued for their high chemical activity. Tannins have several pharmacological effects, including antioxidants, free radical scavenging activity, and antimicrobial, anticancer, anti-nutritional, and cardio-protective properties (SMERIGLIO et al., 2016).

Table 2. Tannin content in aqueous extracts from phyto-compositions and individual ingredients¹.

Tabela 2. Conteúdo de taninos em extratos aquosos de fitocomposições e ingredientes individuais¹.

Composition No.	Tannins of ingredients (µg cm ⁻³)		Arithmetic sum of tannins (µg cm ⁻³)	Tannins phytocompositions (µg cm ⁻³)
	<i>I. obliquus</i>			
1		0.042 ± 0.004	0.374 ± 0.004	0.48 ± 0.04 ^b
2		0.062 ± 0.006	0.52 ± 0.05	0.64 ± 0.05 ^a
3	0.062 ± 0.006	0.083 ± 0.008	0.67 ± 0.07	0.810 ± 0.030 ^b
4		0.64 ± 0.06	0.166 ± 0.017	0.87 ± 0.06 ^a
5		0.125 ± 0.013	0.52 ± 0.05	0.71 ± 0.10 ^b
				0.58 ± 0.06 ^{ab}

1 - Each value in the table represents the mean ± standard deviation. Means within each column sharing different superscripts differ significantly ($\alpha=0.05$) at 95% confidence.

1 - Cada valor na tabela representa a média ± desvio padrão. As médias dentro de cada coluna que compartilham sobrescritos diferentes diferem significativamente ($\alpha = 0,05$) com 95% de confiança.

Among the plant raw materials of tropical and subtropical areas, cinnamon *C. verum* was distinguished by a high content of tannins. The value of this indicator for the extract was 0.64 ± 0.06 µg cm⁻³. A lower content of tannins characterized

aqueous extracts of other plants of warm countries: from 0.042 ± 0.004 µg cm⁻³ in ginger *Z. officinale* to 0.125 ± 0.013 µg cm⁻³ in *H. sabdariffa*. Plants of temperate climates were richer in tannins. The minimum, but

biologically significant level of tannins is recorded in the extract from *Melissa officinalis* ($0.52 \pm 0.05 \mu\text{g cm}^{-3}$). The maximum index was noted for the extract from the leaves of *H. rhannoides* ($0.67 \pm 0.07 \mu\text{g cm}^{-3}$). The joint effects of the ingredients are clearly shown in Figure 2.

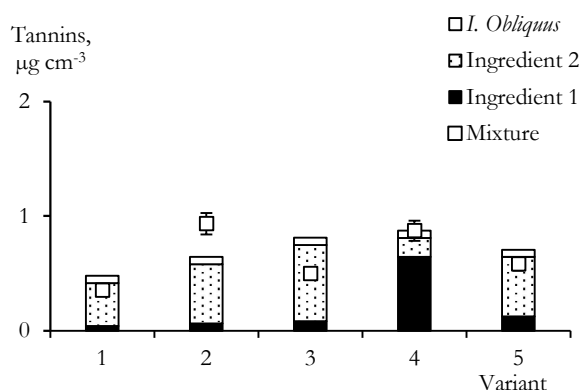


Figure 2. Content of tannins in phytocompositions and individual ingredients.

Figura 2. Conteúdo de taninos em composições FITO e ingredientes individuais.

Some of the regularities identified in the analysis of TPC data were confirmed in the results of the content of tannins in extracts. For example, the maximum synergy of herbal tea components is set for mixture No. 2, which includes chaga *I. obliquus*, lemongrass *C. nardus* and melissa *M. officinalis*. The increase in the content of tannins in the extract from this composition reached 1.5 times compared to the arithmetic sum of tannins in individual ingredients. Also, the effect of composition No. 1 was reduced to the summation of indicators for individual ingredients. Antagonism of chaga

with lemon fruits and sea buckthorn leaves (No. 3) was observed, but was less pronounced than for the TPC indicator: the content of tannins in the aqueous extract with a combination of ingredients decreased by 1.6 times compared to the arithmetic sum of the indicators of chaga, lemon and sea buckthorn separately.

Compositions Nos. 4 and 5, where a simple summation of tannins was recorded, differed from the TPC results.

3.3. Evaluation of antioxidant activity

Antioxidants include substances that can reduce the number of free radicals and regulate their formation in the human body's cells (TRINEEVA, 2017). They protect the body's cells from reactive oxygen species and other oxidizing substances that can cause malfunction and damage to DNA molecules, functional lipids and proteins (PIENIAZEK et al., 2021).

The antioxidant activity of the extract from Chaga *I. obliquus* was $0.129 \pm 0.013 \text{ mg cm}^{-3}$. Extracts from raw plant materials were characterized by a significant variation in the values of the studied indicator in two orders of magnitude. Thus, the AOA of cinnamon *C. verum* was $0.25 \pm 0.03 \text{ mg cm}^{-3}$, and ginger *Z. officinale* was $0.0210 \pm 0.0021 \text{ mg cm}^{-3}$. In extracts from temperate plants, the AOA of apple fruit was 0.065 ± 0.007 , and for other ingredients, $0.227\text{-}0.238 \text{ mg cm}^{-3}$. Figure 3 shows the values of AOA in aqueous extracts from *I. obliquus* and individual ingredients of the developed compositions, as well as AOA extracts from the finished composition.

The analysis results of the extract from compositions Nos. 1 and 3 showed an antagonistic effect, and in No. 5, the effect was of simple summation. The analysis of extracts from mixtures Nos. 2 and 4 showed a synergistic effect: the multiplicity of the increase in the AOA of the mixture compared to the sum of the ingredients was 1.3 times.

Tabela 3. Avaliação da atividade antioxidante em fitocomposições e ingredientes individuais¹.

Table 3. Evaluation of antioxidant activity in phytocompositions and individual ingredients¹.

Composition No.	Evaluation of antioxidant activity of ingredients (mg cm ⁻³)			Arithmetic sum Evaluation of antioxidant activity (mg cm ⁻³)	Evaluation of antioxidant activity of phytocompositions (mg cm ⁻³)
	<i>I. obliquus</i>	1	2		
1		0.0210 ± 0.0021	0.238 ± 0.024	0.39 ± 0.03^b	0.31 ± 0.03^a
2		0.070 ± 0.007	0.227 ± 0.023	0.43 ± 0.04^a	0.56 ± 0.06^b
3	0.129 ± 0.013	0.122 ± 0.012	0.238 ± 0.024	0.49 ± 0.04^b	0.39 ± 0.04^a
4		0.25 ± 0.025	0.065 ± 0.007	0.44 ± 0.05^a	0.56 ± 0.06^b
5		0.046 ± 0.005	0.227 ± 0.023	0.40 ± 0.04^{ab}	0.39 ± 0.04^a

1 - Each value in the table represents the mean \pm standard deviation. Means within each column sharing different superscripts differ significantly ($\alpha=0.05$) at 95% confidence.

1 - Cada valor na tabela representa a média \pm desvio padrão. As médias dentro de cada coluna que compartilham sobrescritos diferentes diferem significativamente ($\alpha = 0,05$) com 95% de confiança.

4. DISCUSSION

4.1. Polyphenolic

Polyphenols are important biologically active substances with antioxidant, antitumor, anticancer, antidiabetic, anti-inflammatory, and cardioprotective effects (ALAM et al., 2022). Daily intake of tannin below the range of 1.5-2.5 g is safe for consumption and does not cause any side effects, but consumption beyond this range is responsible for low absorption of iron from the diet (SHARMA et al., 2019). When the ratio of dry vegetable raw materials and water is 1:20, it is difficult to achieve such values when drinking tea, as shown in this work's results.

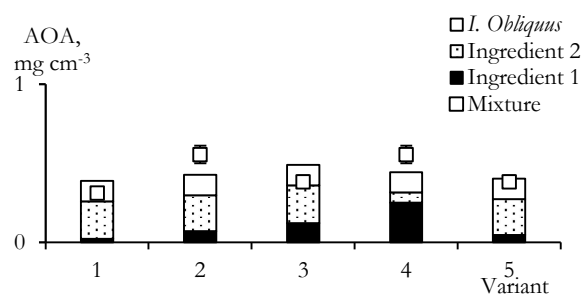


Figure 3. Total antioxidant activity in phytocompositions and individual ingredients.

Figura 3. Atividade antioxidante Total em composições FITO e ingredientes individuais.

It is shown that plants rich in polyphenols grow in temperate and tropical latitudes. According to TPC, extracts from structural parts of plants growing in the tropics and subtropics (lemon and cinnamon) are distinguished among the analyzed raw materials. The leaves of the sea buckthorn *H. rhamnoides*, the area of predominant distribution of which is the temperate zone, approach the lemon fruits by TPC. In the places where *H. rhamnoides* grows, berries are mainly valued. Juice, jams, oils and other products are made from them. However, valuable biologically active substances of bark, leaves and branches (tannins, flavonoids, allantoin) are practically not used (MEZENOVA et al., 2020).

Thus, the aqueous extracts from the developed compositions observed the effects of summation, antagonism, and synergism on TPC.

Antagonism of beneficial properties is an undesirable phenomenon. Therefore, combinations of chaga with lemon fruits, sea buckthorn leaves, and chaga with cinnamon and apple fruits are not recommended for preparing therapeutic and preventive drinks. At the same time, individual ingredients, such as lemon and sea buckthorn leaves, are very valuable. A possible explanation for the antagonism shown is a decrease in the pH level in extracts due to acidic fruits, which negatively affected the subsequent extraction of polyphenols from pH-neutral ingredients. It is known that the strengthening of antioxidant activity is promoted not by an acidic but by an alkaline environment. The pH-dependent increase of the antioxidant activity of the phenols is due to an increase in their electron-donating ability upon deprotonation and their stabilization in alkaline solutions, leading to a polymerization reaction. Such polymerization reactions of polyphenolic antioxidants can form new oxidizable – OH moieties in their polymeric products, resulting in a higher radical scavenging activity (ALTUNKAYA et al., 2016).

On the contrary, the ingredients not led individually by TPC, in combination, showed the effect of summation. For example, the combination of chaga with lemongrass, melissa and karkade led to a synergistic effect of their action.

4.2. Tannins

When analyzing extracts from the selected plant raw materials, it was shown that cinnamon (tropics and subtropics) and sea buckthorn leaves (temperate climate) differ in increased tannins. The values of the indicators for these ingredients are the same ($p > 0.05$). However, a general analysis of the data for “warm” and “cold” areas showed that plants of temperate latitudes accumulate more tannins. Contrary evidence suggests that higher polyphenols, including tannins, are found in more southern regions (RODRIGUEZ-ROMERO et al., 2020).

As shown above, when developing functional herbal compositions for beverage preparation, it is necessary to analyze a heterogeneous product's taste and final effect. The authors of the work also pay attention to this (BASAVEGOWDA; BAEK, 2021).

According to this principle, the best mixtures were No. 2 (fungi *I. obliquus*, lemongrass *C. nardus* and melissa *M. officinalis*) and No. 4 (fungi *I. obliquus*, cinnamon *C. verum* and apple *P. malus*). The health benefits of lemongrass include relief from insomnia, stomach disorders, respiratory disorders, fever, pain, swelling, and infections. The antioxidant activity of the lemongrass herb maintains the immune system and protects against antibiotic-resistant

Staphylococcus aureus. It even helps in maintaining optimum cholesterol levels, managing type 2 diabetes, and promoting healthy skin (MOHAMMED, 2019). Cinnamon has been used for centuries as a culinary ingredient and a traditional medicine. More recently, it has been considered a complementary agent for controlling symptoms of diabetes, metabolic syndrome, and other conditions (KEITH, 2019). These data confirm the value of compositions Nos. 2 and 4.

4.3. Antioxidant activity

Total antioxidant activity consists of the chemical properties of substances that are restorative. Our work shows that plants rich in antioxidants grow in hot and temperate climates. Among the raw materials from the tropics and subtropics, cinnamon was the leader ($0.250 \pm 0.250 \text{ mg cm}^{-3}$). In temperate climates, willow-tea leaves and sea buckthorn leaves are valued; their AOA was the same: $0.238 \pm 0.024 \text{ mg cm}^{-3}$.

When analyzing the compositions, it was shown that the ingredients enhanced each other's effects in 3 out of 5 developed mixtures. We noted the maximum synergy in combining *I. obliquus* with lemongrass, lemon balm, cinnamon, and apple. The mechanism of synergistic antioxidant activity in heterogeneous products is associated with hydroperoxyl radicals and further reduction of quinones to catechols (YAFANG et al., 2020).

4.4. Correlation analysis

Regardless of the category of ingredients, a close relationship was established between the content of polyphenols ($r=0.76$), tannins ($r=0.92$), and antioxidant activity. This is quite natural since polyphenols and tannins are part of the substances that provide the antioxidant potential of plant products.

However, no similar high-level correlations were found for mixtures. This is explained by the phenomena of antagonism, synergy and summation of the action of biologically active substances that pass into the extract from different ingredients. One antioxidant's protective action can also achieve the synergistic antioxidant effect through sacrificial oxidation. The mechanism responsible for synergistic antioxidant activity has not been explained yet, mainly due to the complex nature of the mixture, especially plant extracts. For example, it can be the formation of dimers, adducts, and/or new phenolic products with a greater antioxidant power than the mixture of the parents' compounds. In the literature, one can find several hypotheses explaining the mechanism of antagonistic action in a complex mixture: the stronger antioxidant (including polyphenols) generates a weaker possibility of complex and adduct formation between antioxidants and the possibility of antioxidants polymerization, which causes a decrease in their antioxidant properties. The additive effect often indicates the absence of interactions between the antioxidants (synergism or antagonism) during the oxidation (radical neutralization). This effect occurs when single antioxidants act independently (OLSZOWY-TOMCZYK, 2020).

5. CONCLUSIONS

It was found that plants from geographically distant regions may have similar beneficial properties. Of the five plants grown in tropical and subtropical zones, *Citrus limon* and *Cinnamomum verum* showed the highest levels of biologically active substances. From a set of plants and their

parts selected in a temperate climate, the leaves of *Hippophaë rhamnoides* differed in useful properties. Consequently, finding the most useful raw vegetable materials locally for preparing functional food and drinks is possible in each geographical area.

The trends of globalization have also affected food production. Manufacturers often combine raw materials from different geographical areas. The effects of simple summation, antagonism, and synergism of beneficial properties were established when comparing the arithmetic sum of the BAS content of individual phyto-tea ingredients and experimentally measured composition indicators. Due to the antagonism of all the studied BAS (polyphenols, tannins, and the amount of antioxidants), we do not recommend the combination of *I. obliquus*, *C. limon*, and *H. rhamnoides* to prepare hot drinks. On the contrary, the synergy of BAS was observed in the compositions *I. obliquus*, *C. nardus*, and *M. officinalis* (for polyphenols and tannins), *I. obliquus*, *H. sabdariffa*, and *M. officinalis* (for polyphenols), *I. obliquus*, *C. verum*, and *P. malus* (for the sum of antioxidants). Therefore, these combinations can reasonably be attributed to a superfood.

Combinations of raw plant materials from different geographical locations may differ in advantageous taste properties and a combination or even enhancement of useful properties. When developing a functional phyto-tea, the study of the joint effects of BAS proved to be an effective approach.

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