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RESEARCH OF FLAVOUR CHARACTERISTICS OF BEER WITH ADDITION OF PINE EXTRACT

Об'єктом дослідження є пиво світле, виготовлене за класичною рецептурою (контроль), та пиво «Смарагд», виготовлене за класичною рецептурою з додаванням хвойного екстракту. Одним з найбільш проблемних місць є те, що розширенням асортименту крафтового пива призводить до появи продукції з додаванням численних синтетичних компонентів та ароматизаторів. Дегустаційний шлях оцінки напою є досить суб'єктивним, оскільки не може в повній мірі охарактеризувати вплив якісного складу та кількісного вмісту компонентів використаної сировини. Тому важливо звернути увагу на визначення загальної кількості речовин, які утворюються в процесі бродіння пивного сусла та формують натуральний смак і аромат готового продукту.

Для проведення експериментальних досліджень в якості основної сировини використовували солод ячмінний світлий, гранули хмелю, пивні дріжджі низового бродіння. Додатковою сировиною обрано хвою сосни звичайної. У якості методів дослідження використовувався метод високоефективної газової хроматографії та адаптований метод визначення числа аромату.

Визначено концентрацію смакоароматичних речовин: пиво контроль має 32 компоненти, «Смарагд» — 38, проведено їх розрахунки. Визначено, що до факторів, які визначають смак і аромат пива, відносять побічні продукти спиртового бродіння.

Для повної оцінки виваженості аромату розробленого пива було визначено загальний вміст речовин, що обумовлюють аромат напою. Встановлено, що пиво «Смарагд» має число аромату 2740 мл $Na_2S_2O_3/100$ мл, контроль — 2170 мл $Na_2S_2O_3/100$ мл. Це в повній мірі характеризує вплив на готовий продукт кількісного вмісту ароматоутворювальних речовин додаткової сировини.

Позитивний ефект від розробленого пива «Смарагд» було досягнуто за рахунок: натуральних компонентів, поліпшених органолептичних показників готового напою, цікавості споживача до нового продукту. Також високі антиоксидантні властивості додаткової сировини сприяють зменшенню негативного впливу алкоголю на організм людини. За рахунок часткової заміни хмелю отримано меншу ціну на продукт у порівняння з товаром-аналогом та зменшення негативного впливу хмелю на організм людини.

Ключові слова: пиво з додаванням хвойного екстракту, заміна хмелю, смакоароматичні речовини, число аромату, аромат та смак пива.

1. Introduction

Beer refers to beverages of mass demand, is an alternative to the consumption of strong alcoholic beverages and is targeted at different age groups, especially young people. The brewing industry is expanding due to the introduction of new technologies, modern equipment, original recipes and more than 880 varieties. Now the situation in the beer market can be compared with the industrial revolution. Over the past few years, the development of brewing in the world market is associated with the emergence of a new trend – craft beer [1].

In the alcoholic beverage industry, food additives are widely used. Therefore, the main reason for the increased interest in craft brewing is the opportunity to offer consumers unconventional types of foam beverage that combine all the advantages of classic beer with the addition of original components. Natural ingredients improve the quality of the product, especially their organoleptic properties.

Hop is an indispensable raw material in brewing, because it gives beer a specific flavor and promotes pinholes, stability during storage. But this component is also the

most expensive in the production of beer. In addition, research scientists [2, 3] show that when it is used excessively there are manifestations of the negative influence of hop on the human body. Scientists consider the substitution of hop for vegetable raw materials, will provide beer with certain functional properties [4]. The chemical composition of pine needles is most approximated to the composition of hop cones (polyphenolic, bitter and pectin substances, essential oils, etc.) [5, 6]. Pine needles are a source of natural antioxidants, possess high nutritional and biological value, therefore, can be used as an alternative to hop [7].

Combining different types of malt, hop varieties and other ingredients, brewers create a unique product, boiled in a special recipe and technology. The use of other additives should be aimed at improving the taste or obtaining original taste characteristics. According to the expansion of the assortment of the industry and the growth of minibreweries, the quality requirements for finished products are increasing, which must meet high standards. So, it is actual to search for natural plant raw materials, which could enhance the organoleptic properties of beer and provide it with functional properties.

2. The object of research and its technological audit

The object of research is a light beer made according to the classical recipe and beer «Emerald», made according to the classical recipe with the addition of pine extract.

To carry out the experimental studies, malt barley light, hops granules, brewer's yeast of bottom fermentation were used as the main raw material. When choosing an additional component for beer production, the high antioxidant properties of pine needles were taken into account [7].

Beer «Emerald» was prepared according to the classical technology with the addition of an aqueous extract of pine needles (Pinus sylvestris) at the stage of the main fermentation. In detail with the technology of obtaining pine extract, studies of its quality indicators and developed beer «Emerald» can be found in [7, 8].

The use of pine extract forms the original organoleptic parameters of the quality of the beverage. The taste and aroma of beer is primarily determined by volatile by-products of fermentation, which are the basis in the formation of the sensory profile of beer.

One of the most problematic places is that the expansion of the assortment of craft beer leads to the emergence of products with the addition of numerous synthetic components and flavors. The tasting method for evaluating the beverage is rather subjective, since it can't fully characterize the effect of the qualitative composition and the quantitative content of the components of the used raw materials. Therefore, it is important to pay attention to the determination of the total amount of substances that are formed during the fermentation of beer wort and form the natural taste and aroma of the finished product.

3. The aim and objectives of research

The aim of research is improvement of the organoleptic parameters of beer by adding additional plant material.

To achieve this aim, it is necessary to solve the following tasks:

- 1. To investigate the component composition of the volatile flavors of the experimental beer samples.
- 2. To determine the total amount of substances (the number of flavor) those determine the flavor of beer.

4. Research of existing solutions of the problem

A wide variety of ingredients in the production of craft beer has been widely used. As a flavor additive, mini-breweries use medicinal and spicy-aromatic plant raw materials, it allows to obtain original flavor and aromatic characteristics of the beverage. At the same time, the development of new types of alcoholic products is growing rapidly. Beverages are widely spread, based on numerous synthetic components and flavors. Therefore, it is important to pay attention to the determination of the total amount of substances that are formed during the fermentation of beer wort and form the natural taste and aroma of the finished product.

Researches of scientists testify that partial replacement of hops makes it possible to reduce its negative impact on the human body and expand the assortment of the beer industry by creating new grades of craft beer. Thus, in work [9] a method for the production of health-

improving rice beer was developed, into the formulation of which extracts were introduced: coniferous, ginseng and the root of wild lancelet. The beverage has an original taste and aroma.

The work [10] proposed the production of beer with the addition of a pine additive, the proportion of which is 0.8 % of beer wort. The additive, prepared from bark powder, cones and pine needles, is introduced at the boiling point with hop. This allows to improve the taste properties of beer and increase the biological value and medicinal properties of the finished beverage.

To the factors that determine the taste and aroma of beer include by-products of alcohol fermentation. The tasting method for evaluating the beverage is rather subjective, since it can't fully characterize the effect of the qualitative composition and the quantitative content of the components of the used raw materials. The consumer can evaluate the taste of the beverage if the actual concentration of flavoring substances exceeds the threshold concentration of their taste recognition [11].

A more stable beverage with clear hops taste of beer without hop using mint and basil has been studied [12], which allows reducing the dependence of the beer industry on expensive hops.

In recent years, the European Brewing Convention (EBC) has been successfully used in the world brewing industry with the assistance of the American Society of Brewing Chemists (ASBC) with a timetable and a standard description of the most important tastes and flavors of beer that are accepted and understood internationally [13].

To the international community of brewers, scientists offer generalized indicators of «biological value», «total dose of taste» and «total dose of toxicity» aimed at harmonizing with international norms of nutritional science and introducing a system of quality control and beer safety to the modern methodological level. New approaches to assessing the organoleptic and toxicological characteristics of the main products of fermentation of beer make it possible to identify substances that form the standard of permissible toxicity and flavors of the beverage [14].

The quantitative analysis of aromatic alcohols of beer was developed using gas chromatography [15]. A new and direct method for monitoring the aging of beer has shown the possibility of differentiating its species into ecologically clean to non-ecological [16].

In [17], using a solid-phase microextraction based on the free space associated with gas chromatography-mass spectrometry, quantitative differences of the volatile beer profile were analyzed.

To fully assess the weight balance of the flavor of the developed beer, it was advisable to determine the total content of the substances that determine the flavor of the beverage. This will make it possible to fully characterize the effect on the finished product of the quantitative content of the used aroma-making raw materials [18].

Thus, the results of the analysis make it possible to conclude that a comprehensive assessment of the flavoring substances that form the organoleptic characteristics of the beverage is promising.

5. Methods of research

The identification of flavor substances was determined by standard gas chromatography on an Agilent 7890 GC System gas chromatograph (manufactured by Agilent Technology, USA). Adhere to the appropriate temperature mode of the chromatograph. Since the injection of the raw material into the evaporator of the chromatograph in the thermostat, the columns adhered to the initial temperature of 15 °C, which was gradually increased to 220 °C at a rate of 35 °C/min. The sample volume is (1.0±0.1) $\mu l,$ the experiment time is 16–22 min.

The number of beer aroma was determined by the general method of carrying out physical and chemical studies and adapted the method for determining the number of flavor, given in GOST 8756.7-70.

The method is based on the ability of the chromium mixture to oxidize essential oils. By the amount of expended potassium dichromate, the content of aromatic substances in the test product was established.

The plant (Fig. 1) was assembled, which consists of a flask 1, a funnel 2, which valve 3 is closed, and a reflux condenser 4. The distillation flask containing the contents of distilled water and a sample of beer is heated and the essential oils are distilled into a hopper filled with a chrome mixture. The resulting distillate was heated under reflux in a water bath at a temperature of 95 °C for 1 hour. After cooling, a solution of potassium iodide was added and titrated with a solution of sodium thiosulfate.



Fig. 1. Scheme of installation for determining the number of flavor of beer: 1 – a flask; 2 – funnel; 3 – valve; 4 – reflux condenser

In parallel, the control titration of 5 ml of the original chromium mixture was carried out under the same conditions, replacing the distillate with the essential oil with distilled water. The aromatic content was expressed conditionally in ml of sodium thiosulfate per 100 ml of product (ml $Na_2S_2O_3/100$ ml).

The content of aromatic substances (X) was calculated by the formula:

$$X = \frac{(2 \cdot V_o - V) \cdot K \cdot 100}{G},$$

where V — the amount of 0.1 M sodium thiosulfate solution used for titrating the test solution, ml; V_o — the amount of 0.1 M sodium thiosulfate solution used for control titration with 5 ml of chrome mixture, ml; K — correction factor for 0.1 M solution of sodium thiosulfate; G — the amount of the test product in ml.

6. Research results

The result of complex biochemical processes occurring during fermentation and fermentation of beer is the production of a product with a certain composition, taste and aroma.

The first stage of the research was the study of the component composition of the flavoring substances of beer. Determination of the content of compounds in the samples was carried out by the method of high-performance gas chromatography, which allows the determination of the concentrations of flavor substances, enabling their calculations. Chromatograms of the component composition of the flavors of the test samples are shown in Fig. 2–5.

Beer (control) has in its composition (Fig. 2, 3):

- ethyl ether (peak 19.927);
- acetic acid (peak 7.637);
- methyl ester (peak 8.093);
- heptyl alcohol (peak 16.527);
- aldehyde 2-butynyl (peak 8.806);
- butene acid (peak 18.882);
- aldehyde pentanal (peak 12.995);
- ethane methyl ester (peak 13.732).

In total, in the beer control, 32 taste-aromatic components were identified, the calculation of which is given in Table 1.

According to Fig. 4, 5 in the composition of the beer «Emerald» includes:

- butene acid (peak height 18.882);
- acetic acid (peak 7.639);
- isopentanol (peak 11.365);
- pentanal (peak 12.994);
- diethylamine (peak 14.890);
- methyl ester (peak 8.092);
- ethyl ether (peak 20.719);
- alcohol butantentrol (peak 21.552).

Additionally found substances:

- cyclobutyl alcohol (peak 5.821);
- methylbutane (peak 10.170);
- methylhydrogen sulfate (peak 10.373);
- butane acid (peak 10.731).

Total identified in the beer «Emerald» 38 flavoring components, the calculation of which is shown in Table 2.

In the samples of beer are higher alcohols, ethers, aldehydes, organic acids, etc., which are formed during the enzymatic decomposition of carbohydrates of wort and form the aroma and taste of beer. Organic acids are found in beer as salts. Their main function is suppressing the multiplication and action of many harmful microorganisms.

It is determined that by-products of alcoholic fermentation refer to the factors that determine the taste and aroma of beer. The analysis of the conducted researches testifies that the maintenance of flavoring substances in the developed beverages is more than in the control.

The next stage is the determination of the quantitative content of aromatherapy materials used in the finished product. The determination of the amount of aroma is carried out by a method which is based on the ability of the chromium mixture to oxidize essential oils. The amount of potassium dichromate expended is determined by the content of aromatic substances in the investigated beverage. The results of studies of the number of aroma are given in Table 3.

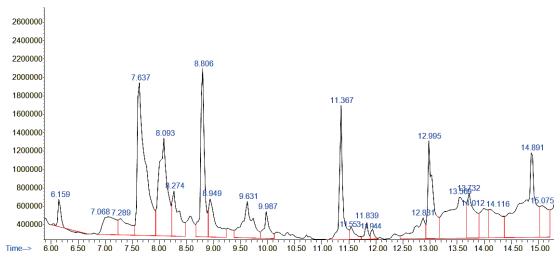


Fig. 2. Chromatogram of the component composition of volatile flavors of beer sample control (15 min)

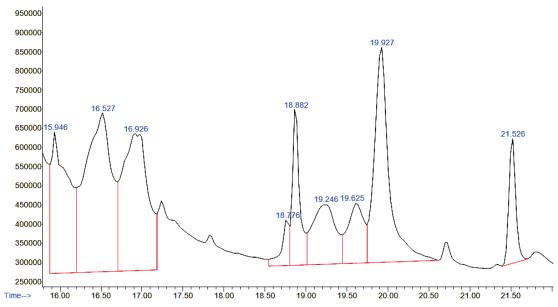


Fig. 3. Chromatogram of the component composition of volatile flavors of beer control samples (16-22 min)

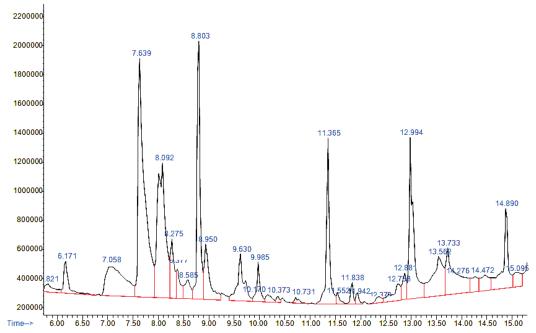
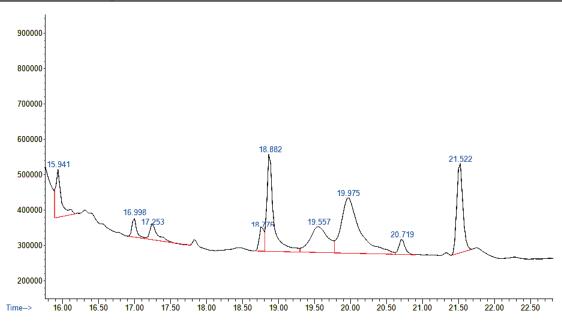


Fig. 4. Chromatogram of the component composition of flavors of the beer sample «Emerald» (15 min)



 $\textbf{Fig. 5.} \ \ \textbf{Chromatogram of the component composition of flavors of the beer sample *Emerald** (16-22 min)$

Calculation of chromatograms of a beer sample (control)

Table 1

Time, min	Component	Peak height	Peak area	Content, %	Content, mg/l
6.159	valeric aldehyde	288102	10061568	0.580	0.000354981
7.068	isopropylamine	193241	31365285	2.42	0.001487208
7.289	ethylene oxide	174444	24385099	1.88	0.001156237
7.637	acetic acid	1631582	169286179	13.05	0.008026826
8.093	methyl ether	1005700	111260410	8.58	0.005275492
8.274	2-rentanol	475055	42677949	3.29	0.002023606
8.806	2-butynyl	1767543	89086437	1.59	0.004224098
8.949	2-butanol	402231	38298042	6.87	0.004224098
9.631	propane	371701	48600937	2.95	0.001815929
9.987	heptanal	275163	18522492	3.75	0.002304448
11.367	gamma-aminobutyric acid	1239120	59792024	1.43	0.000878257
11.553	methyl-1-butanol	133753	9544649	4.61	0.002835981
11.839	bioxiran	172889	7842527	0.75	0.0004538
11.944	furandiol	105049	4906952	0.61	0.000373155
12.881	amyl alcohol	219412	28368611	0.38	0.000234122
12.995	pentanal	989689	68680500	0.11	0.000703531
13.569	2-pentanal	444573	98342441	3.27	0.002013435
13.732	ethane methyl ester	46133	48547951	3.86	0.002375153
14.012	ethane	318429	30574263	2.13	0.001312468
14.116	epoxybutane	307817	48276485	1.2	0.000740219
14.891	vinyl acetate	914407	149194903	1.81	0.001111286
15.075	dimethylamine	341919	36401758	7.69	0.004727687
15.385	acetamide	7468259	521830131	1.81	0.001115536
15.946	2,3-dihydroxypropanal	354305	53514637	2.81	0.001728717
16.527	heptyl alcohol	410718	93369275	5.40	0.003318789
16.926	methyl acetate	355823	77848006	4.62	0.002843052
18.776	acetic aldehyde	118021	7249688	0.36	0.000223375
18.882	butenoic acid	401769	25448034	1.82	0.001118983
19.246	2,3,6,7-tetramethyl-9,10-4-methylphenylsulfonyl	154803	30647641	2.13	0.00130808
19.625	2,3,6,7-tetramethyl-10-4-methylphenylsulfonyl	154568	21186817	1.51	0.00092958
19.927	ethyl ether	552215	70859227	5.31	0.00326724
21.526	1,2,3,4-butatentrol	315649	18782298	1.45	0.000890576

Table 2

Calculation of chromatograms of a sample of beer «Emerald»

Time, min	Component	Peak height	Peak area	Content, %	Content, mg/l
5.821	cyclobutyl alcohol	54884	6814108	0.74	0.000323096
6.171	valeric aldehyde	214824	14337738	1.55	0.000679834
7.058	ethylene oxide	199554	52754440	5.71	0.00250139
7.639	acetic acid	1586526	148342671	16.04	0.007033775
8.092	methyl ether	882420	102301729	11.06	0.00485071
8.275	2-pentanol	390467	21227211	2.30	0.001006503
8.377	propanamide	199374	11313915	1.22	0.000536457
8.585	2-butanol, 3-methyl	129069	10931127	1.18	0.000518307
8.803	2-butanal	1746572	76761667	8.30	0.00363971
8.950	propane	363668	28439044	3.08	0.001348458
9.630	heptanal	304497	28189702	3.05	0.001336635
9.985	gamma-aminobutyric acid	247371	11704147	1.27	0.000554961
10.170	methylbutane	48420	4192348	0.45	0.000198783
10.373	methylhydrogen sulfate	41656	1821650	0.20	8.63748E-05
10.731	butanoic acid	35698	2463443	0.27	0.000116806
11.365	isopentanol	972338	47890585	5.18	0.002270767
11.552	bioxiran	74498	3697617	0.40	0.000175325
11.838	furandiol	139190	5280111	0.57	0.00025036
11.942	amyl alcohol	75341	3313474	0.36	0.000157111
12.370	acetoin	41489	3314469	0.36	0.000157158
12.758	1,5-dietrobiopentane	113691	12393263	1.34	0.000587635
12.881	adenosine	177938	9605098	1.04	0.000455433
12.994	pentanal	1024621	66392776	7.18	0.003148061
13.562	2-heptanol, 6-methyl	260814	40158796	4.34	0.001904158
13.733	ethane	297792	45393626	4.91	0.002152371
14.276	ethanol	101593	10277444	1.11	0.000487312
14.472	vinyl acetate	104349	13482097	1.46	0.000639263
14.890	diethylamine	521704	38823770	4.20	0.001840857
15.095	acetoid	96344	10036624	1.09	0.000475894
15.941	propanal	123257	7742370	0.84	0.00036711
16.998	2-heptanal	50411	2402751	0.26	0.000113928
17.253	methyl acetate	42900	3377856	0.37	0.000160163
18.776	acetic aldehyde	69169	2744418	0.30	0.000130129
18.882	butenoic acid	270408	18486706	2.00	0.00087656
19.557	2,3,6,7-tetramethyl-9,10-4-methylphenylsulfonyl	72832	12436876	1.35	0.000589703
19.975	2,3,6,7-tetramethyl-10-4-methylphenylsulfonyl	156957	28539921	3.09	0.001353241
20.719	ethyl ether	42065	2542084	0.28	0.000120535
21.522	butantentrol	249772	14722821	1.59	0.000698093

Table

The results of determining the number of aroma in the experimental beer samples $(n=5,\ P\geq 0.95)$

Beer name	The amount of a 0.1 M solution of $Na_2S_2O_3$ consumed for titration with 5 ml of a chromium mixture, ml	The amount of aroma (ml Na ₂ S ₂ O ₃ /100 ml)	
Control beer	9.4	2170	
«Emerald»	15.1	2740	

According to the research results it is found that the beer «Emerald» has a flavor number of 2740 ml of $Na_2S_2O_3/100$ ml, beer control – 2170 ml of $Na_2S_2O_3/100$ ml. Thus, the developed sample has a greater amount of sub-

stances that determine the flavor of the beverage by adding a pine extract.

The carried out researches of beer «Emerald» testify that addition of a pine extract positively influences taste and aroma of beer. This makes it possible to obtain a new product with original organoleptic properties, which is attractive to the modern consumer.

7. SWOT analysis of research results

Strengths. The strengths of the developed product include:

– natural components and improved organoleptic characteristics;

- high antioxidant properties;

- reduction of the negative influence of hops and alcohol on the human body;
- consumer's curiosity about the new product;
- reduction of the price for the developed product in comparison with the goods-analogue due to the partial replacement of hop.

Weaknesses. The weaknesses of the developed product include:

- additional costs for preparing the extract;
- poor consumer awareness of the new product.

Opportunities. Additional opportunities to achieve the objectives of the study, are in great potential of this raw material, has high antioxidant properties. Also, coniferous extract can be used in the production of other alcohol products.

Threats. Threats when a new product is released to the consumer market include:

- possibility of the appearance of new analogues;
- growing competitive pressure, as a result of the emergence of new competitors;
- decline in the purchasing power of the population.
 Based on the SWOT analysis, the following strategic solutions are proposed:
 - access to new markets;
 - active role of marketing.

When carrying out marketing activities, it is necessary to focus on the consumer properties of the product, its nutritional value, high organoleptic and antioxidant properties, reduction of the negative influence of alcohol on the human body.

8. Conclusions

- 1. The component composition of volatile flavors of beer is studied by gas chromatography and graphically and tabulated methods are presented graphically. It is determined that beer control has 32 flavoring components, beer «Emerald» 38, their calculations are carried out.
- 2. The total amount of substances that determine the flavor of beer is determined. The flavor of the beer «Emerald» is 2740 ml of $Na_2S_2O_3/100$ ml, beer control 2170 ml of $Na_2S_2O_3/100$ ml. The aroma number technique adopted can be used to determine the assessment of the quality of alcoholic beverages.

On the basis of the conducted researches it is established that one of the ways to improve the organoleptic parameters of beer is the addition of additional plant raw materials, at the same time, by-products of fermentation that form the taste and aroma of beer are formed.

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