STUDY OF BALSAMIC POPLAR EXTRACT OBTAINED FROM DIFFERENT PARTS OF THE PLANT USED AS BIOSTIMULANT

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The qualitative and quantitative composition of extracts obtained from poplar wood waste (buds, leaves, bundles, twigs) generated from logging activities was investigated. Then, the effects of the poplar extract on the morphogenesis, physiological and biochemical parameters, and productivity of white cabbage plants were studied and were found to be significant. Thus, for the plants grown from seeds soaked in a 0.03% aqueous emulsion of the extract, the respiration rate was 2.7 mg CO_2 per 1 g of dry matter, which is 540% higher in relation to control 1 (C_1), and 270% in relation to control 2 (C_2), and a higher yield was obtained compared to other treatment options. The respiration intensity and productivity of plants that received foliar feeding with a 0.003% aqueous emulsion of the extract also exceeded the corresponding indicators in the control samples. Thus, the respiration intensity in the initial growth phase of the rosette and roots was 1.9 mg CO_2 per 1 g of dry matter, which is 380% in relation to control 1 (C_1), and 190% in relation to control 2 (C_2). The yield obtained was 1008 c/ha.

Keywords: *Populus balsamifera*, extraction, poplar, ethanol, cabbage seeds

INTRODUCTION

Biostimulants (BIOs) are defined as "materials that contain substance(s) and/or microorganisms, whose function, when applied to plants or the rhizosphere, is to stimulate natural processes, to enhance/benefit nutrient uptake, nutrient efficiency, tolerance to abiotic stress, and/or crop quality, independently of its nutrient content".[1,2] In this article, an in-depth study was conducted on the effect of plant extract biostimulants on major crops under different environmental conditions.[1]

The poplar extract is a plant growth stimulator due to the presence in its composition of saturated and unsaturated fatty acids, lipids, phenolic and polyphenolic compounds, amino acids, essential oils, and vitamins, which provide a stable growth-stimulating effect.[3] In the current situation, the use of physiologically active substances (PAS), including growth regulators, vitamins, humic substances, antibiotics, organic acids, microelements, *etc*., deserves special attention. Physiologically active substances, in small dosages, improve nutrition, increase crop yields, and improve the quality of the resulting products.[4] The mechanism of the biostimulatory activity of plant extracts is due to the active secondary metabolites they contain. They trigger various internal reactions, helping plants to better withstand environmental stress and ultimately promoting plant growth and development. However, it should be clarified that the benefits of plant extracts on plant growth regulation are not the result of a single active substance, but of the coordinated action of multiple substances, such as plant growth-regulating hormones, antioxidants and osmoprotectants.[1]

The favorable chemical composition of the feedstock allows the use of poplar extract as a cheap and environmentally friendly raw material to produce biological products.

Gibberellins, which are part of poplar buds, are plant growth stimulants, they accelerate the development of foliage and seed ripening. Since

gibberellins cause a sharp acceleration in the growth of green mass of plants, their use should be accompanied by increased plant nutrition.[5,6]

The organic acids that make up the poplar extract have a great influence on the growth and development of plants: malic, tartaric, citric, succinic acids can increase the alkaline reserve of the body and influence metabolic processes. Unsaturated fatty acids of poplar oil, especially linoleic, linolenic and arachidonic acids, play an important role in metabolic processes. Unsaturated fatty acids are part of cell membranes and other structural elements of tissues and participate in metabolic reactions, ensuring the growth process and normal structural functions, which is especially important during tissue processes.[7]

Poplar leaves contain up to 56% of extractive substances, and alcohol-soluble substances account for up to 67% of the extract amount. Flavonoid and salicylic compounds have been identified in the chemical composition of poplar leaves. In addition, the presence of salicortin, tremulacin and chlorogenic acids was confirmed.[8] The presence of various groups of compounds has been established: hydrocarbons, alcohols, acids, ketones, *etc*., including biologically active substances, such as megasterol acetate, 1-sitosterol, sitosterol, 3,7,11,15-tetramethyl-2-hexadecene-1-ol, diethyltoluamide, 4-methoxy-3-nitrobiphenyl and other compounds.[9]

Flavonoids are the most extensive group of phenolic compounds and an important component of the plant body. In plant systems, flavonoids help combat oxidative stress and act as growth regulators. They play an active part in redox processes, the development of immunity, and the protection of plants from the adverse effects of ultraviolet rays and low temperatures. Flavonoids are active metabolites of plant cells. The important biological role of these compounds is evidenced by the nature of their distribution in the plant. Most flavonoids are found in actively functioning organs: leaves, flowers, fruits (color, aroma), seedlings, as well as in integumentary tissues that perform protective functions.[11] Flavonoids play an extremely important functional role in plant-environment interactions. Recent data on the nuclear location of flavonoids (as well as flavonoid biosynthetic enzymes) suggest that flavonoids can modulate the activity of proteins involved in cell growth. Thus, flavonoids may act as transcriptional regulators.[12,13] These reactions are important for plant physiology.

Tannins are a form of reserve nutrients. This is indicated by their localization in underground organs and cortex; tannins perform a protective function, because when plants are damaged, they form complexes with proteins that create a protective film that prevents the penetration of phytopathogenic organisms. They have bactericidal and fungicidal properties; they participate in redox processes and are oxygen carriers in plants.[14]

Coumarins are involved in the actions of plant growth hormones and growth regulators, in the control of respiration, photosynthesis, and protection against infections. It has long been recognized that coumarins have anti-inflammatory, antioxidant, antiallergic, hepatoprotective, antithrombotic, antiviral and anticarcinogenic activity. Hydroxycoumarins are typical phenolic compounds and therefore act as potent metal and free radical chelators. They are powerful chain-breaking antioxidants. Coumarins have a remarkable range of biochemical properties and pharmacological actions; some members of this group of compounds can significantly influence the function of various cellular systems.[15]

Saponins in low concentrations accelerate seed germination, growth and development of plants, and in high concentrations, on the contrary, slow them down. Thus, saponins play the role of plant growth hormones; saponins affect the permeability of plant cells, which is associated with their surface activity.[16]

Amino acids are of key importance for the development of flora. In plant organisms, they appear during the process of photosynthesis and then participate in a huge list of biochemical reactions, supporting optimal growth and development.[17]

Chlorophyll is found in almost all green parts of plants, that is, in leaves and stems, inside the chloroplast, the main organelle containing the largest amount of chlorophyll. Chlorophylls have become the subject of extensive research due to their prominent role in plant physiology and their role as derivatives in the food sector.[18]

Vegetable crops, especially cabbage, are grown as a staple crop over large areas in the southern region of Europe, Central and South Asia, North and South America, Australia and other regions. To meet the population's demand for this product, it is important for agricultural producers to improve the technology for growing it, including optimal planting dates, planting patterns, plant

nutrition, fertilizer and watering rates, as well as the selection of commercial and high-yielding varieties and hybrids.[19]

Faisal Zulfiqar *et al*. have presented examples illustrating the positive effects of biostimulants on plant growth and yield, as well as on a few physiological and biochemical characteristics of several horticultural crops. The mechanism of their action by improving key physiological, biochemical and molecular processes is also discussed. These mechanisms work from increasing soil nutrient availability to improving postharvest fruit quality, even under environmental stress.[10]

In our previous study, balsam poplar essential oil was obtained by the barothermic method. It was found that the use of an aqueous emulsion of poplar bud oil had a significant impact on the morphogenesis, physiological and biochemical parameters of tomato seeds.[20] Biofertilizers promote the growth and development of plants throughout the entire life cycle of the crop from seed germination to plant maturity in different ways, depending on their nature, composition, mode of action, period of application and growing conditions. Organic production and consumption are stable and growing worldwide.[21]

The purpose of the present study is to evaluate the effect of poplar extracts on the growth and development of white cabbage. The qualitative composition of the extracts obtained from various parts of *Populus balsamifera* logging wastes was investigated, supporting the efficient use of industrial residues and circular bioeconomy. Thus, the growth-stimulating activity of balsam poplar extract in relation to white cabbage seeds was studied.

EXPERIMENTAL

Samples for analysis

Buds, twigs, leaves, catkins of balsam poplar (*Populus balsamifera*) were collected in March-May 2023, in the vicinity of the village of Zarechny, North Kazakhstan region of the Republic of Kazakhstan. Freshly collected poplar buds were crushed to a size of 2-5 mm, filled with 96% technical ethanol (GOST 57251-2016) to obtain an extract, and then filtered through a paper filter.

The white cabbage variety “Podarok” was used as the object of the study, the plants were grown in the field zoned in the North Kazakhstan region.

Sample preparation

Balsam poplar extracts were obtained from fresh balsam poplar catkins (harvested in May), fresh buds (collected in March), fresh twigs (collected in March), taken individually. Also, an extract from buds, twigs, catkins and leaves, taken collectively, was obtained from balsam poplar wood processing waste collected in May. The feedstock was crushed, and extraction was carried out with 96% ethanol at room temperature, followed by filtration of the solutions.

The results are presented based on four replicates of four samples, at the level of confidence of 95%. Student's t was used to find the confidence intervals and to compare mean values:[22]

$$\Delta y = t_{\alpha f}\frac{s}{\sqrt{n}} \quad (1)$$

Methods of BAS analysis

Chemical analysis of the studied extracts (poplar buds, catkins, twigs, wood processing waste) was carried out according to known methods and the method of drop analysis was investigated for the presence of the main groups of biologically active substances (BAS).[23]

Quantitative determination

For the quantitative determination of biologically active substances by the spectrophotometric method, the wavelength (max) with the maximum value of the absorption of the test solution was preliminarily determined. The data were taken using a UV Mini-1240 Spectrophotometer in a cuvette with a liquid layer thickness of 1 cm.

Quantitative determination of flavonoids

About 0.450 g (exactly weighed) of the substance (concentrated extract) was placed in a 50 mL volumetric flask, and 25 mL of 96% ethyl alcohol was added and stirred until the extract was dissolved. The volume of the solution was adjusted to the mark with 96% ethyl alcohol. Then, 5.0 mL of the resulting solution was placed in a 10 mL volumetric flask, 1 mL of a 25% aluminum chloride solution was added to it, and the volume of the solution was adjusted to the mark with 96% ethyl alcohol. After 40 minutes, the absorbance of the resulting solution was measured using a spectrophotometer at the wavelength of 430 nm in a cuvette with a liquid layer thickness of 10 mm, using as a comparison the mixture prepared in a 10 mL volumetric flask: 5 mL of the test solution was brought to the mark with 96% ethyl alcohol.

The content of the total flavonoids in terms of quercetin and dry raw materials was determined by the formula:

$$X = \frac{A*100*100*100*25}{764{,}6*m*2*(100-w)} \quad (2)$$

where A is the absorbance of the test solution; 764.6 – specific absorption indicator of the quercetin complex with aluminum chloride at 430 nm; m – mass of raw materials in grams; w – weight loss during drying of raw materials, %.

Quantitative determination of tannins in terms of pyrogallol

About 2.0 g (exactly weighed), unless otherwise specified in the pharmacopoeial monograph, of the crushed test sample, sifted through a sieve with holes measuring 1 mm, was placed in a conical flask with a capacity of 500 mL. Then, 250 mL of water was poured into the flask and it was refluxed on an electric stove, with a closed spiral for 30 minutes, with occasional stirring. The resulting extract was cooled to room temperature and filtered through cotton wool into a 250 mL volumetric flask, ensuring that the particles of the test sample did not get into the flask. The volume of the solution was brought to the mark with water and the solution was mixed. The resulting solution was filtered through an ashless filter with a diameter of about 125 mm, after discarding the first 50 mL of the filtrate.

Solution A. 1.0 mL of the filtrate was placed in a 25 mL volumetric flask, the volume of the solution was adjusted to the mark with water and mixed. The absorbance of solution *A* (A_1) was measured on a spectrophotometer at a wavelength of 266 nm (for hydrolyzed tannins) or 278 nm (for condensed tannins), unless otherwise specified in the pharmacopoeial monograph, in a cuvette with a layer thickness of 1.0 mm relative to the reference solution. Water was used as a reference solution.

Solution B. 0.05 g of plant powder was added to 5.0 mL of filtrate, the resulting mixture was stirred for 30 minutes and filtered through a paper filter with a diameter of about 125 mm. 1.0 mL of the resulting filtrate was placed in a 25 mL volumetric flask, the volume of the solution was adjusted to the mark with water and mixed. The absorbance of solution B (A_2) was measured on a spectrophotometer at a wavelength of 266 nm (for hydrolyzed tannins) or 278 nm (for condensed tannins), unless otherwise specified in the pharmacopoeial monograph, in a cuvette with a layer thickness 10 mm relative to the reference solution. Water was used as a reference solution.

Standard solution. 0.1 g (exactly weighed) of the pharmacopoeial standard sample of pyrogallol (for hydrolyzed tannins) or ((+)-catechin (for condensed tannins), unless otherwise specified in the pharmacopoeial monograph, was placed in a 100 mL volumetric flask, dissolved in water, the volume of the solution was brought to the mark with water and mixed. 2.0 mL of the resulting solution was placed in a 100 mL volumetric flask, the volume of the solution was brought to the mark with water and mixed. The solution used was freshly prepared. The absorbance of the standard sample solution (A_3) was measured on a spectrophotometer at a wavelength of 266 nm (for hydrolyzed tannins) or 278 nm (for condensed tannins), unless otherwise specified in the pharmacopoeial monograph, in a cuvette with a layer thickness of 10 mm relative to the reference solution. Water was used as a reference solution. The content of the total tannins in terms of pyrogallol or (+)-catechin in a dry test sample in percent (X) was calculated using the formula:

$$X = \frac{(A_1-A_2)\cdot a_0\cdot P\cdot 12500}{A_3\cdot a\cdot 100\cdot(100-W)} = \frac{(A_1-A_2)\cdot a_0\cdot P\cdot 125}{A_3\cdot a\cdot(100-W)} \quad (3)$$

where A_1 is the absorbance of solution A; A_2 is the absorbance of solution B; A_3 is the absorbance of a solution of a standard sample of pyrogallol; a – weighed sample of the test sample, g; a_0 is the weight of the pharmacopoeial standard sample of pyrogallol, g; P – content of the main substance in the pharmacopoeial standard sample of pyrogallol, %; W is the humidity of the test sample, %.

Quantitative determination of coumarins

Quantitative determination of coumarins in the plant extracts was carried out according to the "Method in terms of psoralen".[24]

The content of coumarins X (%) in terms of psoralen was calculated using the formula:

$$X = \frac{A*100*100*100*50}{650*a*20*10} \quad (4)$$

where A is the absorbance of the test solution at 290 nm; 650 – specific absorption rate of psoralen at 290 nm; a is the mass of a sample of raw material taken for analysis, g.

Content of total triterpene saponins

Determination of the content of the total triterpene saponins was carried out using the spectrophotometric method after a reaction with concentrated sulfuric acid, because of which triterpenoids are protonated at the double bond to form a carbocation, and in the presence of a carboxyl group at C-28, subsequent lactonization occurs. In this case, a characteristic absorption maximum is observed at 310 nm.[25,26] This method makes it possible to quantify the entire amount of triterpene glycosides, derivatives of oleanolic acid, regardless of the number and structure of carbohydrate residues in their molecules. The reference solution is concentrated sulfuric acid. At the same time, the absorbance of CO of oleanolic acid was determined under similar experimental conditions. The content of total saponins in terms of oleanolic acid was calculated using the formula:

$$X\% = \frac{Ax*m0*250*25*100*100}{A0*mx*650*25*(100-W0)} \quad (5)$$

where A_x is the absorbance of the test solution; m_o is the mass of the standard sample of oleanolic acid in g; m_x – mass of raw materials in g; Ao – absorbance of oleanolic acid; W_o is the loss in mass of raw materials during drying.

Quantitative determination of polysaccharides

To quantify the content of polysaccharides in terms of inulin, a pharmacopoeial method was used.[27] An analytical sample of raw materials was crushed to a particle size that passed through a sieve with 1 mm holes. A sample of 5.0 g of crushed raw material was placed in a flask with a capacity of 250 mL, then 100 mL of water was added and connected to a reflux refrigerator and then boiled on an electric stove (water

bath) for 30 minutes. The extraction was repeated three times with 100 mL each for 30 minutes. The resulting aqueous extracts were combined, then centrifuged at 5000 rpm for 10 minutes and filtered into a 500 mL volumetric flask through five layers of gauze, which was previously moistened with water. The filter was washed with water and the volume of the solution was brought up to the mark with water – thus solution A was obtained. Then, we took 25 mL of solution A, placed it in a centrifuge tube, added 100 mL of 95% ethyl alcohol, heated it in a water bath to 600 °C for 5 minutes, and then centrifuged it for 30 minutes at 5000 rpm. The resulting supernatant liquid was filtered under vacuum, at a residual pressure of 13-16 kPa, through a POR 16 glass filter with a diameter of 40 mm, then, it was dried to constant weight at a temperature of 100-150 °C. Then, the precipitate was transferred to a filter, and washed with 15 mL of a mixture of 95% ethyl alcohol and water (3:1). Then, the filter with the sediment was dried in air at a temperature of 100-105 °C to constant weight. The content of polysaccharides in terms of dry raw materials in % (X) was calculated using the formula:

$$X = \frac{(m2-m1)*500*100*100}{m*25*(100-W)} \quad (6)$$

where m is the mass of raw materials, g; m_1 – filter mass, g; m_2 – mass of the filter with sediment, g; W – weight loss when drying raw materials as a percentage.

Comparative analysis of obtained extracts using IR and UV-vis spectroscopy

IR spectra were collected in the mid-infrared region (4000–400 cm^{-1}) using a Varian 660 IR spectrometer. The samples were prepared in the form of thin films after evaporation of the solvent.

UV-vis spectra of the obtained extract were recorded using a UV-Visible Cary 100 Scan Spectrometer, in the region 190-900 nm, with cuvettes with an internal length of the absorbing layer b = 1 cm. The reference solution was 96% ethanol. For quantitative analysis, the extracts were diluted in 96% ethanol to obtain appropriate absorbance value (below 1) at the studied wavelength.

The dependence of absorbance at a maximum wavelength of 660 nm on the extraction time, in days, was monitored using a spectrophotometer (Spectruquant Prove 300, Merck) with cuvettes with an internal length of the absorbing layer b = 1 cm. The reference solution was 96% ethanol.

Determination of chlorophyll content in various extracts

A sample of crushed raw materials (leaves, buds, twigs, catkins and poplar wood processing waste) weighing 0.1 g was placed in a porcelain mortar, and dry calcium carbonate was added with the tip of the spatula; the mixture was ground with the addition of 2-3 mL of 96% ethanol at first, and then 10 mL of ethanol solution, and grinding was continued for 10 minutes. The resulting extract was filtered into a 25 mL volumetric flask. Extraction was carried out in stages using a pure solvent, achieving complete discoloration of the tissues of the raw materials used. The extract volume was adjusted to the mark.

To calculate the concentrations of chlorophylls a and b in the extract, the absorbance was determined spectrophotometrically at wavelengths corresponding to the maxima of the absorption spectrum of the pigments under study in each solvent.[28] For chlorophyll a in 96% ethyl alcohol, the absorption maximum is at X = 665 nm, for chlorophyll b – at X = 649 nm.

The concentration of chlorophylls a (C_a, mg/L) and b (C_b, mg/L) was calculated using the formula:

$$C_a = 13.70 \times A_{665} - 5.76 \times A_{649} \quad (7)$$

$$C_b = 25.8 \times A_{649} - 7.60 \times A_{665} \quad (8)$$

where A_{665} is the absorbance of the solution at a wavelength of 665 nm; A_{649} is the absorbance of the solution at a wavelength of 649 nm.

Evaluation of the effects of poplar wood processing waste extract on the growth and development of white cabbage

The growth and development of white cabbage plants (variety “Podarok”), in a field in the North Kazakhstan region, was studied. An aqueous emulsion of balsam poplar extract was used for foliar feeding of the plants.

The treatment protocol was differently applied to the sets of samples in the following way: 1) Sowing dry seeds in soil – control 1 (C_1); 2) Sowing in soil seeds soaked in water – control 2 (C_2); 3) Sowing in soil seeds soaked in 0.03% aqueous emulsion of poplar extract; 4) Sowing dry seeds in the soil, with foliar feeding in ontogenesis with 0.003% aqueous emulsion.

Physiological activity of extracts

The physiological activity of the extracts was determined by soaking the seeds in aqueous emulsions (0.3, 0.03, 0.003%) for 18, 24, 36 hours.

The influence of biostimulants on breathing intensity. The respiration rate of the seeds was determined using a respiratory device by I.M. Tolmachev and a titrated solution of barite $Ba(OH)_2$, which absorbs carbon dioxide released by the seeds. The experiment was carried out at a temperature of 20–22 °C. The respiration intensity was calculated from the amount of oxygen absorbed per unit of biomass per unit of time using the formulas given in the workshop on plant physiology.[29]

The influence of biostimulants on chlorophyll content. The total chlorophyll content was determined photometrically using a calibration graph using the Getry reagent. An 80% acetone solution was used for extraction.[30,31]

The influence of biostimulants on water metabolism. The transpiration rate was measured using the rapid weighing method of L.A. Ivanova: change in the weight of the system (part of the leaf) during the exposure time

– 3 min.[32] The measurements were carried out during a period of 2 hours.[33]

RESULTS AND DISCUSSION

To determine the qualitative composition of the studied extracts, known reactions to flavonoids, tannins, coumarins, saponins, amino acids and polysaccharides were used (Table 1). As can be seen from the results presented in Table 1, flavonoids are present in each of the tested extracts. Tannins are present in bud and twig extracts, but absent from the catkin extract. All studied extracts contained coumarins. Saponins were found only in the extracts of buds and twigs. Amino acids can be traced in the extracts of catkins and twigs, but are absent in the extract of buds. The content of polysaccharides was found only in the extract of the twigs; they were absent in the first two.

All classes of the studied substances were found in the extract obtained from poplar wood processing wastes (collectively). Thus, we can conclude that the extract from poplar wood waste is the most promising for study, as it contains flavonoids, tannins, saponins, amino acids and polysaccharides.

When conducting the chemical analysis of the extracts under study using the droplet analysis method, the presence of the main groups of biologically active substances was established. A quantitative analysis was carried out for the detected classes of biologically active substances in accordance with pharmacopoeial monographs. Numerical values of quantitative analysis are expressed as percentages in terms of dry plant raw materials in terms of the dryness coefficient. The moisture content in plant raw materials was 7.66%.[17–19]

The data in Table 2 show that the largest content of flavonoids (9.31 ± 0.34) and amino acids (7.72 ± 0.45) is found in the extract of balsam poplar buds; the extract from poplar twigs demonstrates the highest tannin content (7.69 ± 0.14).

Table 1
Qualitative composition of the extracts

BAS group	Qualitative reaction	Extract of			
		Buds	Catkin	Twig	Wood waste
Flavonoids	Aluminum trichloride	Formation of			
		Yellow precipitate	Yellow-green precipitate	Yellow precipitate	Yellow precipitate
Tannins	Lead acetate solution	Precipitation of curdled sediment			
		Yes	No	Yes	Yes
Coumarins	Lactone test	Formation of a white precipitate			
		Yes	Yes	Yes	Yes
Saponins	Stable foam in HCI solution	Yes	No	Yes	Yes
Amino acids	Ninhydrin reaction	Purple staining			
		No	Yes	Yes	Yes
Polysaccharides	Alcohol precipitation	Formation of a white precipitate			
		No	No	Yes	Yes

Table 2
Contents of main groups of biologically active substances

BAS group	Extract/Content (%)				
	Buds	Catkin	Twigs	Leaves	Wood processing waste
Flavonoids	9.31 ± 0.34	1.74 ± 0.04	0.98 ± 0.05	0.53 ± 0.04	6.31 ± 0.45
Tannins	3.56 ± 0.25	-	7.69 ± 0.14	-	5.33 ± 0.38
Coumarins	0.76 ± 0.03	3.54 ± 0.04	2.34 ± 0.05	0.21 ± 0.04	0.94 ± 0.03
Saponins	1.84 ± 0.17	-	2.28 ± 0.29	1.56 ± 0.33	1.95 ± 0.31
Amino acids	7.72 ± 0.45	5.85 ± 0.19	1.23 ± 0.34	4.93 ± 0.24	6.27 ± 0.43
Polysaccharides	-	-	9.39 ± 0.17	8.39 ± 0.45	7.36 ± 0.23

Coumarins are found in the greatest quantities in the extract of poplar catkins, saponins – in the extract obtained from poplar twigs. Poplar twig extract contains the largest content of polysaccharides. In the extract obtained from poplar wood processing waste, all classes of the

studied substances were found in quantities close to the maximum. Thus, the extract from poplar wood processing waste is of interest for study as a growth-stimulating agent.

Analysis of obtained extracts by IR spectroscopy

Due to their chemical composition, poplar wastes are promising raw materials for the extraction of valuable chemicals. Of particular interest are extractive substances, the main share of which are phenolic compounds. Vegetable tanning extracts, due to their varied composition, give complex IR spectra. The presence of several intense absorption bands was noted. Figure 1 presents the IR spectra, recorded for the five extracts obtained in this study.

The analysis of the IR spectra shows a significant similarity in the absorption bands of the extracts and allows identifying common structural elements. The combination of such absorption bands as aromatic bonds (1580-1500 cm^{-1}), carbonyl groups (1700-1720 cm^{-1}), hydroxyls associated with hydrogen bonds (2500-3000 cm^{-1}), stretching vibrations of hydroxyl groups (3550-3200 cm^{-1}) clearly indicates for the presence of phenolic compounds in the extracts.

From the analysis of data on frequency characteristics presented in Tables 3, it follows that all the studied extracts contain aliphatic CH_3 and CH_2 groups, as evidenced by the strong absorption in the region of 2930–2850 cm^{-1} (stretching vibrations of CH_3 and CH_2 groups) and the region of 1463–1377 cm^{-1} (deformation vibrations). The spectra of all the studied extracts contain absorption bands in the regions of 1611–1617 cm^{-1}, 1505 and 3400 cm^{-1}, characteristic of vibrations of aromatic structures. It should be noted that, along with aromatic compounds, all the extracts contain compounds with conjugated double bonds (such as conjugated dienes), as evidenced by the presence of an absorption band at 1640-1600 cm^{-1} in the spectra. The IR spectra of all the extracts, except those from poplar twigs and catkins, have intense absorption bands in the region of 1700–1735 cm^{-1}, characteristic of stretching vibrations of νC=O groups. A combined consideration of this region and the region of 3800–2600 cm^{-1}, where the bands of stretching vibrations of OH groups are located, suggests that the composition of these extracts includes carboxylic acids. Evidence is provided by the presence of a very broad band with a maximum of ~2650 cm^{-1}, related to the νOH vibrations of carbonyl groups, and an intense band in 1700–1735 cm^{-1}, related to the νC = O stretching vibration of carboxylic acids. Analysis of the observed bands in the region of 1000–1200 cm^{-1} in combination with the absorption at 1735 cm^{-1} in the carbonyl region suggests the presence of 2,3,5 keto-ester compounds in the extracts.

The wide absorption band of the all the extracts in the region of 3200-3600 cm^{-1} is due to the presence of associated hydroxyl groups that overlap the bands of C-H bonds of the aromatic ring at 1450-1650 cm^{-1}. This band is more intensive in the extract of wood processing waste.

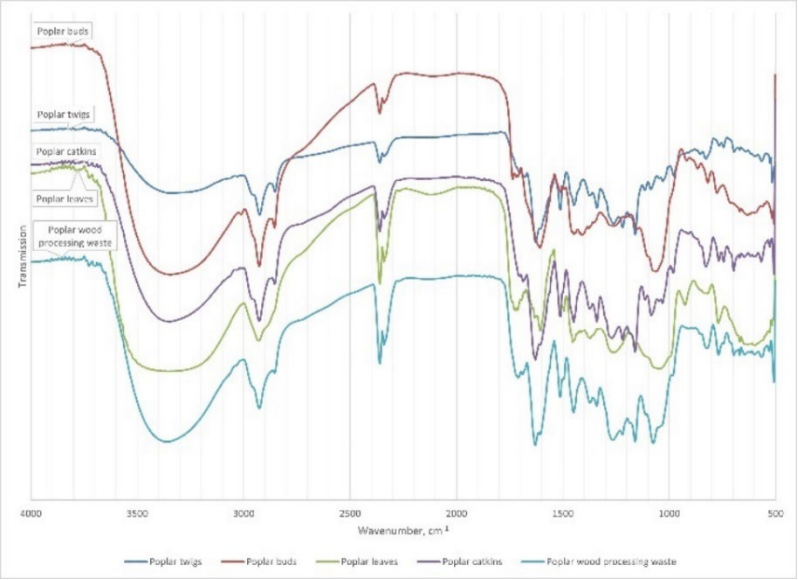

Figure 1: IR spectra of the extracts

Table 3
Absorption bands in IR spectra of the extracts and their assignment

ν, cm $^{-1}$	Intensity	Frequency range of corresponding functional group	Type of vibration and corresponding structural fragment	Functional group
2927-2924	Strong	2975-2860	Valence, CH	$-CH_2$
1454-1447	Average	1470-1430	Deformation asymmetric, CH	$-CH_3$
1376-1374	Average	1380-1370	Deformation symmetrical, CH	$-CH_3$
766-744	Average	750-720	$(-CH_2-)_4$ skeletal	$-CH_2$
1608-1624	Strong	1640-1600	Conjugated double bonds	(C═C-C═C or C═C-C═O)
2926-2924	Strong	2940-2915	Valence asymmetric, CH_2	$-CH_2$
3284-3343	Strong wide	3600-3200	Valence	OH H-bonded Alcohols, phenols,
1114-1112	Average	1116-1105	Valence skeletal	$-CH_2$ -
818-824	Average	840-800	Valence CH	-C=C- cis
1511-1510	Average	1580-1500	Benzene ring	Benzene ring
2856-2853	Strong	2820-2780	Valence	NCH_3
1340		1340-1250	Primary aromatic amines	
1261	Strong	1350-1280	Secondary aromatic amines	CN
1265		1360-1310	Aromatic amines tertiary	
1160 1219 1261	Strong	1300-960	Valence bonded group P=O	P=O Phosphates

The presence of double bonds is indicated by an absorption band in the region of 1620-1680 cm^{-1} in the IR spectrum recorded for poplar twigs. Intense absorption bands at 2850-2920 cm^{-1} characterize the stretching vibrations of the C-H bond in methoxy groups. The presence of carbonyl compounds in the aromatic ring is indicated by absorption bands in the range of 1600-1700 cm^{-1}. A joint examination of this region and the region of 3800-2600 cm^{-1}, where the bands of stretching vibrations of OH groups are located, suggests that the extract contains phenolic compounds. The absence of bands in the region of 1700-1725 cm^{-1} indicates the absence of carboxylic acids in the composition of poplar twigs extract.

In the IR spectrum of the extract from poplar catkins bands are observed in the region of 1340-1250 cm^{-1}, indicating the presence of primary, secondary, and tertiary aromatic amines in the extract. Stretching vibrations in the region of 2820-2780 cm^{-1} confirm the presence of a bond between the methyl group and nitrogen. In the region of 1200-1000 cm^{-1}, deformation vibrations of H-O bonds were recorded. The presence of heterocycles with an oxygen heteroatom is indicated by the absorption bands at 1047 cm^{-1} in the poplar leaf and poplar bud extracts.

The results of IR spectroscopy are in good agreement with the results of UV spectroscopy, indicating the complex chemical composition of poplar extractives and confirming that phenolic compounds belong to tannins in the poplar bud extract. Phenolic compounds, as effective antioxidants, when used in low concentrations, influence various biochemical and physiological processes in plants.[34]

Analysis of extracts by UV-vis spectroscopy

Samples 2 and 4 (poplar leaves and twigs) showed absorbance values of about 2.9 after 2 days of extraction, based on this, we can conclude that the period of residence of the raw material in ethanol is sufficient for its extraction (Fig. 2).

The absorption of the remaining samples increases slightly, and after the fourth day of the extraction of the material in ethanol remains relatively constant (about 1). Therefore, we can conclude that for samples 1, 3 and 4 (buds, catkins and poplar wood processing waste), 4 days of exposure of the raw material to ethanol is sufficient for the extraction. The UV-vis spectra of the obtained extracts are presented in Figure 3.

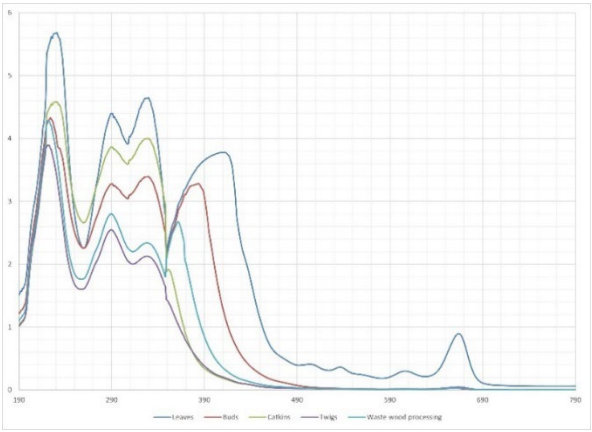

Figure 2: UV-vis spectra of all extracts in the region of 500-750 nm

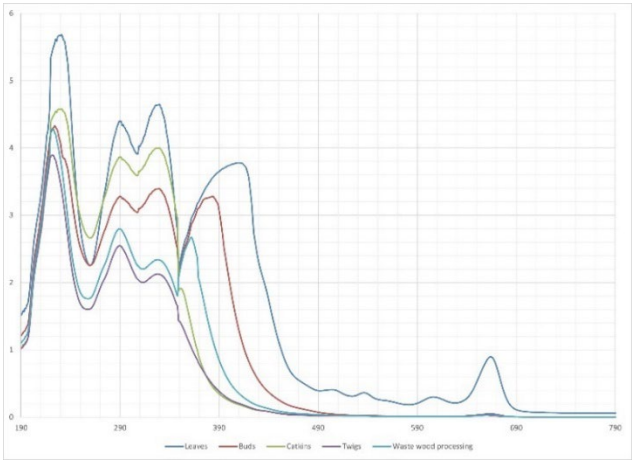

Figure 3: UV-vis spectra of all extracts in the region of 190-900 nm

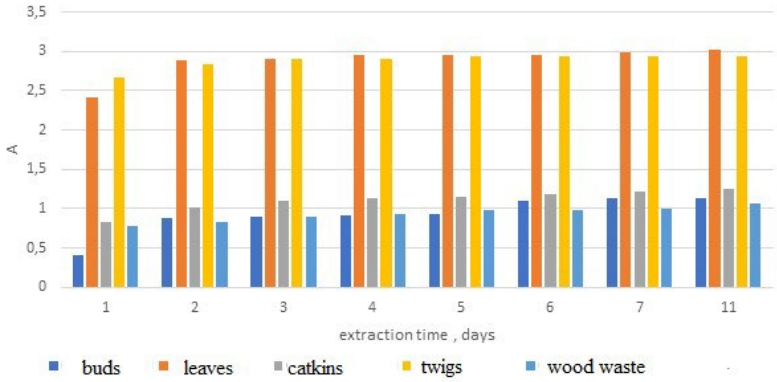

Figure 4: Extraction duration from various raw materials

Table 4
Concentration of chlorophylls a and b

Extract	Chlorophyll a concentration, mg/L	Chlorophyll b concentration, mg/L	Sum of chlorophylls a and b, mg/L
Buds	16.79	25.45	42.24
Leaves	79.55	65.73	145.28
Catkins	35.04	33.82	68.86
Twigs	18.45	12.96	31.41
Waste wood processing	15.40	17.27	32.67

Determination of chlorophyll content in various extracts

According to Table 4, the highest total content of the chlorophylls is observed in the leaf extract (145.279); in the extract obtained from poplar wood processing waste, the chlorophyll content is 32.67 mg/L, which exceeds in content calculated for the extract from twigs only.

Evaluation of the effects of balsam poplar wood processing waste extract on the growth and development of white cabbage

The rate of growth and development of a plant organism, and ultimately its productivity, largely depends on the intensity of energy metabolism, on the intensity of respiration. Research results indicate that the higher the respiration rate of plants, the faster the rate of their growth and development, and the higher the productivity.

The data in Table 5 show that the respiration rate of the white cabbage variety "Podarok" changes during ontogenesis. Moreover, this change leads to a decrease in the intensity of respiration with the onset of subsequent phases of plant growth. Thus, the intensity of respiration in the phase of accumulation of leaf mass and further growth of the root system (age 60 days) is lower than in the phase of initial growth of the rosette and roots (age 30–35 days), in all variants of the experiment. Thus, in plants grown from seeds soaked in 0.03% aqueous emulsion, the respiration rate is 2.7 mg CO_2 per 1 g of dry matter, which is 540% in relation to control 1 (C_1), and in relation to control 2 (C_2) – 270% and higher yield compared to other treatment options. The respiration intensity and productivity of plants that received foliar feeding with 0.003% aqueous emulsion also exceed the corresponding indicators in control samples. Thus, the respiration intensity in the initial growth phase of the rosette and roots is 1.9 mg CO_2 per 1 g of dry matter, which is 380% in relation to control 1 (C_1), and 190% in relation to control 2 (C_2). The yield is 1008 c/ha.

The use of biostimulants also influenced the chlorophyll content in leaves during the ontogenesis of the white cabbage variety "Podarok". The chlorophyll content in the initial growth phase of the plant rosette and roots, and during the accumulation of leaf mass and further growth of the root system increases when using an aqueous emulsion, as can be seen from the data in Table 6. During the growing season, the intensity of transpiration and the content of dry substances were determined.

The data in Table 7 indicates that the water exchange of plants grown from seeds treated with biostimulants and receiving foliar feeding during ontogenesis is not the same.

Table 5

Effect of biostimulants on the intensity of respiration in the ontogenesis of white cabbage variety "Podarok"

Phenophases Treatment option	Initial growth of rosette and roots (age 30-35 days)			Accumulation of leaf mass and further growth of the root system (age 60 days)			Harvest, c/ha
	Respiration rate, mg CO_2/g of dry matter	% relative to C_1	% relative to C_2	Respiration rate, mg CO_2/g of dry matter	% relative to C_1	% relative to C_2	
Sowing dry seeds in soil – C_1	0.5	100	50	0.3	100	75	894
Sowing in soil seeds soaked in water – C_2	1.0	200	100	0.4	133	100	990
Sowing in soil seeds soaked in 0.03% aqueous emulsion	2.7	540	270	2.3	766	75	156
Sowing dry seeds in soil + foliar feeding with 0.003% aqueous emulsion	1.9	380	190	1.7	566	425	1088

Table 6
Effect of biostimulants on chlorophyll content in the ontogenesis of white cabbage variety "Podarok"

Phenophases Treatment option	Initial growth of rosette and roots (age 30-35 days)			Accumulation of leaf mass and further growth of the root system (age 60 days)		
	Chlorophyll content, wt% of dry leaves	% relative to C_1	% relative to C_2	Chlorophyll content, wt% of dry leaves	% relative to C_1	% relative to C_2
Sowing dry seeds in soil – C_1	0.10	100	91	0.09	100	86
Sowing in soil seeds soaked in water – C_2	0.11	110	100	0.10	115	100
Sowing in soil seeds soaked in 0.03% aqueous emulsion	0.16	160	145	0.12	135	117
Sowing dry seeds in soil + foliar feeding with 0.003% aqueous emulsion	0.17	170	155	0.12	145	125

Table 7
Effect of biostimulants on water metabolism

Treatment option	Transpiration rate, mg/cm^2 h	Dry matter content, %
Sowing dry seeds in soil – C_1	1205	6
Sowing in soil seeds soaked in water – C_2	749	7
Sowing in soil seeds soaked in 0.03% aqueous emulsion	628	8
Sowing dry seeds in soil + foliar feeding with 0.003% aqueous emulsion	425	8

The tabulated data show that the plants from the experimental sets have the lowest transpiration rate, which indicates their economical use of water. The intensity of transpiration of control plants was C_1 – 1205 mg/cm^{2}h, C_2 – 749 mg/cm^{2}h. The intensity of transpiration of the plants grown from seeds soaked in a 0.03% aqueous emulsion is 628 mg/cm^{2}h, the same indicator for the plants that received foliar feeding in ontogenesis with a 0.003% aqueous emulsion is 425 mg/cm^{2}h. One of the most important indicators of the biological quality of vegetable crops is their dry matter content. It should be noted that when the seeds were treated with a 0.03% aqueous emulsion, the dry matter content was 8%, which is higher than that determined for the control variants.

CONCLUSION

The qualitative and quantitative composition of extracts of buds, twigs, catkins, and poplar wood processing waste has been established. The most promising for further study was found to be an extract from poplar wood processing waste, as it contained flavonoids, tannins, saponins, amino acids, and polysaccharides. The optimal extraction duration for each sample was determined. The composition of the extracts was analyzed using IR spectroscopy and UV-vis spectroscopy, and the chlorophyll content in the resulting extracts was determined.

The optimal concentration of the balsam poplar extract (0.03%) had a significant effect on the sowing quality of the seeds, in terms of energy and germination, which were 89-90 and 91-93%, respectively, in the treated sets, while in the control: 77-79 and 81-83%. In addition, the use of poplar extract had a significant impact on the morphogenesis, physiological and biochemical parameters, and productivity of white cabbage of the "Podarok" variety, grown in Northern Kazakhstan. The effect of using the extract to treat the seeds and as foliar treatment was traced in plant ontogenesis through changes in important physiological processes, which affected product yield and quality.

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