UDC 544.23.022, 544.478.02

DOI: 10.15372/CSD2023515

EDN: JYFYDX

Qualitative and Quantitative Analysis of Condensates Obtained by Pine Wood Drying

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Abstract

The condensates obtained during wood drying are dilute aqueous solutions of volatile substances and are the closest in their properties to widely known hydrolates. The analysis of the composition of hydrolates is often carried out using their preliminary extraction with organic solvents. However, some hydrophilic compounds may be lost during this procedure, so that analysis results might not correspond to the native composition of the hydrolate. In this work, we have developed a modified technique of GLC analysis with preliminary sample preparation, including the extraction of condensate with an organic solvent, by using an internal standard of hydrophobic nature in the analysis. The developed technique has been tested on hydrolates of ordinary pine wood (*Pinus sylvestris* var. *mongolica* Litv.), but it can also be applied both to condensates obtained during drying of other wood materials and other plant material, and to hydrolates.

Keywords: condensates, hydrolates, *Pinus sylvestris*, quantitative analysis, gas-liquid chromatography

INTRODUCTION

The condensate of vapour formed from wood drying in the warm air flow at a temperature of 40–65 °C has appeared on the market during the recent years [1]. This product is distilled water with the dissolved volatile compounds extracted along with water from wood into the heated air flows. This condensate has been registered in the International Nomenclature of Cosmetic Ingredients (INCI) under the name of water extract for use in cosmetic industry. The composition and properties of the condensate obtained by wood drying resemble those of so-called hydrolates, often called hydrosol, floral water, aromatic water, or herbal water [2], which are florentine water remaining after essential oil settling [3] during steam- or hydrodistillation of plant raw material [4]. Unlike for condensates under discussion, hydrolates are colloid solutions composed of the continuous phase – distilled water with the components dissolved in it (oxygen-containing compounds) and the dispersed phase – emulsion of essential oil drops. Hydrolates are no more related exclusively as the wastes from essential oil production, and at present they are considered as valuable products in extensive demand for perfume and cosmetic industry [5–7], as well as for food industry [8, 9] and in medicine [10–13].

Hydrolates are manufactured mainly for use as cosmetic products or their ingredients, so currently there are no regulatory documents formulating the methods to control their quality, and every specific manufacturer includes a method considered to be necessary into the technical specifications of a product (TU). Many manufac-

turers only include the gravimetric determination of the mass fraction of essential oil present in the hydrolate [14]. This creates a vast opportunity for adulteration [15, 16]. In addition, some manufacturers fail to mention the fact that their products contain additional ingredients such as preservation agents or co-solvents, which would not cause any harm when the hydrolate is used in perfumes and cosmetics, or in household chemicals, but may be unsafe when used by ingestion. Another issue is in hydrolate concentration, which may vary substantially from one batch to another, not only as a result of intentional dilution but also due to the features of production process (the quality of raw material involved, total distillation time, the flow rate of steam passing through the raw material, efficiency of cooling system). All the above-mentioned considerations indicate that quality control is extremely necessary for these kinds of products, but no methods have been developed at present for the analysis of aqueous condensates.

The goal of the present work is to develop a procedure for the quantitative determination of essential oil content in the condensates obtained from wood drying, and for the qualitative analysis of their composition.

EXPERIMENTAL

Condensate samples

The condensates used in the work were obtained as a result of condensation of the vapour formed by drying the lumber of common pine (*Pinus sylvestris* var. *mongolica* Litv.) growing at the eastern side of Lake Baikal, using a set-up described in patent literature [1]. Drying time for one lumber batch was 5–7 days; one cycle included sequential drying of several lumber batches. The condensate was sampled: a) at the start of the cycle after the first batch; b) at the end of the cycle after the last batch; c) during the entire drying cycle (the condensate from all batches was collected in one vessel and mixed). The following samples were investigated:

Sample 1 – sawn wood: profiled log, batten; drying term: 18.12.2020 – 30.01.2021 (44 days); condensate was sampled at the end of cycle.

Sample 2 – sawn wood: profiled log, batten; drying term: 04.02.2021 – 11.03.2021 (36 days); condensate was sampled at the start of the cycle.

Sample 3 – sawn wood: profiled log, batten; drying term: 04.02.2021 – 11.03.2021 (36 days); condensate was collected generally.

Sample 4 – sawn wood: profiled log, batten; drying term: 17.03.2021 – 24.04.2021 (39 days); condensate was sampled at the start of the cycle.

Sample preparation

Condensate sample (100 mL) was extracted with 2 mL of the hexane solution of *iso*-octanol (17.0 mg of *iso*-octanol with purity ≥98% in 100 mL of hexane) for 2 h. The extract was separated, dried over Na_2SO_4, and analysed by means of gas-liquid chromatography (GLC).

Identification of components

The qualitative analysis of extracts by gas chromatography – mass spectrometry was carried out with Hewlett Packard G1081A instrument, composed of HP 5890 gas chromatograph of series II and a mass-selective detector HP MSD 5971. Column: HP5 (5% diphenyl, 95% dimethylsiloxane), 30 m × 0.25 mm × 0.25 μm; two modes of temperature column programming were used: mode A – 2 min at 50 °C, temperature rise at a rate of 4 °C/min, 5 min at 280 °C; mode B – 2 min at 50 °C, temperature rise at a rate of 10 °C/min, 5 min at 280 °C. Helium was used as carrier gas, flow rate 1 mL/min. The energy of ionising electrons was 70 eV; ion source temperature 173 °C; the data were acquired at a rate of 1.2 scans/s within the mass range 30–650 a.m.u.

Component identification was based on comparing the mass spectra and retention indices with the corresponding data for reference compounds [4].

Quantitative analysis

The quantitative determination of component concentrations in the extracts was performed with a gas chromatograph Agilent 7890A (Agilent Technologies) equipped with flame ionisation detector (FID). Column: HP-5MS (Agilent 19091S-433), 30 m × 0.25 mm × 0.25 μm; column temperature programming mode – 2 min at 50 °C, temperature rise at a rate of 4 °C/min, 10 min at 240 °C. Carrier gas – helium, 1 mL/min. An aliquot (1 μL) of solution was injected with an automatic liquid sampler (7693A, Agilent Technologies), input with split ratio (1 : 50); injector temperature – 280 °C. The FID temperature – 300 °C; air and hydrogen flow rates in the detector – 300 and 30 mL/min, respectively. Measurements for all samples were carried out in triplicate.

Analysis of the data of gas chromatography – mass spectrometry (GC-MS) and chromatography

TABLE 1

Composition of aqueous condensates formed from drying common pine wood (*Pinus sylvestris* var. *mongolica* Litv.)

Component name	RI	Concentrations of organic compounds in condensate samples obtained at different time moments, mg/L			
		Sample 1	Sample 2	Sample 3	Sample 4
Hexanol-1	865	0.10±0.009	0.25±0.013	0.27±0.014	0.09±0.007
Heptanone-2	891	–	0.06±0.004	0.09±0.008	0.11±0.006
1-Octen-3-ol	979	0.04±0.003	0.05±0.003	0.06±0.005	0.03±0.003
Octanone-3	987	0.03±0.003	0.07±0.004	0.07±0.006	0.04±0.003
Octanone-2	995	–	0.08±0.004	0.15±0.008	0.35±0.014
α-Fenchol	1113	0.20±0.010	0.33±0.010	0.39±0.020	0.14±0.007
Trans-pinocarveol	1138	0.09±0.007	0.15±0.008	0.21±0.011	0.09±0.007
Cis-β-terpineol	1144	0.43±0.022	0.58±0.029	0.72±0.036	0.05±0.005
Camphene hydrate	1148	0.11±0.006	0.16±0.008	0.23±0.012	0.03±0.003
Borneol	1166	0.42±0.021	0.66±0.033	0.69±0.035	0.26±0.013
Terpinen-4-ol	1177	2.65±0.080	4.87±0.146	5.87±0.117	1.22±0.037
p-Cymen-8-ol	1186	0.10±0.005	0.10±0.005	0.13±0.007	0.06±0.005
$C_{10}H_{18}O$	1187	0.38±0.019	0.42±0.021	0.49±0.025	0.12±0.006
α-Terpineol	1191	5.03±0.101	4.99±0.100	5.45±0.109	1.88±0.075
Myrtenol	1197	0.08±0.006	0.10±0.005	0.10±0.005	0.06±0.005
Verbenone	1210	0.13±0.007	0.14±0.007	0.19±0.010	0.11±0.006
Trans-carveol	1219	0.05±0.005	0.07±0.006	0.07±0.006	0.05±0.004
Methyl eugenol	1406	0.79±0.040	0.85±0.043	1.21±0.048	0.48±0.024
Copaborneol	1605	0.10±0.008	0.06±0.004	0.14±0.007	0.09±0.008
δ-Cadinol	1649	0.19±0.010	0.11±0.006	0.16±0.008	0.14±0.007
Total content of minor compounds		0.25±0.013	Traces	0.13±0.007	0.20±0.012
Total content		**11.17±0.37**	**14.10±0.46**	**16.82±0.50**	**5.60±0.26**
Gravimetry (control)		**13.0±0.7**	**12.0±0.6**	**17.5±0.8**	**9.0±0.5**

Note. The values of linear retention indices (RI) were determined from the data recorded by gas chromatography – mass spectrometry in mode A (see Experimental).

was performed using specialised software MSD ChemStation E.02.02.1431 (Agilent Technologies).

The internal standard for quantitative analysis was *iso*-octanol. Response factors of all classes of compounds detected in the samples (fatty alcohols and ketones, monoterpene alcohols and ketones, sesquiterpene alcohols, phenol compounds) with respect to *iso*-octanol were accepted to be 1.

Gravimetric determination of the content of volatile compounds in condensates (control)

Sodium chloride NaCl was added in the amount of 5 g to the condensate sample (200 mL), the mixture was shaken till complete salt dissolution, and extracted four time with 10 mL hexane portions (with intense mixing for 1 h with each hexane portion). The united hexane extract was dried over Na_2SO_4, evaporated with a rotary evaporator in the vacuum of water-jet pump at water bath temperature 30–35 °C, and weighted. Results are shown in Table 1.

RESULTS AND DISCUSSION

No methods have been developed yet to analyse condensates obtained from wood drying. These condensates are similar to hydrolates in their composition and properties, so it is reasonable to develop analysis methods relying on the achievements in the analysis of hydrolates. Hydrolate is a diluted aqueous solution of volatile organic compounds, obtained in the production of essential oil by steam- or hydro-distillation of a plant or any its part. The composition of volatile compounds present in hydrolate is to some extent similar to the composition of the corresponding essential oil, except for increased fraction of water-soluble components.

It may seem that hydrolates can be analysed in a similar way as essential oil: by gas chromatography (GC) or gas chromatography in combination with mass spectrometry (GC-MS), which simplifies identification of the detected compounds [4]. However, it should be taken into account that there is a great difference between pure essential oil and hydrolate: the later is always an aqueous solution, so that the concentration of volatile organic components present in it is very low (usually from 0.001 to 0.2%).

Some researchers apply direct GC analysis of hydrolates [14, 17]. This method has a number of advantages. First of all, at present it is the simplest method among those in use, because it excludes the necessity of additional procedures to prepare a sample for analysis. Second, it requires a minimal amount of the sample for analysis. Third, which is substantially more important, it allows obtaining true ratios for the compounds dissolved in water, in particular strongly hydrophilic ones, and simplifies detection of co-solvents added into initial sample. This method has also some disadvantages: poor reproducibility of results [17], difficulties in the case if hydrolate turns out to be too diluted. However, the main reason why the direct GC analysis is rarely used by researchers is poor compatibility of aqueous samples with GLC-columns.

Limitations of the direct gas chromatographic analysis of hydrolates make researchers apply preliminary isolation of organic components from the aqueous solution. During the extraction of hydrolates, most of organic compounds present in them are readily isolated from the aqueous solution because they possess good affinity to organic solvent. Unlike initial hydrolate, the extract may be concentrated if necessary. However, this is not as perfect as it sounds. The major disadvantage of the extraction of hydrolates by organic solvents as a method of sample preparation for analysis is inevitable loss of a part of hydrophilic compounds. The latter compounds are the major organic components of hydrolate, so the determined total concentration of organic compounds in the aqueous solution turns out to be underestimated in comparison with the actual value.

The quantitative determination of component content in the mixture through its GLC-analysis is usually based on the application of internal standard method, that is, on the comparison of the chromatographic peaks of each component with the peak of internal standard added into the mixture in the known amount. The major criteria in choosing the internal standard are the purity of this compound, its chemical neutrality, its absence from the mixture under analysis, and the absence of superposition of the standard peak on the peaks of components in the mixture under analysis. One of *n*-alkanes is often used as the internal standard for GLC analysis of hydrolates by adding an aliquot of the standard to the hydrolate extract prepared for analysis.

The above-listed requirements to the substances that are potential internal standards may be supplemented by one more item: the structural similarity of the molecules of internal standard and the major components of the reaction mixture. Hydrolates are composed mainly of oxygen-containing monoterpenoids (mainly alcohols), while the content of monoterpene hydrocarbons in them is very low because of their poor solubility in water. The use of an alcohol with molecular mass close to that of monoterpene alcohol would allow reducing the contribution from the error related to the loss of hydrophilic reagents of hydrolate at the stage of extraction. In this case, the standard is to be added not to the final extract but to initial hydrolate.

Extraction does not provide 100% isolation of the components into the organic phase, which is further on subjected to analysis. A part of the substance remains in the aqueous phase and is lost for analysis. The higher are the hydrophilic properties of a component, the larger is this lost portion. For instance, if *n*-nonane (a widely used internal standard for the quantitative analysis of essential oils) is added before extraction, the fraction of the standard passing into the organic phase would exceed the fraction of less lipophilic terpenoids due to the difference in the coefficients of distribution for *n*-nonane and oxygen-containing terpene derivatives in water – hexane system. The use of *iso*-octane provides levelling of this different to a substantial extent because distribution coefficients in water – hexane system are comparable for *iso*-octanol and monoterpene alcohols and ketones.

To test the correctness of this assumption, we analysed the composition of a series of condensates obtained by drying the lumber of common pine (*Pinus sylvestris* var. *mongolica* Litv.) growing at the eastern side of Lake Baikal, and performed quantitative determination of the concentrations of these components. Qualitative analysis was carried out by GC-MS by comparing the full electron-impact mass spectra and linear retention indices of the detected components (mode A of

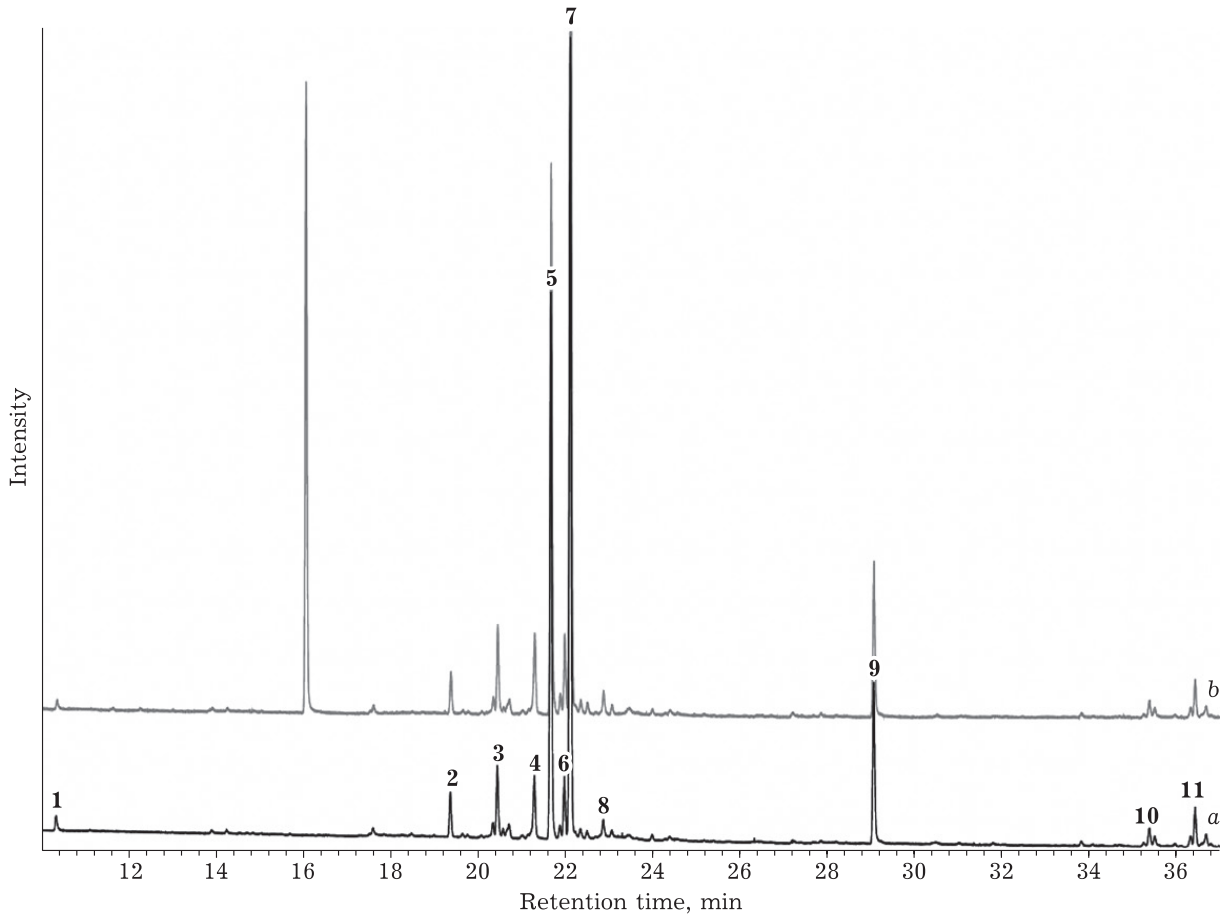

Fig. 1. Typical chromatogram of the extract from the condensate obtained by drying the wood of *Pinus sylvestris* var. *mongolica* Litv. without the addition of the standard (*a*) and with the addition of *iso*-octanol as the internal standard (*b*). Figures indicate the peaks of the following components: **1** – hexanol-1, **2** – α-fenchol, **3** – *cis*-β-terpineol, **4** – borneol, **5** – terpinen-4-ol, **6** – non-identified monoterpene alcohol with the composition $C_{10}H_{18}O$, with retention index RI = 1187 [electron-impact mass spectrum, 70 eV, m/z (I_{rel}, %): 154 (1), 139 (8), 136 (18), 121 (16), 93 (100), 81 (30), 67 (18), 59 (62), 43 (20)], **7** – α-terpineol, **8** – verbenon, **9** – methyl eugenol, **10** – copaborneol, **11** – δ-cadinol.

column temperature programming, see Experimental) with the parameters of volatile substances reported in [4]. To make sure that low-intensity peaks of minor components are not superimposed on the peaks of dominating components, GC-MS measurement was also carried out in mode B. Analysis has shown that the major components of the studied condensates are terpinen-4-ol, α-terpineol, methyl eugenol, and some other alcohols, mainly of monoterpene series. A typical chromatogram of the extract of condensate, with the indication of major components, is shown in Fig. 1, while a complete list of the detected organic components is presented in Table 1.

The standard for the quantitative analysis of extracts was chosen to be *iso*-octanol, which meets all the above-listed requirements to internal standards. The standard was introduced not into the mixture prepared for GLC analysis but as early as at the stage of extraction. The structural similarity of the substance used as the standard and the components of the mixture under analysis allows us to assume that their physicochemical properties would also be similar, in particular the coefficients of distribution between the aqueous and organic phases. Therefore, the error related to the incomplete isolation of organic components from the aqueous phase will be taken into account.

To check the completeness of organic component isolation from condensates, we determined the concentration of essential oil in the samples with the help of gravimetric method based on multiple extraction of the condensates with an increased amount of the solvent, followed by extract concentrating and weighting. Analysis of the results of control experiments (see Table 1) shows that the total concentration of volatile components in the condensates under investigation, determined with the GLC procedure, differs

from the value determined with the help of the gravimetric control procedure by not more than 4 mg/L. In general, there is rather good correlation between them, and one may expect that the difference between results obtained by GLC and control measurements would be smaller when the method is applied to the condensates with higher content of organic components.

All samples under study are similar to each other in composition (see Table 1) both in the set of detected components and in the relative concentrations of major components. Differences between samples 1–3 depict variations of the properties of different lumber batches. Sample 4 has the chemical composition similar to that of samples 1–3, but it contains substantially lower concentration of volatile substances because it was obtained from drying the pine profiled log alone but not a combination of profiled log with batten as it was the case for samples 1–3. Wood drying is accompanied by the diffusion of water and volatile substances from the deep layers of the material to the surface, and water molecules should diffuse at a substantially higher rate. The volatile substances are present in pine-tree resin, which is localised in the so-called gum ducts, thin channels filled with resin. Most probably, the major fraction of volatile substances gets into the condensate not through diffusion from deep layers but due to evaporation from resin emerging on the sawn face as a result of gum duct destruction. Profiled log is characterised by a smaller ratio of surface area to volume, therefore, the number of sawn faces (gum duct destructions) per unit volume is smaller. So, a smaller amount of volatile substances from the resin gets into the condensate during profiled log drying.

CONCLUSION

The developed GLC-based procedure allows us to determine the composition and concentrations of organic components in the condensates. The use of a moderately hydrophobic compound as the internal standard for the quantitative analysis promotes a decrease of determination error related to the incomplete isolation of hydrophobic components from hydrolates. This procedure was tested with a series of condensates obtained from wood drying, and the composition of volatile substances detected in the condensates correlates with the composition of essential oil [18]. All the detected components of condensates are well known natural compounds, and most of them are used as fragrances in perfumery, cosmetics and food industry [19]. In addition to many useful properties, many volatile substances of plant origin also possess substantial toxicity when they are present in high concentrations [20]. One can see in the data presented in Table 1 that the highest concentration of volatile components in the samples does not exceed several milligrams per litre, which is substantially lower than the toxic concentrations. Thus, according to the data of electronic resources [19, 20], the recommended maximal concentration of α-fenchol in aromatic compositions is 5 mg/kg, while α-fenchol content in the samples studied by us does not exceed 1.4 mg/kg, and borneol is present in the studied products in the amount not more than 0.7 mg/kg. Finally, for α-terpineol, the major component of the studied extracts, its use in cosmetic products is permitted in the concentration up to 28.5 g/kg, while its content in the condensates is lower by almost four orders of magnitude and does not exceed 5.5 mg/kg.

The described procedure may be applied either to the condensates obtained by drying other wood materials and other plant raw materials or to hydrolates because, as a rule, organic components present in them are similar in their chemical nature. However, detection of readily volatile components of condensates, such as co-solvents, and determination of their amounts remain the issues to be solved.

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