Research Article

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Influence of pre-drying storage time on essential oil components in dried hops (*Humulus lupulus* L.)

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Abstract

BACKGROUND: It is well known that duration of pre-drying storage impacts on hop quality. However, little knowledge exists regarding its actual effects on valuable hop components. To investigate these effects, fresh hop cones were stored for 5 or 24 h and dried for 210 min at 65 °C thereafter. Furthermore, to understand the effect of freezing hop cones on the essential oil content, both fresh and stored samples were frozen before and after drying.

RESULTS: The results from gas chromatography analysis show an increase in linalool, β-caryophyllene, humulene, geraniol content and decrease in myrcene content dependent on the period of storage. Total colour difference ΔE values of 4.61 and 5.27 were obtained for fresh and stored hops respectively, indicating discoloration of hops during storage. Modelling of moisture curves revealed the Wang and Singh model to be suitable, with R^2_{adj} values of 0.978 and 0.989 and root-mean-square error values of 0.037 and 0.019 for fresh and stored hops respectively.

CONCLUSION: The results from this study provide an in-depth understanding on the changes occurring within the hop cones both during storage and drying and will further help hop processors optimize their storage times.

Keywords: hops drying; gas chromatography; storage; moisture content; essential oils

INTRODUCTION

Hop (*Humulus lupulus* L.) is an essential raw material for the brewing industry as it provides an increased shelf life, bitterness, and aroma to the beer.[1,2] The bitter taste and hoppy aroma to the beer originate from the bitter α- and β-acids and aroma compounds that are present in the lupulin glands of the hop cones.[3,4] The aroma compounds responsible for the characteristic aroma of dried hop cones are part of the essential oil fraction that measure ~0.5–3.0% of the total dry weight.[1] Hop essential oil is a mixture of several hundred volatile substances, with 60–80% of the total weight of essential oils belonging to terpenic hydrocarbons like myrcene, β-caryophyllene, and α-humulene.[2] The amount of oil extracted depends on various factors, such as environmental conditions during cultivation (light, precipitation, soil quality), variety, maturity, optimum harvest times, and drying.[3,4] Currently, there are more than 200 varieties of hops globally, each characterized by its specific aroma composition.[5] Traditionally, α-acids were regarded as the most valuable components. However, with the increase in craft beer production, the core of interest has shifted towards essential oil components of hops so as to achieve a wide range of flavours without any further additions to the traditional ingredients.[6,7]

Freshly harvested hops cannot be stored for a long time because of their high moisture content, which promotes microbial spoilage, colour degradation, and chemical reactions, thus reducing the hop quality.[8,9] In order to increase the stability of hops, convection drying is a commonly used preservation technique. The process of hop drying is as old as hop cultivation itself. Over the years, the traditional hop drying method (single-stage drier) has been replaced by new improved multistage kiln driers. These kilns are commonly used as the design allows the achievement of the maximum drying capacity per square metre of the drying surface.[9] They consist of three or four superimposed perforated trays whose louvre-like floors can be opened to allow the

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drying material to fall into the next section. Fresh hops are filled in the topmost tray, and the hops are shifted to the lower trays as the drying progresses.

Drying of hops is a complex process mainly due to the structure of the hop cone, which consists of strig as its main axis and a bract that is made up of sheets of fine petals covering the main axis. The bract also consists of lupulin glands, which contain the essential oils. Even though the bract encompasses most of the hop cone structure, the strig has the highest water content. During the drying process, the strig does not come into contact with the hot air due to the bract, which leads to the outer layers of the hop cone drying faster than the inner core,[10] thus leaving 4–7% of moisture content on the bract and 25–35% in the strig.[11] The optimal moisture content of dried hops is in the range 8–10%.[11] If the hops are overdried (i.e. reaching a moisture content of <7%), the hop cones tend to shatter, thus increasing the lupulin losses. At the same time, hop cones with a moisture content >13% are at a risk of microbial deterioration.[2] In order to overcome this issue, each tray is equipped with temperature and humidity sensors to allow processors to determine the moisture content of hops, which in turn aids in maintaining the quality of dried hops. After completion of the drying process, hops are placed in a conditioning chamber so as to establish a constant moisture equilibrium between the strig and the bract.[9] In addition to obtaining the desired moisture content, it is also important to maintain the colour of hops. Fresh hops cones when harvested within the optimum harvesting period have a green–yellow combination of colour, which also varies with the variety. Fresh hops, when stored for a long time, not only tend to develop a glossy unappealing colour but also undergo changes in the chemical components, and hence the need for drying right after harvesting.[12] According to Rybáček,[13] the temperature of the drying process also has an effect on the lupulin colour within the hops. At an optimum drying temperature, the lupulin colour tends to remain lemon yellow; however, at higher temperatures and longer exposure time this colour changes towards brown. Rybáček also states that the change in the lupulin colour is a good indicator for the chemical content of the hops. As for the exterior of the hop cone, the colour of the bract also undergoes slight discoloration, which can be reduced by controlling the termination period of the drying process. The length of exposure time depends on the experience of the processor and the information obtained from the temperature and humidity sensors. In order to aid hop processors to non-invasively monitor quality parameters such as colour, Crichton *et al.*[14] and Sturm *et al.*[15] implemented the use of an RGB camera to measure the chromaticity of the hops during the drying process. The results obtained from those investigations show the possibility for novel real-time monitoring of the hops during the drying process. A recent investigation on the application of a thermal imaging camera in kilns is also showing promising results to optimize hop drying using non-invasive measurement techniques.[16]

The drying process has a significant influence on the hop oil content, with losses measured up to 30–40%,[17] particularly of the aromatic compounds and essential oils that tend to degrade prior to, during, and after the drying process. This is mainly due to the high water vapour volatility.[18] One of the key components responsible for the overall hop aroma is myrcene, which accounts for up to 63% of the total oil fraction.[19] Hops are usually dried between 55 °C and 65 °C, and with myrcene being highly volatile at high temperatures the losses are significant. Myrcene losses of 25–30% have been reported during the drying process.[6,20]

With harvest periods lasting for a maximum of 8 weeks per year (2 weeks per variety), post-harvest processing technologies usually run 24/7 without any significant downtimes. Hence, it is essential to ensure the supply of undried hops to the quasi-continuous drier. Prior to the drying process, freshly harvested hop cones are collected in silos until a large enough bulk is harvested to fill the uppermost tray of the multistage kiln drier. Studies performed on stored, dried hops have shown that α-acids and β-acids are prone to significant degradation, as they oxidize rapidly during storage.[21] Furthermore, Münsterer[9] also observed the degradation of colour and gloss for fresh hop cones that were exposed to ambient air humidity >70% in a storage container prior to drying. However, up until now there have been few studies on the effect of storage of fresh harvest on the essential oil components, both prior and after drying. Therefore, this study aims to identify the effect of storing the fresh harvest for a significant period of time on the essential hop oil components, moisture content, and overall colour changes. Furthermore, owing to the short harvest periods and large number of varieties from varying hop gardens, both fresh and dried hop cones are often frozen so as to enable gas chromatography (GC) analysis at a later stage and further understand the variations in the valuable chemical components within hops. Hence, this study also aims to determine the effect of freezing fresh and dried hop cones for both fresh and stored conditions prior to GC analysis.

MATERIALS AND METHODS

Materials

Experimental investigations were performed at the Hop Research Centre, Hüll, Wolnzach, Germany. Aroma hops of the Mandarina Bavaria variety were freshly harvested and mechanically de-vined using a commercial hop cone harvester (Hopfenpflückmaschine Wolf WHE 220, Geisenfeld, Germany) at the research centre and were further dried in a laboratory-scale drier (Heindl GmbH, Nuremberg, Germany) at 65 °C at a constant air velocity for a period of 210 min so as to achieve 10% as the final moisture content. For storage tests, plastic sacks were filled with 2 kg of hops and placed in a temperature-controlled room at 25 °C for 5 or 24 h. After completion of storage periods, the stored samples were dried in the laboratory-scale drier. In order to understand of effect of freezing hop cones prior to chemical analysis on the essential oil components, additional samples of both fresh and stored hops prior to and after drying were vacuumed at 130 mbar and frozen in a freezer at −21 °C.

Methods

Drier set-up and sampling

The Heindl laboratory-scale drier consists of a stainless-steel container with a perforated bottom plate that allows for a constant airflow through the bulk. The container is placed on the drier and set at the desired air temperature. Temperature and humidity of the inlet and the outlet air were recorded using Testo 174 H data loggers (accuracy ±0.5 °C and ±3% relative humidity; Testo SE & Co. KGaA, Lenzkirch, Germany) placed on the top and the bottom of the container unit.

Samples for the moisture content analysis were collected after 0, 20, 40, 60, 90, 150, and 210 min for fresh hops and hops stored 5 h and at 0 and 210 min for hops stored 24 h. These samples were further placed in an oven at 105 °C for 24 h to obtain the

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final moisture content of the samples.[22] In order to ensure accurate measurements for analysis, only one bulk of 2 kg was used per time interval, and hence three laboratory-scale driers were used simultaneously. Furthermore, in order to mimic the commercial drying process, the bulk was thoroughly mixed prior to sample collection. Three repetitions were performed for each of the fresh hops, 5 and 24 h stored hops. For distillation of essential oils, two sets of samples were collected at 0 and 210 min each. The first set was distilled immediately after collection, whereas the other set was vacuumed and frozen for a minimum period of 24 h. The frozen samples were thawed in a refrigerator at 4 °C for a period of 5 h prior to the distillation process.

Imaging set-up

To analyse the effect of drying on colour change, hop samples were placed in a photo box (Life of Photo 60 cm LED Light Cube; Weiwa Foto, Gummersbach, Germany) that includes an array of 160 LED lamps as the illumination and a camera window. The camera was placed at a distance of 29 cm from the surface of the product for image capture. An RGB camera (model 61BUC02) in combination with IC Capture software (The Imaging Source Europe GmbH, Bremen, Germany) was used to capture the images of hops.

The captured RGB images were further processed using MATLAB® (The MathWorks Inc., Natick, MA, USA) to convert RGB values to CIE $L^*a^*b^*$ values, where L^* indicates the lightness (+)/darkness (−), a^* defines the red (+)/green (−) coordinate, and b^* defines the yellow (+)/blue (−) coordinate. The values obtained from each of the repetitions were further averaged in order to calculate the total colour difference $\Delta E^*_{\mathrm{Lab}}$ using the following formula:

$$\Delta E^*_{\mathrm{Lab}} = \sqrt{\left(L^*_2 - L^*_1\right)^2 + \left(a^*_2 - a^*_1\right)^2 + \left(b^*_2 - b^*_1\right)^2} \tag{1}$$

where L^*_2, a^*_2, and b^*_2 are the lightness, red/green, and blue/yellow respectively at the specific time interval and L^*_1, a^*_1, and b^*_1 are the initial lightness, initial red/green, and initial blue/yellow respectively.

GC analysis

Essential oils were distilled using a Clevenger apparatus, which consisted of a round flask (500 mL), condenser, and a standard heating mantle with heat regulator (LABHeat series KM-G, SAF Wärmetechnik GmbH, Mörlenbach, Germany). The quantity of hops distilled was calculated with an assumption of 80% initial moisture content, thus resulting in an estimation of 400 g of dry substance (DS) per 2 kg of initial hop weight. The amount of hops thus required for distillation was determined using the following formulas:

$$\mathrm{MC_{wb}}\,(\%) = \frac{m - \mathrm{DS}}{m} \times 100 \tag{2}$$

where $\mathrm{MC_{wb}}$ is moisture content and m is the weight of hops measured after drying. Hence, DS in measured sample equates to

$$\mathrm{DS_{sample}} = 100 - \mathrm{MC} \tag{3}$$

In each batch, 18 g hops (dry matter) was used for distillation. Thus, the quantity of sample required was

$$M_{\mathrm{sample}} = \frac{18\ \mathrm{g}}{\mathrm{DS_{sample}}} \times 100 \tag{4}$$

where M_{sample} (g) is the mass of initial sample.

One complete distillation process takes 2 h, during which, in the first stage, the water evaporates and condenses, followed by essential oils that float on the condensed water due to the difference in their densities and polarity. With the aid of a three-way valve, the oil was extracted from the water into a 5 mL volumetric flask and stored with tridecane (internal standard with a concentration of 1 g per 50 mL for GC analysis) and *n*-hexane. To bind the existing water in the extracted oil, anhydrous sodium sulfate was added to the solution. The samples were further filled into small vials and then placed in the GC unit for further analysis.

GC analysis was performed using a Dani gas chromatograph equipped with flame ionization detection (FID; DANI Instruments SpA, Milan, Italy) with the following temperature programme: 60 °C for 5 min, 1 °C min^{-1} to 75 °C, 2 °C min^{-1} to 150 °C, 1 °C min^{-1} to 160 °C, 4 °C min^{-1} to 230 °C, and 230 °C for 5 min. The injector temperature and detector temperature were set to 200 °C with helium as the carrier gas. The gas flow was divided in a ratio of 1:25 (split). A 60 m × 0.25 mm fused silica column crosslinked and bonded with polyethylene glycol from the company Macherey–Nagel GmbH (Düren, Germany) was used. The GC unit was also calibrated using an internal standard. Five major components of hop oil, namely myrcene, linalool, β-caryophyllene, humulene, and geraniol, were analysed in the GC unit.

Modelling drying curves

The Page and the Wang and Singh models[23] were used to fit the drying curves for the experimental moisture content values. The Page and the Wang and Singh models are represented by Eqns (5) and (6) respectively:

$$\mathrm{MR} = \exp\left(-Kt^n\right) \tag{5}$$

where K, t (min), and n are the drying constant, drying time, and the drying exponent respectively;

$$\mathrm{MR} = 1 + at + bt^2 \tag{6}$$

where a and b are the drying constants and t (min) is the drying time.

The moisture ratio MR for hops was calculated using the following equation:

$$\mathrm{MR} = \frac{M - M_{\mathrm{e}}}{M_0 - M_{\mathrm{e}}} \tag{7}$$

where M is the moisture content at any given time, M_0 is the initial moisture content, and M_{e} is the equilibrium moisture content. As M_{e} is relatively small compared with M and M_0,[24] the moisture ratio was simplified to

$$\mathrm{MR} = \frac{M}{M_0} \tag{8}$$

The method similar to that described by Sturm *et al.*[15] was used to perform the non-linear regression analysis for changes in moisture content. GraphPad Prism 8.0 (GraphPad Software, San Diego,

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CA, USA) was used to determine the adjusted R^2 (R^2_{adj}), root-mean-square error (RMSE), and the Akaike information criterion corrected for small sample sizes (AICc) and to further evaluate the appropriateness of the models. Additionally, the basic requirements for the application of non-linear regression in the form of residual analysis were also examined.

RESULTS AND DISCUSSION

Modelling of the drying curve

The non-linear regression analysis was performed only for the fresh hops and the hops stored for 5 h. For the hops stored 24 h, moisture content analysis was only conducted for 0 min and 210 min as this extreme case of storage was performed in order to prove the effect of extended storage times on the essential oil components.

Table 1 provides details for the models of the experimentally observed drying curves. For both fresh and 5 h stored hops, R^2_{adj}, RMSE, and AICc were used to estimate the goodness of fit. AICc was used to analyse the data as it allows estimation of the most suitable model for the selected data. Furthermore, owing to the assumption of non-nested models, AICc rather than the *F* test was performed to assess the comparison of fits.[15] As observed in the table, high R^2_{adj} values and low RMSE values were obtained for the Wang and Singh model. AICc values for the models ranged between −124.7 and −139.4 for the Page model and the Wang and Singh model respectively. The lower the AICc values, the better the conformity of the model. Thus, based on the results obtained from the non-linear regression analysis, it can be concluded that the Wang and Singh model[23] is a representative model for the selected hops data.

The fit for the predicted and measured values for moisture content analysis is presented in Fig. 1.

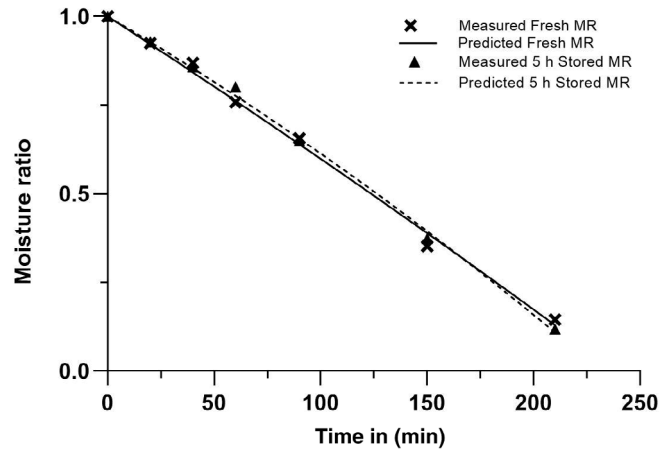

Figure 1. Measured and predicted moisture ratio (MR) values for fresh and stored hops using non-linear regression analysis.

Colour measurement

Colour is an important quality parameter in the drying process, as changes within a product can be identified through the change in colour.[15] Figure 2(a) compares the total colour change ΔE of hops over the drying period, and Fig. 2(b) compares the a^* values for the fresh hops and hops stored for 5 h.

The results obtained show an increase in the ΔE as a function of moisture ratio. For the fresh hops, a clear trend of increasing ΔE can be observed until 0.50 moisture ratio (equivalent to first 90 min of drying). In the case of the stored hops, the ΔE values are higher than those for fresh hops, as the change in ΔE is calculated by using the corresponding initial values of the fresh hops. Hence, the observed shift of ΔE in stored hops before drying (moisture ratio 1.0) equals to 5.5. However, in both cases, a dip in the ΔE is observed after 90 min. The a^* values obtained for hops also show a similar trend as ΔE, with stored hops decreasing in the overall greenness over the drying time. According to Münsterer[25] and Sturm *et al.*,[10] during the initial period of the drying process the hop cones are colder than the surrounding air, which leads to recondensation of water in the upper layers of the hop stack from the air at high relative humidity, thus promoting discoloration in the hop cones. For the stored hops, the high water content in combination with the warm environment inhibits changes within the cones, which when further dried has a significant influence on the colour. Furthermore, the natural heterogeneity in colour and varying hop cone sizes also significantly influence the colour measurements. Fresh hops, depending on the harvest time and the variety, vary between slight dark green to yellow–green colour on the exterior side of the cone. For imaging, the samples were collected in a small bulk, which in turn meant that the size and orientation of the hop cones captured were random. During image analysis, the variations in orientation and the size lead to colour reflection of varying wavelengths, hence affecting the CIE $L^*a^*b^*$ parameters during image processing and resulting in the observed scattering in Fig. 2(a).

Chemical analysis

Mandarina Bavaria is a hop variety with a distinct fruity and exotic aroma of mandarin and orange.[26,27] During drying of stored hops, especially for those stored for 24 h, a distinct foul odour and a sliminess on the surface of the hop cones was observed. This can be linked to the increase in the dimethyl disulfide component within the hops. Dried hops that have been stored for a long time are known to contain high levels of 2-methylpropionic acid, 2-methylbutyric acid, and 3-methylbutyric acid.[19] It is likely that a similar development occurred in the storage of fresh hops, though at a much higher rate due to the high moisture content and, thus, increased enzymatic activity. Increased amounts of dimethyl disulfide contents were also observed for late harvest dates. Since the hops used in the experiment were harvested at

Table 1. Details of the Page model and Wang and Singh (W&S) model analysed for both fresh and stored hops (hops dried at 65 °C)

	R^2_{adj}		RMSE		AICc		Probability (%)	
Sample	Page	W&S	Page	W&S	Page	W&S	Page	W&S
Fresh	0.977	0.978	0.044	0.037	−124.7	−125.5	40.2	59.8
Stored 5 h	0.983	0.989	0.038	0.019	−130.3	−139.4	1.0	98.9

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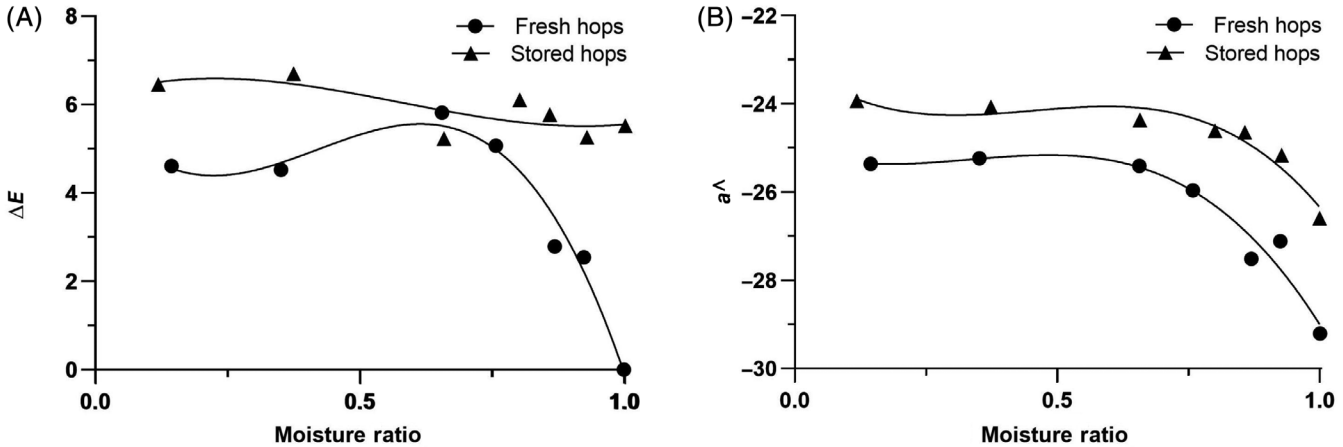

Figure 2. (a) ΔE *versus* moisture ratio for fresh and stored hops. (b) Change in a^* (greenness) values for both fresh and stored hops over moisture ratio.

a later stage (end of September), it is possible that this also had an influence on the increased content of dimethyl disulfide,[6,20] which became noticeable during 24 h storage.

The amount of oil reported in the literature for Mandarina Bavaria ranges from 0.015 to 0.022 L kg^{-1} for fresh hops.[27] In the experimental analysis performed, the average amount of oil extracted was 0.025 L kg^{-1} for fresh hops, which is higher than reported in the literature. Among the factors strongly influencing the amount of oil content is the harvest period and the growing conditions at the location.[28,29] In 2012, an investigation performed on the Mandarina Bavaria variety revealed that the amount of oil extracted varied each week during the seven-week harvesting period. The study also reveals the variations in the amount of oil when compared between two different hop gardens. The experimental analysis for this study was performed in the last weeks of the harvesting season. Although the hop cones were harvested from the same hop garden, the late harvest period is believed to have an effect on the oil content, thus resulting in the higher values.[6,28]

Figure 3 shows the results obtained before and after drying for varying pre-drying storage conditions. Before drying, the highest hop oil content is observed for fresh frozen samples (0.026 L kg^{-1} hops) followed by samples stored 24 h (0.025 L kg^{-1} hops). Ihloff,[30] in a study on the effect of freezing *Anethum graveolens* L. (dill), suggested that the enzymes present can cause conversions and degradation within the product. Ihloff also stated that the process of freezing potentially retains catalytic activity of these enzymes, which after thawing are present to a larger extent. A similar phenomenon is believed to have occurred for the fresh frozen samples in this study. After drying, the hop oil content for fresh, fresh frozen, and samples stored for 5 h decreases as expected. However, the same trend was not observed for stored frozen and samples stored for 24 h, with the latter showing a significant rise of 12.7%. This increase could also be associated with the significant degradation and conversion of oils during the drying process.

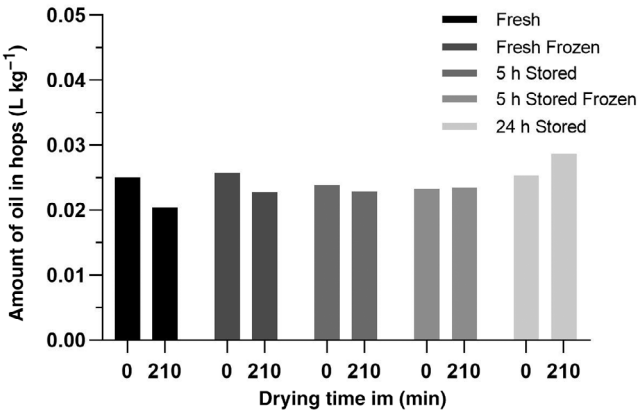

Figure 3. Amount of oil in hops extracted for samples extracted at 0 and 210 min for varying conditions.

Analysis of essential oil

Figure 4 shows a chromatogram of the five major components obtained for fresh hops using a GC-FID. The results obtained for five major components from the gas chromatography analysis are represented in Fig. 5 and summarized in Table 2.

Prior to drying, the myrcene content was highest within fresh samples (10.49 g kg^{-1} hops) followed by samples stored 5 h (8.59 g kg^{-1} hops). Both fresh frozen samples and samples stored 5 h showed the lowest myrcene content, at 6.62 g kg^{-1} hops and 6.87 g kg^{-1} hops respectively. After drying for 210 min, all samples showed a significant decrease in the myrcene content. The samples stored 24 h had the highest loss (40.22%), followed by the fresh samples (38.17%). The lowest degree of loss, at 1.62%, was observed in the samples stored 5 h. Myrcene is a key component of hop aroma,[1] and making up about 17–37% of the total hop oil.[31] It is a terpene hydrocarbon and a significant contributor to the aroma of fresh hops. During the storage and drying process, myrcene undergoes oxidation and polymerization.[32] The autoxidation of myrcene leads to cyclic reactions forming various products, such as α-pinene, β-pinene, camphene, and terpenoids, which include linalool and geraniol.[32] Increased oxidation, which has an influence on the biogenesis of myrcene during storage, leads to significant loss of myrcene.[33] Additionally, harvest time has also been reported to have an influence on the total myrcene content, with late harvest periods, such as those for Mandarina Bavaria, resulting in a significantly high myrcene content compared with an early harvest variety.[7]

Linalool, much like myrcene, is also one of the key components for hop aroma. It has a low odour threshold value[6] and provides a floral and citrusy aroma to the beer.[1] In this study, linalool showed a different trend compared with myrcene. At 0 min, the highest

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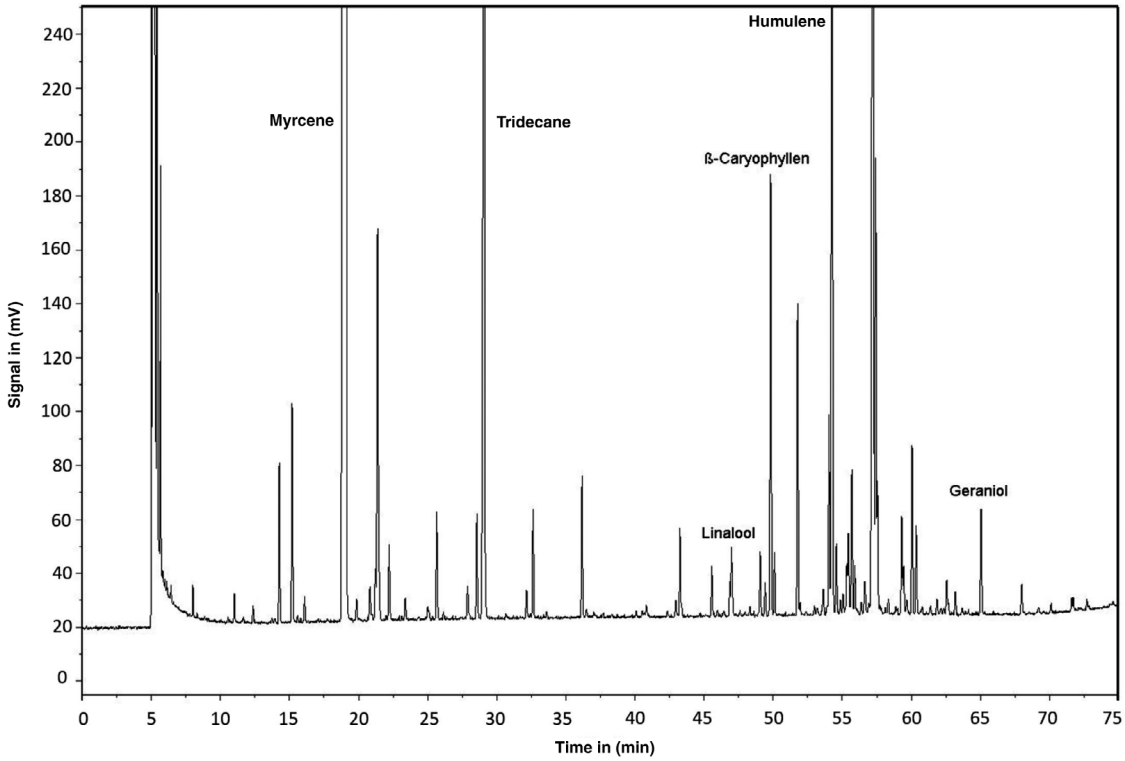

Figure 4. Gas chromatogram of fresh hops (var. Mandarina Bavaria) using GC-FID.

amount of linalool was observed for samples stored24 h (0.08 g kg^{-1} hops) followed by the samples fresh frozen, stored for 5 h, and stored frozen for 5 h, with the amount ranging between 0.06 g kg^{-1} hops and 0.07 g kg^{-1} hops. The linalool content was found to be lowest in the fresh samples, with 0.05 g kg^{-1} hops. After drying, the linalool content decreased for fresh and fresh frozen samples but increased in the stored, stored frozen, and stored for 24 h samples. A 10.9% of rise in the linalool content was observed in the stored frozen samples. As mentioned previously, the degradation of myrcene leads to the formation of additional products, such as linalool. This is especially observed for the stored samples (5 and 24 h), thus explaining the increase in the linalool content and corresponding decrease in the myrcene content. Furthermore, experimental investigations performed by Haslbeck *et al.*[34] revealed that glycosidically bound linalool could be released by enzyme Rapidase F64, resulting in an odour-active aglycone (linalool) and sugar residue. The authors further revealed that 2 μg of linalool could be released for every gram of hops (DS). The release of the glycosidically bound linalool also depends strongly on the hop variety.[35] Glycosides are water soluble, odourless, and can be split either through enzymes or heat into sugar or odour-active components.[36] For the storage experiments, the samples were placed in a warm environment, which could have promoted enzyme activity. Additionally, as the 24 h samples were stored for a longer time, it can be presumed that the enzymes had more time for reactions, thus resulting in a higher linalool content. As for the increase in the linalool content in the stored samples after drying, high temperatures during the drying process aided the conversion of the glycosides mentioned earlier. The process, which started during the storage, was further triggered during the drying process, hence increasing the overall linalool content.

β-Caryophyllene provides the beer a lilac-like, flowery, and musty smell.[20] For the various samples, large variations in the β-caryophyllene content can be observed. At 0 min, the highest β-caryophyllene content was observed for the stored frozen samples (0.78 g kg^{-1} hops), whereas the lowest β-caryophyllene was observed for fresh samples (0.36 g kg^{-1} hops). After drying, the β-caryophyllene content showed a decreasing trend for all the samples, except for samples stored 24 h. The decrease in the β-caryophyllene content could be due to the oxidation of β-caryophyllene to caryophyllene oxide, 14-hydroxy-β-caryophyllene, and other such components within the oil.[32] As for the samples stored 24 h, it is believed that additional conversion processes within the hop cones could have taken place during storage. However, specific reasons for these changes in the β-caryophyllene content have not yet been identified.

Prior to drying, humulene shows a similar trend to that of β-caryophyllene, with the stored frozen samples having the highest content (1.97 g kg^{-1} hops) and the fresh samples having the lowest amount (1.04 g kg^{-1} hops). After drying, the total amount of humulene increased by 16.12% in the samples stored 24 h. For all other samples, losses ranging from 9.96% to 25.37% were observed. Humulene, like β-caryophyllene, belongs to the sesquiterpenes group.[31] As both components belong to the same group, it is possible that they undergo similar degradation and conversion processes during pre-drying storage, thus increasing the overall amount for the stored samples. As the fresh samples are dried immediately after harvest, the conversion process is inhibited, thus accounting for the lowest amount of the component.

Geraniol is a monoterpene and provides an impression of a lilac-like, floral, cedar smell to the beer.[20] In line with the other components, the highest content of geraniol at 0 min was observed for

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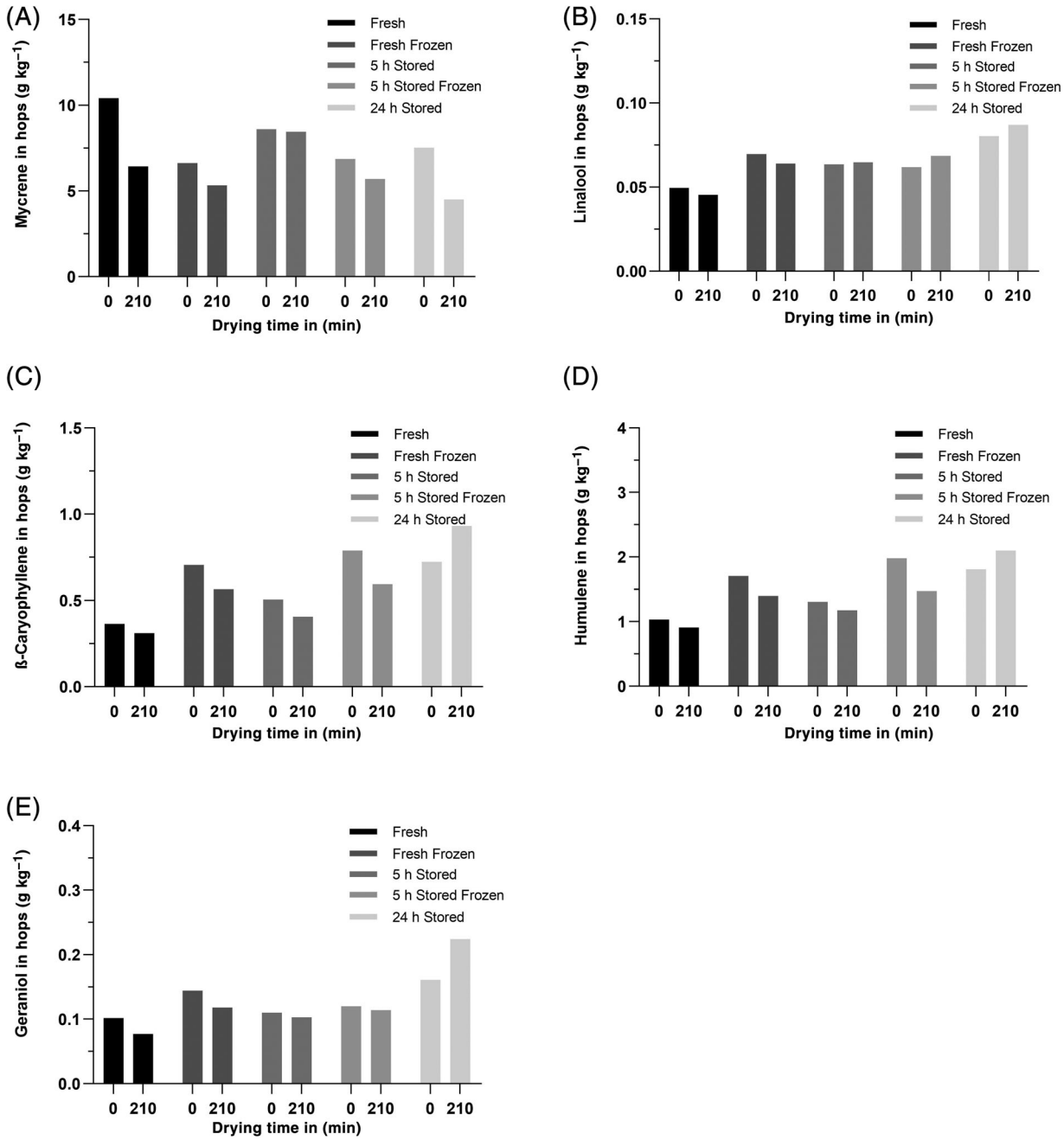

Figure 5. Oil components analysed from GC analysis at 0 and 210 min for varying conditions: (a) myrcene; (b) linalool; (c) β-caryophyllene; (d) humulene; (e) geraniol.

Table 2. Amount of essential oil content obtained for different sample preparation

	Essential oil content (%)									
	Myrcene		Linalool		β-Caryophyllene		Humulene		Geraniol	
Sample	0 min	210 min	0 min	210 min	0 min	210 min	0 min	210 min	0 min	210 min
Fresh	10.41	6.43	0.049	0.045	0.364	0.310	1.035	0.909	0.102	0.077
Fresh frozen	6.62	5.32	0.069	0.063	0.704	0.564	1.705	1.396	0.144	0.118
Stored 5 h	8.60	8.46	0.063	0.064	0.506	0.405	1.307	1.177	0.110	0.102
Stored frozen 5 h	6.87	5.68	0.061	0.068	0.787	0.593	1.977	1.475	0.120	0.114
Stored 24 h	7.51	4.49	0.080	0.087	0.722	0.932	1.807	2.099	0.161	0.224

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samples stored 24 h and the lowest was for fresh samples. After drying, the amount of geraniol further increased by 29.3% in samples stored 24 h and showed a decreasing trend in all the other samples. Geraniol is also a product of the oxidation process of myrcene during storage, which explains the increase in the amount of geraniol in stored hops. Geraniol, like linalool, has glycosidic bonds,[35] which could also explain the increase in the geraniol content in hops stored 24 h due to an increase in enzymatic conversion. However, in contrast to linalool, the glycosidic bonds of geraniol are heat stable,[36] and hence it is possible that other processes that still remain unexplained were involved in the significant rise of geraniol in samples stored 24 h after drying.

CONCLUSION

In conclusion, we show that an increased pre-drying storage period has a significant effect on the oil content both prior to and after drying, especially in the case of hops stored 24 h. An increase in the amount of oil through prolonged storage and the associated foul odour indicate the degradation of valuable aromatic compounds in hops. Furthermore, freezing of hop cones for both fresh and stored conditions prior to and after drying also showed variations in the oil components, which indicate that hop samples should be analysed as soon as possible so as to identify the exact quantities of valuable components within the hop cones. Chemical analysis that could indicate an increase in the dimethyl disulfide content was not conducted and can potentially be included in future investigations. Thus, based on the results obtained, it is recommended to maintain the storage period to a minimum. Regression analyses that were performed for moisture content data indicate a goodness of fit with the Wang and Singh model. Significant colour changes between fresh and stored hops were observed during drying, indicating discoloration of hops both due to storage and drying.

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CONFLICT OF INTEREST

There are no conflicts of interest to declare.

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