Acta Univ. Sapientiae, Alimentaria, **17** (2024) 120–135

DOI: 10.47745/ausal-2024-0007

Preliminary study of lavender-flavoured beer production methods

M. Kelemen[1]
e-mail: markelemen24@yahoo.com

Cs. Albert[1]
e-mail: albertcsilla@uni.sapientia.ro
ORCID:
https://orcid.org/0000-0001-6882-6691

Cs. D. András[1]
andrascsaba@uni.sapientia.ro
ORCID:
https://orcid.org/0000-0001-5220-832X

R. V. Salamon[1]
salamonrozalia@uni.sapientia.ro
ORCID:
https://orcid.org/0000-0002-3497-6859

[1] Sapientia Hungarian University of Transylvania,
Faculty of Economics, Socio-Human Sciences and Engineering,
Department of Food Science, RO-530104 Miercurea Ciuc, 1 Libertății Sq.

Abstract. The aim of our research was to produce a lavender-flavoured beer belonging to the Blonde Ale category, which can have a positive physiological effect on the consumer. Two flavouring methods were investigated: the addition of lavender flowers (control) during hop boiling and the addition of β-CD lavender essential oil microcapsules (LEO-β-CD) obtained from the essential oil of the lavender flower before the final beer bottling.
The physicochemical and sensorial properties of beer samples were studied in terms of the concentration of β-CD microcapsule that gave the most pleasant taste to our consumer target group, as well as the amount of LEO-β-CDs required during dosing and safety for the consumer. The effectiveness of the preparation of microcapsules and the dosage concentrations were examined with GC-MS method using the concentration of linalool as the main essential oil component.

Keywords and phrases: β-cyclodextrin, microcapsule, linalool, sensorial properties

1. Introduction

There is a growing interest in and consumption of various beers and a growing demand for flavoured beers. For this reason, the aim of the investigation was to prepare a beer flavoured with herbs and a beer flavoured with microencapsulated essential oil to investigate which dosing method is more effective and which one provides the most beneficial sensory properties for the consumer. Based on some surveys, it can be stated that the consumers of flavoured beers are mainly women, followed by young

adults over 18 years of age. Thus, the target consumer community for this product is made up of women and young adults (*Núñez-Caraballo et al.*, 2019).

1.1. Lavender as a medicinal plant, its active substances, fields of application, and physiological effects

According to the *European Pharmacopoeia*, 11th edition (*Ph. Eur.* 11, 2022), the drug can be the essential oil produced by steam distillation from the dried flowers of lavender and the fresh flower buds and less frequently from the flowering shoots of lavender species. The pharmacopoeial-grade drug shall contain at least 13 ml/kg of essential oil, containing 25–47% of the ester expressed as linalyl acetate. The French lavender flower drug contains 0.5–3% essential oil, mainly linalyl acetate (30–60%) and linalool (20–50%). The hybrid lavender flower contains essential oil (0.9–5%), but the proportions of the components vary.

According to *Oroian et al*'s research, the ratio of essential oil components in *Lavandula angustifolia* fresh stalked flower and stalkless flower is as follows: linalool 25.3–43.0% and 25.5–42.1%, linalyl acetate 3.6–37.4% and 8.0–47.2%, and camphor 0–9.8% and 0–11.4% respectively (*Oroian et al.*, 2019).

Lavandula angustifolia has sedative, bile-secreting, and antibacterial effects, and the decoction made from the drug is used for sleep disorders, restlessness, "indigestion", flatulence, and gallstones (*Mardani et al.*, 2022; *Batiha et al.*, 2023; *Firoozeei et al.*, 2021). Lavender essential oil (LEO) is used externally in ointments or as a rubbing agent in an alcohol solution to treat rheumatic and nerve pains and reduce skin aging. The largest user is the cosmetics and perfume industry (*Prusinowska & Śmigielski*, 2014; *Henriques et al.*, 2020). It has been used recently in food production for the natural flavouring of (mainly alcoholic) beverages, ice creams, confectionery, bakery products, and chewing gum *(Da Porto et al.*, 2009).

1.2. Characterization of Blonde Ale beers

Blonde Ale beers are easy-drinking, approachable, malt-centric American craft beers, often with interesting fruity, hoppy, or malty notes. Well-balanced and clean, it is a refreshing beer without intrusive flavours. The aroma ranges from light to moderately sweet malty, with possible slight bread or caramel notes appearing. Low to moderate fruitiness may be present, which is acceptable, and low to medium intensity hop aroma; almost any hop variety may be present, although citrus, floral, fruity, and spicy notes are common. The colour can range from light yellow to dark gold, clear to bright, with little to medium white foam, and the persistence ranges from acceptable to good. The flavour shows a soft, malty sweetness to start but may optionally show light, malty flavours (e.g. bread, biscuits, toast, wheat). Caramel flavours are typically absent, but if present, pale caramel notes are typical

characteristics. Few to moderate amounts of esters are optional but acceptable; hop flavours (of any type) range from light to moderate and are not overly aggressive. There is a medium to low bitterness, but the balance leans towards malt or more malty rather than hoppy. From medium dry to malty sweet finish – the impression of sweetness is often due to the perceived bitterness being lower than the actual residual sweetness (*Gordon & Kristen*, 2022).

1.3. Dosing options for lavender in beer

Lavender has a relatively strong aroma, so the amount to be administered should be determined accordingly. Dosing can be done in several ways, first considering the plant or the essential oil made from it. Having studied the process used by several small producers, the conclusion is that the plant itself is preferred for flavouring. Dosing options also offer two possibilities: addition during the hop boiling process or fermentation process. Based on a previous thesis (*Ignácz*, 2013), dosing during boiling was found to be better based on the sensory analysis of the final product, and this method was used to produce lavender-flavoured control beer. Another possibility was explored in parallel: the dosing of essential oil mentioned above, which is introduced into the product using a carrier material that can also be used in the food industry immediately before bottling. For this purpose, β-cyclodextrin was used, a carrier with recognized and accepted properties in the food industry but not yet used by the brewing industry.

1.4. Cyclodextrin (CD) as a food carrier

Despite the importance of CDs in food, the last complete study on their use in food science was published in 2009. The article covers the characteristics of the most important industrial-grade CDs (α-CD, β-CD, and γ-CD) and their main technological properties such as solubility and ability to form inclusion complexes. It also includes the current technology for using these compounds in the food industry (*Matencio et al.*, 2020). CDs are torus-shaped oligosaccharides composed of α-(1,4)-type glucose units produced from the breakdown of starch by the enzyme cyclodextrin glucosyltransferase (CGTAse). The CD ring is an amphiphilic cone-shaped cone with a hydrophilic outer part (by hydroxyl groups) and a predominantly lipophilic cavity that may contain water (*Pereva et al.*, 2019). CDs have a promising future due to consumer demand for healthy and functional products (*Pereira et al.*, 2021).

1.5. Metabolism and toxicology of cyclodextrin

In nature, only CGTAse can convert starch into CDs, although other enzymes can help in their industrial production (*Matencio et al.*, 2020); however, different

enzymes or processes in our body can break down CDs into glucose derivatives. In the mouth (if we consume something containing CDs), salivary α-amylase can rapidly hydrolyse dextrins although rapid transport to the stomach means that the degradation rate is insignificant. Of the three natural CDs (α-, β- and γ-CD), the first two are essentially stable in the presence of α-amylase, whereas γ-CD is rapidly digested (*Matencio et al.*, 2020).

Specific pH-dependent degradation can occur in the stomach, but lower degradation is observed in the presence of complexes (*Matencio et al.*, 2020). After the stomach, in the neutral pH environment, the pancreatic amylase of the small intestine continues the hydrolysis reaction. While α- and β-CDs are mainly digested by bacteria in the large intestine (where α-CD is more rapidly degraded than β-CD), γ-CD is almost completely digested in the gastrointestinal tract. Finally, the undegraded CDs are metabolized by microbiota in the last stage of the digestive tract, where they are almost completely degraded, being used as prebiotics in their life functions (*Fenyvesi et al.,* 2016), the rest being excreted in the faeces.

Generally, the bioavailability of natural and more relevant CD derivatives is very low, making them safe for oral administration (*Matencio et al.*, 2020). As their molecular weight increases, linear dextrins and CDs are increasingly excreted in the urine. Indeed, molecules smaller than 15 KDa are almost entirely excreted in the urine with almost no modifications. More than 90% of CDs are excreted in the urine.

When used as food additives, natural CDs are classified as such (E457, E458, and E459) and are accepted as "Generally Recognized as Safe" (GRAS). The recommendation of the Joint FAO/WHO Expert Committee on Food Additives (JECFA) has set a maximum recommended level of 5 mg/kg/day for β-CD in food. On the other hand, due to their favourable toxicological profile, there is no Acceptable Daily Intake (ADI) for α and γ-CD. The European Food Safety Authority (EFSA) has confirmed the positive health effects of α-CD as dietary fibre and its suitability for reducing post-prandial glycaemic responses (*EFSA Panel on NDA*, 2012).

The dose of β-CD was reassessed in 2016 and accepted without modification at 5 mg/kg/day (*Matencio et al.*, 2020; *Fenyvesi et al.*, 2016; *EFSA ANS Panel*, 2016), with the safety of CDs being assessed and consumption levels in food being established, supporting the view that there was no need to modify the already established levels. Generally recognized as safe (GRAS) molecules, they are directly approved for use as excipients (such as natural CDs). In this regard, the US Food and Drug Administration (FDA) has published a list of inactive pharmaceutical ingredients, which includes recommended minimum and maximum consumption concentrations. A question-and-answer document on CDs and their use (*Committee for Human Medicinal Products*, 2017) provides information on their safety: for example, the consumption threshold for β-CD for oral use is 20 mg/kg/day (*Committee for Human Medicinal Products*, 2017).

2. Materials and methods

2.1. Description of the beer recipe

To produce 50 L of wort, 7.5 kg of Pale Ale malt (Weyermann), 63 g of Cascade hops (Yakima chief) with 5.7% alpha acid content, 11.5 g of Safale US-05 yeast (Lallemand), and 55 L of pre-boiled and cooled tap water were used.

2.2. Description of the wort brewing process

In order to set up the experiment, a Ziptech NaNo small-scale brewing machine was used to brew approximately 50 L of wort. The mashing was carried out using 7.5 kg of Pale Ale malt and 25 L of pre-boiled and cooled mashing water. For mashing, the following temperature programme was used: holding at 65 °C for 60 min, holding at 72 °C (1 °C/min) for 30 min, increasing to 75 °C for 5 min, and holding at 78 °C for 1 min. 30 L water was used for the crawling.

2.2.1. Preparation of lavender-flavoured beer

At the start of the hop boil, 25 g of lavender flowers and 42 g of Cascade hops with 5.7% alpha bittering acid are added. At the end of the 60-minute boil, 50 g of lavender flowers and 21 g of Cascade hops are added. The resulting wort was settled for 30 minutes and, after cooling (28 °C), it was pumped into the fermentation tank, where it was inoculated with 11.5 g Safale US-05 pre-hydrated yeast and fermented at 22 °C for seven days. After the primary fermentation, the temperature of the young beer was reduced to 4 °C, and after two days, the yeast was removed, and the maturation continued for fourteen days, after which the finished beer was bottled in 0.5 L bottles and stored in a beverage refrigerator at 4 °C.

2.2.2. Preparation of beer flavoured with β-CD lavender essential oil microcapsules (LEO-β-CD)

For the dosing of the LEO-β-CD described in section 2.2.3, again, the beer recipe described above was used, dosing the microcapsules at the end of the primary fermentation before bottling. Dosing was carried out at several concentrations, summarized in *Table 1*. After dosing the LEO-β-CD, the flavoured beers were stored at 4 °C. The quantities of LEO-β-CD added were determined considering the expected essential oil content of the beer containing lavender flowers. The recipe shows that 75 g of flowers were added to 50 L of beer. Since the essential oil was previously extracted from lavender flowers by steam distillation, it can be observed

that the essential oil content of the flowers is 2.7%, so the amount of essential oil added together with the flowers can also be determined, which was 2.02 grams. The hop flower beer contained approximately 0.0434 g of essential oil per litre. Knowing the amount of essential oil added in the production of the LEO-β-CD (3.6 mL), taking into account the density of lavender essential oil (0.885 g/cm^3), we determined the mass of essential oil added (3.186 g) and then the number of grams of essential oil contained in one gram of LEO-β-CD (0.155 g).

Based on the above data, it was estimated that 0.287 g of LEO-β-CD is required to obtain 0.0434 g/L of essential oil in beers produced with LEO-β-CD. Since the above calculation involves estimating several values, five beers produced with LEO-β-CD at different concentration levels (two lower, one similar, and one higher volatile oil concentration) were prepared using the amounts of LEO-β-CD shown in *Table 1*.

Table 1. The LEO-β-CD dosage amounts

Sample ID	**1**	**2**	**3**	**4**	**5**
LEO-β-CD concentration [g/g]			0.155		
Amounts of LEO-β-CD in beer [g/L]	0.016	0.072	0.107	0.144	0.21

2.2.3. Preparation of LEO-β-CD

For the preparation of LEO-β-CD, the recipe published by *García-Segovia et al.* (2011) was used. In the first step as per the recipe, a 30 mL, 12% essential oil-ethanol solution was prepared. For this purpose, 3.6 mL of lavender essential oil obtained with steam distillation and 26.4 mL of ethanol (96 v/v%, Sigma Aldrich) were poured and mixed using a magnetic stirrer. Next, a 10% β-CD solution was prepared using 220 ml of ethanol-water solution in a 1:2 ratio and 22 g of β-CD (Cyclolab). The ethanol-water solution contained 146.6 ml distilled water and 73.3 ml ethanol (96 v/v%, Sigma Aldrich).

β-CD was added to the ethanol-water mixture with constant stirring using a magnetic stirrer with heating. To the 10% β-CD solution, set at 55 °C, the 30 mL essential oil-ethanol solution was slowly added, drop by drop, with constant stirring. Following the addition, mixing continued for four more hours, then cooled and maintained at 4 °C for 12 hours. After settling, the microcapsules were separated from the liquid, using a vacuum filter with 0.40 µm porosity and dried at room temperature (22 °C) for 24 hours. In order to ensure a longer shelf life of the LEO-β-CD prepared, they were dried in a drying oven at 50 °C for another 24 hours and stored in an airtight glass container until use.

2.3. Sensory analysis of the beer

The sensory analysis was conducted in the Fermentation Laboratory of the Department of Food Science, Sapientia Hungarian University of Transylvania, which meets the conditions described in the Romanian SR 13355-1/1997 standard, with light-coloured furniture, white walls, and a room free of foreign odours and noises. Natural light was available during the tasting, without direct sunlight, which allowed the correct assessment of the colour of the beer. At each tasting point, a plate with bread was placed to eliminate any residual flavour, and a bowl was placed to collect the leftovers from the tasting.

A focus group carried out sensory evaluations of beer samples with 26 people (14 male and 12 female) aged 18–25 years, who were also selected as the target consumer group. The tasters evaluated our beer samples using a tasting questionnaire that was compiled to assess the external appearance, colour, smell, taste, CO_2 content, foam appearance, and durability, using a 0–5 hedonic scale (0: very bad, 5: excellent). The values obtained were weighted by a constant: external appearance (0.6), colour (0.8), smell (0.2), taste (1.4), CO_2 content (0.6), and foam stability (0.4). The acceptance test's final scores were based on the classification provided in SR 13355-1/1997, and the following categories were distinguished: excellent (20.0–18.1), good (18.0–15.1), agreeable (15.0–12.1), unsatisfactory (12.0–7.1), and bad (7.0–0.0).

Carbon dioxide impregnation was evaluated simultaneously with the evaluation of appearance and taste. The odour was assessed immediately after opening the bottle. Six beer samples were subjected to sensory analysis, which were analysed in the following order: 1st sample: 0.016 g/L LEO-β-CD; 2nd sample: 0.072 g/L LEO-β-CD; lavender-flower-flavoured; 3rd sample: 0.107 g/L LEO-β-CD; 4th sample: 0.144 g/L LEO-β-CD; 5th sample 0.21 g/L LEO-β-CD. A 2-minute break was taken between tasting the samples. Tasting participants recorded their ratings on a tasting form developed online (Sensory evaluation of lavender-flavoured beers).

2.4. Determination of the physicochemical properties of beer

The original extract, sugar, alcohol, CO_2, O_2, density, and beer turbidity were determined using an Anton Paar PBA-B (Packaged Beverage Analyzer for Beer) analyser according to EBC 9.43.2/2004. For the pH and colour measurements (EBC 9.35/2004), CO_2 was removed from the beer by shaking. Since our sample was an unfiltered beer before the colour determination, the samples were filtered with kieselguhr filter powder according to EBC 9.6/2000 and then measured with a spectrophotometer (Hach Lange DM 6000) at $\lambda = 275$ nm.

The bitterness of the beer was determined according to EBC 9.8/2004. The sample was centrifuged at 4,000 rpm for 20 min at 20 °C. 1 mL of 3 M HCl and 20 mL of isooctane were added to 10 mL of beer sample, shaken for 25 min

on an IKA shaker at 450 rpm, left in the dark for 30 min, and measured with a spectrophotometer at λ = 275 nm.

2.5. Determination of linalool content

The main component of lavender oil, linalool, was determined using the method published by *Kishimoto et al.* (2006). Hexane (3:1) was added to the beer samples and shaken at 4 °C for 24 hrs at 160 rpm. The supernatant was removed by pipette, dried with Na_2SO_4, and 1 µL was injected into the GC-MS. Lavender essential oil and LEO-β-CD were also dissolved in hexane. The GC-MS data were the following: Agilent 7890 A-5975C GC-MS, HP-5MS capillary column (30 m × 0.25 mm × 0.25 µm). Carrier gas was helium at a flow rate of 1 mL/min. MS settings were: ionization voltage (EI), 70 eV; ionization temperature, 250 °C; quadrupole temperature, 150 °C; quantitative scanning range: 35–350 u. For qualitative analysis of linalool MS NIST database was used (retention time 7.43 min). For the quantification of linalool, a calibration was performed over a concentration range of 0–2 mg/L, where the equation of calibration line was A = 33350.0 ·Clinalool + 303.1 (correlation coefficient R2 = 0.9970), obtained by linear regression.

2.6. Statistical analyses

All data were expressed as the means of the measurements ± standard deviation and were made in triplicates. The one-way analysis of variance (ANOVA) with Tukey's test was employed to evaluate the significant differences at $p < 0.05$. Statistical tests were made using XLSTAT software for the Excel 2021 version (Addinsoft, New York, NY, USA).

3. Results and discussions

3.1. Results of the sensory test and their evaluation

The test was carried out according to the Romanian standard SR 13355-1/1997, using an online questionnaire, which we designed following the questions of the standard. During the tasting, 26 participants gave their opinions on beer samples containing different concentrations of LEO-β-CD and beer samples produced with lavender flower dosage. The participants were between 18 and 25 years old, and the gender distribution was 55% men and 45% women.

From the general questions (How often do you drink beer? What characteristics do you consider when choosing a beer? Which type of beer do you prefer based on taste? Which type of beer do you prefer based on alcohol content?), it was found

that men drink alcohol more often and base their choices on taste and flavour, prefer less bitter beers, and mainly drink beers with 4-5% alcohol. Women's beer consumption was less frequent, once a month, and, like men, women chose beer based on taste and aroma. Opinions were divided on the classification by flavour, but the majority preferred flavoured beers. In terms of alcohol consumption, women prefer beers with an alcohol content of 4-5% v/v.

After the tasting, the acceptance test results (*Table 2*) showed that the beer samples produced were suitable for consumption and contained minor defects. These are the low CO_2 content and the low foam, which are mostly due to the fact that the laboratory does not yet have bottling technology to adjust the CO_2 content in the product. The low CO_2 content also negatively affects foam stability (*Figure 1*).

At the acceptance test, lavender flower beer (13.5) and a beer sample containing 0.072 g/L LEO-β-CD (13.51) achieved the highest scores. Otherwise, this result is promising, as this feedback confirms that LEO-β-CDs can be a good option for flavouring beers (*Table 2*).

Table 2. Acceptance test results

Properties	**Beer samples**					
	1	2	Lavender flower	3	4	5
	Amount of LEO-β-CD in beer, g/L					
	0.016	0.072	-	0.107	0.144	0.210
	Weighted average score values of beer samples					
External appearance	2.03 ± 0.67[a] *	2.26 ± 0.76[a]	2.42 ± 0.73[a]	2.17 ± 0.75[a]	2.22 ± 0.54[a]	1.98 ± 0.78[a]
Colour	3.20 ± 0.99[a]	3.17 ± 0.89[a]	2.03 ± 1.66[b]	2.95 ± 0.91[a]	3.14 ± 0.81[a]	3.17 ± 1.03[a]
Smell	0.72 ± 0.18[a]	0.77 ± 0.23[a]	0.78 ± 0.29[b]	0.64 ± 0.21[a]	0.75 ± 0.21[a]	0.74 ± 0.26[a]
Taste	5.28 ± 1.29[a]	5.55 ± 1.50[a]	5.12 ± 1.70[b]	5.17 ± 1,64[a]	5.22 ± 1.38[a]	4.95 ± 1.85[a]
CO_2 content	1.25 ± 0.86[a]	1.22 ± 0.64[a]	2.08 ± 0.60[b]	1.29 ± 0.76[a]	1.15 ± 0.68[a]	1.52 ± 0.92[a,b]
Foam	0.43 ± 0.39[a]	0.54 ± 0.44[a]	1.08 ± 0.39[b]	0.68 ± 0.52[a]	0.68 ± 0.52[a]	0.83 ± 0.48[c]
Sum	12.90	13.51	13.50	12.90	13.15	13.20
Category	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable

Notes: * Results represent mean values ± standard deviation (SD), n = 26; letters *a*, *b* indicate significant differences between the beer samples at $P \leq 0.05$ level; letter *c* indicates significant differences between sample 1 and sample 5 (at $P \leq 0.05$ level).

The external appearance of lavender-flavoured beer is that of a clear, bright liquid, without suspensions or sediment, with foam and carbonation, and is appreciated by both male and female tasters. As for the colour, amber was also the highest-scoring colour for men, while it was light yellow for women. The tasters found that the beer samples had the characteristic lavender flavour but needed to be more pronounced and balanced. The characteristic lavender aroma was present in the samples, although some tasters did not detect it. As noted in the taste assessment, the aroma has the characteristic lavender scent, as confirmed by the tasters, with one glaring case of a featureless scent.

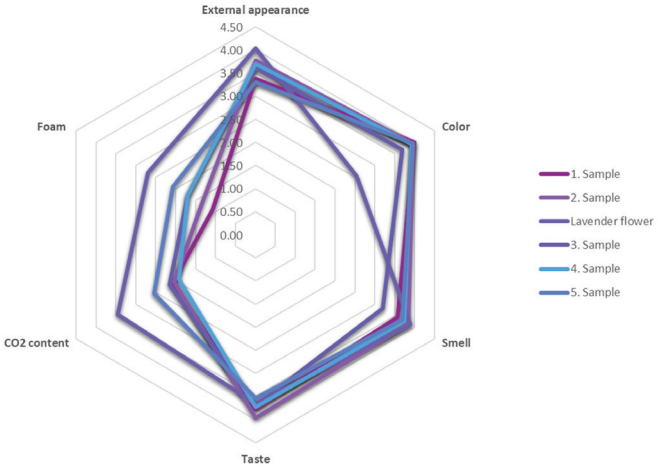

Figure 1. Results (expressed as the median) of the sensory analysis

The external appearance of the LEO-β-CD 0.072 g/L sample was acceptable, with most tasters describing it as clear and free of suspension (*Figure 2*). However, three tasters thought it contained small particles, which we attribute to its unfiltered nature. Regarding colours, most male tasters considered the sample containing 0.072 g/L β-CD amber, while the female tasters considered it more typical ruby amber. Discrepancies may be due to differences in colour perception between the sexes or individuals. The tasters did not experience any uncharacteristic beer flavour despite the β-CD used for flavouring, and the majority considered the lavender flavour to be noticeable. As with the flavour, the sample smelled of lavender, with one glaring opinion that the sample smelled of lavender was featureless, again a function of the tasters' different perceptions.

Based on the responses to the final question of the questionnaire, "Which sample number did you like the most?", women overall preferred the beer sample containing 0.072 g/L LEO-β-CD, while men who participated in the tasting preferred the lavender flower-flavoured beer sample.

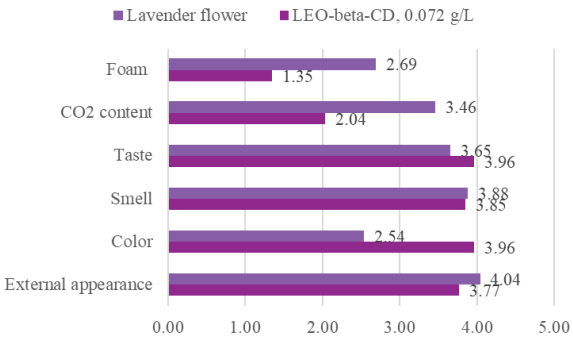

Figure 2. The acceptance test results for most liked samples

The one-way ANOVA results show (*Table 2*) statistically significant differences in the sensory scores for colour, smell, taste, CO_2 content, and foam among the six beer samples. The results of the Tukey HSD test present significant differences between the beer with lavender flowers and the beers with LEO-β-CD. Regarding the CO_2 content, the beer sample with 0.210 g/l LEO-β-CD did not show statistically significant differences from the other samples. In terms of foam, the beers with the smallest and largest amounts of LEO-β-CD microcapsules showed significant differences.

3.2. Main physicochemical properties of the base beer

Taking into account the BJCP – Beer Judge Certification Program standard, the lavender beers we produce (since the base beer is the same, and therefore the physicochemical properties are the same except for the aroma), as evidenced by the physicochemical properties defined (*Table 3*), can be classified as Blonde Ale.

Table 3. Physicochemical properties of beer samples

Properties	**Lavender beer**	**Blonde Ale**[1]
Original extract [Plato]	8.91 ± 0.02	11.00
Apparent extract [%]	1.30 ± 0.02	–
Alcohol content [% v/v]	3.96 ± 0.01	3.80-5.50
Density [g/cm^3]	1.00345 ± 0.0001	-
CO_2 [g/l]	1.368 ± 0.002	-
O_2 [mg/l]	3.20 ± 0.01	-
Calorie [kJ/100ml]	132.23	-
pH	5.13 ± 0.01	-
Colour [EBC]	13.90 ± 0.02	5.90–11.80
Bitterness [IBU]	24.60 ± 0.10	15.00–28.00

Note: [1] BJCP – Beer Judge Certification Program.

3.3. Results of the linalool content of beer samples

The GC-MS measurement showed that the linalool content of lavender flower essential oil used for LEO-β-CD production was 33%, which fit within the limits found in the literature (25.3–43.0% – *Kara & Baydar*, 2012), and the linalool content of LEO-β-CD was 45 mg/g microcapsule.

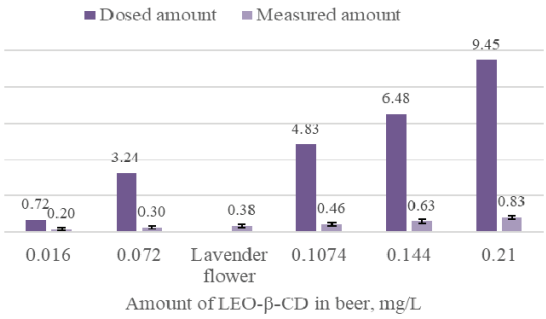

Figure 3. Linalool concentration calculated based on the dosed (45 mg linalool/g LEO-β-CD microcapsule) and measured amount in different beer samples

The measured values of linalool concentration are around 10% of the dosed amount, and increasing the dosed amount does not increase the measured value, so further experiments are needed to confirm this by examining whether the amount of solubles does not increase with higher amounts of LEO-β-CD.

The following graph shows the amount of linalool already present in the samples, expressed as a percentage.

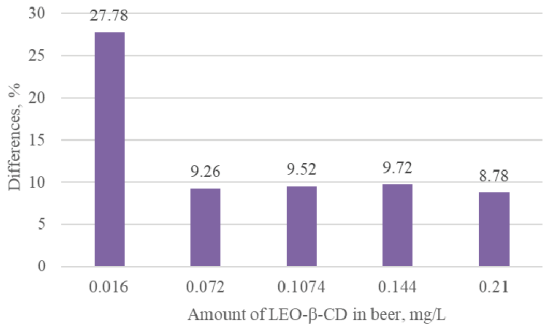

Figure 4. Percentage differences between the calculated and the determined linalool concentration

Concerning the β-CD microcapsule dosing of the beer samples, the recovery efficiency of linalool was around 9% on average. The lowest concentration of LEO-β-CD beer (0.016 g/L) has a very high recovery of 27.7%, which was attributed to the sample preparation method, but further measurements are needed to confirm this.

3.4. Safety aspects of the consumption of CD-microcapsule-flavoured beers

The safe consumption limit for β-CDs is 5 mg/kg/day (EFSA, 2016) or 20 mg/kg/day for oral use (EMA, 2017). The daily consumption of β-CDs can vary between 325 mg/day and 1,300 mg/day for a person with an average body weight of 65 kg. Our beer samples with LEO-β-CD contained between 0.013 g β-CD/L and 0.178 g β-CD/L of β-CD, so we can state that none of the samples exceeded the safe daily intake of β-CD. In the light of this, it was calculated that 1.8 L of beer containing the highest amount of LEO-β-CD (0.178 g β-CD/L) can be safely consumed in one day.

4. Conclusions

Based on our results, both dosing methods can be used for flavouring. However, in the case of beers flavoured with cyclodextrin microcapsules, the influencing factor is the amount of microcapsules added. Acceptability tests show that consumers preferred lavender-flavoured beer to the same extent as beer containing 0.072 g/l LEO-β-CD.

Our beers, containing lavender flowers and LEO-β-CD, are suitable for consumption based on their organoleptic and physicochemical properties. The sensory tests have shown that the beer samples containing lavender flowers and 0.072 g/L LEO-β-CD best meet the requirements of SR 13355-1/1997.

None of our beer samples exceeds the safe daily intake of β-CD, and at least nine (0.5 L) bottles of beer (4.5 L) containing the most preferred beer sample with LEO-β-CD (0.072 g β-CD/L) could be safely consumed per day.

Further research is needed to develop recipes for beer samples containing 0.072 g/L of LEO-β-CD, focusing on balancing the hop and lavender flavours.

Acknowledgments

The cofounder, *András Lénárd*, and the staff of Tiltott Csíki Sör Manufaktúra are thanked for providing the laboratory facilities for the physicochemical analysis of the beer samples.

References

[1] Batiha, G. E. S., Teibo, J. O., Wasef, L., Shaheen, H. M., Akomolafe, A. P., Teibo, T. K. A., Al-kuraishy, H. M., Al-Garbeeb, A. I., Alexiou, A., Papadakis, M., A review of the bioactive components and pharmacological properties of Lavandula species. *Naunyn-Schmiedeberg's Archives of Pharmacology*, 396. 5. (2023) 877–900. Springer Science and Business Media Deutschland GmbH. https://doi.org/10.1007/s00210-023-02392-x.

[2] Committee for Human Medicinal Products, *Questions and answers on cyclodextrins used as excipients in medicinal products for human use*. EMA/CHMP/495747/2013.9/10/2017 [accessed 24/09/2024]. Available at: https://www.ema.europa.eu/en/documents/scientific-guideline/questions-and-answers-cyclodextrins-used-excipients-medicinal-products-human-use_en.pdf.

[3] Da Porto, C., Decorti, D., Kikic, I., Flavour compounds of *Lavandula angustifolia* L. to use in food manufacturing: Comparison of three different extraction methods. *Food Chemistry*, 112. 4. (2009) 1072–1078. https://doi.org/10.1016/j.foodchem.2008.07.015.

[4] EFSA ANS Panel (EFSA Panel on Food Additives and Nutrient Sources Added to Food), Mortensen, A., Aguilar, F., Crebelli, R., Di Domenico, A., Dusemund, B., Frutos, M. J., Galtier, P., Gott, D., Gundert-Remy, U., Leblanc, J.-C., Lindtner, O., Moldeus, P., Mosesso, P., Parent-Massin, D., Oskarsson, A., Stankovic, I., Waalkens-Berendsen, I., Woutersen, R. A., Wright, M., Younes, M., Boon, P., Chrysafidis, D., Gürtler, R., Tobback, P., Arcella, D., Rincon, A. M., Lambré, C., Re-evaluation of b-cyclodextrin (E 459) as a food additive. *EFSA Journal*, 14. 12. (2016). https://doi.org/10.2903/J.EFSA.2016.4628.

[5] EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA); Scientific opinion on the substantiation of health claims related to alpha-cyclodextrin and reduction of post-prandial glycaemic responses (ID 2926, further assessment) pursuant to Article 13(1) of Regulation (EC) No 1924/2006. *EFSA Journal*, 10. 6. (2012) 17. doi:10.2903/j.efsa.2012.2713.

[6] European Medicines Agency (EMA). Community herbal monograph on lavender oil (04/2023:2972). *European Pharmacopoeia (Ph. Eur.), 11th ed.*; EDQM Council of Europe: Strasbourg, France. (2022) 4627–4628.

[7] Fenyvesi, É., Vikmon, M., Szente, L., Cyclodextrins in food technology and human nutrition: Benefits and limitations. *Critical Reviews in Food Science and Nutrition*, 56. 12. (2016) 1981–2004. https://doi.org/10.1080/10408398.2013.809513.

[8] Firoozeei, T. S., Feizi, A., Rezaeizadeh, H., Zargaran, A., Roohafza, H. R., Karimi, M., The antidepressant effects of lavender (*Lavandula angustifolia* Mill.): A systematic review and meta-analysis of randomized controlled clinical trials. *Complementary Therapies in Medicine*, 59. (2021). Churchill Livingstone. https://doi.org/10.1016/j.ctim.2021.102679.

[9] Gordon, S., Kristen, E. *2021 Style guidelines*; BJCP, Inc.: Boulder, CO, USA. (2022) 33.

[10] Henriques Ribeiro Machado, A. C., Gonsales Spíndola, D., Rodrigues Leite Silva, V., Rocha Oliveira, C., Efeitos anti-idade dos óleos essenciais de lavanda, alecrim e melaleuca em fibroblastos dérmicos humanos. *Revista Científica de Estética e Cosmetologia*, 1. 1. (2020) 1–7. https://doi.org/10.48051/rcec.v1i1.15.

[11] Ignácz, K., *Levendulával ízesített sör gyártástechnológiájának kidolgozása*. Diploma thesis. Budapest. (2013). https://stud.mater.uni-mate.hu/3198/.

[12] Kara, N., Baydar, H., Essential oil contents and composition of lavenders and lavandins cultivated in Turkey. *Res. on Crops*, 13. 2. (2012).

[13] Kishimoto, T., Wanikawa, A., Kono, K., Shibata, K., Comparison of the odor-active compounds in unhopped beer and beers hopped with different hop varieties. *Journal of Agricultural and Food Chemistry*, 54. 23. (2006) 8855–8861. https://doi.org/10.1021/jf061342c.

[14] Mardani, A., Maleki, M., Hanifi, N., Borghei, Y., Vaismoradi, M., A systematic review of the effect of lavender on cancer complications. *Complementary Therapies in Medicine*, 67. (2022). Churchill Livingstone. https://doi.org/10.1016/j.ctim.2022.102836.

[15] Matencio, A., Navarro-Orcajada, S., García-Carmona, F., López-Nicolás, J. M., Applications of cyclodextrins in food science. A review. *Trends in Food Science and Technology*, 104. (2020) 132–143. Elsevier Ltd. https://doi.org/10.1016/j.tifs.2020.08.009.

[16] Núñez-Caraballo, A., García-García, J. D., Ilyina, A., Flores-Gallegos, A. C., Michelena-Álvarez, L. G., Rodríguez-Cutiño, G., Martínez-Hernández, J. L., Aguilar, C. N., Alcoholic beverages: Current situation and generalities of anthropological interest. *Processing and Sustainability of Beverages*, (2019) 37–72. Elsevier. https://doi.org/10.1016/b978-0-12-815259-1.00002-1.

[17] Oroian, C., Odagiu, A., Racz, C. P., Oroian, I., Mureşan, I. C., Duda, M., Ilea, M., Braşovean, I., Iederan, C., Marchiş, Z., Composition of *Lavandula angustifolia* L. cultivated in Transylvania, Romania. *Notulae Botanicae Horti Agrobotanici Cluj-Napoca*, 47. 3. (2019) 643–650. https://doi.org/10.15835/nbha47311565.

[18] Pereira, A. G., Carpena, M., Oliveira, P. G., Mejuto, J. C., Prieto, M. A., Gandara, J. S., Main applications of cyclodextrins in the food industry as the compounds of choice to form host–guest complexes. *International Journal of Molecular Sciences*, 22. 3. (2021) 1–23. MDPI AG. https://doi.org/10.3390/ijms22031339.

[19] Pereva, S., Nikolova, V., Angelova, S., Spassov, T., Dudev, T., Water inside β-cyclodextrin cavity: Amount, stability and mechanism of binding. *Beilstein Journal of Organic Chemistry*, 15. (2019) 1592–1600. https://doi.org/10.3762/bjoc.15.163.

[20] Prusinowska, R., Śmigielski, K. B., Composition, biological properties and therapeutic effects of lavender (*Lavandula angustifolia* L.). A review. *Herba Polonica*, 60. 2. (2014) 56–66. https://doi.org/10.2478/hepo-2014-0010.