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# Foaming properties and olfactory profile of fermented chickpea aquafaba and its application in vegan chocolate mousse

Lea Mehren, Lena Elliger, Hanna May, Andreas Schieber , Nadine Schulze-Kaysers 0

Institute of Nutritional and Food Sciences, Molecular Food Technology, Faculty of Agricultural, Nutritional and Engineering Sciences, University of Bonn, Friedrich-Hirzebruch-Allee 7, 53115, Bonn, Germany

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#### ABSTRACT

Aquafaba, the cooking water of chickpeas and other pulses, is used as a vegan egg white substitute because of its favorable technofunctional properties. Nonetheless, its application is often restricted by a "beany" flavor and poor foaming properties compared to egg white. To overcome these limitations, aquafaba was fermented with two edible basidiomycetes. During the fermentation process, foaming properties were measured and analyzed. Furthermore, the aroma change was described by sensory experts. Based on these results, the optimal fermentation day was selected for each mushroom and sensory profiling was conducted. Subsequently, chocolate mousse was prepared from fermented aquafaba and profiled as well as tested in an acceptance test. Aquafaba profiling revealed significantly lower scores ( $\alpha=5\%$ ) for "beany" odor in fermented samples. Chocolate mousse produced with fermented aquafaba was described as less "beany" but more "chocolatey" and "cocoa-like" in smell and taste, and more "sweet" in taste. The texture of mousse prepared with fermented aquafaba was more "fluffy/light/porous" and "soft" but less "homogenous" than mousse with unfermented aquafaba. The consumer test showed high overall liking for all mousses. The research described in this study revealed for the first time promising aroma changes based on fermentation in aquafaba and demonstrated improved foaming properties. Thus, fermentation can be considered a useful tool to enhance the quality of aquafaba and thus expand its fields of application.

# 1. Introduction

Plant-based alternatives have the potential to enhance sustainability and decrease greenhouse gas emissions (Xu et al., 2021). Furthermore, a plant-based diet is associated with both health and environmental benefits compared to an animal-based diet. Plant protein sources contain higher levels of fiber and lower levels of saturated fat, which may contribute to the reduction of chronic diseases (Ewy et al., 2022). Aquafaba, derived from the Latin terms "aqua" for water and "faba" for the Fabaceae family, is the liquid produced when pulses are boiled. Whereas aquafaba has mostly been discarded previously, it now offers a valuable and cost-effective byproduct with the potential to serve as an alternative protein source. In addition, it shows a wide range of advantages because its plant origin. In 2014, Joël Roessel discovered that aquafaba from canned pulses can be used to produce a foam similar to egg white (The Official Aquafaba Website, 2023). Subsequently, Goose

Wholt shared a recipe on social media for vegan meringue using chickpea cooking water and sugar (The Official Aquafaba Website, 2023). This was the beginning of a growing interest in aquafaba within the vegan community and in academic circles (The Official Aquafaba Website, 2023; Alkin, 2023; He et al., 2021).

Because of its technological characteristics such as foaming, gelling, and emulsifying properties, aquafaba can be used to replace egg white or dairy proteins in vegan and sustainable food systems (He et al., 2021). Current research has focused primarily on the chemical composition, physicochemical properties and food application of aquafaba as an egg replacer (He et al., 2021; Alsalman et al., 2020; Bekiroglu et al., 2023), and most studies deal with aquafaba from chickpeas. The use of aquafaba as an egg white substitute may be limited by its beany aroma (Noordraven et al., 2021) and its reduced foam stability (Mustafa et al., 2018). To overcome these constraints, one promising approach is the fermentation of aquafaba. During fermentation, substances can be

Abbreviations: AXA, Antrodia xantha; CATA, check all that apply; FC, foaming capacity; FS, foam stability; LED, Lentinula edodes.

<sup>\*</sup> Corresponding author.

E-mail addresses: lmehren@uni-bonn.de (L. Mehren), lelliger@uni-bonn.de (L. Elliger), may\_hanna@web.de (H. May), schieber@uni-bonn.de (A. Schieber), nadine.schulze@uni-bonn.de (N. Schulze-Kaysers).

metabolized, resulting in the generation of new compounds that affect sensory and technological properties (Fritsche, 2016). According to a recent study, the foaming and emulsifying properties of aquafaba can be improved by fermentation with *Lactobacillus plantarum* (Bekiroglu et al., 2023). Fungi from the division Basidiomycota are interesting for fermentation processes as well because they are known to produce a wide range of aroma-active substances (Bouws et al., 2008; Lomascolo et al., 1999; Fraatz et al., 2011) and can utilize numerous agricultural by-products as substrates. For example, *Lentinula edodes* has successfully been cultivated on beer wort (Zhang et al., 2014) and pea-rice protein isolate (Clark et al., 2022). Furthermore, many species of basidiomycetes are edible (Bouws et al., 2008). Thus, they have promising potential for the fermentation of aquafaba to enhance flavor.

The objective of this study was to investigate the fermentation of aquafaba by Basidiomycota, which, to the best of our knowledge, has not been studied yet. As the aim was to use aquafaba in sweet aerated desserts, where it can act as a vegan egg white replacer, the focus was on the following targets: Firstly, to improve the aroma of aquafaba by reducing the typical beany and increasing sweet or fruity flavors through fermentation. Secondly, the foam stability should be improved. The latter is usually inferior to that of egg white, while the foaming capacity is comparable. Lentinula edodes and Antrodia xantha were selected for their successful growth on aquafaba in screening tests with a range of fungi, and for their ability to produce sweet, floral, and fruity odors while reducing the beany aroma. To assess the suitability of these fermentation processes, sensory experts evaluated the odor throughout the fermentation period. In addition, the foaming capacity and foam stability were analyzed. Based on the results, the optimal fermentation duration was determined, and the fermented aquafaba was used to produce a vegan chocolate mousse, which belongs to the most common aerated desserts. It is crucial to characterize the sensory properties of fermented aquafaba and chocolate mousse. Moreover, consumer tests are necessary to assess the acceptability of vegan chocolate mousse prepared with aquafaba. In general, these analyses are important in order to assess the potential of application of fermented aquafaba in desserts and other sweet products. Nonetheless, this study can only deliver first insights into the feasibility of the utilization of aquafaba as vegan food ingredient. Overall, this study contributes to the value-added use of aquafaba for the development of innovative plant-based and sustainable food products.

## 2. Materials and methods

## 2.1. Materials

Canned chickpeas from Suntat (Baktat International, Mannheim-Neckarau, Germany), chocolate (72 %, Schokoliebe Edition, Maxhütte-Haidhof, Germany), and sugar (Südzucker AG, Mannheim, Germany) were obtained from local supermarkets.

Two species of basidiomycetes, *Antrodia xantha* (AXA) and *Lentinula edodes* (Berk.) Pegler (LED), were used. LED (strain number 389.89) and AXA were kindly provided by the Institute of Food Chemistry and Food Biotechnology (Justus-Liebig-University, Gie $\beta$ en, Germany). The malt extract and agar-agar were purchased from Carl Roth GmbH + Co. KG (Karlsruhe, Germany) and Merck (Darmstadt, Germany), respectively.

## 2.2. Sample preparation

# 2.2.1. Fermentation

The basidiomycetes used in the fermentation, AXA and LED, were selected on the basis of screening tests and preliminary trials. The fermentation was performed according to Ahlborn et al. (2019) with some modifications. In brief, AXA and LED were grown on malt extract agar plates (20 g/L malt extract, 15 g/L agar-agar) at 24 °C in an incubator (Binder, Tuttlingen, Germany) until they covered 80 % of the agar plate, which was achieved after seven and eleven days,

respectively. The inoculum was prepared by transferring an agar clot (approximately  $1.0~\rm cm^2$ ) to 2~% malt extract solutions ( $100~\rm mL$  in  $250~\rm mL$  flasks). The main cultivation took place at  $24~^\circ \rm C$  on a rotary shaker (Stuart SSL 1, VWR, Mannheim, Germany) at  $150~\rm rpm$  for seven days. Submerged fermentation was performed on a rotary shaker at  $150~\rm rpm$  and  $24~^\circ \rm C$  in  $250~\rm mL$  flasks containing  $98~\rm mL$  aquafaba and  $2~\rm mL$  of a sucrose solution ( $100~\rm g$  sugar in  $100~\rm mL$  water). Homogenized inoculum ( $10~\rm mL$ ) was added to the mixture, and fermentation was carried out for nine days. On each day of fermentation, starting from inoculation, the foaming properties were analyzed, and a sensory analysis was performed. The optimal fermentation time for each mushroom was determined based on superior foaming capacity and overall likings of the aroma that were at least acceptable or even better. For the fermentation with AXA day 4 and for LED day 3 were selected and used for the application in chocolate mousse.

## 2.2.2. Preparation of chocolate mousse

The suitability of fermented aquafaba was tested to produce chocolate mousse, which is one of the most common aerated food products. For its preparation, 600 mL of unfermented or fermented aquafaba was stirred with a MultiMix M 830 M (Braun, Procter & Gamble Service GmbH, Kronberg im Taunus, Germany), speed level 3, for 10 min. After 2 min of stirring, 90 g sugar was added. Meanwhile, 525 g chocolate was melted in a water bath at 65  $^{\circ}\mathrm{C}$  and then cooled down to 44  $^{\circ}\mathrm{C}$ . The melted chocolate was carefully folded into the foamed aquafaba with a dough scraper. Subsequently, the chocolate mousse was stored in a refrigerator.

# 2.3. Foaming properties

Foaming capacity (FC) and foam stability (FS) were determined daily. For this purpose, 110 mL of fermented or unfermented aquafaba ( $V_{sample}$ ) was mixed in a beaker (1000 mL) for 90 s at 9000 rpm using a Dynamix 160 mixer equipped with a beater (Dynamic Professional, Kehl Auenheim, Germany). After whipping, the initial foam volume ( $V_0$ ) was calculated. For FS, the drainage water was removed after 60 min, and the final foam volume ( $V_{60}$ ) was determined. FC and FS were calculated using the following equations (1) according to Alsalman et al. (2020) and (2) according to Martinez et al. (2016), respectively.

$$FC [\%] = \left(V_0 - V_{sample}\right) / V_{sample} * 100 \tag{1}$$

$$FS \ [\%] = V_{60} / V_0 * 100 \tag{2}$$

In addition, foaming properties were analyzed using a DFA100 dynamic foam analyzer (Krüss GmbH, Hamburg, Germany) to obtain more detailed information about foam morphology and to explain differences in foam stability. Mean bubble count and mean bubble diameter were recorded. Aliquots of 50 mL of either fermented or unfermented aquafaba were filled into a glass column (CY4572 Krüss GmbH, Hamburg, Germany) and whipped with a spiral agitator (SR4501 Krüss GmbH, Hamburg, Germany) at 5000 rpm for 5 min. The measurements were conducted for 35 min.

## 2.4. Sensory evaluation for identification of fermentation time

During fermentation over the nine days, four trained experts performed the sensory bench tasting. The expert team conducted a descriptive examination with unfermented and fermented aquafaba to identify appropriate food references according to their aroma profile for later profiling (Table 1a). To this end, aquafaba samples were placed in neutral plastic containers (Huhtamaki Group, Espoo, Finland) and sealed with a lid 30 min before the sensory evaluation to ensure an accumulation of aroma compounds in the headspace. Simultaneously, a vocabulary was developed to describe the smell of aquafaba samples. For this purpose, a descriptive examination with unfermented and

**Table 1**a) Attributes for olfactory aquafaba profiling and reference materials/chemicals;
b) attributes for chocolate mousse profiling regarding appearance, smell, taste, and texture.

1a Attributes for olfactory aquafaba profiling	References presented in training sessions $\binom{a}{b}$ and during profiling $\binom{b}{b}$				
beany/chickpea	- chickpeas <sup>a, b</sup> - unfermented aquafaba <sup>a</sup> - cooked green beans <sup>a</sup> - nonanal <sup>a</sup> - 1-octanol <sup>a</sup>				
hay/potato	- cooked potatoes <sup>a, b</sup>				
musty/earthy	- wet forest soil <sup>a, b</sup>				
	- 1-octen-3-ol <sup>a</sup>				
malty	- malt beer without alcohol <sup>a, b</sup>				
sweet odor	- honey <sup>a</sup>				
	- rose water <sup>a</sup>				
roasty/caramel	- caramel from sugar <sup>a, b</sup>				
broth	- meat broth <sup>b</sup>				
sweaty	no reference				
1b Attributes for chocolate mou	isse profiling				
Appearance	- brown				
	- fluffy/light/porous				
Smell	- beany/chickpea				
	- cocoa/tangy				
	- chocolatey				
	- sweet odor				
	- roasty				
Taste	- chocolatey				
	- sour				
	- sweet				
	- salty				
	- cocoa/sharp				
	- beany/chickpea				
m .	- roasty				
Texture	- homogenous				
	- melting				
	- soft				
	- fluffy/light/porous				
	- heavy/dense				

fermented aquafaba was conducted. For aroma description, the expert team used the aroma wheel of the German Agricultural Society (DLG e. V.) and added additional attributes as suggested in the respective norm DIN 10964 (DIN 10964:2014-11).

The sensory evaluation of the daily samples included overall liking on a hedonic scale from 1 "dislike extremely" to 5 "like extremely" and rating of the aroma intensity on a scale from 0 "no perception" to 10 "very strong perception".

## 2.5. Panel training

For the analytical profiling, 12 panelists were recruited (mean age = 29; range: 21-42 years; 8 females, 4 males). They were trained in ten sessions, including selection tests, training of recognition and differentiation of stimuli, development and use of descriptors, and specific product training according to DIN EN ISO 8586 (DIN EN ISO 8586) as well as an olfactory training with the food references mentioned in 2.4. Taste sensitivity was determined by threshold tests according to DIN ISO 3972 (DIN ISO 3972) for sweet, bitter, salty, and sour taste. Only participants who met the requirements were included in the panel. Texture training was carried out by a descriptive examination (DIN 10964). Based on the basic sensory vocabulary ("German Agricultural Society"), attributes related to chocolate mousse and suitable reference foods were selected, tasted, described, and discussed. The following attributes were evaluated in combination with the corresponding reference food: "heavy/dense" with a cream pudding, "homogeneous" with mashed potatoes, "light" and "soft" with whipped cream, "fluffy" with beaten egg whites and "porous" with aerated chocolate. Finally, the panel developed a list of descriptors through consensus, which is presented in Table 1b.

#### 2.6. Profiling

The sensory profiling according to DIN EN ISO 13299 (DIN EN ISO 13299) was carried out in the sensory laboratory at the Institute of Nutritional and Food Sciences of the University of Bonn in two sessions (n = 12). First, trained judges evaluated the odor of the three aquafaba samples: unfermented aquafaba, aquafaba fermented with AXA (after 4 days) and aquafaba fermented with LED (after 3 days). The evaluated attributes and reference materials used in the training or during the profiling are listed in Table 1a. In a second session, the three corresponding chocolate mousses were profiled. Aquafaba (20 mL) or chocolate mousse (20 g) were served randomly and coded with three-digit numbers in neutral glass containers. The chocolate mousse was freshly prepared, refrigerated for 1 h and removed 15 min before being served. The panel evaluated the intensity of all attributes (Table 1b) on an interval scale from 0 "not perceptible" to 5 "very strongly perceptible". Between the samples, the panelists were asked to rest for 60 s and neutralize the sense of smell using coffee beans and the sense of taste by drinking water. Data were collected using the sensory software Red-Jade® (RedJade Sensory Solutions, LLC, Pleasant Hill, California, USA). Panel performance for aquafaba and chocolate mousse profiling was monitored, and no further trainings were recommended based on the panel metrics summary, since most panelists were well performing in all categories. The sensory study was approved by the Ethics Committee of the University of Bonn (June 24, 2022, file number 122.22). The participants have been informed about their privacy rights and have given their written consent to participate in the study.

#### 2.7. Consumer tests

The consumer tests were conducted in the city center of Bonn (Germany), at the Campus Poppelsdorf (Bonn, Germany) and at a company in Euskirchen (Germany). The study involved 120 untrained participants (n = 120; mean age = 31; range: 8-75 years; 72 females, 48 males). Sequential monadic evaluation of the three mousse samples (20 g) was conducted following a complete block design. The chocolate mousses were served coded and randomized in neutral glass containers. Consumers had a spoon for tasting and water to neutralize their taste. Between the samples, the consumers had to wait for 10 s and to rinse their mouth with water before they were redirected to the next questionnaire. By scanning a QR code, consumers got access to an online questionnaire, which started with a consumer acceptance test combined with check-all-that-apply (CATA) questions. Overall liking as well as likings of the appearance, odor, taste, and texture were evaluated on hedonic scales from 1 "dislike extremely" to 7 "like extremely". The CATA questions included the same attributes as the chocolate mousse profiling. After completion, the consumers continued with a hedonic ranking test according to DIN ISO 8587 (DIN ISO 8587), where they had to rank their preferred sample in first place and the least favored sample in third place. At the end of the questionnaire, consumers were asked to submit their gender and age. Moreover, they were asked about their dietary preferences: vegan, vegetarian, flexitarian (occasional meat consumption), meat eater (regular meat consumption) or another alternative. All samples were prepared as described in section 2.6. Data collection was carried out using the sensory software RedJade® (Sensory Solutions, LLC, Pleasant Hill, California, USA).

#### 2.8. Statistical analyses

Statistical analyses were performed using Microsoft Excel, version 2016 and XLStat Sensory, version 2019.2.1 (Lumivero, Denver, Colorado, USA). One-way analysis of variance (ANOVA) followed by Tukey's honest significance test (p < 0.05) were used to compare the mean bubble count and size. For the profiles of the chocolate mousse prepared with unfermented and fermented aquafaba, a two-way analysis of variance (ANOVA) followed by Tukey's honest significance test (p < 0.05)

0.05) was conducted. Mousse profiling was further analyzed by principal component analysis to identify the relation between samples and attributes. The first two dimensions were considered and explained 100 % of the variance. Overall liking and acceptance of appearance, smell, taste and texture were analyzed by means of one-way ANOVA followed by Tukey's honest significance test (p < 0.05). CATA data were analyzed by Cochrans Q-Test (p < 0.05) and subjected to a correspondence analysis and a principal coordinates analysis to evaluate similarities in consumer liking. Hedonic rankings were evaluated by means of a Friedman test, followed by pairwise comparisons by means of Nemenyi's procedure.

## 3. Results and discussion

#### 3.1 Fermentation

Based on the results of the foaming properties and sensory bench tasting of the daily fermentation samples, the optimal fermentation times for both mushrooms were determined (4 days for AXA, 3 days for LED; see chapter 3.2 and 3.3). Aquafaba produced according to these protocols was used for the application in chocolate mousse, followed by sensory profiling and consumer tests.

## 3.2. Foaming properties

The results of foaming capacity and foam stability for aquafaba fermented with AXA and LED, depending on the fermentation time, are presented in Table 2.

The foaming capacity of unfermented aquafaba (415 %) is comparable to previously published findings, where FC varied from 182 % to 510 % for aquafaba derived from canned chickpeas (Alsalman et al., 2020; Mustafa et al., 2018; Shim et al., 2018). Different chickpea cultivars and canning conditions may explain the variation (He et al., 2021). The foaming capacity of fermented aquafaba revealed an overall variation of 49 % and 116 % for AXA and LED, respectively. However, no discernible trend during the fermentation process was observed. Fermented aquafaba showed superior foaming capacity (531-647 %) during the entire fermentation period compared to the unfermented sample. Mustafa et al. showed that increasing whipping time from 2 to 15 min improved FC and FS (Mustafa et al., 2018). However, in this study, similar or better results were observed for FC and FS after 90 s of whipping. It may be assumed that the foaming properties of aquafaba are based on low molecular weight proteins, carbohydrates and saponins (Mustafa et al., 2018; Serventi, 2020). The higher foaming capacity of fermented aquafaba may be attributed to a second sterilization step, whereas unfermented aquafaba is only sterilized once during the canning process. The high temperature (121 °C) used for sterilization may lead to further denaturation of proteins, possibly improving surface activity and thus foaming properties (Zhu and Damodaran, 1994; Belitz et al., 2008). Furthermore, the process of fermentation may lead to additional protein degradation (Zhao et al., 2016), which may result in the formation of low molecular weight proteins such as albumins, which are known for their good foaming properties (Belitz et al., 2008; Kapp and Bamforth, 2002). Different basidiomycetes are able to produce exopolysaccharides (García-Cruz et al., 2020), which are recognized for their foaming potential (Oluwa, 2020). The production of a high-molecular weight homopolysaccharide, namely lentinan (Stoica et al., 2023), has already been demonstrated for LED under conditions comparable to those in this study (García-Cruz et al., 2020; Bisko et al., 2020). In general, exopolysaccharide production depends on temperature, pH and composition of the media (Lin and Sung, 2006). For LED and Antrodia cinnamomea, glucose and peptone have been shown to be important sources of carbon and nitrogen for the production of exopolysaccharides (Stoica et al., 2023; Lin and Chen, 2007). Since glucose, amino acids and peptides have already been detected in aquafaba (Shim et al., 2018), it is possible that both LED and AXA used these metabolites for exopolysaccharide production. In addition, brown and white rot fungi can degrade cellulose and hemicellulose (Andlar et al., 2018), which are also present in aquafaba (He et al., 2021). They might serve as substrates for exopolysaccharide production, as recently described for bacteria (Tsutsui et al., 2022). The results obtained for foam stability are consistent with those reported in the literature (Mustafa et al., 2018).

The initial and final mean bubble count and mean bubble size were determined using a foam analyzer and are presented in Fig. 1.

The initial differences for both parameters were not significant between unfermented and fermented samples over the fermentation period. However, measurements revealed that the final mean bubble count was significantly higher in unfermented aquafaba and aquafaba fermented with LED on day 0, whereas the mean bubble size was significantly lower for these samples. Significant differences were observed between the different fermentation days, but no clear trend was detected over the fermentation period. The results of the aquafaba samples used for the production of chocolate mousse including unfermented aquafaba, aquafaba fermented with AXA for 4 days, and aquafaba fermented with LED for 3 days are shown in Fig. 2. During the analysis, the mean bubble count decreased, whereas the mean bubble size increased for all samples. Furthermore, a low mean bubble count is associated with a high mean bubble size. Based on these results, the higher foam stability of unfermented aquafaba can be explained because unfermented aquafaba showed a significantly higher final bubble count and a lower final bubble size (Fig. 1b and d), which indicates a more stable foam with less Ostwald ripening (Damodaran, 2005). Due to Ostwald ripening and increased drainage, the opposite trend was observed for the fermented samples, where a lower bubble count resulted in a larger bubble size (Fig. 2). For fermented aquafaba, the final bubble count and size differed over the fermentation period (Fig. 1b and d) as well as FC and FS (Table 1).

# 3.3. Aroma changes during fermentation

The olfactory evaluation by the experts showed a shift in the aroma profile, which was different between the two mushrooms. Until day three of the fermentation, the smell of AXA-fermented aquafaba was described by attributes of the aroma classes "vegetal" (e.g., chickpea) and "flowery" (e.g., sweet). During days four to nine of the fermentation, the smell was associated with attributes of the aroma classes "roasty" (e.g. caramel) and "spicy/aromatic" (e.g. malty). LED-fermented aquafaba was characterized by attributes of the aroma classes "roasty", "spicy/

**Table 2**Foaming capacity (FC) and foam stability (FS) for aquafaba fermented by AXA and LED during the fermentation period. FC of unfermented aquafaba = 415 % and FS of unfermented aquafaba = 97 %.

AXA	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9
FC (%)	564	555	578	569	604	569	587	555	591	580
FS (%)	92	92	92	92	93	92	91	92	92	91
LED	Day 0	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9
FC (%)	580	580	580	647	633	584	598	531	551	545
FS (%)	92	93	93	94	93	92	92	92	92	92

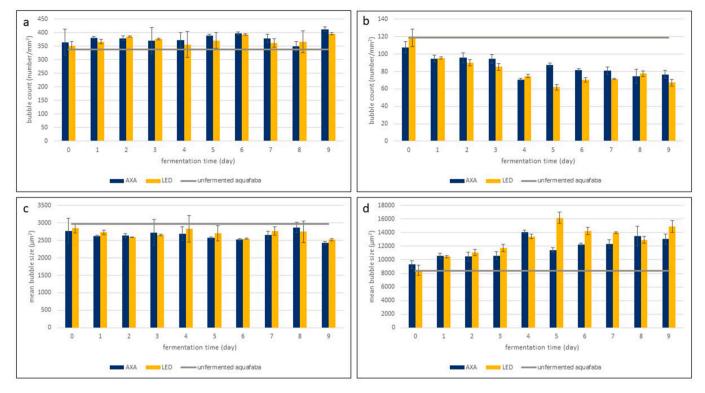


Fig. 1. Initial and final mean bubble count (a–b) and initial and final mean bubble size (c–d) of unfermented aquafaba and aquafaba fermented with AXA and LED during the fermentation period. The values of unfermented aquafaba are shown by the grey line.

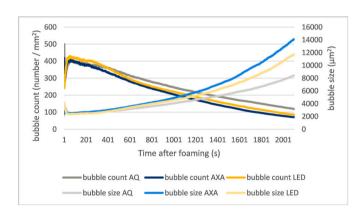
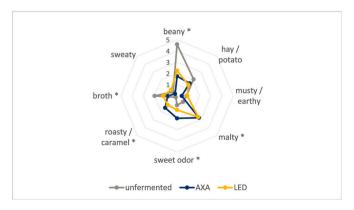


Fig. 2. Bubble count and bubble size of unfermented aquafaba and aquafaba fermented with AXA or LED.

aromatic", and "earthy" from inoculation to day three of fermentation. The aroma class "flowery" was predominant on fermentation days five to nine. All experts noticed a "microbial, animal" off-flavor on day nine. These findings are consistent with those of previous studies, where the fermentation of beer wort with various basidiomycetes led to different aroma profiles (Zhang et al., 2014). While the literature concerning the fermentation of food with AXA is scarce, for LED, an almond-like and a sweetish, fruity, plum-like, sourish aroma was observed after fermentation of green tea and wort, respectively (Zhang et al., 2014; Rigling et al., 2021). Hence, the production of different transformation products influencing the aroma profile is dependent not only on the mushrooms used but also on the substrate (Zhang et al., 2014; Liao, 2007). To the best of our knowledge, there are no studies regarding the fermentation of aquafaba by basidiomycetes; thus, no information on aroma changes is available. During the fermentation process, a promising aroma shift was observed for aquafaba application in chocolate mousse. The "roasty" and "malty" odor impressions found for AXA and LED were expected to harmonize with the chocolate, as these flavors are known to occur in dark chocolate (Ullrich et al., 2022). Furthermore, the overall liking of the samples described with these odor impressions was up to 4.0: "like". This represents the highest overall liking of the AXA samples, which was scored on day zero and day four. During the fermentation period, the liking varied between "neither like nor dislike" and "like". For LED-fermented samples, the overall liking ranged from 2.5 (day 4) to 4.0 (day 0).

#### 3.4. Aroma profile of unfermented and fermented aquafaba

Fig. 3 shows the results of the sensory smell profiling of aquafaba and provides first insights into the aroma profiles of aquafaba fermented with AXA and LED. The data show mean values of the perceived intensities for the aroma descriptors, with the marked attributes being



**Fig. 3.** Olfactory profiles of unfermented aquafaba and aquafaba fermented with LED and AXA, respectively. The intensity was evaluated on an interval scale from 0 "not perceptible" to 5 "very strongly perceptible".

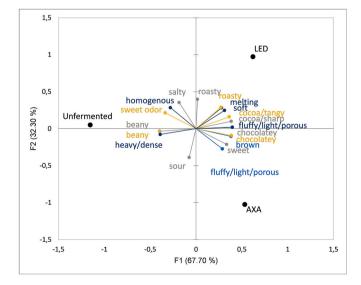
significant ( $\alpha = 0.05$ ).

As indicated in Fig. 3, the profiles of the two fermented samples are comparable, whereas the unfermented sample shows clear deviations. Remarkably, the perception of the "beany" smell, which is characteristic for aquafaba, was significantly less pronounced in fermented samples. Furthermore, the AXA-fermented aquafaba had significantly lower ratings for the attribute "broth" compared to the unfermented aquafaba. Similar observations have been reported regarding the fermentation of a pea-rice protein isolate using LED, where a decrease in "beany" and "earthy" off-notes was monitored in fermented samples (Clark et al., 2022). Product-characteristic odors can either be reduced or covered by new odor-active compounds synthesized by the mushrooms (Clark et al., 2022; Rigling et al., 2021; Zhang et al., 2015). Regarding new aroma impressions, the results of the aquafaba profiling and the odor characterization by experts during fermentation are in agreement. The experts described fermented aquafaba by using the attributes "roasty", "caramel" and "malty", which is consistent with significantly higher ratings for these attributes through the sensory panel. The perception of a "sweet odor" is more pronounced for fermented samples, in particular for AXA, where it is significantly higher than in the unfermented sample. For fermentation of beer wort with LED, a sweetish aroma was already observed (Zhang et al., 2014). Possibly, the "caramel" and "malty" notes, which are significantly more intense in fermented samples, implied a sweet aroma perception, leading to higher scores for "sweet odor" perception (Beal and Mottram, 1994; Paravisini et al., 2012). No significant differences were found for the attributes "hay/potato", "musty/earthy" and "sweaty". In conclusion, the fermentation of aquafaba with AXA and LED resulted in the reduction of unpleasant odors like "beany" and "broth", while interesting new odor impressions occurred.

## 3.5. Profiles of chocolate mousses

To determine whether the changes in aroma occurring during the fermentation process are also perceived in a food product and to analyze the effect on consumer acceptance, the fermented aquafaba was used in a sample food product, namely chocolate mousse.

The profiling data of chocolate mousses revealed differences mainly between mousses prepared with fermented aquafaba and mousse made with unfermented aquafaba. The results are illustrated in the principal component analysis (PCA) in Fig. 4, conducted to identify underlying



**Fig. 4.** PCA plot of the sensory attributes for the samples with unfermented and fermented aquafaba. Attributes for appearance are displayed in dark blue, smell attributes are displayed in yellow, taste attributes are displayed in grey and texture attributes are displayed in light blue.

factors that explain correlations within the observed attributes. Two principal components, namely F1 and F2, were identified, which accounted for 67.7~% and 32.3~% of variance, respectively. A clear differentiation between the three chocolate mousse samples is shown. The mousse prepared with unfermented aquafaba is characterized by negative loadings on F1, whereas the mousses with fermented aquafaba have positive loadings. The fermented samples are separated based on the F2 axis, with LED having positive loadings, whereas AXA is characterized by negative loadings on the F2 axis.

In chocolate mousses produced with fermented aquafaba, the intensity of the appearance attribute "brown" was significantly higher than in those prepared with unfermented aquafaba. Comparable to the aroma profile, the mousse produced with unfermented aquafaba was characterized by its "beany" odor, whereas the fermented mousse samples were described by the attributes "cocoa/tangy", "chocolatey" and "roasty", with the latter being perceived significantly more intensely in mousse prepared from LED-fermented aquafaba. Unlike the aquafaba profiling, where fermented samples had a "sweet odor", the odor of chocolate mousse produced with unfermented aquafaba was perceived as sweeter. Concurrently, the enhanced perception of the attribute "chocolatey" in mousse samples with fermented aquafaba might lead to the higher perception of the attribute "cocoa/tangy", and hence lower intensities of the "sweet" odor. Nonetheless, concerning taste, fermented samples were perceived more "sweet". LED contains amino acids that can contribute to a sweet flavor (Zhang et al., 2021) and may have led to the sweet impression. The other attributes used for the description of smell and taste, such as "beany", "roasty", "cocoa/tangy/sharp", and "chocolatey", showed clear positive correlations. This is probably based on the retronasal perception of these attributes. Moreover, the chocolate mousse prepared with AXA-fermented aquafaba was perceived as significantly more "sour". Most probably, the perceived sourness is based primarily on the formation of organic acids (Da Ramos Conceicao Neta et al., 2007). Therefore, the ability of AXA to produce oxalic acid may lead to a higher perception of this attribute (Vlasov et al., 2023). Furthermore, the pH of aquafaba fermented with AXA is slightly lower (pH = 6.0) than the pH of the sample fermented with LED (pH = 6.2) and the unfermented aquafaba (pH = 6.2). A decrease in pH due to oxalate accumulation has already been observed for brown-rot fungi, which also include AXA (Dutton et al., 1993). However, it should be noted that the differences in the aroma of chocolate mousses prepared with fermented and unfermented aquafaba were smaller than those observed in the profiling of the aquafaba. This phenomenon may be explained by the predominance of the chocolate aroma, which masks other flavor impressions like "beany" or "roasty" in all mousse samples.

With regard to the texture, the chocolate mousse produced with unfermented aquafaba was described as significantly more "homogenous" than the AXA-fermented sample. The perception of the attribute "fluffy/light/porous" in texture was found to be significantly higher when mousse was prepared with LED-fermented aquafaba. The outcomes of the texture description can be related to the foaming properties. Mousses produced with fermented aquafaba exhibited a higher foaming capacity, resulting in higher ratings for the attributes "melting", "soft" and "fluffy/light/porous". In contrast, the lower foaming capacity of unfermented aquafaba resulted in a texture characterized as "heavy/ dense". The foam analyzer measurements revealed information about the mean bubble count and the mean bubble size. The final bubble count of aquafaba fermented with AXA for 4 days and with LED for 3 days was significantly lower than that of unfermented aquafaba. The mean bubble size showed an opposite pattern: bubbles were significantly larger in fermented samples than in unfermented aquafaba. Thus, it is assumed that fewer but larger bubbles lead to a lighter and fluffier texture. It has already been observed that the airy mouthfeel increased with increasing bubble size in semi-solid formulations (Minor et al., 2009) and in chocolate (Haedelt et al., 2007). Furthermore, larger bubbles resulted in a faster melting of chocolate in the mouth and a less hard perception (Haedelt et al., 2007), which was observed for fermented samples in this study, being described as more "melting" and "soft". Nonetheless, the chocolate with larger bubbles was perceived as less creamy (Haedelt et al., 2007), since large bubbles and variations in bubble size might lead to an irregular texture (Bennion et al., 1997). This may result in a lower perception of the attribute "homogenous" in the mousse made from fermented aquafaba. Furthermore, for the attribute "smoothness", which is comparable to "homogenous", higher ratings were reached in another study for foamed milk samples comprised of significantly smaller bubbles (Hatakeyama et al., 2019). Accordingly, unfermented aquafaba, with many small bubbles, was perceived as more "homogenous".

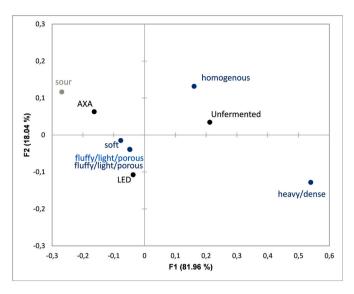
#### 3.6. Examination of chocolate mousses by consumers

In total, 120 consumers, including 72 females and 48 males, aged between 8 and 75 years, participated in the tastings. Among the participants, 69 % ate meat, whereas 23 % were vegetarians and 8 % were vegans.

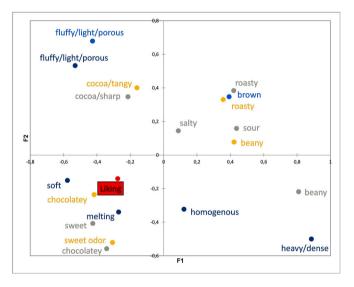
The acceptance test revealed that the overall liking scores ranged from 5 "like a little" to 6 "like" for all chocolate mousses. The liking scores for appearance, smell, taste, and texture were rated in the same range. Significant differences were observed only for texture, where chocolate mousse prepared with LED-fermented aquafaba received significantly lower ratings for liking than mousse made with unfermented aquafaba. However, there were only small differences between the samples, indicating that consumers were unable to distinguish between the different mousses. As previously stated for the profiling data, it is possible that the chocolate used for mousse production may have masked other flavor impressions. The smaller differences between samples might also be attributed to the fact that consumers were less sensitive to small changes in aroma than the trained panel. In another study, a chocolate mousse made from aquafaba was compared to a traditional mousse made from egg white, where no significant differences ( $\alpha = 5\%$ ) were observed in terms of the acceptance of the mousses (Donatus et al., 2023).

The minor differences in the acceptance test can be related to the results of the ranking test. The Friedman test indicated a significant difference between the samples (F = 18.3). However, a subsequent sample comparison showed significant differences only between chocolate mousse prepared with LED-fermented aquafaba and mousse prepared with AXA-fermented aquafaba, as well as unfermented aquafaba. The LED mousse sample was ranked significantly lower. No significant differences were observed between mousses produced with AXA-fermented aquafaba and unfermented aquafaba; therefore, they share the first rank. The lower ranking of the mousse prepared with LED-fermented aquafaba may be explained by a significantly lower acceptance of its texture.

Analysis of the CATA data revealed significant differences for six attributes, which are displayed in a correspondence analysis in Fig. 5. Based on the frequency with which attributes were selected by consumers for each aquafaba sample, relative frequencies are calculated and displayed in a two-dimensional space. This allows relationships to be shown both between attributes and between attributes and samples. The texture of the chocolate mousse prepared with fermented aquafaba tended to be "fluffy/light/porous" and "soft", whereas the mousse of unfermented aquafaba was found to be more "homogenous". These results are in line with the profiling data collected by the trained panel (Figs. 3 and 4). Furthermore, both analyses indicated that mousse prepared with AXA-fermented aquafaba was characterized as "sour". The correspondence analysis further revealed that the CATA attributes with significant differences were mainly related to texture, suggesting that texture may be crucial for the liking of the samples. As expected, the principal coordinates analysis (Fig. 6), comprising all CATA attributes, associates the overall liking of chocolate mousses with the attributes "sweet" and "chocolatey" in both smell and taste, and with "melting"



**Fig. 5.** Biplot of the correspondence analysis of the significant sensory attributes for the unfermented and fermented aquafaba samples. Attributes for appearance are displayed in dark blue, taste attributes are displayed in grey and texture attributes are displayed in light blue.



**Fig. 6.** Extended internal preference map for overall liking and sensory appearance, flavor and texture descriptors for F1 and F2. Attributes for appearance are displayed in dark blue, smell attributes are displayed in yellow, taste attributes are displayed in grey and texture attributes are displayed in light blue.

and "soft" in texture. These attributes were more pronounced for mousse with fermented aquafaba in the profiling data, but no significant differences were detected by consumers in the CATA analysis. For texture, the attributes "soft", "melting", and "homogenous" indicated pleasantness. Among these attributes, only "soft" and "homogenous" showed significant differences in the correspondence analysis (Fig. 5). Whereas "soft" characterized both mousses of fermented aquafaba, "homogenous" is more characteristic for the unfermented aquafaba mousse. This may have a balancing effect on the overall liking, considering that the smell and taste attributes related to pleasantness did not show significant differences. In general, the texture is one of the most important quality attributes of chocolate mousse, and the importance of homogeneity for chocolate mousse was already demonstrated in a previous study where whey was used to produce chocolate mousse. The results

were compared to a commercial mousse prepared with egg white. One third of the participants preferred the commercial mousse and justified their decision with an increased homogeneity, among other factors (Bouizar et al., 2021).

In this study, the fermented aquafaba, which was distinguished by a sweet, caramel-like and roasty aroma, was used in chocolate mousse as an example for an aerated dessert. The superior foaming capacity of fermented aquafaba would also benefit the production of other aerated, foam-based foods, such as meringue. The changes in aroma observed were considered to be well-suited for the use in the chocolate mousse. Further research is required to select suitable organisms for fermentation and to determine the optimal fermentation time, depending on other intended applications.

#### 4. Conclusion

Aquafaba was fermented using basidiomycetes for the first time. It was shown that it is an efficient substrate, providing all nutrients that the fungi need for their growth. The fermentation process resulted in a superior foaming capacity, but no increase in foam stability was achieved. However, a promising change in the flavor profile was observed for application in products where beany flavors would be considered an off-flavor. The characteristic but unpleasant aroma impressions, commonly associated with aquafaba, were significantly reduced by fermentation. These findings are crucial since the application of aquafaba in food is often limited by its aroma. The change observed in aroma towards "roasty" and "malty" notes may be attributed to the type of organism used in fermentation. It would therefore be of interest to investigate the potential for aquafaba fermentation with other basidiomycetes, fungi, or bacteria to facilitate specific applications, depending on the change in flavor. Furthermore, future research should focus on the characterization of the aroma-active compounds.

In this study, fermented aquafaba was successfully used in an innovative plant-based chocolate mousse, which gained high consumer acceptance and was perceived as more "fluffy/light/porous", "melting" and "soft" than mousse prepared with unfermented aquafaba. In conclusion, first insights into the aroma profiles of fermented aquafaba and chocolate mousse produced with aquafaba have been provided, which may play an important role in future aquafaba research. Fermentation has also been shown to be a useful tool for reducing beany off-flavors in aquafaba, which may facilitate its use in foods without masking ingredients. Furthermore, it can offer the possibility to raise new aromas in innovative food products while the technological properties are maintained or even improved.

# CRediT authorship contribution statement

Lea Mehren: Conceptualization, Methodology, Investigation, Formal analysis, Writing – original draft. Lena Elliger: Methodology, Investigation, Formal analysis. Hanna May: Methodology, Investigation, Formal analysis. Andreas Schieber: Resources, Supervision, Writing – review & editing. Nadine Schulze-Kaysers: Conceptualization, Supervision, Methodology, Writing – review & editing.

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# Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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#### Data availability

I have shared the link to my data at the Attach File step. doi: 10.17632/4v7vymddwv.1

Foaming properties and olfactory profile of fermented chickpea aquafaba and its application in vegan chocolate mousse (Original data) (Mendeley Data)

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