Impact of processing and storage on changes in the volatile compounds of whole chickpeas: an untargeted headspace fingerprinting study

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Abstract

In the present research the impact of processing (soaking, cooking and sterilisation) and oxygen availability during storage (up to 40 weeks at 20 ºC) on the volatile compounds in whole chickpeas was investigated. Volatile profiles of the differently processed chickpeas were obtained and compared. Soaking resulted in an increased number of aldehydes and alcohols, while thermal processing (e.g. cooking and sterilisation) originated compounds like furan derivatives, sulphur compounds and aromatic compounds. More hydrocarbons, ketones and sulphur compounds were formed in sterilised chickpeas after storage at higher oxygen availability, while more alcohols were present in the chickpeas stored at lower oxygen availability. The results showed that both processing and oxygen availably during storage significantly affected the volatile profile of intact chickpeas.

Keywords: Cicer arietinum L., volatile analysis, HS-SPME-GC-MS, untargeted fingerprinting approach, MVDA

Introduction

Aroma is an important attribute that can partly determine the sensory shelf life of shelf-stable food products and thus can influence their marketability. Legumes, such as chickpeas, have become increasingly important in the last few years but are often associated with an unfamiliar and unpleasant 'beany' aroma. The volatiles classified as contributing to this 'beany' aroma can be formed at different stages in the pulse processing chain. Chickpeas are most commonly available as processed ready-to-eat chickpeas. Yet, the volatile compounds in whole processed chickpeas and their evolution during storage are only very scarcely described in scientific literature. Therefore, in this study the headspace volatile components in differently processed chickpeas as well as in chickpeas stored between 0 and 40 weeks, in different packaging materials, were investigated.

Experimental

Preparation of soaked and cooked chickpeas

Dried kabuli chickpeas were soaked for 16 hours in an excess of demineralised water to obtain the 'raw, soaked chickpeas'. These chickpeas were cooked in semi-closed recipients in a water bath at 95 °C for 40 min to obtain the 'home-cooked chickpeas'. In order to obtain the industrially 'sterilised chickpeas', the same chickpeas were soaked in standardised production water and consecutively sterilised at 116 ºC in a Steriflow pilot retort (Barriquand, Paris, France) to obtain a F_0-value of 15.6 min. Two types of pouches were used during sterilisation, aluminium and plastic pouches, with an oxygen permeability of 0.05 and 1 $cm^3/(m^2 day.bar)$, respectively. The sterilised chickpeas were stored at 20 ºC and samples were taken at 12 time points between 0 and 40 weeks (sampling weeks: 0, 1, 2, 3, 4, 8, 12, 16, 20, 24, 28, 32, 40). At the specific sampling points, pouches were opened and chickpeas (without aquafaba) were transferred into odourless tubes, frozen and stored at -40 ºC until analysis.

HS-SPME-GC-MS fingerprinting

All samples were analysed using an untargeted headspace solid-phase micro extraction-gas chromatography (HS-SPME-GC-MS) fingerprinting approach, adapted from Kebede *et al*. [1] and Vervoort *et al*. [2].

Chickpeas were mashed with demineralised water (ratio dry chickpea to water 1:3.4) using an Ultra-Turrax T25 (Janke & Kunkel, IKA Labortechnik, Staufen, Germany) at 8000 rpm for 1 min. 3 ± 0.05 g chickpea puree and 3 ml saturated NaCl solution were added to amber glass vials (20 ml, Macherey-Nagel, Düren, Germany). A GC (Agilent Technologies, Santa Clara, CA, United States) coupled to an MSD 5977A (Agilent Technologies, Santa Clara, CA, United States) was used to analyse the volatiles and six replications per sample were analysed. Samples were incubated and extracted at 40°C for 30 and 40 minutes, respectively, under 500 rpm agitation. Desorption took place at 230 ºC for 2 minutes at the GC inlet where a splitless injection was performed. The GC was equipped with a capillary HP Innowax column (60 m x 250 µm x 0,25 µm, Agilent Technologies, Santa

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Clara, CA, United States) with helium as the carrier gas at a constant flow of 1.1 ml/min and a pressure of 124.9 kPa. The temperature profile of the GC oven consisted of a holding step (40 °C, 2 min), a first heating ramp (40 °C to 80 °C at 3 °C/min), a second holding step (80 °C, 1 min), a second heating ramp (80 to 220 °C at 6 °C/min) and a final heating ramp (220 to 250 °C at 50 °C/min). MS detection was obtained in electron ionisation mode at 70 eV with a scanning range of 35 – 400 m/z and a scanning speed of 3.8 scans per second. The MS ion source and quadrupole were 230 °C and 150 °C, respectively.

Data analysis

GC-MS fingerprinting data were analysed using the multivariate data analysis (MVDA) approach as described by Kebede *et al.* [1] and Vervoort *et al.* [2], to obtain PLS and PLS-DA models and to calculate variable identification coefficients (VID). Depending on the volatile data, volatiles with an |VID| value between 0.7 or 0.8 to 1.0 were considered as discriminant components for a certain class of samples (PLS-DA models) or to be significantly changing over shelf life (PLS models). To identify the volatile components, the spectral library of NIST (NIST14, version 2.2, National Institute of Standards and Technology, Gaithersburg, MA, USA) was used as well as comparison of retention indices to those reported in literature or obtained by analytical standards.

Results and discussion

Impact of processing method on chickpea volatiles

A total of 121, 72 and 109 volatiles were found in the headspace of the soaked, cooked and sterilised chickpeas, respectively. A PLS-DA model with 2 LVs was obtained, explaining 99.8% of the *Y*-variance. In Figure 1, a biplot of this PLS-DA model explaining the difference between the three processing conditions on the volatile compounds is shown. In this biplot the differently processed samples are denoted as the coloured objects, the volatile compounds as the open circles. The vectors represent the correlation loadings of the classes (processing conditions). From the biplot it is clear that processing conditions significantly impacted the volatile profile of chickpeas, as the three different classes are clearly separated.

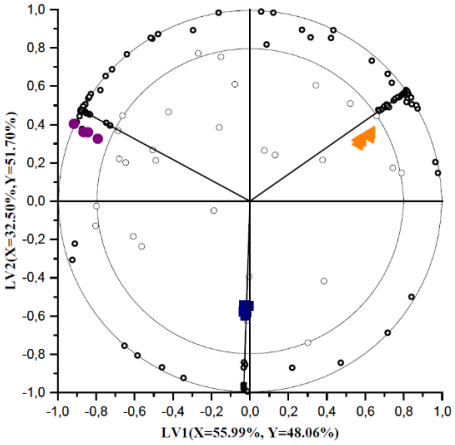

Figure 1*: Biplot (LV axes 1 vs 2) of the PLS-DA conducted on the volatile profile of raw (soaked) (●), cooked (■) and sterilised (◀) chickpeas. Volatile components are represented as open circles and markers with |VID|>0.8 are represented in bold. The vectors represent the correlation loadings for the Y-variables (classes). The outer and inner circles on the biplot represent the correlation coefficient of 1.0 and 0.8 respectively, indicating the area where the volatiles that are characteristic for a certain class are present.*

Discriminant compounds were determined for the different classes using the VID criterion (|VID|>0.8), indicating that these compounds were present in higher (positive value) or lower (negative value) concentration in a certain class compared to the other classes. These discriminant compounds are presented as the bold open circles in Figure 1. For the soaked, cooked and sterilised chickpeas, 66, 13, and 53 discriminant components with a positive and 2, 12, and 5 discriminant components with a negative VID coefficient were found, respectively. Of these components, 43, 5, and 31 of the discriminant components with a positive and 2, 7, and 2 of the discriminant components with a negative VID coefficient could be (tentatively) identified, respectively.

In raw, soaked chickpeas the identified discriminant components with a positive VID coefficient consisted mainly of aldehydes (45%) and alcohols (30%). Possibly, these compounds resulted from enzymatic lipid breakdown in the chickpeas during the soaking step or at the beginning of the supply chain, prior to drying of the chickpeas. In raw chickpeas, aldehydes and alcohols have previously been described to be the most abundant volatile classes [3] and these types of compounds can potentially be associated with a ‘beany’ type of aroma [4].

In contrast, in the thermally processed chickpeas (i.e. cooked and sterilised), other volatile classes were present at higher concentrations. In the cooked chickpeas, two sulphur compounds, a furan derivative, an organic compound and an alcohol were found. High intensity processing (sterilisation) mainly resulted in more aromatic compounds (23%) and furan derivatives (19%). The sulphur compounds potentially resulted from degradation of sulphur containing amino acids during cooking, the aromatic compounds from lipid oxidation or degradation of aromatic amino acids, while furan derivatives potentially formed during the early stages of Maillard reactions [5].

Impact of oxygen availability during ambient storage on chickpea volatiles

Headspace volatiles of the sterilised chickpeas stored in both aluminium and plastic pouches were analysed at several time points between 0 and 40 weeks. PLS models with 3 and 4 LVs were obtained, explaining 94.7% and 96.3% of the *Y*-variance for chickpeas stored in the aluminium and plastic pouches, respectively. Biplots of the first two LVs of the PLS models on the volatile profile of these samples are shown in Figure 2. In these biplots, the samples stored at different time points are presented as the coloured objects, the volatiles as the open circles and the volatiles significantly changing over time are presented as the bold open circles. The vectors represent the correlation loading for the *Y*-variables (time). In total, 121 volatiles were found in the chickpeas stored in the aluminium pouches, 156 in the chickpeas stored in the plastic pouches. The higher number of volatiles formed in the plastic pouches is probably caused by an increased amount of oxidation reactions during storage due to the higher oxygen permeability of this packaging material.

Comparing the two biplots, some differences can be observed. In the biplot of the aluminium pouches (Figure 2A), the first LV is explaining the major part of the effect of storage time, as a clear chronological trend is found from the left to the right in the biplot. In this biplot, most of the volatile compounds were located closer to the centre of the biplot. This indicates that most volatiles did not significantly change during the storage period of 40 weeks. In the biplot of the chickpeas stored in plastic pouches, a less clear chronological trend is observed in the biplot, indicating that the volatile profile of the chickpeas stored in the plastic pouches were not largely affected by storage time either. However, based on the VID criterion (|VID|>0.7), in both chickpeas stored in the different packaging materials, some compounds were found to be significantly changing during storage time.

At lower oxygen availably, acetyl thiazole significantly decreased during storage time, while ethyl acetate significantly increased. At higher oxygen availability, different changes took place. Methyl benzaldehyde decreased over time and methyl cyclohexane increased over time. This indicates that the oxygen availability related to packaging permeability during storage not only changed the amount of chemical reactions, but also the type of chemical reactions taking place.

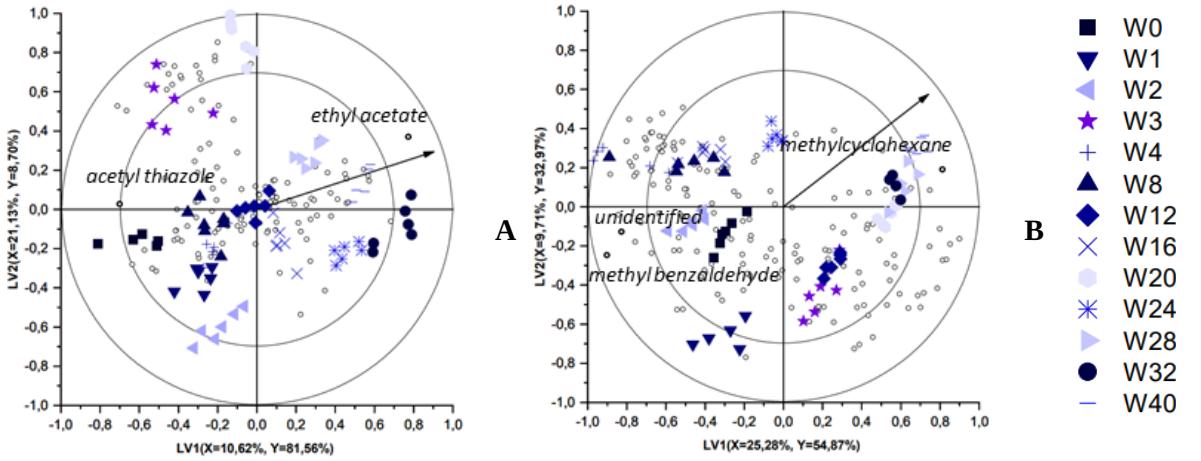

Figure 2*: Biplots (LV axes 1 vs 2) of the PLS models representing volatile changes of chickpeas stored in aluminium (A) and plastic (B) pouches. Volatile components are represented as open circles and markers with |VID|>0.7 are represented in bold. The vectors represent the correlation loadings for the Y-variable (time). The outer and inner circles on the biplot represent the correlation coefficient of 1.0 and 0.7 respectively, indicating the area where volatiles changing significantly during storage are present. W=weeks of storage.*

In order to investigate the differences in the volatile changes at different oxygen availably in more detail, the headspace volatiles of chickpeas stored in aluminium and plastic pouches for 0 weeks and 40 weeks were compared. A PLS-DA model with 4 LVs, explaining 95.9% of the *Y*-variance was obtained. A biplot of the first two LVs of the PLS-DA model is presented in Figure 3. Four classes can be seen in this biplot. In the chickpea samples at the beginning of storage (0 weeks), for the aluminium and plastic pouches, 12 and 3 discriminant compounds with a positive and 5 and 7 discriminant compounds with a negative VID coefficient were found, respectively (|VID|>0.7). These compounds consisted of an aromatic compound and an aldehyde for the plastic pouches, and for the aluminium pouches, they mostly consisted of aromatic compounds (40%) and of sulphur

compounds (30%) formed during the sterilisation process. These results indicate that, even though the samples had undergone the same sterilisation process, the oxygen permeability of the pouches impacted the volatile profile of the sterilised chickpeas during the process. After a 40-week storage period, the volatiles profiles of the chickpeas stored in the different packaging materials were more different from each other. In the plastic pouches, the discriminant compounds present in higher concentrations were mainly hydrocarbons (33%), ketones (22%) and sulphur compounds (22%), the former two probably resulting from lipid oxidation during storage at higher oxygen availability. In contrast, the chickpeas stored in the aluminium pouches showed a higher concentration of alcohols (33%) compared to the other samples. Probably more alcohols were found in these samples, because less oxidation of alcohols to other compounds, like ketones, took place at lower oxygen availability [6]. This figure also confirms the storage effect for a given packaging.

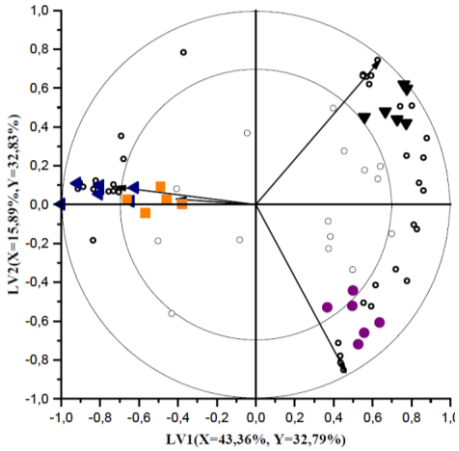

Figure 3*: Biplot (LV axes 1 vs 2) of the PLS DA-model on the volatile profile of chickpeas sterilised and stored for 0 weeks (plastic (■), aluminium (◀)) and 40 weeks (plastic (▼), aluminium (●)). Volatile components are represented as open circles and markers with |VID|>0.7 are represented as bold circles. The vectors represent the correlation loadings for the Y-variables (classes). The outer and inner circles on the biplot represent the correlation coefficient of 1.0 and 0.7 respectively, indicating the area where the volatiles that are characteristic for a certain class are present.*

Conclusion

It was concluded that processing conditions significantly influence the volatile profile of whole chickpeas. Since the soaked chickpeas were found to contain more 'beany' related compounds, it could be stated that thermal treatment can potentially be used to contribute to a more pleasantly perceived chickpea aroma. However, additional sensory testing needs to be performed to confirm which processing conditions give the most desirable aroma.

Moreover, it was concluded that oxygen availability, depending on permeability of the packaging material, significantly impacts the volatile profile of chickpeas during a 40-week storage period at 20 ºC. However, additional sensory testing is required to understand if these volatile differences are actually observed by the consumer and if so, which packaging material is most desirable.

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