Characterisation of the aroma developed during fermentation and roasting of jackfruit seeds

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Abstract

When jack fruit seeds are fermented with banana leaves, and dried and roasted in a process similar to that used in the production of roasted cocoa nibs, a chocolate aroma develops. By using SPME GC-olfactometry, we have shown that compounds such as 2- and 3-methylbutanal, trimethylpyrazine and phenylacetaldehyde, which are key components of cocoa aroma, are present in the headspace of roasted fermented jack fruit (FJS) seeds at similar levels to that of a typical Brazilian cocoa powder. However, a series of less desirable higher molecular weight pyrazines with branched chain substituents was also found in the headspace of the FJS, but not in the cocoa. The valine-derived substituents imparted carbolic and cardboard aromas typical of the jackfruit seeds. Minimisation of these is important for improving the flavour of this sustainable and inexpensive cocoa substitute.

Introduction

The flesh of the jackfruit (*Artocarpus heterophyllus* Lam.) is popular in tropical countries where it grows in abundance in the wild. The seeds, which are usually discarded, account for 15-18% of the weight of the fruit and they are an under-utilised waste stream which could be exploited by local communities in Brazil. Recently, Spada et al. [1] showed that when jackfruit seeds are fermented and roasted using a process similar to that used for cocoa beans, a distinctive chocolate aroma develops. Various novel applications are currently being developed for the use of ground and roasted jackfruit seeds as a partial substitute for cocoa powder in cakes, cappuccino and cosmetics.

The development of the desirable chocolate aroma is very dependent on the post-harvest treatment, and the subsequent drying and roasting processes. Response surface methodology was used to compare fermentation and acidification steps prior to roasting, and to identify optimum roasting conditions to maximise the chocolate aroma of the ground roasted seeds. Twenty-seven different roasted jackfruit seed powders were assessed for "chocolate aroma" by a sensory panel (n=162) using ranking tests [1]. Optimum roasting conditions from each of the three processes (dried, acidified and fermented) were selected and the corresponding powders analysed by GC-MS and GC-olfactometry and compared to a standard Brazilian cocoa powder. In this paper, we focus on the fermented product (FJS) which had the highest ranking score for chocolate aroma.

The aim of the work was to confirm the presence of key chocolate aroma compounds in the FJS powder and to identify those compounds responsible for the less desirable "jackfruit seed" aromas.

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Experimental

Materials

The jackfruit were collected from the local countryside, the flesh discarded and the seeds, pulp and banana leaves were placed in a closed container for 3 days to encourage anaerobic fermentation and production of alcohol. Over 5 days, the container was opened daily and the fermentating mass was turned over manually to encourage oxidation and production of acetic acid. After 8 days the pulp and banana leaves were removed and the fermented seeds dried at 60 ºC for 24 h. The seeds were then roasted at 154 ºC for 35 min and ground to a powder.

The cocoa powder was obtained from Cargill, Brazil. It was of Brazilian origin and the cocoa beans had been fermented and roasted. All reference standards were obtained from Sigma Aldrich, Gillingham, UK.

Model reactions

Equimolar amounts (0.1 mM) of glucose, glycine and another amino acid (valine, leucine or isoleucine) were adjusted to pH 7 and heated in an autoclave at 125 °C for 30 min. They were diluted 10 times prior to GC-MS analysis on two columns under identical conditions to the analysis of the powders.

Analysis of the volatile compounds by SPME and GC-MS

FJS powder (3 g) or Brazilian cocoa powder (3 g) were mixed with HPLC grade water (3 ml) in an SPME vial and vortexed for 2 min. After equilibrating the sample at 45 °C for 15 min, the triple phase fibre (65 µm PDMS/DVB/Carboxen from Supelco) was exposed to the headspace for 55 min as previously described [1]. SPME extracts were analysed by GC-MS on an Agilent HP5890 Series II GC, coupled to a 5975 MSD. The GC was equipped with either a Zebron ZB-wax column or a Zebron DB5 column (both Phenomenex® 30 m x 0.25 mm x 0.25 µm film thickness) and a standard 5 °C/min temperature ramp programme was used.

Analysis of the volatile compounds by SPME and GC-Olfactometry

SPME extracts were also analysed on the same two columns using an Agilent HP5890 Series II GC-FID system coupled to an ODO 2 odourport (SGE). The outlet was split between a flame ionisation detector and a sniffing port, each with a flow of 1 ml/min. The contents of the SPME fibre were desorbed for 3 min in a split/splitless injection port, in splitless mode, onto five small loops (5 cm diameter) of the column in a coil, which were cooled in solid carbon dioxide, contained within a 250 mL beaker. After 3 min the beaker was removed and a standard 5 °C/min temperature ramp programme employed. The eluting aroma regions were described and scored by two assessors in duplicate on a scale of 0 (none) to 7 (strong). Mean values are reported in Table 1.

Results and discussion

GC-MS

The extract contained ~200 volatile compounds, most of which were identified and at least 70 of the identities were confirmed by comparison with the appropriate standard reference compound. Of the 200 volatiles, we believe that >60 are pyrazines, although reference standards were only available for 10 of these. For that reason, model reactions were prepared in order to distinguish the many pyrazines that were generated during the roasting process. By using either valine, isoleucine or leucine in a simple glucose/-glycine

Maillard reaction, it was possible, in conjunction with mass spectra and LRI data on two columns, to confirm the identity of the substituents on many pyrazines as 2-methylpropyl, 2-methylbutyl or 3-methylbutyl respectively. However, the position of the substituents could not be determined unless there were authentic standards available for some of the isomers. This information was vital in attributing the aroma regions in the GC-Olfactometry to particular compounds.

GC-Olfactometry

Over 50 aroma regions were detected by the assessors on the DBWax column. Of these, 40 were assigned to compounds, most of which were also found in the GC-MS. The identities of these 40 compounds were further confirmed by carrying out GC-O on a DB5 column. Most of the aromas detected on the DBWax were found at the correct LRI on the DB5 column. Many of these were generic aroma compounds found in most foods as described by Dunkel et al. [2].

In this paper, the main focus is on the aroma regions which obtained high scores from the GC-O assessors, particularly those which are known to be important in the aroma of chocolate [3] or cocoa powder [4], or those which resembled the less desirable character of the jackfruit seeds which dominated the aroma of some of the earlier trial samples. These are summarised in Table 1.

Table 1: Comparison of GC-Olfactometry scores for selected compounds for roasted fermented jackfruit seeds (FJS) and Brazilian cocoa powder, mean score of 2 assessors in duplicate where 0 = none and 7 = strong

LRI on DBWax				LRI on DB5			GC-O Score	
GC-O expt	GC-MS expt	GC-MS au	Identity of compound	GC-MS au	GC-O expt	GC-MS expt	FJS	Cocoa
i)Compounds typically found to be important in chocolate or cocoa aroma, detected in both FJS and cocoa								
909	*911*	*925/ 928*	2/3-methylbutanal	656/ 665	<600	651/ 662	7	6
1390	*1394*	*1394*	trimethylpyrazine	1008	coelute	1003	7	6
1628	*1624*	*1624*	phenylacetaldehyde	1058	1053	1049	6	5
ii) Compounds only found in roasted jackfruit seeds, typically with jackfruit, carbolic, cardboard aroma								
1487	*1489*	*1489**	methyl-2-methyl-propylpyrazine 1	1134*	1133	1140	5	0
1494	*1495*	*1495**	methyl-2-methyl-propylpyrazine 2	1134*	1145	1149	5	0
1553	*1553*	*1555**	a dimethyl 2-methyl-propylpyrazine	1206*	1205	1206	3	0

**found in corresponding reaction mixture*

The upper half of Table 1 shows compounds which are typically associated with chocolate or cocoa aroma and have been reported by GC-Olfactometry, and deemed to be important, in many cocoa based products including milk chocolate [3] and cocoa powder [4]. These compounds were detected in both the Brazilian cocoa and the FJS,

suggesting that these too might be important in the chocolate component of the FJS aroma.

The lower half of the table shows the compounds which had relatively high GC-O scores and were detected in FJS, but not in the cocoa powder. These were 2-methylpropyl substituted pyrazines which were far more abundant in the FJS chromatograms compared to the cocoa, and were described by the GC-O assessors with less desirable terms such carbolic, cardboard and "roasted jackfruit seed flour". All three isomers of methyl-2-methylpropylpyrazine were detected in the FJS by GC-MS, and in the valine model system, two of which corresponded to the very characteristic jackfruit seed aromas which were detected in the GC-O at the corresponding LRIs. Similarly, all three isomers of dimethyl-2-methylpropylpyrazine were detected by GCMS in both FJS and the valine model system, but only the most abundant isomer was detected by GC-O. The LRI of this isomer on a DB5 column matches the LRI of one of the two isomers (2,5- and 2,6-dimethyl-3-(2-methylpropyl)pyrazine) synthesised in our lab and reported previously [5].

Further development of the flavour of this potential cocoa substitute needs to focus on removing or decreasing the contribution from the branched chain substituted pyrazines, particularly those which are likely to be derived from valine during the roasting process.

References

1. Spada, F.P., Masson Zerbeto, L., Cabreira Ragazi, G., Roel Gutierrez, E., Coelho Souza, M., Parker, J. K., Canniatti-Brazaca, S. (2017) J. Ag. Food Chem. 65: 1196-1208.
2. Dunkel, A., Steinhaus, M., Kotthoff, M., Nowak, B., Krautwurst, D., Schieberle, P., Hofmann, T. (2014) Angew. Chem. Int. Ed Engl. 53(28):7124-43.
3. Schnermann, P., Schieberle, P. (1997) J. Agric. Food Chem. 45: 867–872.
4. Frauendorfer, F., Schieberle, P. (2006) J. Agric. Food Chem. 54: 5521- 529.
5. Elmore, J.S., Dodson, A.T., Muttucumaru, N., Halford N.G., Parry, M.A.J., Mottram, D.S. (2010) Food Chem. 122:753-760.