

Effects of Roasting Temperature on the Aroma Components of Carob (*Ceratonia siliqua* L.)

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The analysis of the volatile fraction of carob bean pulp during a roasting process between 10 and 60 min was performed. In all, 137 components (comprising ca. 98.0% of the total isolate) were positively identified. Acids, alcohols, and aldehydes represent 91.4% of the total of the identified compounds in raw carob. The levels of these compounds decreased steeply during the roasting process, representing 51.2% of the total as a result of the increasing amounts of furans, esters, and pyrroles in roasted carob, whose odor is more pleasant than that of raw carob and reminiscent of chocolate or cocoa and which could be used in foods as a flavoring agent.

Keywords: Carob; *Ceratonia siliqua* L.; roasting process; volatiles

INTRODUCTION

Carob is a fruit from a dark evergreen tree (*Ceratonia siliqua* L.) that belongs to the family Leguminosae and which originated in the Middle East and later spread to the Mediterranean and other southern European countries (Albanell *et al.*, 1993). It grows uncultivated in many areas where it has traditionally been fed to livestock, and, in times of scarcity, it has been used for human consumption (Harvey, 1984).

The fruit, harvested in late summer or early autumn, forms a flat leathery pod that contains 5–15 hard brown seeds surrounded by edible pulp. The pod consists of about 90% pulp, 8% kernels (seeds), and 2% other materials (Carlson, 1980). Carob beans have very high sugar and low protein contents, whereas the seeds contain little sugar and relatively more protein (Bravo *et al.*, 1994; Albanell *et al.*, 1993; Saura-Calixto, 1987; Saura-Calixto and Canella, 1982). The chemical composition of carob pods is very variable, depending on the variety, the stage of maturity, area, climate, and cultural techniques (Binder *et al.*, 1959; Davies *et al.*, 1971).

Due to the valuable thickening and gelling properties of the locust bean gum from seeds, they contribute to more than 60% of the pod market price (Canellas *et al.*, 1989). Nevertheless, it is important to develop new and more attractive uses for the remaining 90% of the carob pods used until now as a feedstuff (Lanzani, 1982).

The present work has focused on the analysis of the volatile fraction of carob bean pulp, after a roasting process of 10–60 min, in order to find out whether the new generated aroma compounds could be used in foods as flavoring agents.

MATERIALS AND METHODS

Materials. Carob pods (*C. siliqua* L.) for the present study were obtained in different locations from southeastern Spain and provided to us by Deprovesa Company, Carcaixent, Valencia (Spain). The samples used in all analyses were mixtures of pods of the principal commercially available carob varieties, which are cultivated in Castellón (cultivar varieties Costilla de asno, Negra and Rojal), Valencia (cv. Melar,

Matalafera, and Banya de cabra), and on the island of Mallorca (cv. Negra and Durayó).

Isolation of Volatile Compounds. Raw and roasted volatiles were isolated by well-established procedures (Cronin, 1982; Parliment, 1986). The distillation apparatus were provided by Wild GmbH, Heidelberg, Germany. All reagents were analytical grade from Protochem GmbH, Wesel, Germany (pentane, dichloromethane); Aldrich Chemie, Steinheim, Germany (ethyl acetate), and Bundesmonopolverwaltung, Offenbach, Germany (ethanol). For the isolation and fractionation of roasted carob volatiles, raw carob pods were roasted in a roasting apparatus (MVS GmbH, Garching, Germany) at 120 °C for 10–60 min. The analyses were carried out on dry carob pod pulp homogenized and ground (Moulinette, Moulinex) to pass through a 0.5 mm sieve. A mixture of 1 kg of carob pods and 1.7 L of water was stirred and distilled at 40 °C under a pressure of 5×10^3 Pa. From 1 kg of carob pods was obtained 110–120 mL of extract.

The distillate was pressed through a Chromabond HR-P cartridge (polystyrol–divinylbenzol, 40–120 μ m particle size) (Fa. Macherey-Nagel, Düren, Germany) for adsorption and eluted with a pentane–dichloromethane (2:1) solvent system to perform the extraction. The extract was concentrated to 100–200 μ L of a residual colorless liquid on a water bath at 40 °C using a Vigreux column.

Gas Chromatography–Mass Spectrometry (GC–MS). The analyses were performed on concentrated extracts of raw and roasted carob and carried out using a GC Hewlett-Packard 5890 Series II coupled with a MSD-HP 5985. 1 μ L of aroma extract was injected onto a Carbowax 20 M capillary column (50 m \times 0.25 mm i.d., film thickness 0.4 μ m) (CS-Chromatographie Service GmbH, Langerwehe, Germany) using a split injector. The column inlet pressure was 10^5 Pa, and hydrogen was used as a carrier gas (split 1:30). The oven temperature was programmed as follows: the first 5 min at 50 °C, then from 50 to 110 °C at 3 °C/min, followed by 110 to 230 °C at 5 °C/min, and finally remaining at 230 °C for 40 min; the detector temperature was 230 °C. The conditions of the programmable temperature vaporizer (PTV) injector (Fa. Gerstel) were as follows: from 40 to 250 °C at 10 °C/s. A mass spectrometer was used as a detector. The MS parameters were as follows: temperature of ion source, 200 °C; mass interval, 40–350 m/e^- ; ionization energy, 70 eV.

The carob aroma compounds were identified using Kovats indices and mass spectra (comparison with reference data) (Kovats, 1965; MS Data Centre, 1974; Stenhagen *et al.*, 1974; Braw *et al.*, 1979; Heller and Milne, 1978).

Detection and Identification of γ -Lactones in Carob. GC and MS operating conditions were as follows: the GC was carried out with a 5890 HP Series II instrument equipped with a flame-ionization detector and a Carbowax 20 M fused-silica capillary column (CS-Chromatographie Service GmbH, Langerwehe,

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Table 1. Aroma Compounds Identified by GC-MS in the Extract Obtained by High-Vacuum Distillation in Raw and Roasted Carob Pods^a

no.	compounds	raw carob		carob roasted for <i>n</i> min															
				10 min	20 min	30 min	40 min	50 min	60 min										
acids																			
1	2-methylbutyric acid	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
2	4-methylpentanoic acid	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
3	butanoic acid	+	9.8	+	7.9	+	5.1	+	3.6	+	2.4	+	2.2	+	1.8	+	1.8	+	1.8
4	2-methylbutanoic acid	+	14.1	+	11.2	+	8.8	+	7.4	+	6.8	+	6.5	+	6.4	+	6.4	+	6.4
5	acetic acid	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
6	propionic acid	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06
7	<i>n</i> -butyric acid	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3
8	3-methyl-2-butenic acid	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
9	methylpropanoic acid	+	27.1	+	43.4	+	54.3	+	51.7	+	44.4	+	37.0	+	30.0	+	30.0	+	30.0
10	hexanoic acid	+	19.9	+	16.6	+	13.2	+	10.7	+	9.5	+	8.4	+	7.2	+	7.2	+	7.2
11	<i>n</i> -valeric acid	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
12	2-hexenoic acid	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
13	heptanoic acid	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
14	octanoic acid	+	0.7	+	0.7	+	0.7	+	0.7	+	0.7	+	0.7	+	0.7	+	0.7	+	0.7
15	7-octenoic acid	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
16	nonanoic acid	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03
17	decanoic acid	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
18	dodecanoic acid	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05
19	hexadecanoic acid	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01
alcohols																			
20	2-pentanol	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
21	isobutyl alcohol	+	4.2	+	0.5	+	0.3	+	0.3	+	0.2	+	0.02	+	0.01	+	0.01	+	0.01
22	<i>n</i> -butyl alcohol	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
23	2-butanol	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
24	isoamyl alcohol	+	6.6	+	0.9	+	0.1	+	0.03	+	0.02	+	0.01	+	<0.01	+	<0.01	+	<0.01
25	1-penten-3-ol	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01
26	2-methyl-1-propanol	+	2.1	+	1.7	+	1.5	+	1.3	+	0.9	+	0.5	+	0.3	+	0.3	+	0.3
27	<i>n</i> -amyl alcohol	+	0.9	+	0.9	+	0.9	+	0.9	+	0.9	+	0.9	+	0.9	+	0.9	+	0.9
28	2-heptanol	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
29	<i>cis</i> -4-hexenol	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
30	<i>n</i> -hexanol	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06	+	0.06
31	2-octanol	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08
32	1-octen-3-ol	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
33	linalool	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
34	1-heptanol	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
35	1-octanol	+	0.4	+	0.07	+	0.03	+	0.02	+	0.01	+	0.01	+	<0.01	+	<0.01	+	<0.01
36	4-terpineol	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
37	α -terpineol	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01
38	1-nonanol	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
39	benzyl alcohol	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
40	phenethyl alcohol	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
41	cumin alcohol	+	0.03	+	0.02	+	0.02	+	0.01	+	0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
42	6-nonen-1-ol	+	0.5	+	0.3	+	0.2	+	0.1	+	0.08	+	0.06	+	0.04	+	0.04	+	0.04
43	2-decanol	+	0.06	+	0.02	+	0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
44	6,6-dimethyl-2-hydroxymethylbicyclo-[3.1.1]hept-2-ene ^b	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
aldehydes																			
45	myrtenal	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02	+	0.02
46	2-butenal	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
47	2-methyl-2-butenal	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03
48	tiglic aldehyde	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03
49	benzaldehyde	+	1.7	+	1.7	+	1.7	+	1.7	+	1.7	+	1.7	+	1.7	+	1.7	+	1.7
50	3-methoxybenzaldehyde	+	0.2	+	0.1	+	0.08	+	0.07	+	0.05	+	0.03	+	0.02	+	0.02	+	0.02
51	neral	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08	+	0.08
52	geranial	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05	+	0.05
53	2-hydroxy-6-methylbenzaldehyde	+	0.2	+	0.1	+	0.08	+	0.07	+	0.05	+	0.03	+	0.03	+	0.03	+	0.03
54	phenylacetaldehyde	+	0.6	+	0.4	+	0.2	+	0.1	+	0.08	+	0.03	+	<0.01	+	<0.01	+	<0.01
55	vanillin	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
hydrocarbons																			
56	camphene	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01	+	0.01
esters																			
57	butanoic acid, methyl ester	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
58	ethyl-2-hydroxyvalerate	+	<0.01	+	–	+	–	+	–	+	–	+	–	+	–	+	–	+	–
59	butanoic acid, ethyl ester	+	0.1	+	0.4	+	0.8	+	1.8	+	2.3	+	4.0	+	5.5	+	5.5	+	5.5
60	butanoic acid, ethenyl ester	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04	+	0.04
61	2-methylpropanoic acid, 2-methylpropyl ester ^b	+	<0.01	+	0.06	+	0.1	+	0.2	+	0.3	+	0.3	+	0.3	+	0.3	+	0.3
62	isoamyl isobutyrate	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
63	methyl 2-methylbutyrate	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
64	ethyl (iso)butyrate	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1	+	0.1
65	ethyl 2-methylbutyrate	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
66	2-methylpropanoic acid, 3-methylbutyl ester ^b	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
67	methyl cinnamate	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01	+	<0.01
68	phenethyl butyrate	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03	+	0.03
69	phenylethyl acetate	+	<0.01	+	0.01	+	0.01	+	0.02	+	0.03	+	0.04	+	0.05	+	0.05	+	0.05
70	ethyl benzoate	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2	+	0.2
71	benzyl isobutyrate	+	0.03	+	0.03	+													

Table 1. (continued)

no.	compounds	raw carob	carob roasted for <i>n</i> min					
			10 min	20 min	30 min	40 min	50 min	60 min
ketones								
77	2-pentanone	+ <0.01	+ 0.01	+ 0.02	+ 0.08	+ 0.2	+ 0.3	+ 0.4
78	2-methyl-3-pentanone	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2
79	2,3-butanedione	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
80	3-penten-2-one	+ <0.01	+ 0.02	+ 0.04	+ 0.06	+ 0.09	+ 0.1	+ 0.1
81	2,4-dimethyl-3-hexanone	-	+ <0.01	+ 0.01	+ 0.03	+ 0.06	+ 0.07	+ 0.09
82	2-heptanone	+ <0.01	+ 0.3	+ 0.5	+ 0.6	+ 1.0	+ 1.3	+ 1.9
83	3-hydroxy-2-butanone	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02
84	2,4-pentanedione	+ 0.4	+ 0.4	+ 0.4	+ 0.4	+ 0.4	+ 0.4	+ 0.5
85	3-methyl-2-butanone	-	+ <0.01	+ <0.01	+ 0.01	+ 0.02	+ 0.06	+ 0.1
86	acetoin	+ 0.08	-	-	-	-	-	-
87	3-hepten-2-one	+ <0.01	+ 0.01	+ 0.01	+ 0.04	+ 0.05	+ 0.07	+ 0.08
88	6-methyl-5-hepten-2-one	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
89	3-octen-2-one	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
90	2-methyl-5-(1-methylethenyl)-2-cyclohexen-1-one ^b	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04
91	2-octen-4-one	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2	+ 0.2
92	6-methyl-3,5-heptadien-2-one	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04
93	2-nonanone	+ 0.8	+ 0.6	+ 0.3	+ 0.2	+ 0.09	+ 0.03	+ 0.02
94	3-nonen-2-one	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08
95	2-nonen-4-one	+ 0.09	+ 0.09	+ 0.09	+ 0.09	+ 0.09	+ 0.09	+ 0.09
96	2,4-dimethyl-1,3-cyclopentanedione	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
97	acetophenone	+ 0.1	-	-	-	-	-	-
98	2-undecanone	+ 0.06	-	-	-	-	-	-
99	ketoisophorone	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01
100	methyl corylone ^b	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
101	1-phenylethanone	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04
102	1-phenyl-1-butanone	-	+ <0.01	+ 0.01	+ 0.02	+ 0.03	+ 0.04	+ 0.05
103	methylcyclopentenolone	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02
104	3-cyclohepten-1-one	+ <0.01	+ <0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.02	+ 0.03
105	D-verbenone	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01
lactones								
106	γ -valerolactone	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01
107	γ -butyrolactone	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
108	γ -hexalactone	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05
109	γ -octalactone	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
110	γ -decalactone	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
furans								
111	α,α -2-5-ethenyltetrahydro-2-furanmethanol ^b	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
112	furfural	+ 1.4	+ 2.3	+ 3.5	+ 7.1	+ 13.2	+ 19.2	+ 26.2
113	dihydro-2-methyl-3-(2 <i>H</i>)-furanone	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05
114	2-acetylfuran	+ 0.01	+ 0.03	+ 0.07	+ 0.3	+ 0.5	+ 0.6	+ 1.2
115	α -methylfurfural	+ 0.1	+ 0.2	+ 0.3	+ 0.7	+ 1.1	+ 1.6	+ 2.7
116	furfuryl alcohol	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08	+ 0.08
117	2-methyltetrahydrofuran-3-one	+ <0.01	+ 0.01	+ 0.04	+ 0.08	+ 0.1	+ 0.3	+ 0.8
118	hydroxymethylfurfural	-	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
phenols								
119	<i>p</i> -allylphenol (chavicol)	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
120	guaiacol	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02
121	phenol	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
122	2-methoxy-4-[1(2)-propenyl]phenol ^b	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04	+ 0.04
123	3(4)-methyl phenol	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01
124	<i>p</i> -vinylguaiacol	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02	+ 0.02
pyrroles								
125	1-ethyl-1 <i>H</i> -pyrrole-2-carboxaldehyde ^b	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05	+ 0.05
126	2-acetylpyrrole	+ 0.1	+ 0.1	+ 0.2	+ 0.3	+ 0.6	+ 0.9	+ 1.1
127	1 <i>H</i> -pyrrole-2-carboxaldehyde	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03	+ 0.03
pyridines								
128	2-acetylpyridine	<i>c</i>	<i>c</i>	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
pyrazines								
129	2,6-dimethylpyrazine	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
130	2-ethylpyrazine	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01	+ <0.01
131	2-ethyl-3,5-dimethylpyrazine	+ 0.2	+ 0.09	+ 0.07	+ 0.05	+ 0.03	+ 0.02	+ 0.01
132	2-acetyl-3-ethylpyrazine	<i>c</i>	<i>c</i>	<i>c</i>	+ <0.01	+ <0.01	+ <0.01	+ <0.01
thiazoles								
133	benzothiazole	+ <0.01	+ 0.01	+ 0.02	+ 0.04	+ 0.05	+ 0.06	+ 0.08
sulfur compounds								
134	dimethyl disulfide	<i>c</i>	<i>c</i>	<i>c</i>	+ 0.02	+ 0.05	+ 0.07	+ 1.1
135	dimethyl trisulfide	<i>c</i>	<i>c</i>	<i>c</i>	+ 0.01	+ 0.03	+ 0.1	+ 0.4
other nitrogen compounds								
136	4(3 <i>H</i>)-pyrimidone ^b	+ 0.01	+ 0.02	+ 0.04	+ 0.06	+ 0.08	+ 0.1	+ 0.2
137	3-methyl-1 <i>H</i> -pyrazole ^b	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01	+ 0.01
identified peaks (%)		98.1	96.7	99.2	96.4	93.7	93.3	97.7

^a "+" and "-" indicate presence and absence of aroma compound, respectively. All values are given in % area. ^b Tentatively identified. ^c Not quantifiable.

wehe, Germany) as precolumn (25 m \times 0.25 mm i.d., film thickness 0.25 μ m). The precolumn inlet pressure was 292 \times 10³ Pa, and helium was used as the carrier gas (2.5 mL/min). The oven temperature was programmed as follows: the first 2 min at 80 $^{\circ}$ C, from 80 to 110 $^{\circ}$ C at 5 $^{\circ}$ C/min, and from 110

to 230 $^{\circ}$ C at 8 $^{\circ}$ C/min; the FID detector temperature was 250 $^{\circ}$ C. The conditions of the PTV injector (Fa. Gerstel) were as follows: the first 2 s at 35 $^{\circ}$ C, from 35 to 230 $^{\circ}$ C at 12 $^{\circ}$ C/s.

A FS-Cyclodex- γ -lactone (CS-Chromatographie Service GmbH, Langerwehe, Germany) capillary column (50 m \times 0.25

mm i.d., film thickness 0.4 μm) was used. The column inlet pressure was 271×10^3 Pa, and helium was used as the carrier gas (2.5 mL/min). The oven temperature was programmed with the first 24 min at 140 °C and then from 140 to 180 °C at 1 °C/min. A MSD 5970 Hewlett Packard (Quadrupole) was used as a detector. The MS parameters were as follows: temperature of ion source, 200 °C; mass interval, 40–350 m/e^- ; ionization energy, 70 eV.

RESULTS AND DISCUSSION

A survey of the literature shows only two previous reports of certain volatile components derived from carob (Stubbs *et al.*, 1985; MacLeod and Forcen, 1992). In the present research, the main contributors to the differences in the odors of raw and roasted carob are shown in Table 1, in which carob aroma constituents are listed according to their chemical structure.

The yields and appearance of the products were greatly affected by the method of isolation employed. From the comparison of the high-vacuum (HV) extract with the simultaneous distillation (SD) extract of carob, the SD extract showed thermal effects during the distillation and extraction process; yields were much lower with SD extraction than with HV distillation. This latter was found to give more reproducible results and include higher amounts of isolated compounds than with the other method of extraction. Likewise, Table 1 refers to the aroma compounds isolated only by HV distillation.

In all, 137 compounds (comprising ca. 98.0% of the total isolate) were positively identified and the mass spectra agreed with those in the literature (MS Data Centre, 1974; Stenhagen *et al.*, 1974; Braw *et al.*, 1979; Heller and Milne, 1978).

Unidentified components (ca. 1.5–2%) were present in such low amounts that either no mass spectrum could be recorded or the spectrum was too poor to be interpreted.

Of the fully identified components derived from raw carob beans, aliphatic acids represented an extraordinarily high level (72.6%) of the isolate, the major contributors being methylpropanoic acid (No. 9) (27.1%), hexanoic acid (No. 10) (19.9%), 2-methylbutanoic acid (No. 4) (14.1%), and butanoic acid (No. 3) (9.8%). In addition, traces of acetic acid (No. 5) (0.2%) and propionic acid (No. 6) (0.06%) were detected. The identified acids originate largely from the high sugar content of carob pulp (37–52%) with a lesser contribution from the lower content of lipids present (0.5–1.0%). These results agree, at least in part, with the data published by MacLeod and Forcen (1992) and Stubbs *et al.* (1985). The high concentrations of these acids, coupled with their known odor qualities, are largely responsible for the rancid/sweaty quality that is characteristic of the carob pod and its aroma extract.

Among alcohols which represented 15.9% in raw carob, isoamyl alcohol (no. 24) (6.6%), isobutyl alcohol (no. 21) (4.2%), and 2-methyl-1-propyl alcohol (no. 26) (2.1%) are the major contributors, whereas in the case of aldehydes (2.9% in total), benzaldehyde (no. 49) plays the main role (1.7%) in raw carob.

These three groups (acids, alcohols, and aldehydes) represent 91.4% of the total of the identified compounds in raw carob. Esters and ketones represent 2.3% each, with smaller representation from furans (1.6%), whereas lactones, phenols, pyrroles, pyrazines, thiazoles, sulfur compounds, and other nitrogen components represent less than 0.7% of the total (Figure 1A).

Most of the aliphatic alcohols and aliphatic ketones identified originated from sugars and/or lipids; 6-meth-

yl-5-hepten-2-one (no. 88) and 6-methyl-3,5-heptadien-2-one (no. 92) derive from carotenoid precursors (MacLeod and Forcen, 1992).

On the other hand, the majority of the volatiles identified in roasted carob as well as the fact that carob pods contain very high carbohydrates and low lipid and protein contents (Mitrakos, 1968; Artik and Erbas, 1988) suggest the possibility of flavor formation via the oxidation of flavor precursors by enzymatically and non-enzymatically formed oxidizing agents. Other thermal reactions such as the pyrolytic degradation of the main components of foods including sugars, amino acids, dipeptides, vitamins, as well as the oxidative degradation of lipids, contribute not only to the formation of heterocyclic compounds responsible for the flavor of foodstuffs but also to the reduction in the amounts of aldehydes (Hodge *et al.*, 1972; Vernin and Párkányi, 1982).

The levels of the main compounds in raw carob decreased steeply during the roasting process, representing 51.2% of the total as a result of the increase in esters, ketones, pyrroles, pyrans, thiazoles, and sulfur compounds in roasted carob (Figure 1B).

In the case of acids, these are reduced from 72.6% in raw carob to 47.1% after 60 min of roasting: methylpropanoic acid (no. 9) increases from 27.1% in raw carob to 54.3% after 20 min of roasting, and decreases from 30 min onward until reaches 30.0% after 60 min of roasting, perhaps as a result of its reactivity with esters (MacLeod and Forcen, 1992); 2-methylbutanoic acid (no. 4) decreases from 14.1% in raw carob to 6.4% after 60 min of roasting; and butanoic acid (no. 3) decreases from 9.8% in raw carob to 1.8% in carob roasted for 60 min.

The main components of the monoterpenes identified were D-verbenone (no. 105), myrtenal (no. 45), and linalool (no. 33). Another important group of roast aroma compounds are pyridines. They can be formed via a cyclization of suitable amino sugar derivatives. Some pyridines like 2-acetylpyridine (no. 129) seem to be found generally when amino acids and sugars are treated under the conditions of Maillard reaction.

In the case of alcohols, they decrease from 15.9% in raw carob to 2.3% after 60 min of roasting: isobutyl alcohol (no. 21) and isoamyl alcohol (no. 24) decrease from 4.2% and 6.6%, respectively, in raw carob to 0.01% in the case of isobutyl alcohol and to <0.01% isoamyl alcohol after 60 min of roasting. Aldehydes also decrease from 2.9% in raw carob to 2% in carob roasted for 60 min.

Phenols and lactones do not change concentration along the roasting process. Also according to the chiro-specific analysis, the mean enantiomeric ratios [(*R*)-(+)]%:[(S)-(-)]% of the identified lactones γ -hexa (28.9%:71.1%), γ -octa (43.2%:56.8%), and γ -decalactone (42.2%:57.8%) remain constant.

Aldehydes and ketones are precursors of many heterocyclic compounds that contribute to roasted flavors in cooked foods. Heterocyclic compounds increase both in number and concentration when carob pulp was roasted, creating an odor more reminiscent of cocoa or chocolate. Furans are quite important to the palatability of roasted foods as they provide warm, sugary flavor notes. Furans can be the result of the cyclization of the sugar moiety after sugar activation and Amadori rearrangements via interaction with amine compounds (Izzo and Ho, 1993). The relative amounts of furans increase with temperature from 1.6% in raw carob to 31.0% in carob roasted for 60 min. Furfural (no. 112)

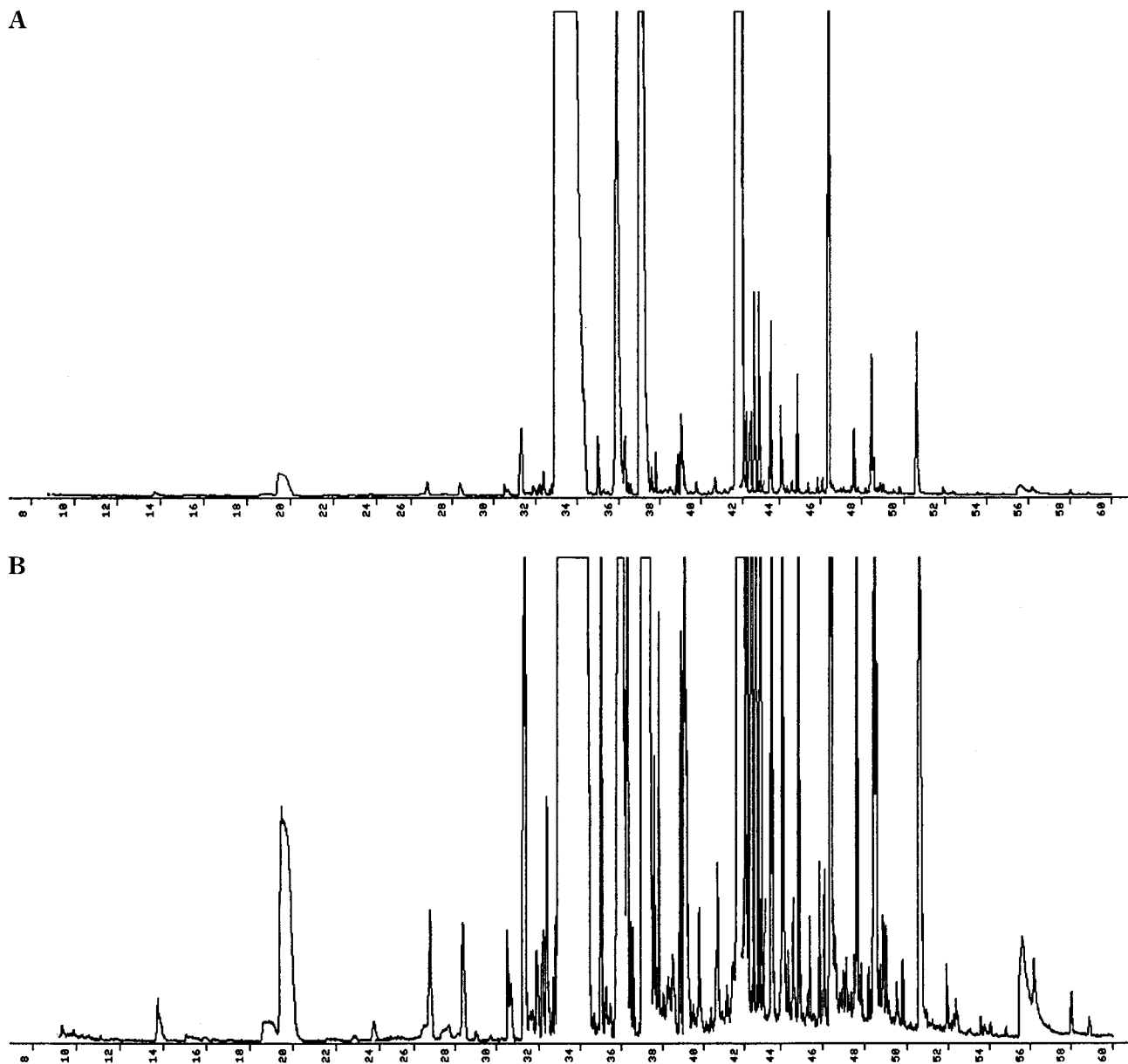


Figure 1. (A) Gas chromatogram of the aroma compounds in the extract obtained by high-vacuum distillation in raw carob. (B) Gas-chromatogram of the aroma compounds in the extract obtained by high-vacuum distillation in 60 min roasted carob.

and α -methylfurfural (no. 115) increase characteristically as a function of time from 2.3 and 0.2%, respectively, at 10 min of roasting to 26.2% in the case of furfural and to 2.7% α -methylfurfural at 60 min of roasting. Furans are formed mainly by thermal degradation of carbohydrates and ascorbic acid and from sugar–amino acids interactions during food processing (Maga, 1981).

Sulfurous notes in the isolate are explained by the presence of dimethyl disulfide (no. 134) and dimethyl trisulfide (no. 135), which have been described to have cabbage, fermented radish, black mushroom-like flavor (Tu and Ho, 1995).

Apart from the furans, esters, ketones, and pyrroles, the roasting process has led to an increase of the amount of esters from 2.3% in raw carob to 7.9% after 60 min, from 2.3 to 4.2% in the case of ketones, and from 0.2 to 1.2% in the case of pyrroles.

Concerning the possible influence of the high content of condensed tannins of carobs (ca. 18.5%) in the formation of volatile compounds, most of the condensed polyphenols of carob remained in the same form after

heat treatment. Only a minor part of them (the free polyphenols) were partially solubilized. The tannins are characterized by their ability to interact with and precipitate proteins. The condensed tannins are the predominant type of tannin in carob pods. Tannins bind to enzymes and other proteins by hydrogen bonding to amide groups to form insoluble complexes. Generally, hydrolyzable tannins are more reactive than condensed tannins, and their oxidized derivatives form covalent bonds with proteins that are resistant to enzymatic and heat treatment.

The low solubility of carob-condensed tannins in organic solvents, their ability to aggregate to complex carbohydrates and proteins, as well as the high concentration of sugars in carob complicate the task of separation and identification. Roasting did not modify the condensed polyphenol content or composition regardless of the time of roasting.

The high content of condensed tannins seems to interfere in the profile of aromas obtained during carob roasting; for instance, the high content of sugar does not appear reflected in the results obtained. Likewise,

the protein of carob pulp is strongly linked with tannins through the hydroxyl groups of the tannins, by means of hydrogen bonds with the active centers of peptidic linkages (Haslam, 1989). Such binding is quite powerful and difficult to reverse. This could be one of the reasons why the binding of flavors with the food matrix affected flavor release in carob. Moreover, the liberation of proteins from condensed tannins did not seem to have occurred during the roasting of carob. As a result, little or no ammonia was liberated and the subsequently enhanced activation of glucose leading to the formation of more Maillard-specific volatile compounds (i.e., pyrazines) did not seem to have taken place. Only few pyrazines (nos. 129–132) were identified, and they could have been originated either directly or indirectly, from the high sugar content of the carobs, and are likely to contribute to the caramel note of the roasted carob.

On the other hand, nonenzymatic browning reactions can lead to the formation of numerous flavor-producing compounds, including pyrroles (Hodge *et al.*, 1972). Likewise, it would appear that the main contribution of lipid–Maillard interactions to the aroma of roasted carob is a modification of the balance of aroma compounds, reducing the amounts of certain potent compounds like aliphatic acids in raw carob and increasing the levels of others like furans, esters, and pyrroles in roasted carob, whose odor is more pleasant and reminiscent of chocolate or cocoa (Benk, 1989) and which is particularly suitable for use in a wide range of confectionary products such as biscuits, cakes, and beverages, among others.

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