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Identification of the Volatile Constituents of Raw Pumpkin (*Cucurbita pepo* L.) by Dynamic Headspace Analyses

by

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Summary

The volatile constituents of Raw Pumpkin, (*Cucurbita pepo* L. subsp. *pepo* - Howden Pumpkin cultivar – a Connecticut Field type) were determined by dynamic headspace GC-MS analyses. In excess of 110 volatile constituents were identified. Analysis of the fibrous stringy and/or seedy portions gave primarily (Z)-3-hexenol and 1-hexanol while the “meat or flesh” portion was dominated by 2-propanol and dimethyl disulfide. Significant quantities of dimethyl disulfide and dimethyl trisulfide were found in all samples.

Background:

The genus Cucurbita is a member of the Cucurbitaceae plant family which includes squash (including pumpkin), zucchini, and some gourds. These were first cultivated in the Andes and Mesoamerica. The Cucurbita genus includes five major species which comprise the majority of pumpkins and squashes - *Cucurbita pepo, Cucurbita moschata, Cucurbita maxima, Cucurbita argyrosperma* and *Cucurbita ficifolia*. Many hundreds of varieties and cultivars of these are grown around the world and are major agricultural commodities.

In 2012, The Food and Agriculture Organization of the United Nations (FAOSTAT) estimates that 24,616 million tons of pumpkins, squash and gourds were produced, with the following countries being the major producers: China (7,063 mil. tons), India (4,900 mil. tons), Russian Federation (1,081 mil. tons), Iran (965 mil. tons) and the USA (901 mil. tons).

Pumpkins***** are grown primarily for processing with a lesser percentage being grown for festivals (e.g. Halloween) or ornamental sales. Uses for foods include everything from pumpkin pie to pumpkin bread and even curries. Roasted pumpkin seeds are used for snacks and the pressed oil of roasted seeds is popular for salad dressings, especially in Europe.

Previous work:

No analyses of the volatile constituents of raw pumpkin (*Cucurbita Pepo* L.) has been previously published other than the work of Bowman et al. in 2012 on raw dried pumpkin seed varieties (1) and from the specific search and isolation of 2-sec-butyl-3-methoxypyrazine in various raw vegetables by Murray & Whitfield in 1975 (2).

In 1981, Parliment and coworkers (3) reported on the volatiles of freshly boiled pumpkin (*Cucurbita pepo* L. - Connecticut Field cultivar) and compared these to the volatiles of commercially canned pumpkins (presumably that from the Dickinson pumpkin cultivar of *Cucurbita moschata*).

***Note - The term “pumpkins” actually has no botanical meaning, as they are all varieties of winter squash.**

Parliment et al. found relatively high amounts of (Z)-3-hexenol and 1-hexanol (in freshly cooked pumpkin) similar to that reported in several of our analyses herein, while Anderson (4) also found high amounts of these two materials in the steam distillate of *Cucurbita maxima Duchesne* cultivar 'Blue Hubbard' flower petals.

Roasted pumpkin seeds (and the expressed oil thereof) have become increasingly popular in recent years as the seeds & oil have a pleasant nutty flavor and are considered to have a number of nutraceutical benefits as the seed oil is rich in antioxidants and polyunsaturated fatty acids (5-9). In particular, *Cucurbita pepo* subsp. *pepo* var. Styriaka is valued in Europe. Several reports have appeared on the volatiles of roasted pumpkin seeds (10-14).

Experimental:

Plant Name: *Cucurbita pepo* L. subsp. *pepo* (Howden Pumpkin cultivar – a Connecticut Field type)
Source: Commercial Halloween pumpkin; purchased at Walmart in Louisville, KY, September 2014
Plant Part: See experimental

Sample Preparation: Four analyses of raw pumpkin samples were conducted as described below.

SAMPLE-1.D: A 4-5 gram sample of "raw" pumpkin (consisting of the flesh, along with seeds and the fibrous stringy portion) cut from the mesocarp portion was placed in 16 fl. Oz. glass jar fitted with a metal lid having both inlet and outlet gas tubes. The inlet tube was connected to a helium tank and the outlet tube connected to a stainless tube with Swagelok nuts & ferrules attached. The top of the tube was fitted with a 1/4'" Swagelok union with the top nut hole diameter increased to allow the easy introduction of a Agilent injection port liner (Agilent part 5181-3316) and sealed with an injection port O-Ring. The injection port liner had been packed with 100 mg of Tenax TA (20-35 mesh) (glass-wool plugs on top and bottom) and previously baked out at 260C for 2.5 hours. Helium (30 cc/min) was introduced into the bottom of the glass jar containing the pumpkin sample. The sample was purged for 100 minutes trapping the volatiles on the Tenax injection port liner. The Tenax liner was then placed directly into the GC injection port for thermal desorption of the volatiles onto the GC column.

SAMPLE-2.D: In a manner similar to that described for sample 1.D, a 4-5 gram sample of "raw" pumpkin (consisting of seeds and the fibrous stringy portion, but with minimal flesh) was purged for 260 minutes.

SAMPLE-3.D: In a manner similar to that described for sample 1.D, a 4-5 gram sample of "raw" pumpkin (consisting of only the flesh portion) was purged for 120 minutes.

SAMPLE-4.D: In a manner similar to that described for sample 1.D, a 4-5 gram sample of "raw" pumpkin (consisting primarily of the fibrous stringy portion) was purged for 300 minutes.

The four samples from the same pumpkin were successively purged on the same day.
GC and GC/MS: The GC-MS was an Agilent 7890A/5975C High Performance combination. An Agilent 60m X 0.32mm I. D. fused silica column coated with a 0.25 micron film thickness of HP-5MS (HP part No. 19091S-416M), a DB-5 equivalent column, was used in all analyses. The column was held isothermally at 30^{o}C for 1.5 minutes, then programmed from 30^{o}C to 260^{o}C at 2^{o}C/min, with a final hold time of 28.5 minutes to give a total analysis time of 145

minutes. The Injection port was held at 260°C. Helium Carrier Gas was used with a flow rate of 3.403 ml/min. Split ratio of 15:1.The Mass spectrometer was scanned in the EI mode from 26m/z to 350m/z using 70eV ionizing voltage. Additionally, FID chromatograms were obtained and a PFPD detector was employed for sulfur compound detection. Percentages are TIC percentages, corrected for known experimental artifacts, but without correction for response factors. Where overlapping component peaks were present, the NIST AMDIS program was utilized to estimate peak percentages, when possible.

Analysis was done on both the HP MS Enhanced Chemstation program Version D.03.00.611 and Version E.02.02.1431 employing both normal and selective ion modes. The NIST AMDIS deconvolution program (Version 2.71) was also employed in both normal and high resolution modes using the MSP file format from data imported from the Wiley and NIST MS libraries as well as the authors libraries.

Component Identification: Identifications were based on mass spectra from the Wiley 6 and NIST 05 MS libraries as well as from the authors MS library. Standard classic (isothermal) Kovats Indices (KI) based on n-Alkanes (15) were calculated using the formula:

$$I_x = 100n + 100[\log(t_x) - \log(t_n)] / [\log(t_{n+1}) - \log(t_n)]$$

Additionally Linear Retention Indices (LRI), sometimes referred to as the Arithmetic Index (AI), based on n-alkanes using the methodology of Van den Dool and Kratz (16) were calculated using the formula:

$$LRI_x = 100n + 100(t_x - t_n) / (t_{n+1} - t_n)$$

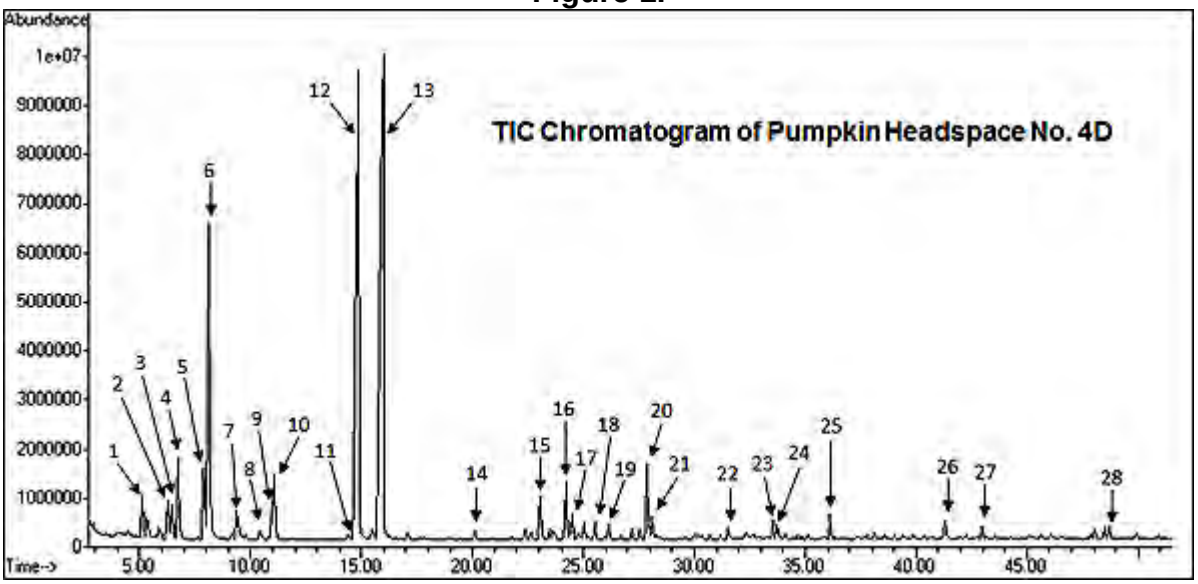
Where available, retention time comparisons were used employing primarily the Retention Indices compilations of the NIST Mass Spec Data Center (17), as well as those of Boelens (18) and Adams (19). Both the calculated Kovats KI and the LRI retention indices are provided in our tables because some of the reported literature does not specify which RI calculations were actually employed. The NIST and Adams values used for comparison were both the van den Dool and Kratz temperature programmed LRI values and the Kovats (KI) values. The Boelens values in the ESO 2000 (Update 2006) database that were derived from the literature do not specify the method of RI calculation. In most cases the RI values by both methods are quite similar, but can vary by 1-9 units. As with all such reported retention index values, those below 500-700 KI/LRI must be considered as somewhat approximate as such values are subject to greater instrumental experimental fluctuations.

Results:

A series of purge and trap headspace analyses of Raw Pumpkin were conducted using different portions cut from the inner cavity for sample collection. Figure 1 illustrates the GC profile of sample 4.D.

Table 1 presents the major constituents present in the four headspace samples analyzed while Table 2 provides the detailed analysis of sample 4.D with both Kovats retention indices and the Linear retention Indices (LRI).

Figure 1.



1. 3-Methylbutanal; **2.** 3-Pentanone; **3.** 3-Pentanol; **4.** 3-Hydroxy-2-butanone; **5.** 3-Methylbutanol; **6.** Dimethyl disulfide; **7.** 1-Pentanol; **8.** 2,3-Butanediol; **9.** (Z)-3-Hexenal; **10.** Hexanal; **11.** (E)-3-Hexenol; **12.** (Z)-3-Hexenol; **13.** 1-Hexanol; **14.** α-Pinene; **15.** Dimethyl trisulfide; **16.** 1-Octen-3-ol; **17.** 3-Octanone; **18.** Decane; **19.** δ-3-Carene; **20.** Limonene; **21.** 2-Ethyl-1-hexanol + 1,8-Cineole; **22.** 1-Octanol + unknown; **23.** Undecane; **24.** Linalool; **25.** Artifact; **26.** Dodecane; **27.** β-Cyclocitral; **28.** Tridecane

Table 1. – Major Raw Pumpkin Constituents

SAMPLE No.	1D	2D	3D	4D	Avg.
Compound	%	%	%	%	%
2-Propanol	9.09	0.27	26.46	0.44	9.06
3-Methylbutanal	ND	0.12	ND	1.00	0.28
3-Pentanone	2.54	1.97	ND	1.15	1.41
3-Pentanol	1.01	0.90	1.06	1.02	1.00
3-Hydroxy-2-butanone	5.60	1.65	<0.01	2.49	2.44
3-Methylbutanol	0.12	0.43	ND	1.53	0.52
Dimethyl disulfide	3.56	24.13	13.85	10.85	13.10
(Z)-3-Hexenal	1.73	1.01	ND	0.62	0.84
Hexanal	2.62	1.70	0.42	1.99	1.68
(Z)-3-hexenol	21.51	22.45	1.15	20.76	16.47
1-Hexanol	8.79	12.95	0.80	23.82	11.59
α-Pinene	3.01	0.38	2.97	0.33	1.68
Dimethyl trisulfide	2.25	2.29	1.86	1.35	1.94
1-Octen-3-ol	0.40	0.16	0.21	1.34	0.53
β-Pinene	0.67	0.16	0.64	0.40	0.47
3-Octanone	<0.01	0.33	<0.01	0.76	0.27
3-Octanol	<0.01	0.03	<0.01	0.22	0.06
Limonene	7.46	1.70	6.75	1.88	4.45
2-Ethyl-1-hexanol	2.33	0.50	2.49	0.33	1.41
Linalool	0.98	0.33	0.80	0.52	0.66
Nonanal	0.85	0.13	0.82	0.20	0.50
Decanal	0.50	0.01	0.85	0.13	0.37
β-Cyclocitral	<0.01	0.08	ND	0.35	0.11
β-Ionone	<0.01	<0.01	<0.01	0.14	0.04
% of Total	**75.02**	**73.67**	**61.14**	**73.41**	**70.78**

Table 2. - GC-MS Analysis of Pumpkin Sample 4.D

LRI - KI Lit.	HP-5MS (calc) Kovats RI	HP-5MS (calc) LRI	Compound	Actual R.T.	TIC %	Ident.
381-412	412	412	Acetaldehyde[h,i]	2.451	0.01	MS,RI
423[(25)]-473	425	422	Methanethiol[e,h]	2.624	<0.01	MS,RI
426-489	438	434	Ethanol[d,h]	2.793	0.63	MS,RI
491-503	496	496	2-Propanone[h]	2.726	<0.01	MS,RI
500-515	514	512	2-Propanol	2.910	0.44	MS,RI
505-529	530	526	Dimethyl sulfide[c,d,h]	3.078	<0.01	MS,RI
550-560	561	557	2-Methylpropanal[c,d,e,f,i] + unknown	3.443	0.01	MS,RI
585-613	592	591	Diacetyl[a,b,c,d,e,h]	3.838	0.01	MS,RI
592-606	592	592	2-Butanone[c,d,h]	3.897	<0.01	MS,RI
596-609	599	598	2-Butanol	4.028	0.01	MS,RI
598-626	605	603	2-Methylfuran[c]	4.136	<0.01	MS,RI
605-628	608	605	Ethyl acetate[a,b,d,h]	4.191	0.03	MS,RI
600-663	613	610	Acetic acid[e]	4.299	0.50	MS,RI
614-635	623	618	2-Methylpropanol[h]	4.494	0.01	MS,RI
642-666	651	646	3-Methylbutanal[b,c,d(isomer not identified),e,f,h,i]	5.112	1.00	MS,RI
640-670	661	656	2-Methylbutanal[b,c,e,f,h,i]	5.347	0.50	MS,RI
680-688	682	680	1-Penten-3-ol[c,g,h]	5.900	0.32	MS,RI
678-688	684	683	2-Pentanone[c,d,h]	5.952	<0.01	MS,RI
687-705	703	702	3-Pentanone[a]	6.254	1.15	MS,RI
695-712	705	704	2-Ethylfuran[a]	6.329	0.09	MS,RI
703-712	708	706	3-Pentanol	6.458	1.02	MS,RI
701-720	716	712	3-Hydroxy-2-butanone[d]	6.737	2.49	MS,RI
731-742	738	731	3-Methyl-3-buten-1-ol	7.628	0.08	MS,RI
737-759	743	737	3-Methylbutanol[c]	7.875	1.53	MS,RI
730-763	747	740	2-Methylbutanol[c,d]	8.050	0.70	MS,RI
730-756	749	742	Dimethyl disulfide[h]	8.132	10.85	MS,RI
732-850	752	745	1,2-Propanediol	8.283	0.02	MS,RI
745-759	762	755	(E)-2-Pentenal[a]	8.733	0.05	MS,RI
758-781	774	769	1-Pentanol[a,c,h]	9.385	1.06	MS,RI
763-783	777	772	(Z)-2-Pentenol	9.528	0.04	MS,RI
770-789	781	777	3-Methyl-2-buten-1-ol (Prenol)	9.757	0.17	MS,RI
767-806	793	791	2,3-Butanediol	10.426	0.39	MS,RI
800	800	800	Octane	10.848	0.09	MS,RI
785-806	802	802	(Z)-3-Hexenal	10.962	0.62	MS,RI
795-808	804	803	Hexanal[a,c,d,e,f,g,h]	11.059	1.99	MS,RI
800-828	819	816	Butyl acetate	11.914	0.05	MS,RI
835[(21)]	837	832	2,3-Dimethylbutan-1-ol (TID)	12.988	0.02	MS,RI
830-851	840	834	Furfural[a,c,d,h]	13.167	<0.01	MS,RI
850-864	858	852	(E)-3-Hexenol	14.408	0.25	MS,RI
851-861	860	854	(E)-2-Hexenal[a,c,d,g,h]	14.544	<0.01	MS,RI

LRI - KI Lit.	HP-5MS (calc) Kovats RI	HP-5MS (calc) LRI	Compound	Actual R.T.	TIC %	Ident.
848-870	865	859	(Z)-3-Hexenol[a,b,g]	14.873	20.76	MS,RI
858-888	880	875	1-Hexanol[a,c,d,g]	15.972	23.82	MS,RI
900	900	900	Nonane	17.635	0.10	MS,RI
888-915	903	902	(Z)-4-Heptenal[e,i]	17.821	0.06	MS,RI
899-907	905	905	Heptanal[f,h]	17.997	0.03	MS,RI
NA	916	914	Methylthiocyclopentane (TID)	18.709	0.03	MS
912-924	919	916	Methyl allyl disulfide (TID)	18.934	0.03	MS,RI
915-923	920	917	Anisole	18.999	0.04	MS,RI
928-941	935	931	α-Pinene[e,h]	20.104	0.33	MS,RI
954-963	960	955	6-Methyl-2-heptanone	21.986	0.07	MS,RI
965-975	972	969	Dimethyl trisulfide[b,c,e,h]	23.049	1.35	MS,RI
969-978	975	972	Sabinene	23.289	0.06	MS,RI
967-977	977	974	1-Heptanol	23.464	0.27	MS,RI
973-985	979	976	β-Pinene	23.646	0.40	MS,RI
974-989	986	983	1-Octen-3-ol[a]	24.215	1.34	MS,RI
985[(20)]	988	986	(Z)-6-Octen-2-one	24.387	0.02	MS,RI
984-990	989	987	3-Octanone	24.509	0.76	MS,RI
989-993	991	990	β-Myrcene	24.720	0.02	MS,RI
991-994	992	991	2-Pentylfuran[a,c,d,h]	24.795	0.30	MS,RI
992-997	993	992	2-Octanone[h]	24.866	<0.01	MS,RI
993-1003	997	996	6-Methyl-5-hepten-2-ol	25.214	0.12	MS,RI
1000	1000	1000	Decane	25.510	0.30	MS,RI
993-1004	1001	1001	3-Octanol	25.557	0.20	MS,RI
994-1013	1007	1006	Octanal[e,h]	25.976	0.02	MS,RI
997-1013	1007	1006	α-Phellandrene	25.986	0.09	MS,RI
1005-1020	1009	1008	δ-3-Carene	26.162	0.47	MS,RI
1007-1018	1016	1014	Hexyl acetate	26.680	0.14	MS,RI
1010-1026	1019	1017	α-Terpinene	26.856	0.02	MS,RI
1020-1029	1028	1025	p-Cymene	27.532	0.31	MS,RI
1027-1035	1032	1029	Limonene[h]	27.872	1.88	MS,RI
1026-1039	1034	1031	β-Phellandrene	27.979	0.24	MS,RI
1029-1045	1035	1032	2-Ethyl-1-hexanol	28.105	0.33	MS,RI
1031-1046	1036	1032	1,8-Cineole	28.122	0.24	MS,RI
1035-1040	1039	1036	2,6,6-Trimethylcyclohexanone[f]	28.423	0.16	MS,RI
1024-1051	1044	1040	Benzyl alcohol[c,g]	28.749	0.09	MS,RI
1054-1065	1062	1059	γ-Terpinene	30.265	0.01	MS,RI
1065-1076	1072	1069	Acetophenone	31.038	0.03	MS,RI
1069-1080	1077	1075	1-Octanol + unknown	31.529	0.41	MS,RI
1082-1092	1087	1086	α-Terpinolene	32.416	0.01	MS,RI
NA	1091	1089	Sorbic acid (E,E) (TID)	32.699	0.36	MS
1088-1095	1093	1092	1-Isopropenyl-4-methylbenzene	32.895	0.14	MS,RI
1091-1096	1094	1093	2-Nonanone	32.967	<0.01	MS,RI
1100	1100	1100	Undecane	33.547	0.50	MS,RI
1097-1103	1103	1102	Linalool	33.733	0.52	MS,RI

LRI - KI Lit.	HP-5MS (calc) Kovats RI	HP-5MS (calc) LRI	Compound	Actual R.T.	TIC %	Ident.
1098-1109	1108	1107	Nonanal[c,d,f,g,h]	34.101	0.20	MS,RI
1110-1123	1119	1117	2-Phenylethyl alcohol[a,c,d,e] + C4 benzene	34.874	0.05	MS,RI
1150[(22)]	1146	1143	(E)-5-Ethyl-6-methyl-3-hepten-2-one	36.906	0.08	MS,RI
1160-1172	1168	1166	Benzyl acetate	38.656	0.07	MS,RI
1168-1183	1177	1176	Nonanol	39.418	0.16	MS,RI
1175-1186	1185	1183	Terpinen-4-ol	40.037	0.04	MS,RI
1182-1190	1188	1187	Naphthalene	40.334	0.18	MS,RI
1186-1194	1193	1192	1-Phenylethyl acetate	40.703	0.15	MS,RI
1190-1201	1197	1196	Methyl salicylate	41.046	0.03	MS,RI
1190-1207	1199	1199	α-Terpineol	41.235	0.02	MS,RI
1200	1200	1200	Dodecane	41.322	0.64	MS,RI
1195-1213	1209	1209	Decanal[g]	41.966	0.13	MS,RI
1210-1233	1221	1219	Dimethyl tetrasulfide (TID)	42.756	<0.01	MS,RI
1214-1226	1224	1222	β-Cyclocitral[f,g]	42.978	0.35	MS,RI
1242-1255	1250	1248	Carvone	44.896	0.05	MS,RI
1247-1259	1253	1251	Linalyl acetate	45.086	0.07	MS,RI
1253-1261	1261	1259	2,6,6-Trimethyl-1-cyclohexene-1-acetaldehyde[f]	45.653	0.06	MS,RI
1291-1315	1299	1299	2-Methylnapthalene	48.660	0.01	MS,RI
1300	1300	1300	Tridecane	48.706	0.39	MS,RI
1299-1325	1316	1315	1-Methylnapthalene	49.766	<0.01	MS,RI
1341-1356	1351	1349	α-Terpinyl acetate	52.131	0.01	MS,RI
NA	1390	1390	Methyl 2-methylphenyl disulfide (TID)	54.975	0.11	MS[(23)]
1400	1400	1400	Tetradecane	55.687	0.21	MS,RI
1481-1496	1483	1483	β-Ionone[f,g]	61.129	0.14	MS,RI
1484-1497	1486	1486	5,6-Epoxy-β-ionone	61.333	<0.01	MS,RI
1500	1500	1500	Pentadecane	62.281	0.15	MS,RI
1525-1546	1537	1536	Dihydroactinidiolide	64.528	<0.01	MS,RI
1600	1600	1600	Hexadecane	68.525	0.17	MS,RI
1700	1700	1700	Heptadecane	74.446	0.09	MS,RI
1800	1800	1800	Octadecane	80.082	0.02	MS,RI
1900	1900	1900	Nonadecane	85.431	<0.01	MS,RI
2000	2000	2000	Eicosane	90.544	<0.01	MS,RI
2100	2100	2100	Heneicosane	95.431	<0.01	MS,RI
2200	2200	2200	Docosane	100.041	<0.01	MS,RI
			Total % Identified		86.14	

<0.01% = trace; TID = tentative identification

Compounds previously identified in:

a - Freshly cooked pumpkins [water/steam distilled] (Parliment et al. - Ref 3.)

b - commercial canned pumpkin [water/steam distilled] (Parliment et al. - Ref 3.)

c - roasted pumpkin seeds [headspace-SPME] (Siegmund and Murkovic - Ref 10.)

d - raw & roasted pumpkin seeds [headspace in water] (Bowman and Barringer - Ref 1.)

e - roasted pumpkin seed oil [extraction and separation techniques] (Poehlmann and Schieberle - Ref 12.)

f - heated pumpkin slices [headspace-SPME] (Kebede et al. - Ref 13.)

g - Cucurbita maxima Duchesne [steam distilled blossoms] (Andersen - Ref 4.)

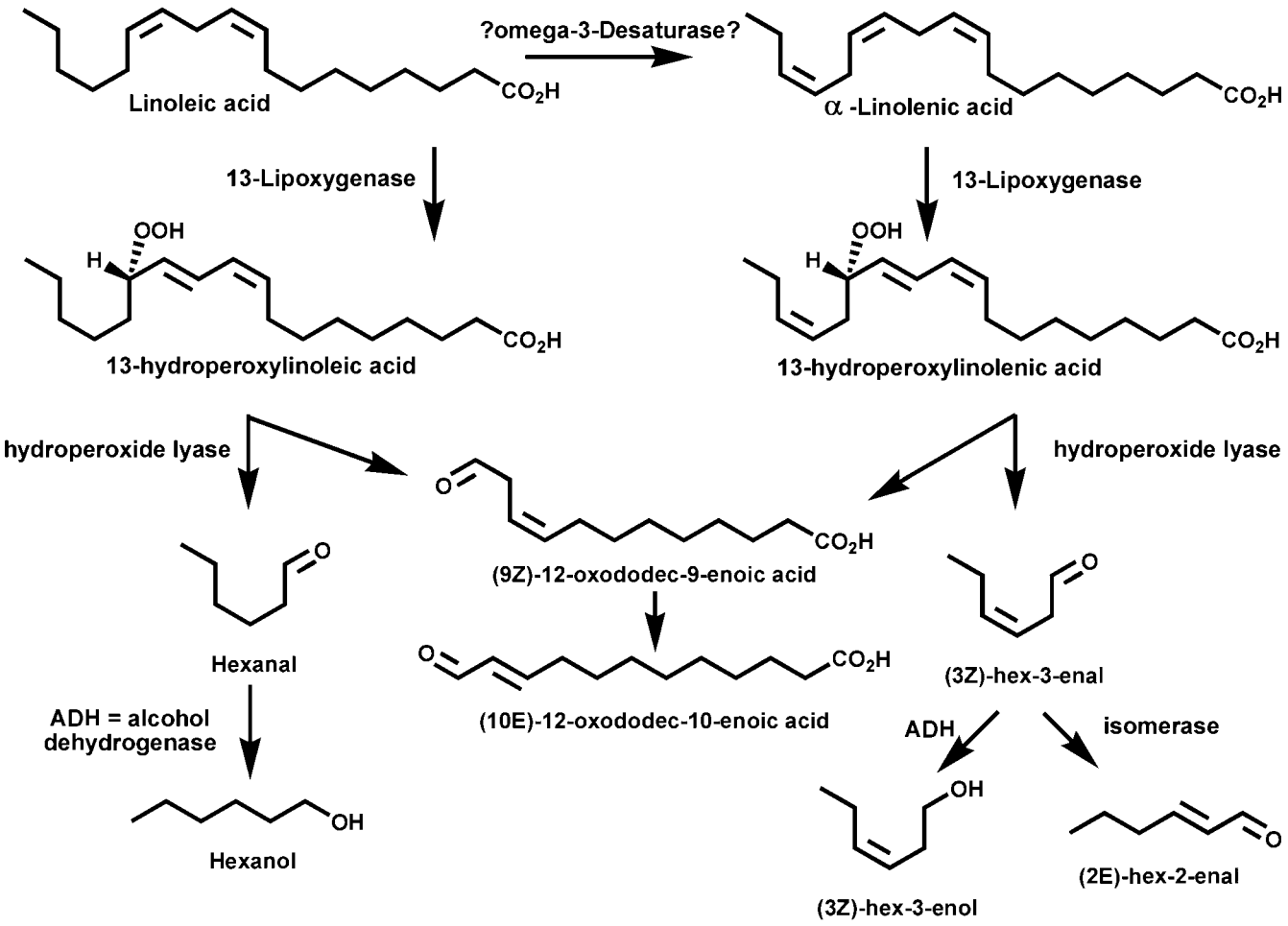
h - roasted pumpkin seed oil [commercial samples] (Procida et al. - Ref 14.)

i - roasted pumpkin seed oil [headspace] (Matsui et al. Ref 11.)

Discussion:

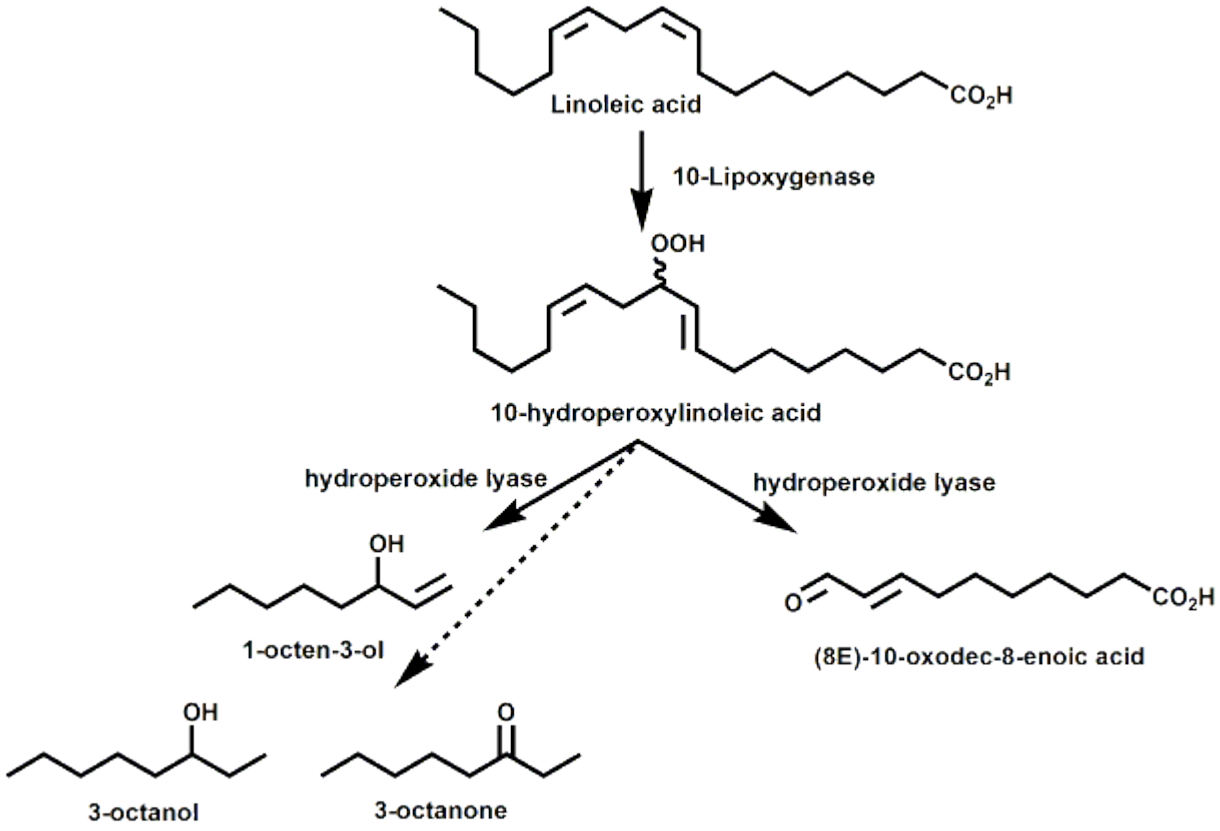
The presence of the green vegetative notes, e.g. (3Z)-hex-3-enal and (3Z)-hex-3-enol, are derived from the 13-lipoxygenase pathway from linolenic acid while the linoleic acid 13-lipoxygenase pathway produces hexanal and hexanol (24-28).

Figure 2. – 13-Lipoxygenase Pathway to Green Aroma Volatiles



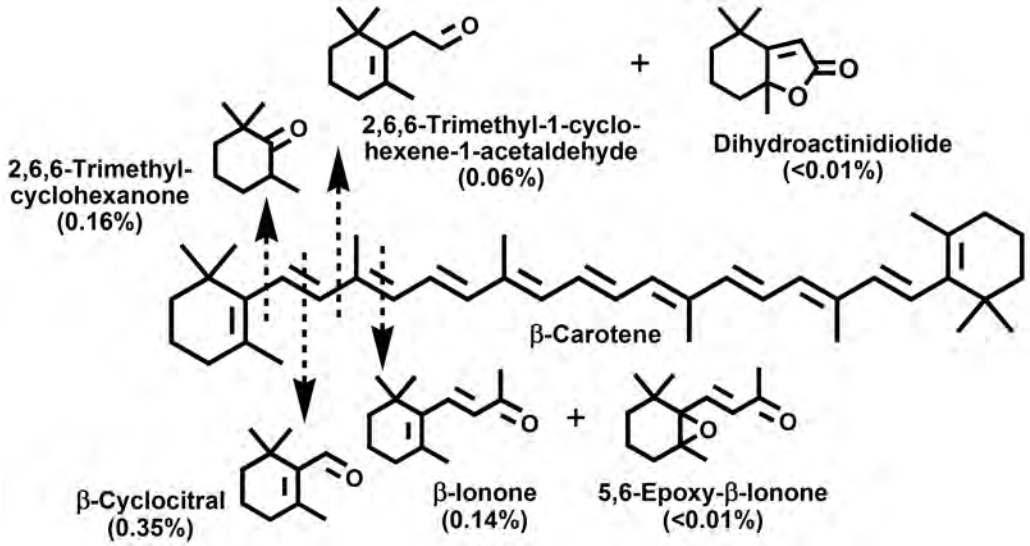
Similarly, the presence of 1-octen-3-ol, and to a lesser extent 3-octanone and 3-octanol, presumably arise from the 10-lipoxygenase pathway (29-32) as shown in Figure 3.

Figure 3. – 10-Lipoxygenase Pathway to 1-Octen-3-ol



Among the major carotenoid pigments found in pumpkin varieties is β-carotene which is known to form a number of constituents (33-39) found in our pumpkin analyses (Figure 4.).

Figure 4. – Oxidative Degradation Products of β-Carotene found in Sample 4.D



While the oxidative degradation of β-Carotene can occur by light mediated photolytic singlet oxygen (35-37) to form the products identified in Figure 4, these may also be formed by carotenoid cleavage dioxygenase enzymes (38-39).

Finally, while the exact mechanism for formation of dimethyl disulfide and dimethyl trisulfide remains somewhat unclear (40-43), it is known that these are formed from cysteine and/or methionine as the precursors.

Conclusions:

More than 110 constituents were identified and the primary constituents present suggest that the 13-lipoxygenase pathway is responsible for much of the fresh vegetative pumpkin aroma.

The dynamic headspace analyses of raw pumpkin by GC-MS provided a convenient method for the identification of the complex mixture of volatiles. The use of the NIST AMDIS program allows the automated and accurate identification of overlapping constituents in the various overlapping GC peaks.

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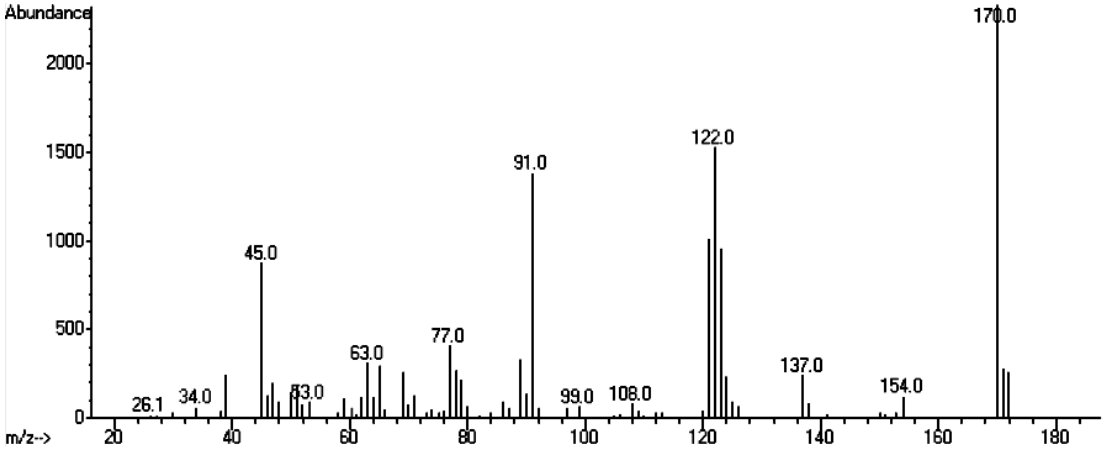
Supplementary Material

Mass Spectrum of Material Identified as Methyl 2-methylphenyl disulfide (syn. Methyl o-Tolyl Disulfide) at LRI = 1390 (R.T. 54.975)



Photos of the Primary *Cucurbita* Species / Cultivars Discussed

Cucurbita pepo **– Connecticut Field Pumpkin**

Cucurbita moschata **– Dickinson Field Pumpkin**

Cucurbita maxima Duchesne **–**
'Blue Hubbard' Blossoms

Cucurbita pepo **var. Styriaka –**
Styrian Seed Pumpkin