FORMULATIONS: A STABLE CHEMICAL / SENSORY EQUIVALENT TO NATURAL PRODUCTS FOR PERMEATION / PACKAGE TESTING

R. Bleiler, A. Iwasinska, F. Kuhrt, D. Wright
MOCON®, Round Rock, TX

Introduction

Natural products (such as fruits, vegetables, etc.) are composed of an abundance of chemical compounds that can stimulate the human olfactory senses. These volatile and semi-volatile organic molecules are responsible for creating the characteristic flavor (scent and/or taste) of many things around us, natural and man-made.

During the lifetime of natural products, their appealing flavors can change [as a function of time] either moving toward a rotting, decomposing odor or loosing the desirable intensity. The packaging materials designated for these types of products face many challenges, as they need to protect the quality of the packaged goods; preserving their freshness, color, aroma and taste. A stable chemical formulation mimicking the aroma/flavor of certain foods (fruits, vegetables, etc.) can be useful as a challenge material for a variety of testing of packaging materials such as: permeation, sensory evaluation and remediation [in the case of odors].

Multi-Dimensional Gas Chromatography / Mass Spectrometry / Olfactometry (MDGC/MS/O) is an analytical technique that combines sensory and mass spectral detection of odorous molecules emitted by different materials (simultaneous double detection). This methodology can be utilized to identify the aroma notes found in the headspace generated from a sample and find their impact on the total flavor of the material – the crucial information in the process of creating an artificial flavor substitute (formulation).

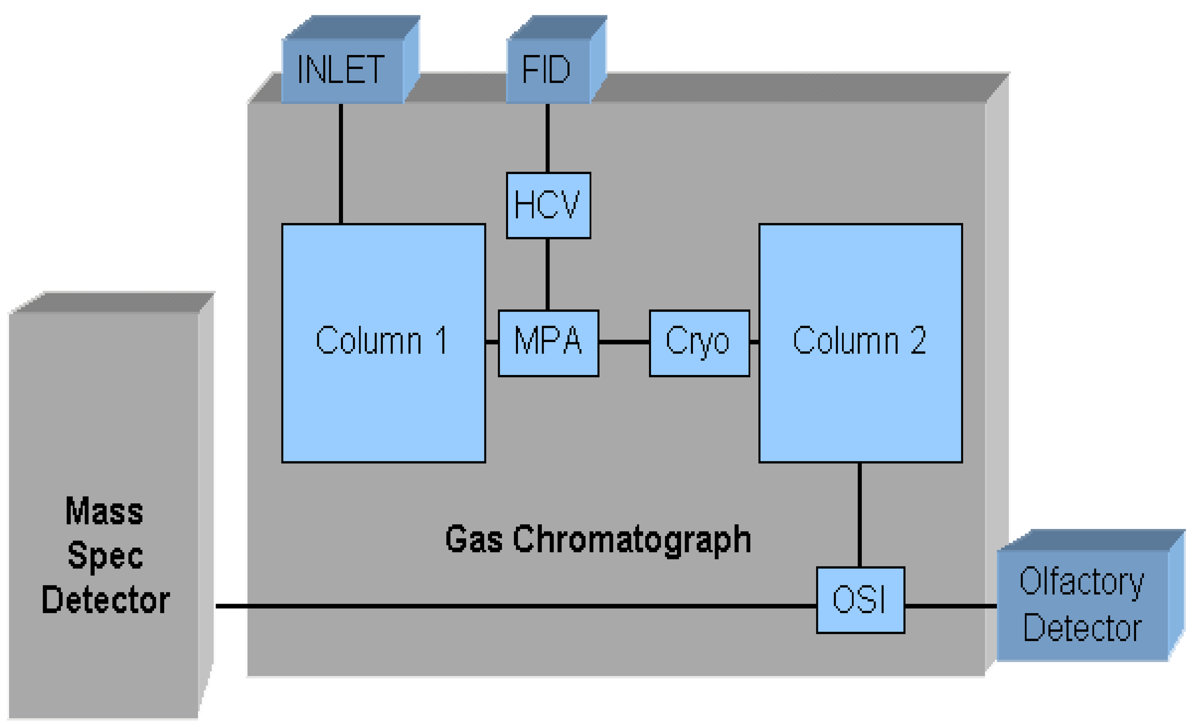
The preparation of the formulation using MDGC/MS/O is demonstrated here for a natural, organic food item → red onion (Allium cepa). The characteristic aroma notes emitted by the chopped red onion detected at certain retention times at the olfactory port were prioritized based on their individual olfactory impact. These odor characters were then correlated to their respective chemical compounds through the results obtained (simultaneously) from the mass spectrometer detector. It is well known that many flavor compounds have extremely low odor threshold [which means they are responsible for high impact notes while present at extremely low concentrations] and their identification may be quite challenging. The use of multidimensional heart-cutting and cryo-trapping techniques combined with olfactory detection may help to identify these hidden flavor contributors.

In the case presented in this poster the chemical compounds recognized as major "players" in total odor of red onion were combined in an inert matrix at the same ratio as detected in the natural sample material. This process yields a formulation which provides the same sensory experience as the original natural product but is far more stable and can be prepared reproducibly to be used in different types of testing (for example, in permeation testing).

Experimental

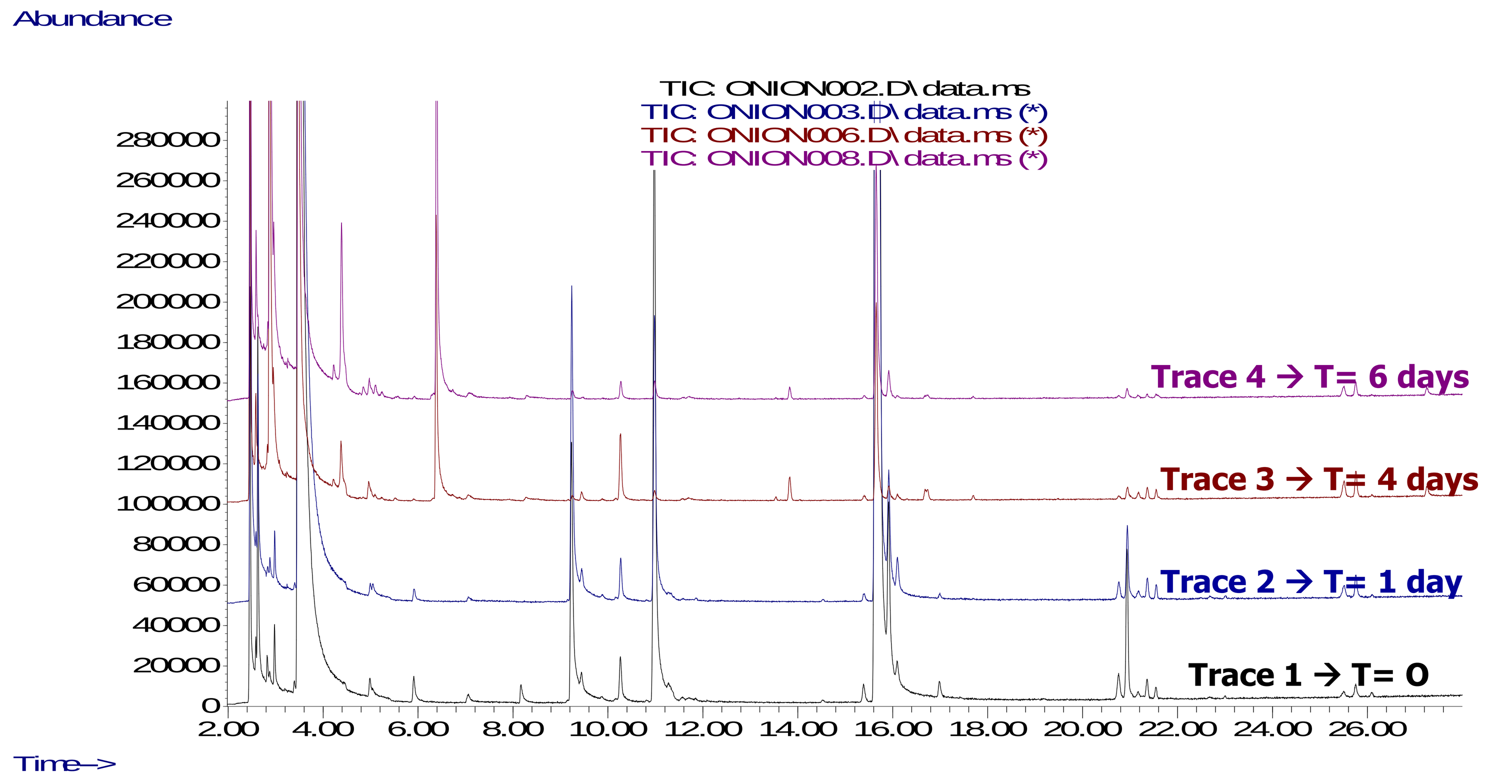
- 50 grams of chopped red onion enclosed in a 240 ml glass vessel for headspace equilibration
- Volatile and semivolatile compounds collected from headspace using Solid Phase MicroExtraction (SPME) fibers (75 µm carboxen/polydimethylsiloxane)
- MDGC / MS / O analysis performed using AromaTrax® instrument (7820 Agilent GC / 5975 Agilent MSD / "Sniff Port" (Olfactory Detector)
- Temperature program: 40ºC (hold for 3 minutes), 7ºC/min ramp to 240º C and 240ºC (hold for 8.4 minutes)

MDGC/MS/O Instrument Schematic



GCMS Total Ion Chromatograms

Comparison of GCMS total ion chromatograms acquired from the red onion sample as a function of time (**Trace 1→ T = 0, Trace 2→ T = 1 day, Trace 3→ T = 4 days (10x collection time), Trace 4→ T= 6 days, (10x collection time)**).

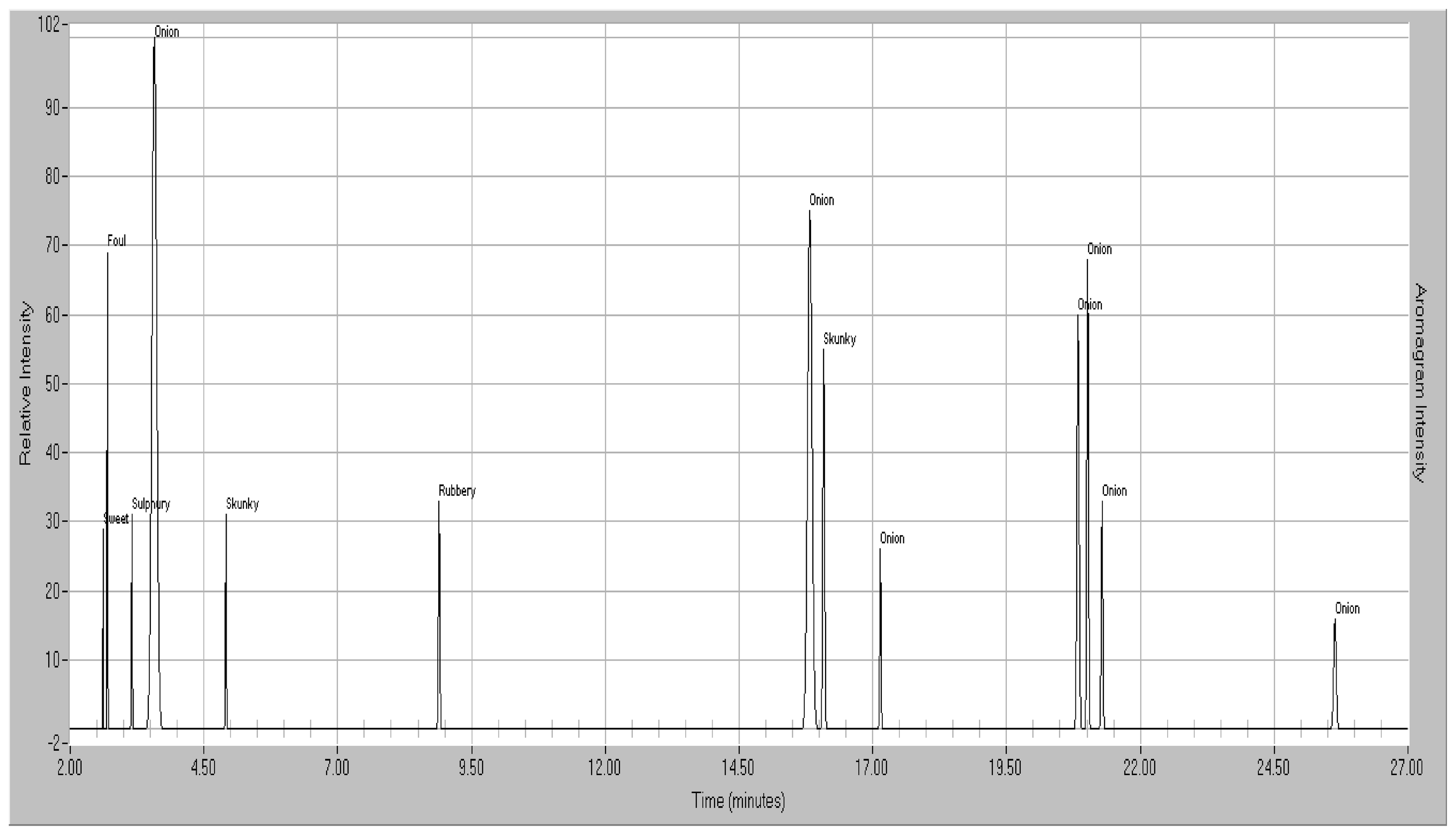


The results presented here show the differences in the chemical composition of the red onion headspace that were observed with time – illustration of the aging process.

Summary of GCMS Results – Headspace Composition

RT (min)	Tentative Compound Identification	T=0	1 day	4 days	6 days
2.59	**acetaldehyde**	X	X	X	X
2.63	methanethiol	X	X	X	X
2.82	propanal	X	X		
2.84	acetone				X
2.88	ethanol		X	X	X
2.96	methyl acetate				X
2.98	**carbon disulfide**	X	X	X	
3.46	**1-propanethiol**	X	X	X	X
4.22	2-methyl-1-propanol			X	X
4.38	1-methoxy-3-methyl butane			X	X
4.96	methyl propyl sulfide			X	X
4.98	methylthiol acetate	X	X		
5.91	dimethyl disulfide	X	X	X	
6.38	3-hydroxy-2-butanone			X	X
8.17	2-methyl-2-pentenal	X			
9.24	n-propyl thiol acetate	X	X	X	X
9.45	x,y-dimethyl thiophene	X	X	X	X
10.26	x,y-dimethyl thiophene	X	X	X	X
10.97	**methyl propyl disulfide**	X	X	X	X
11.27	1-propenyl methyl disulfide	X			
13.83	3,7-dimethyl-1,3,6-octatriene			X	X
15.38	cis-2-propenyl propyl disulfide	X	X	X	X
15.70	**dipropyl disulfide**	X	X	X	X
15.92	**trans-2-propenyl propyl disulfide**	X	X	X	X
20.76	dipropyl disulfide	X	X	X	X
20.93	**dipropyl trisulfide**	X	X	X	X
21.17	unknown	X	X	X	X
21.36	**(cis or trans) propenyl propyl trisulfide**	X	X	X	X
21.55	**(cis or trans) propenyl propyl trisulfide**	X	X	X	X
23.00	unknown	X	X		
25.49	**unknown**	X	X	X	X
25.75	**unknown**	X	X	X	X
26.09	unknown	X			
27.25	unknown			X	X
29.50	unknown	X			
30.03	unknown	X			

Olfactory Analysis – Aromagram of Red Onion at T = 0



Olfactory analysis was performed simultaneously during the GC/MS analysis to detect the compounds that contribute to the aroma of the red onion. An aromagram is a graphical representation of this olfactory analysis. Each peak indicates a presence of an aroma compound that was sensed at the Sniff Port (olfactory detector) by the scientist at the time of compound's elution.

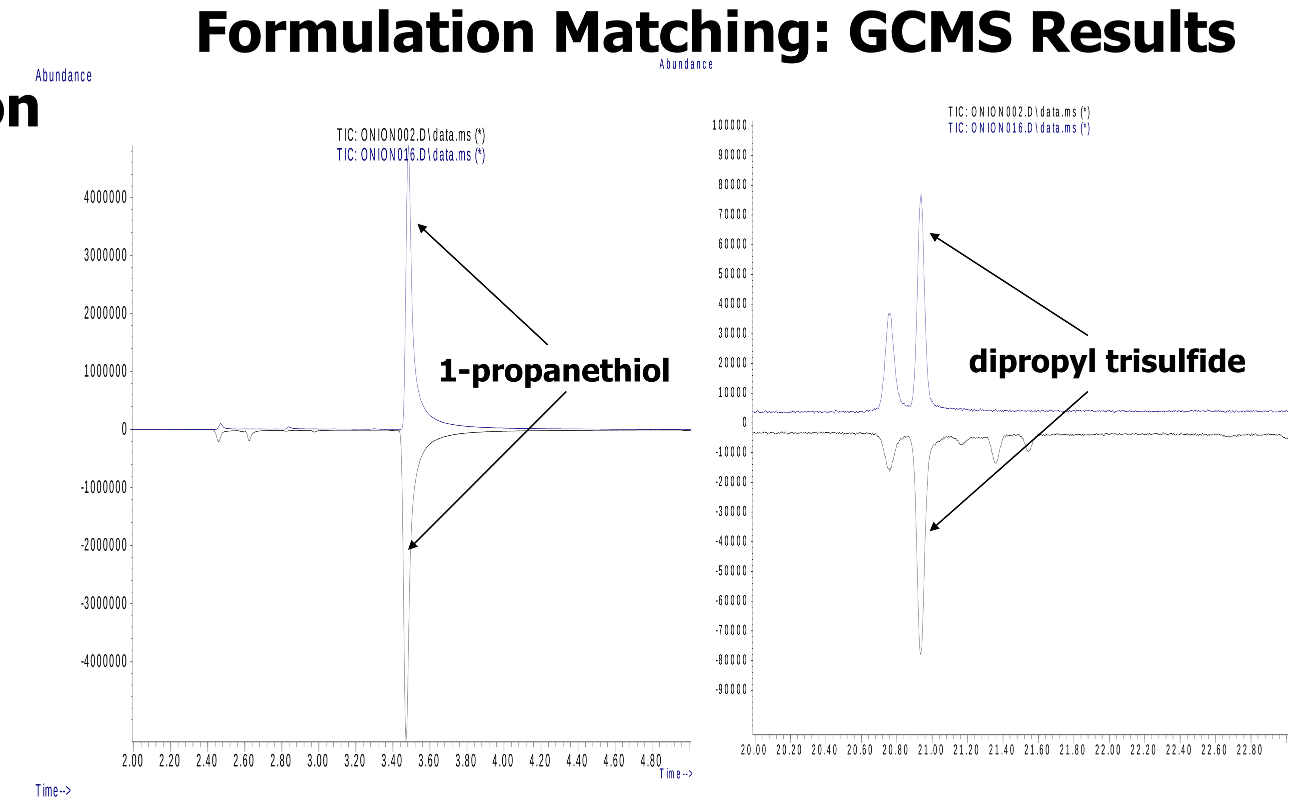
First & Second Tier Aroma Impact Compounds

Rank	RT (min)	Aroma Character	Tentative Identification
Group 1	3.45	onion	**1-propanethiol**
	15.71	onion, sulfury, skunky	**dipropyl disulfide**
	20.97	onion, skunky	**dipropyl trisulfide**
Group 2	16.04	skunky, foul	**trans-2-propenyl propyl disulfide**
	21.24	onion, sharp	**propenyl propyl trisulfide (cis, trans)**
	21.43	onion, sharp	**propenyl propyl trisulfide (cis, trans)**

Additional Aroma Modifier Compounds

Rank	RT (min)	Aroma Character	Tentative Identification
Group 3	2.68	foul, sulfury, sharp	**methanethiol**
	3.13	sulfury, foul	**carbon disulfide**
	11.04	onion, foul	**methyl propyl disulfide**
	17.11	onion	unknown
	25.51	onion, sharp	unknown
	25.85	onion, sharp	unknown
	26.20	onion, skunky	unknown

Formulation Matching: GCMS Results



The **figure on the left** displays the retention time region for the **primary odorant compound → 1-propanethiol**. The **figure on the right** displays the retention time region for **another 1st tier odorant compound → dipropyl trisulfide.**

In both figures: the bottom curve represents the headspace concentration in the natural red onion sample, while the top curve represents the headspace concentration created in the formulation sample using propylene glycol as the matrix. It has to be noted that the ratio of the relative concentration levels (represented by peak areas) of the two compounds are the same in the formulation as observed in the headspace of natural onion sample.

Conclusions

- The utilization of Solid Phase Microextraction (SPME) headspace collection and the MDGC/MS-Olfactometry analytical technique is presented here as an accurate way of preparing a flavor formulation of a natural food product → organic red onion (Allium Cepa). The GC Total Ion Chromatograms and Aromagrams were obtained simultaneously from the SPME-collected headspace volatiles emitted by the red onion.
- GCMS results from the fresh and aged red onion show differences in the volatiles composition as the onion ages. This is especially evident in the older two samples where collection time was increased by an order of magnitude. For many of the compounds, the overall odor intensity decreases with time. At the same time a number of new compounds emerge and increase in concentration. These two facts indicate instability of the total flavor of natural food products such as vegetables and fruits.
- 12 major aroma notes were identified based on the olfactory data acquired from the headspace of the fresh red onion sample (at T = 0) – three compounds in this group represented the highest impact odorants (odor defining components, core components of the flavor formulation).
- The addition of lower impact odor contributors helps to increase the fidelity rating of this flavor simulant → the resulting final formulation provides a sensory equivalent to the original red onion.
- Use of MDGC/MS-Olfactometry methodology enabled identification and prioritization of flavor compounds. The ratio of relative concentration levels of the compounds of interest present in the headspace was established based on the collected chromatographic and olfactory data. The collected information was used to prepare a synthetic red onion flavor formulation (using pure chemicals) → a solution that mimics sufficiently organoleptic properties of natural product and is stable during extended period of time.
- The synthetic odor/flavor formulations should be considered as a challenge material in many cases of permeation testing due to their time stability and reproducible organoleptic properties.