AppBrief

GERSTEL
MAKING LABS WORK

Determining Flavor Compounds in Shisha Tobacco using Dynamic Headspace GC-MS

Highlights

- Dynamic headspace GC-MS allows sensitive determination of flavor compounds in Shisha tobacco
- No sample preparation other than dissolution necessary
- High throughput VOC analysis due to full automation
- Aroma Office software improves flavor compound recognition and identification

Introduction

Shisha tobacco is a moist tobacco mix that is smoked in a type of waterpipe known as "hookah" or "shisha." It is made by fermenting tobacco with molasses, glycerine and fruit essence, producing a tough, pliable mixture challenging to analyze for flavor compounds.

Thoroughly determining the VOCs and semi-VOCs in the final product is vital for ensuring quality control, batch consistency and successful product development studies.

Experimental

Instrumentation

For this application, a GERSTEL LabWorks Platform, supplemented with dynamic headspace (DHS) and an Agilent GC-MS, was used. The GERSTEL LabWorks Platform is a universal system for sample introduction and offers unrivaled capabilities and flexibility to solve your critical challenges. Liquid, headspace, and thermal desorption are all included without the need for additional bench space.

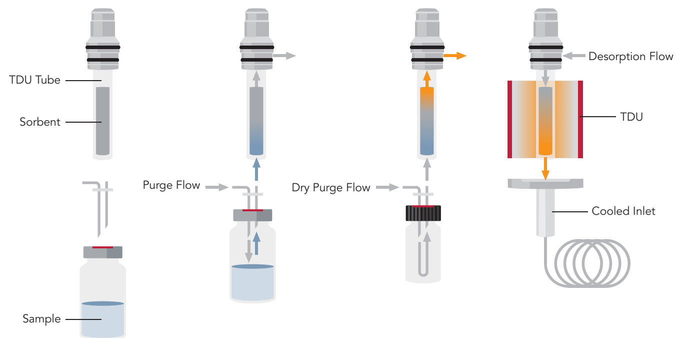
Sample Preparation

A slurry of 0.1 g of the sample in 20 mL of either distilled water or methanol was prepared. No further sample preparation steps were necessary.

Sample Introduction

Dynamic headspace was selected as the sample introduction technique. The dynamic headspace technique enriches volatile and semi-volatile analytes from the gas phase above a liquid or solid sample on a sorbent-filled tube. Analytes are continuously removed from the sample's headspace and transferred to a sorbent-filled tube using a permanent gas flow.

This leads to a constant shift in the equilibrium between the headspace and the sample, providing exhaustive analyte extraction. This is in contrast to the classical headspace technique, where only a finite amount of headspace is sampled.



The dynamic headspace workflow from sample insertion to thermal desorption

Purging the entire vial content to dryness leads to almost quantitative extraction of the flavor compounds with matrix independent and close to reality chromatograms. The sorbent-filled tube traps the analytes, whereas the solvents in use are vented. Non-volatile residues are left in the vial. This technique is known as Full Evaporation Dynamic Headspace (FEDHS).

The original idea of overcoming matrix effects by dispensing a small aliquot of sample into a headspace vial, thermally equilibrate and then analyze it is from Michael Markelov back in the early 90's [1]. The use of dynamic headspace overcomes the disadvantage of low sensitivity originally associated with static headspace.

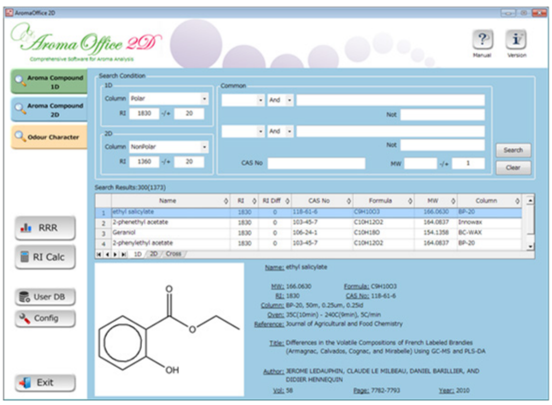
The dynamic headspace technique is automated using the GERSTEL MPS. Incubation, extraction, and, if necessary, a drying step of the sorbent are performed in a DHS station mounted on the MPS. The adsorption is carried out on sorbent-filled tubes, which can be packed with different sorbents. Following extraction, these are desorbed in a GERSTEL thermal desorption system and re-focused in a cold injection system (CIS 4) for subsequent temperature-programmed transfer to the GC-MS system. There are no valves, transfer lines, or active sites in the sample path, ensuring the best possible recovery of all analytes.

Analysis

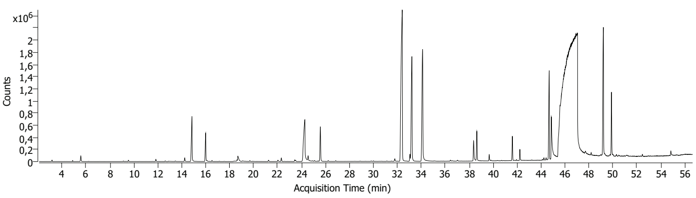
A filtered aliquot of 50 µL of the sample slurry was introduced into a 20 mL headspace vial and incubated at 80 °C. A purge flow of 3000 mL in total extracted the entire volatile fraction from the vial and transferred it to the sorbent-filled tube. Subsequently, the tube was thermally desorbed and analytes were refocused in the cooled injection system (CIS 4) with a Tenax® liner at a trap temperature of 20 °C.

Results and Discussion

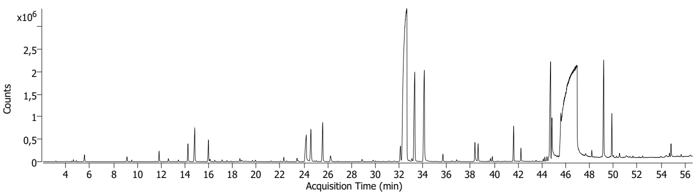
Detection was performed with a mass selective detector from Agilent Technologies in scan mode. The verification of the detected compounds was carried out using deconvolution with MassHunter Unkowns analysis and a NIST library. Furthermore, the flavor compounds were identified with Aroma Office, matching GC-MS data with retention indices and CAS numbers.



Aroma Office



Chromatogram of water aliquot



Chromatogram of methanol aliquot

The following table lists the compounds identified with Aroma Office. The compounds highlighted are found in the methanol extract in addition to the water extract.

Compounds identified with Aroma Office

RT	compound	descriptor	RT	compound	descriptor
4.48	methyl butyrate	apple	24.59	hexyl caproate	apple
5.57	ethyl 2-methyl butyrate	anise	24.91	methyl benzoate	flowery
6.23	hexanal	apple	25.58	menthol	mentholated
7.17	amyl acetate	apple	26.24	(Z)-3-hexenyl hexanoate	fruity
9.04	isopentyl propionate	fruity	26.86	2-methylbutyric acid	acidic
9.14	d-limonene	citric	28.89	benzyl acetate	floral herbal
11.55	isopentyl butanoate	banana	32.11	hexyl octanoate	green
11.84	hexyl acetate	apple	32.64	anethole	aniseed
12.62	isoamyl isovalerate	fruity	33.07	caproic acid	acidic
13.46	Z-3-hexenyl acetate	banana	34.12	benzyl alcohol	aromatic
14.25	hexyl propanoate	apple	36.44	2-ethylhexanoic acid	rancid fatty
14.83	hexanol	alcoholic	39.67	octanoic acid	acid
15.98	(E)-3-hexenol	floral	41.63	4-decanolide	candy
17.13	hexyl butyrate	apple	44.12	γ-undecalactone	apricot
17.54	hexyl 2-methyl butyrate	apple and grape-fruit-like	44.73	methyl dihydro jasmonate	flowery
18.75	acetic acid	acetic	47.68	benzoic acid	aqueous
19.68	menthone	dry	49.21	ethyl vanillin	vanilla-like
21.25	benzaldehyde	almond	49.88	vanillin	fruity
22.03	propanoic acid	acidic	54.85	palmitic acid	creamy
22.32	linalool	althea			

Conclusions

- DHS is an excellent, fully automated technique for the determination of flavors in shisha tobacco.
- Full Evaporation Dynamic Headspace (FEDHS) enables almost quantitative extraction of flavor compounds across a wide volatility range.
- FEDHS leads to results that are closer to the actual flavor composition than with other commonly used techniques.
- Apart from a slurrying step, no sample preparation was necessary.

References

[1] M. Markelov, J.P. Guzowski, Jr., Analytica Chimica Acta, 276 (**1993**) 235-245