Turkish Journal of Agriculture and Forestry

Volume 44 | Number 4 Article 9

1-1-2020

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TÜBİTAK

Turkish Journal of Agriculture and Forestry

http://journals.tubitak.gov.tr/agriculture/

Research Article

Turk J Agric For
(2020) 44: 408-416

doi:10.3906/tar-1910-26

Variety differences in garlic volatile sulfur compounds, by application of selected ion flow tube mass spectrometry (SIFT–MS) with chemometrics

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Received: 11.10.2019 • **Accepted/Published Online:** 06.03.2020 • **Final Version:** 02.08.2020

Abstract: Garlic releases a strong odor after tissue disruption by chewing or other mechanical effects. This odor is strongly associated with the volatile sulfur compounds that can be used for the characterization of garlic. In this study, different varieties of garlic were examined for their volatile sulfur compound concentration, which may be used for purposes of quality control as well as for authentication and classification of garlic varieties. 2- or 3-vinyl-4H-1, 2- or 3-dithiin, allicin, allyl mercaptan, allyl methyl disulfide, allyl methyl sulfide, allyl methyl tetrasulfide, allyl methyl thiosulfide, allyl methyl trisulfide, diallyl disulfide, diallyl sulfide, diallyl tetrasulfide, diallyl trisulfide, dimethyl disulfide, dimethyl thioether, dimethyl thiosulfide, dimethyl trisulfide, and methyl mercaptan were measured in the headspace of garlic for 30 min by selected ion flow tube mass spectrometry (SIFT–MS). German White was determined to be the garlic variety with the highest volatile sulfur compound concentration. Korean Mountain, Music, and Godfathers Italian varieties were discriminated well based on interclass distance (ICD) values. Also, softneck and hardneck garlic were clearly differentiated with chemometrics.

Key words: Selected ion flow tube mass spectrometry, garlic, sulfur volatiles, chemometrics

1. Introduction

Garlic (*Allium sativum* L.) is one of the most valuable vegetables in the world. Overall harvested yield of dry bulbs is 1,468,811 ha with a yearly production of 28,164,055 tons (FAO, 2017).

Garlic has been commonly used as both food and medicine all over the world since ancient times. Garlic is consumed as a raw or dried vegetable, oil, extract, and powder. It has several benefits in culinary, therapeutic, and medicinal areas. As well as its bioactive content, garlic has a high amount of sulfur compounds that are responsible for its distinctive odor and flavor (Lawson, 1996). The formation of the characteristic flavor of garlic is a consequence of several biochemical responses that produce volatile organosulfur compounds and nonvolatile amino acids (Amagase et al., 2001; Martins et al., 2016). Alliin is the main volatile producing compound present in raw garlic. Diallyl disulfide, allyl mercaptan, allyl methyl disulfide, and allyl methyl sulfide are volatile components responsible for garlic breath (Cai et al., 1995; Suarez et al., 1999; Rosen et al., 2001; Tamaki et al., 2008). The development of these volatiles starts immediately after garlic is crushed, chopped, or chewed, which leads to the conversion of alliin to allylsulfenic acid by the enzyme alliinase, followed by condensation into allicin. Once allicin is produced, it contributes to the formation of allyl methyl disulfide and diallyl disulfide (Negishi et al., 2002). Allyl methyl disulfide is formed from allicin and does not react further to produce other garlic volatiles (Suarez et al., 1999). Diallyl disulfide is reduced to form allyl mercaptan (Iciek et al., 2009). The formation of allyl methyl sulfide can occur either outside or inside of the body (Yu et al., 1989; Kim et al., 1995; Lawson, 1998; Hansanugrum and Barringer, 2010; Castada and Barringer, 2019)

There are two main categories of garlic: hardneck and softneck (Bayan et al., 2014). Hardneck varieties (*Allium sativum var. ophioscorodon*) generate a flower stalk. Typically, hardneck garlic varieties have 4–12 cloves surrounding the flower stalk. Because of the hard flower stalk, they are difficult to braid. Softneck varieties (*Allium sativum var. sativum*), also referred to as "artichoke" varieties, do not generate a flower stalk. Softneck varieties are the most commonly domesticated varieties, because they direct all of their energy to producing a bulb. Also, softneck garlic generally has a longer shelf life (Rosen et al., 2016).

Selected ion flow tube mass spectrometry (SIFT–MS) is an analytical technique for the real time analysis of volatiles

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in air and breath, or static and dynamic headspace that does not require complex preanalysis sample preparation or separation. SIFT–MS relies on the soft chemical ionization of volatile compounds by H_3O^+, NO^+, and O_2^+ reagent ions in a flow tube system using helium as a carrier gas (Spaněl and Smith, 2011; Smith and Spaněl, 2005).

Gas Chromatography Mass Spectrophotometry (GC/MS) is one of the most common used techniques for volatile organic compounds; however, it is better to monitor samples containing large numbers of VOCs at high concentrations. SIFT–MS demonstrated simple usage and wider concentration range determination than GC/MS (Langford et al., 2014).

The objective of this study was to classify different varieties, types, and phenotypes of garlic grown in the same location, based on their volatile sulfur compound formation after blending, using selected ion flow tube mass spectrometry (SIFT–MS) and chemometrics. Application of SIFT–MS and chemometrics for discrimination of softneck and hardneck garlic varieties was also evaluated.

2. Materials and methods

Garlic bulbs were obtained from a local farm (Mad River Garlic Growers, Yellow Springs, Ohio) and stored at room temperature until use. Twenty-five garlic varieties were used in this study (Table 1).

2.1. Headspace volatile measurements

Five grams of peeled garlic cloves and 50 mL of HPLC grade water were blended (Magic Bullet model MB1001B, Ningbo Great Height Commodity, Ningbo, China) for 30 s. This blend was transferred to a 500 mL Schott glass bottle and capped with a Teflon sealed screw cap right before the measurement. The headspace volatiles of the mixture were analyzed using selected ion flow tube mass spectrometry (SIFT–MS) immediately. Volatile analysis was done at room temperature (24 ± 1 °C) from 0–30 min at 5 min intervals with an analysis time of 2 min.

2.2. Selected ion flow tube mass spectrometry (SIFT–MS)

Measurements were taken using selected ion mode, with a SIFT–MS (model Voice 200; Syft Technologies Ltd., Christchurch, New Zealand). This technology uses chemical ionization of sample compounds with 3 different precursor ions, H_3O^+, NO^+, or O_2^+, which are generated by microwave discharge. The concentrations of volatile compounds were detected from their reactions with these precursors using predetermined reaction rate constants for the volatiles with the selected precursor ions and accounting for the dilution of the sample gas into the carrier gas in the flow tube (Smith and Spanel, 1996; Spaněl and Smith, 1999; Smith and Spaněl, 2005). Selected compounds were measured on human breath after garlic consumption and by headspace analysis with Syft VOICE-200 software (v.1.4.9.17754; Syft Technologies Ltd).

Table 1. Cultivar type and phenotypes of garlics used in the study.

Name	Cultivar type	Phenotype
Armenian- A	Hardneck	Porcelain
Chesnok Red- CR	Hardneck	Purple Stripe
Georgian Crystal- GC	Hardneck	Porcelain
Georgian Fire- GF	Hardneck	Porcelain
German Red- GR	Hardneck	Rocambole
German White- GW	Hardneck	Porcelain
Korean Mountain- KM	Hardneck	Asiatic
Music- M	Hardneck	Porcelain
Metachi- Me	Hardneck	Marble Purple Stripe
Russian Giant- RG	Hardneck	Purple Stripe
Romanian Red- RR	Hardneck	Porcelain
Russian Red- RU	Hardneck	Rocambole
Siberian- S	Hardneck	Marble Purple Stripe
Shantung Purple- SP	Hardneck	Turban
Spanish Roja- SR	Hardneck	Rocambole
Thai Purple- TP	Hardneck	Turban
Uzbek- U	Hardneck	Turban
Xian- X	Hardneck	Turban
Burgundy- B	Softneck	Creole
God Fathers Italian- GFI	Softneck	Artichoke
Inchelium- I	Softneck	Artichoke
Nootka Rose- NR	Softneck	Silverskin
Polish White- PW	Softneck	Artichoke
Red Toch- RT	Softneck	Artichoke
Transylvanian- T	Softneck	Artichoke

Table 2 outlines the volatile compounds analyzed in this study with their corresponding precursor ions, ion product, reaction rates, and mass–to–charge ratios (m/z) used in SIFT–MS. The concentration (M) of volatile compounds were obtained via product count rate (Ip), reaction rate constant (k), precursor ions count rate (I), and reaction time (t) as follows: (M) = Ip/Ikt (Spaněl and Smith, 1999). Conflicts were removed by selecting different masses or different precursor ions for each compound in the method. The instrument was validated to quantify properly via linearity, range, accuracy, precision, and limit of detection using a pressurized mixture of certified gas standards (benzene, ethylene, isobutane, octafluorotoluene, hexafluorobenzene, toluene, p-xylene, and 1,2,3,4-tetrafluorobenzene) each having a concentration of 2 ppm (±5%) in nitrogen (Air Liquide America Specialty Gases LLC, Plumsteadville, PA, USA) and regulated to a pressure of 21 kPa (3 psi).

Table 2. Kinetic parameters for SIFT–MS analysis of measured volatile compounds in garlic

Compound	Precursor ion	Product ion	k (10^{-9} cm^3/s)	m/z
2-or 3-vinyl-4H-1 2or3-dithiin	NO^+	$C_6H_8S_2^+$	2.4	144
Allicin	NO^+	$C_6H_{10}OS_2^+$	2.4	162
Allyl mercaptan	NO^+	C_3H_6S	2.4	74
Allyl methyl disulfide	NO^+	$C_4H_8S_2$	2.4	120
Allyl methyl sulfide	H_3O^+	$C_4H_8S.H^+$	2.6	89
Allyl methyl tetrasulfide	NO^+	$C_4H_8S_4^+$	2.4	184
Allyl methyl thiosulfide	H_3O^+	$C_4H_8S_2O.H^+$	2.6	137
Allyl methyl trisulfide	NO^+	$C_4H_8S_3^+$	2.4	152
Diallyl disulfide	NO^+	$(C_3H_5)_2S_2^+$	2.4	146
Diallyl sulfide	H_3O^+	$(C_3H_5)_2S.H^+$	2.9	115
Diallyl tetrasulfide	NO^+	$C_6H_{10}S_4^+$	2.4	210
Diallyl trisulfide	H_3O^+	$C_6H_{10}S_3.H^+$	2.6	179
Dimethyl disulfide	NO^+	$(CH_3)_2S_2^+$	2.4	94
Dimethyl thioether	NO^+	$(CH_3)_2S^+$	2.2	62
Dimethyl thiosulfide	NO^+	$C_2H_6OS_2^+$	2.4	178
Dimethyl trisulfide	H_3O^+	$C_2H_6S_3H^+$	2.8	127
Methyl mercaptan	H_3O^+	$CH_4S.H^+$	1.8	49

2.3. Statistical analysis

The concentrations of volatile compounds were analyzed in triplicate. One-way analysis of variance (ANOVA) using Tukey's procedure with a 95% confidence interval was performed to determine statistical differences among samples; significance was defined as $P \leq 0.05$ using SPSS (version 25, SPSS Inc., Chicago, IL., U.S.A.). Chemometrics used in this study were provided by Pirouette software for Windows Comprehensive Chemometrics Modeling, version 4.0 (Infometrix Inc., Bothell, Wash., USA).

Soft independent modeling of class analogy algorithm (SIMCA), which is a classification process depending on principal component analysis (PCA), was used to group garlic samples based on their variety. The discriminating power in SIMCA contributes a diagnostic tool that displays how important a volatile compound is to differentiating sample classification. When interclass distances were determined higher than 3, classes were acknowledged as statistically different from each other (Aykas and Rodriguez-Saona, 2016).

3. Results and discussion

3.1. Volatile sulfur compound profile of garlic

A total of 18 volatile compounds were measured in the twenty-five different garlic varieties grown in Ohio, USA (Table 3). Diallyl disulfide had a noticeably higher concentration among all cultivars followed by allyl mercaptan. Similarly, Molina-Calle et al. (2016) investigated the volatile profile of different varieties of garlic and determined that diallyl disulfide had the highest relative percentage with 26.4, 24.8, and 21.0 among all measured compounds in Chinese, White and Purple varieties, respectively. Diallyl disulfide was also determined to be the main aroma compound in both Kastamonu and Chinese types of garlic (Keles et al., 2014).

Allyl methyl tetrasulfide, diallyl tetrasulfide, and dimethyl thiosulfide were at a very low percentage of all aroma compounds. Block (1984) also identified lower amounts of diallyl tetrasulfide in garlic oil, compared to the abundant diallyl disulfide content.

2- or 3-vinyl-4H-1, 2 or 3-dithiin and diallyl sulfide concentration was not significantly different between varieties ($P \leq 0.05$). Allyl mercaptan, allyl methyl disulfide, allyl methyl sulfide, allicin, diallyl disulfide, dimethyl disulfide, and dimethyl thioether were the compounds with the highest concentration in German White, while allyl methyl thiosulfide, and allyl methyl trisulfide were the highest concentration compounds in Burgundy. The concentration of allyl methyl tetrasulfide, diallyl tetrasulfide, and dimethyl trisulfide were at a high level in Shantung Purple. While diallyl trisulfide amount was high in Inchelium, dimethyl thiosulfide and methyl mercaptan were high in Romanian Red ($P \leq 0.05$) (Table 3).

Table 3. Percentage concentration of garlic volatiles from different varieties at 30 min.

	Variety Code/ Compound	2 or 3 vinyl-4H-1 2 or3-dithiin	Allyl mercaptan	Allyl methyl thiosulfide	Allyl methyl disulfide	Allyl methyl sulfide	Allyl methyl tetrasulfide	Allyl methyl trisulfide	Diallyl tetrasulfide	Diallyl trisulfide	Dimethyl thiosulfide	Allicin	Diallyl disulfide	Diallyl sulfide	Dimethyl disulfide	Dimethyl thioether	Dimethyl trisulfide	Methyl mercaptan
HARDNECK	A	0.21	10.94	0.15	11.48	1.81	0.04	0.12	0.02	0.90	0.09	1.46	68.28	0.68	0.41	1.53	0.08	0.68
	CR	0.20	8.67	0.15	10.16	1.01	0.05	0.12	0.02	1.06	0.09	1.37	73.66	0.56	0.31	1.17	0.10	0.50
	GC	0.17	10.01	0.17	11.29	0.63	0.05	0.14	0.02	1.00	0.09	1.26	71.87	0.52	0.33	0.92	0.08	0.80
	GF	0.28	8.58	0.21	8.69	0.78	0.05	0.12	0.02	1.17	0.09	2.21	74.41	0.65	0.21	1.01	0.11	0.42
	GR	0.26	11.24	0.17	8.85	4.51	0.06	0.27	0.03	2.78	0.07	1.77	66.36	0.92	0.23	0.77	0.13	0.41
	GW	0.20	11.72	0.09	9.57	21.95	0.03	0.07	0.01	0.69	0.04	0.87	50.43	1.78	0.34	0.77	0.04	0.23
	KM	0.22	7.61	0.13	5.35	1.41	0.07	0.10	0.04	1.75	0.09	2.60	77.22	0.73	0.11	1.11	0.13	0.21
	M	0.11	14.33	0.12	6.19	25.38	0.03	0.11	0.01	1.38	0.04	1.33	48.82	0.39	0.14	0.69	0.05	0.24
	Me	0.15	8.63	0.20	8.99	1.71	0.05	0.15	0.02	1.27	0.08	1.67	71.71	0.55	0.23	1.03	0.08	0.58
	RG	0.28	13.65	0.16	10.33	9.51	0.04	0.21	0.02	1.62	0.08	1.58	59.12	0.72	0.32	0.80	0.08	0.40
	RR	0.15	8.33	0.17	13.91	0.92	0.04	0.20	0.02	1.09	0.10	1.50	69.34	0.61	0.55	1.38	0.09	0.65
	RU	0.19	7.29	0.11	7.13	0.55	0.05	0.10	0.02	1.15	0.07	1.68	79.11	0.54	0.15	0.76	0.11	0.30
	S	0.35	8.85	0.22	11.27	1.45	0.07	0.25	0.02	1.97	0.09	2.15	68.45	1.00	0.35	0.91	0.12	0.49
	SP	0.29	5.30	0.16	7.32	1.04	0.10	0.27	0.04	3.54	0.09	2.69	75.08	1.16	0.17	0.80	0.15	0.28
	SR	0.29	9.79	0.20	4.65	0.76	0.06	0.11	0.03	2.34	0.07	2.17	76.50	0.74	0.09	0.89	0.09	0.27
	TP	0.23	10.96	0.09	5.06	0.66	0.07	0.26	0.03	4.62	0.08	1.53	73.20	0.81	0.10	0.77	0.12	0.53
	U	0.24	7.58	0.07	5.57	0.61	0.07	0.18	0.04	3.45	0.07	1.39	77.80	0.78	0.10	0.72	0.15	0.34
	X	0.17	10.40	0.14	7.75	0.71	0.05	0.31	0.03	3.53	0.07	1.75	72.23	0.61	0.17	0.73	0.09	0.54
SOFTNECK	B	0.16	16.83	0.31	7.02	16.88	0.05	0.29	0.02	2.62	0.08	2.07	50.38	0.67	0.22	0.71	0.08	0.41
	GFI	0.17	11.26	0.07	1.65	0.63	0.06	0.08	0.04	3.52	0.06	1.19	78.68	0.64	0.06	0.95	0.08	0.14
	I	0.21	10.97	0.08	2.12	3.06	0.06	0.08	0.03	4.48	0.05	1.59	71.81	3.12	0.05	0.82	0.10	0.10
	NR	0.27	9.72	0.21	7.89	0.83	0.08	0.14	0.03	2.30	0.08	1.63	72.93	0.66	0.19	1.08	0.13	0.58
	PW	0.17	14.68	0.08	1.56	7.05	0.04	0.06	0.02	3.38	0.05	1.74	68.37	0.73	0.03	0.84	0.07	0.10
	RT	0.13	15.82	0.09	2.25	15.03	0.05	0.06	0.02	2.20	0.05	1.61	60.11	0.63	0.04	0.84	0.07	0.12
	T	0.24	7.58	0.11	3.34	0.99	0.09	0.11	0.05	3.48	0.09	2.85	77.31	1.01	0.05	0.90	0.13	0.15

A: Armenian, CR: Chesnok Red, GC: Georgian Crystal, GF: Georgian Fire, GR: German Red, GW: German White, KM: Korean Mountain, M: Music, Me: Metachi, RG: Russian Giant, RR: Romanian Red, RU: Russian Red, S: Siberian, SP: Shantung Purple, SR: Spanish Roja, TP: Thai Purple, U: Uzbek, X: Xian, B: Burgundy, GFI: God Fathers Italian, I: Inchelium, NR: Nootka Rose, PW: Polish White, RT: Red Toch, T: ransylvanian.

2- or 3-vinyl-4H-1, 2- or 3-dithiin and dimethyl trisulfide were one of the most abundant volatile compounds present in *Allium ursinum,* which is one of the wild garlic varieties (Ivanova et al., 2009).

German White and Metachi had the lowest diallyl disulfide concentration of all garlics (Table 3), while having the highest total volatile concentration of all garlics (Figure 1). In addition, concentration of allyl methyl sulfide in these varieties were the highest among all (Table 3). This means that they had a much higher enzymatic activity, of converting diallyl disulfide to allyl methyl sulfide, than the other varieties.

Based on the calculated interclass distance (ICD) values, Shantung Purple had the highest discrimination from other hardneck varieties, while German Red showed the most similarities with others (Table 4). Godfathers Italian had the highest discrimination from other softneck varieties (Table 5). Red Toch had the lowest ICD value, which means it is the most similar garlic to other softneck varieties based on volatile sulfur composition.

Alliin is enclosed in a large membrane bound vesicle inside the cell and protected from interaction with the alliinase enzyme, which is found in the cytoplasm. When garlic tissue is disrupted, the alliinase enzyme has access to alliin and the formation of allicin and the specific garlic aroma occurs (Holub et al., 2002). The volatile composition of garlic and concentration of bioactive metabolites are dependent on the type of cultivar, the geographic origin, and growing conditions (Baghalian et al., 2006; Khar et al., 2011; Martins et al., 2016). Total volatile concentration of garlic varieties at 0, 5, 10, 15, 20, 25, and 30 min were analyzed (Figure 1). The results revealed that German White had the highest amount of volatile sulfur concentration at all times. Five minutes after tissue disruption, the Music variety had the second highest concentration, while Nootka Rose had the lowest concentration until 20 min.

3.2. Chemometrics classification

Dimethyl thiosulfide and allyl mercaptan were the most discriminant compounds for 0 and 30 min respectively according to the discriminating power plots (Figure 2).

In order to differentiate softneck and hardneck varieties at the end of 30 min in terms of the volatile sulfur compounds associated with each variety, chemometrics (principal component analysis (PCA)) was used (Figure 3). PCA provides a visual relationship between the garlic varieties and their volatile compounds. This method makes the interpretation of the multivariate analysis easy. Interclass distances (ICDs) greater than 3 indicate that samples are significantly different (Kvalheim and Karstang, 1992; Aykas and Rodriguez-Saona, 2016), also greater ICD indicates greater difference between

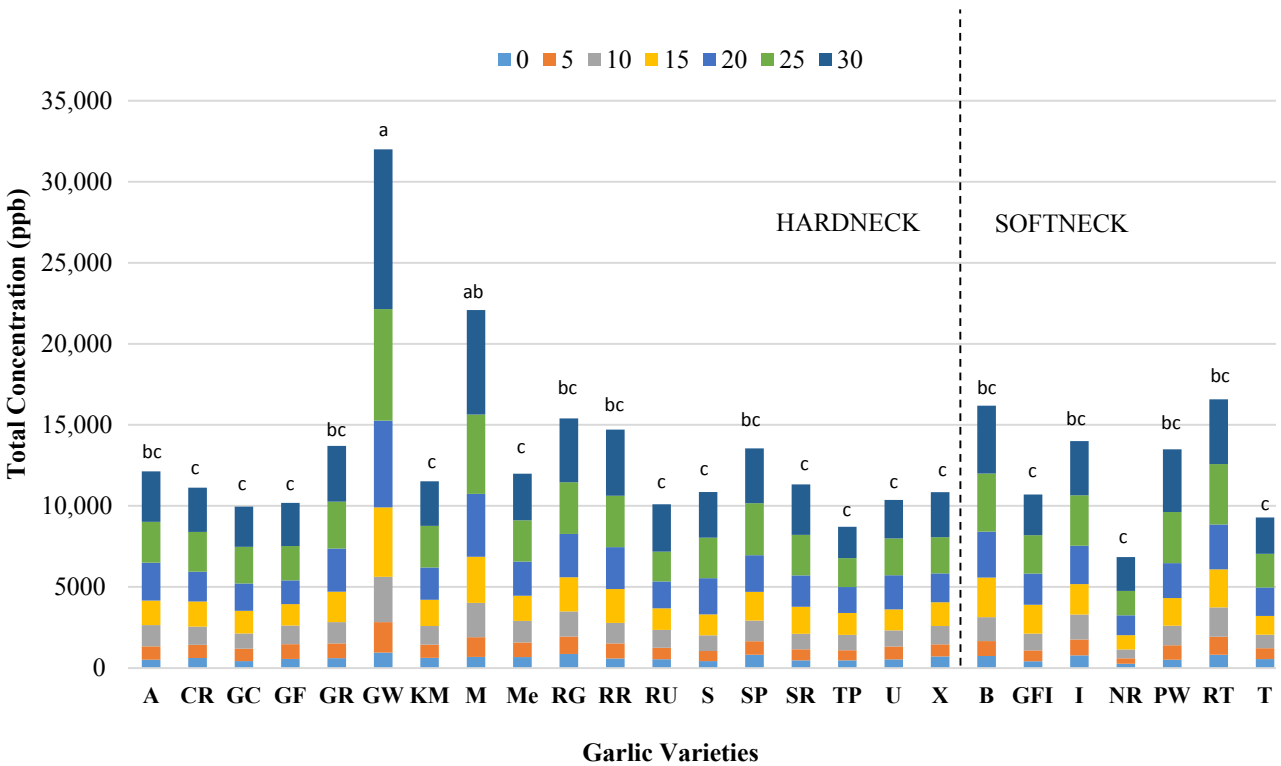

Figure 1. Total volatile concentration of garlic varieties at different times (minutes).

Table 4. Interclass distances (ICD) between hardneck garlic varieties

	A	CR	GC	GF	GR	GW	KM	M	Me	RG	RR	RU	S	SP	SR	TP	U	X
A	-																	
CR	6.76	-																
GC	4.86	9.16	-															
GF	9.57	21.08	7.18	-														
GR	1.49	0.70	1.70	0.76	-													
GW	3.55	4.15	4.46	4.62	4.02	-												
KM	14.48	29.16	8.69	17.21	3.60	5.72	-											
M	5.04	8.20	4.54	6.80	5.02	3.19	5.68	-										
Me	4.35	1.93	3.27	0.44	0.74	4.58	3.19	6.73	-									
RG	3.24	8.16	7.03	10.14	3.43	2.68	13.68	5.16	6.44	-								
RR	1.31	1.15	3.41	3.05	1.85	3.63	5.99	7.31	2.82	2.64	-							
RU	10.62	9.96	7.33	4.17	1.89	5.09	3.78	7.67	0.81	11.81	4.75	-						
S	7.45	2.51	10.20	17.07	0.63	4.19	28.27	8.27	1.64	8.39	1.38	9.13	-					
SP	22.52	70.69	21.04	53.17	4.94	6.07	18.38	9.11	5.00	19.01	9.15	10.58	62.75	-				
SR	9.83	9.92	5.20	4.01	2.24	5.30	4.30	6.45	1.45	11.72	5.93	1.23	9.24	6.70	-			
TP	13.40	16.82	13.10	11.67	2.11	5.04	9.39	8.15	1.08	12.23	4.27	2.15	15.16	15.42	0.75	-		
U	14.39	18.33	11.86	10.93	3.23	5.47	9.42	7.98	2.67	13.99	6.76	4.30	17.03	6.32	2.61	2.59	-	
X	10.96	15.10	8.52	5.47	1.24	4.83	9.34	7.13	0.28	11.20	3.91	2.12	13.31	24.55	1.91	4.86	7.21	-

A: Armenian, CR: Chesnok Red, GC: Georgian Crystal, GF: Georgian Fire, GR: German Red, GW: German White, KM: Korean Mountain, M: Music, Me: Metachi, RG: Russian Giant, RR: Romanian Red, RU: Russian Red, S: Siberian, SP: Shantung Purple, SR: Spanish Roja, TP: Thai Purple, U: Uzbek, X: Xian

Table 5. Interclass distances (ICD) between softneck garlic varieties.

	B	GFI	I	NR	PW	RT	T
B	-						
GFI	16.81	-					
I	6.60	13.91	-				
NR	8.31	30.68	3.12	-			
PW	4.52	8.39	1.28	1.16	-		
RT	1.56	9.11	2.77	3.31	1.42	-	
T	12.62	42.89	1.73	12.85	3.64	5.32	-

B: Burgundy, GFI: God Fathers Italian, I: Inchelium, NR: Nootka Rose, PW: Polish White, RT: Red Toch, T: Transylvanian.

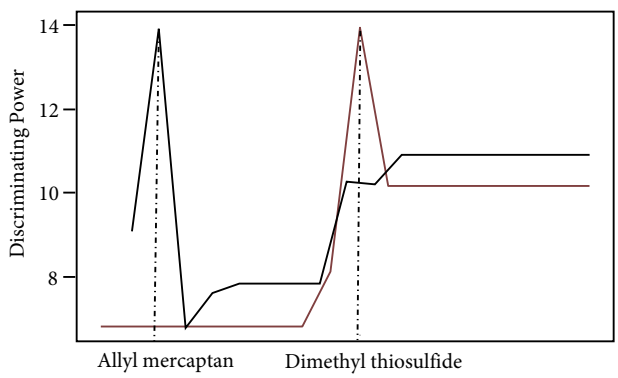

Figure 2. SIMCA discriminating power plot for 0 min and 30 min.

clusters (Dunn and Wold, 1995). Better separation of garlic varieties is achieved when there are higher interclass distances between garlics. When softneck and hardneck garlic varieties were compared in terms of general sulfur volatile compound content after 30 min of tissue disruption, interclass distance was found to be 3.2, which indicates that garlic samples can be discriminated as softneck or hardneck based on their volatile content.

Beyond the division as hardneck or softneck, some garlic growers also classify based on phenotypes (Engeland, 1991, 1995). In phenotypic classification,

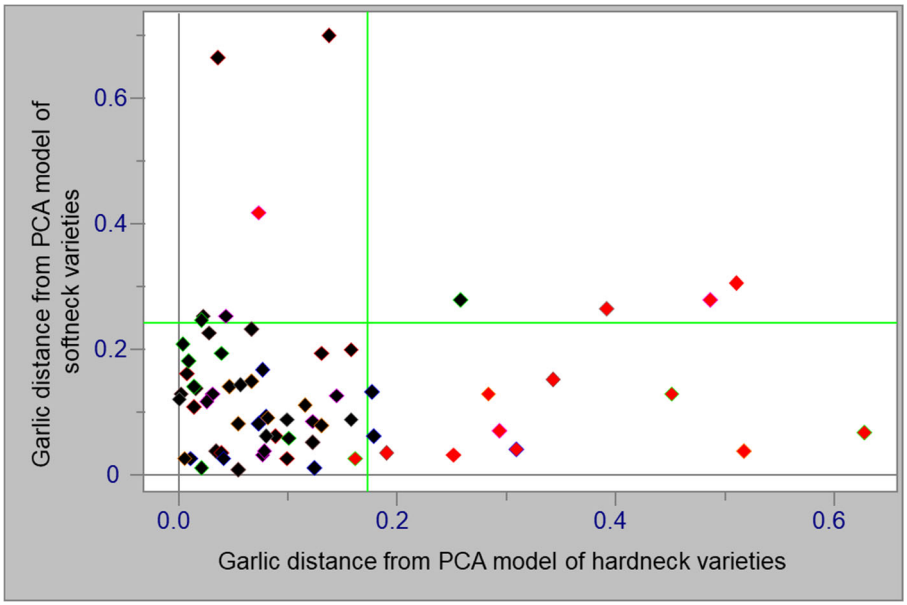

Figure 3. Class distances obtained for discriminating varieties (hardneck, softneck).

Table 6. Interclass distances (ICD) between garlic phenotypes

	Silverskin	Artichoke	Rocambole	Porcelain	Purple Stripe	Marble Purple Stripe	Asiatic	Turban	Creole
Silverskin	-								
Artichoke	**7.9**	-							
Rocambole	**9.4**	1.6	-						
Porcelain	**12.2**	2.3	**3.7**	-					
Purple Stripe	**23.3**	**3.4**	**3.9**	2.7	-				
Marble Purple Stripe	**5.7**	**3.1**	2.7	**4.3**	**7.2**	-			
Asiatic	**8.3**	**3.0**	2.7	**6.6**	**10.0**	**4.5**	-		
Turban	**3.7**	1.9	1.6	**4.9**	**6.0**	2.4	0.8	-	
Creole	**56.7**	**5.5**	**15.7**	**3.4**	**10.6**	**28.8**	**33.6**	**13.3**	-

Hardneck Varieties: A: Armenian, CR: Chesnok Red, GC: Georgian Crystal, GF: Georgian Fire, GR: German Red, GW: German White, KM: Korean Mountain, M: Music, Me: Metachi, RG: Russian Giant, RR: Romanian Red, RU: Russian Red, S: Siberian, SP: Shantung Purple, SR: Spanish Roja, TP: Thai Purple, U: Uzbek, X: Xian. Softneck Varieties: B: Burgundy, GFI: God Fathers Italian, I: Inchelium, NR: Nootka Rose, PW: Polish White, RT: Red Toch, T: Transylvanian.

morphological features are used. Table 6 shows the ICD values based on phenotypical classification. Silverskin and Creole had the highest ICD values, which indicate good separation from other phenotypes. However, some phenotypes had no significant separation.

4. Conclusion

SIFT–MS analyses were employed to elucidate the volatile sulfur compound concentrations of different varieties of garlic. The German White variety had the highest amount of measured volatile sulfur compounds at all measurement times. Inchelium and Burgundy are softneck garlics, which were high in diallyl trisulfide, allyl methyl thiosulfide, and allyl methyl trisulfide. German White, Shantung Purple, and Romanian Red, which are hardneck type, were high in allyl mercaptan, allyl methyl disulfide, allyl methyl sulfide, allicin, diallyl disulfide, dimethyl disulfide, dimethyl thioether, allyl methyl

tetrasulfide, diallyl tetrasulfide, dimethyl trisulfide, dimethyl thiosulfide, and methyl mercaptan. Korean Mountain and Music varieties were discriminated from all other hardneck garlics, while Godfathers Italian variety had the highest discrimination between softneck types. Garlic samples can be discriminated by their type as softneck or hardneck and their phenotype based on their volatile composition. Silverskin and Creole showed the best discrimination from other phenotypes.

Acknowledgement
Gülşah ÖZCAN-SİNİR acknowledges the Scientific and Technological Research Council of Turkey (TÜBİTAK) for 2219–postdoctoral scholarship.

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