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Identification and Quantification of Active Chemical Compounds from Daily Used Spices by GC-MS

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ABSTRACT

In our day-to-day life, we used many spices in food as a colouring agent, flavouring agent or as a preservative. These spices are the rich source of chemical compounds. The study was conducted to determine the concentration and identification of biologically and chemically active compounds in daily used spices. Gas chromatography is very effective method for identification and with the mass detector, we can quantify them as well. The previous report proved that spices have health benefits against the various types of chronic disease like Cancer, Alzheimer, Blood pressure and most of the infectious diseases. Comparative study of biologically active components of spices namely Garlic (*Allium sativum*), cardamom (*Elettaria cardamomum*), Nutmeg (*Myristica fragrans* houtt), Cinnamon (*Cinnamomum zeylanicum* nees) was investigated. Analysis of these spices has shown that great source of biological and chemical active compounds which are used for medicinal purposes as well as in foodstuff for colouring and flavouring agents.

1. INTRODUCTION

Spices and herbs have played a crucial role in the civilization of humankind and in the history of particular nations. The delightful flavour and pungency of spices make them indispensable in the preparation of palatable food. In addition, they are reputed to possess several medicinal and pharmacological properties and hence find the position in the preparation of a number of medicines [2]. Spices are the dried substance or sometimes it's a fresh part of specific plants. Spices either whole use or in the form of extract. It has been using frequently throughout history in the Ayurvedic system (ancient Indian medicine system) and Traditional Chinese medicine, for its good medicinal benefit. In ancient times, spices blend with other herbal material depending upon the desired medicinal properties [4], [5].

The spices production and consumption rate in India is highest, usually Indian and Indian sub-continent people, they like spices in their foodstuff, it increases the flavour, the texture of the food and sometimes it used as a preservative [6], [7]. India is a leading edge in the production of spices, most of the spices produced in only India, along with it few percentage of India's economy depends upon the spices. In the worldwide, they used some common spices like Nutmeg, Garlic, Onion, Paper, Ginger, Cardamom, Cinnamon, Turmeric etc.

The spices which we used in our day to day life in food for colouring, flavouring and increasing the food texture also it has some health benefits [3]. The spices have a lot of biological and chemical active compounds, which is used in various diseases and disorders like cardiovascular disease, cancer, diabetes, blood pressure, atherosclerosis, and hyperlipidaemia and highly praised by several authors [8] as well as it is used as herbicidal [9], [5].

The biological name of Garlic is *Allium sativum* and belonging to the family *Amaryllidaceae*, the Garlic contains 33 types of sulphuric group mainly which is used as antimicrobial and herbicidal [9], [3], also it has wide variety of antioxidant agents which is mainly used against cancer [10],[11],[12]. The common pharmacological uses of Garlic is anti-hyperlipidaemic, anti-hypertensive, reduce blood sugar level, atherosclerosis and various cardiovascular diseases [8]. Moreover, the Nutmeg also has health benefits, The nutmeg contains myristicin which is the active chemical ingredient and it is used as psychotropic agent [13], [14]. Cinnamon and cardamom are world recognized spice in foodstuff and it has some good

medicinal properties like anti-inflammatory, antioxidant, protect heart health etc. [15],[16],[17].

It is the significant challenge to an analytical chemist to identify the biological and chemical active compounds by using instrumental analysis. This spices having the very rich amount of chemical compound with great biological activity and often used in pharmaceutical industry for preparation of medicine.

MATERIALS AND METHODS

All chemicals and solvents obtained from the department of biology Vytautas Magnus University Lithuania (Kaunas).

- Methyl Alcohol
- Distilled water

All the solvent and chemicals were analytical grade. The sample of spices was collected from the local supermarket (Maxima, Kaunas, Lithuania).

Preparation of samples

The dried spices cardamom, nutmeg, cinnamon (0.5gm) and garlic (1gm) were crushed into small pieces of 2-6 mm using cylindrical crusher

Analysis of listed spices

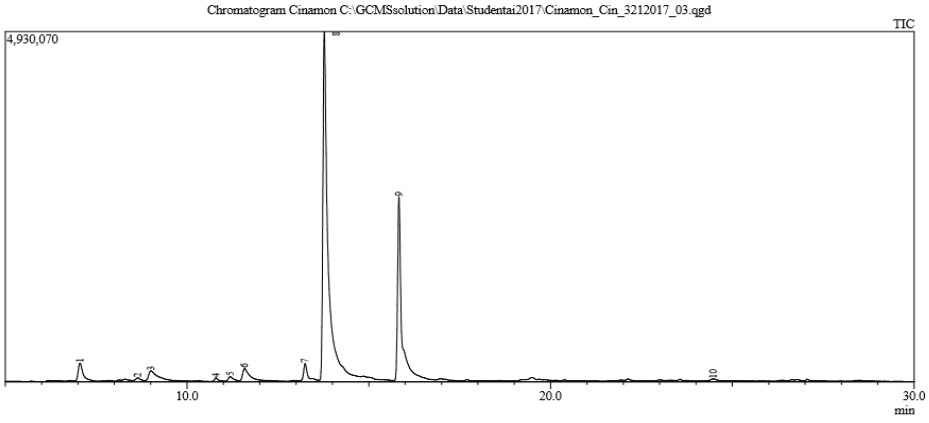
The spices contain volatile compounds were determined and quantified by using GC-MS. In this method, we used spices in the powder form about 10 mg and put into the small bottles which are dedicated for this instrument only. The analysis was carried out using a GC-MS system (GCMS-QP2010, Shimadzu, Tokyo, Japan). A Restec (Bellefonte, USA) RTX-5MS (30 m × 0.25 mm i.d. × 0.25 μm film thickness) GC column was used. The oven temperature gradient was started at 30 °C and raised to 200 °C at 5 °C/min, and then raised to 280 °C at 20 °C/min and was held for 2 min. Helium (99.999%, AGA Lithuania) was used as carrier gas with a constant flow rate of 1.2 mL/min. The injector temperature was kept at 230 °C in split mode (1:10). The mass detector was operated in electron impact mode (70 eV). The ion source and interface temperatures were set at 220 and 260 °C correspondingly. Identification of compounds was performed according to their mass spectra (NIST v1.7). Positive

identification was assumed when good matches (90% and more) of mass spectra were achieved.

RESULT AND DISCUSSION

The total chemically active compounds in spices like Cinnamon, Nutmeg, Cardamom, Garlic were determined using GC-MS. However around more than ten chemically active compounds were found in each spice, among that most of volatile oils in various alpha and beta form. They vary from the different concentration, some in very minute concentration and some of them are in highest concentration. In this spices cinnamaldehyde, Linalool, terpinyl acetate, limonene, sulfide, disulfide, propyl cyanide, butane, myristicin respectively. This chemically active compound has a vitalness to the body as well as various health benefits against various chronic disease and disorders like antibacterial, antifungal, anticholinergic agents, anti-inflammatory etc. The second spice Nutmeg which has several clinical application. The nutmeg contains Myristicin which is the active chemical ingredient and it is used as psychotropic agent [15].

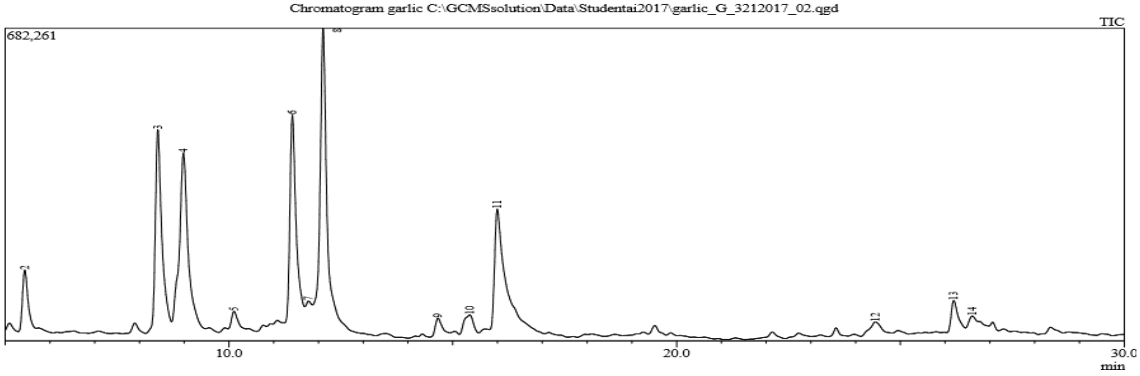
From the following result, there is the highest concentration of volatile oils in all spices. From the below Chromatogram we can easily focus on active chemical compound, here height indicates the concentration.



Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height	Height%	A/H	Mark	Name
1	7.056	6.908	7.575	2394054	2.99	253184	2.99	9.46		Limonene
2	8.643	8.500	8.842	319012	0.40	39477	0.47	8.08		Terpinolene
3	9.006	8.842	9.642	2241722	2.80	136762	1.62	16.39		Linalool
4	10.794	10.658	11.017	320102	0.40	48116	0.57	6.65		Borneol
5	11.185	11.058	11.433	542497	0.68	60390	0.71	8.98		Terpinen-4-ol
6	11.582	11.433	12.200	2153735	2.69	170606	2.02	12.62		Terpineol <alpha->
7	13.248	13.067	13.400	1573401	1.97	240778	2.84	6.53	V	Linalyl formate
8	13.776	13.633	15.217	49623637	62.04	4905135	57.95	10.12	S	Cinnamaldehyde <(E)->
9	15.828	15.692	16.842	20499129	25.63	2583586	30.52	7.93		Terpinyl acetate <alpha->
10	24.474	24.233	24.750	323145	0.40	26958	0.32	11.99		Tridecane <n->
				79990434	100.00	8464992	100.00			

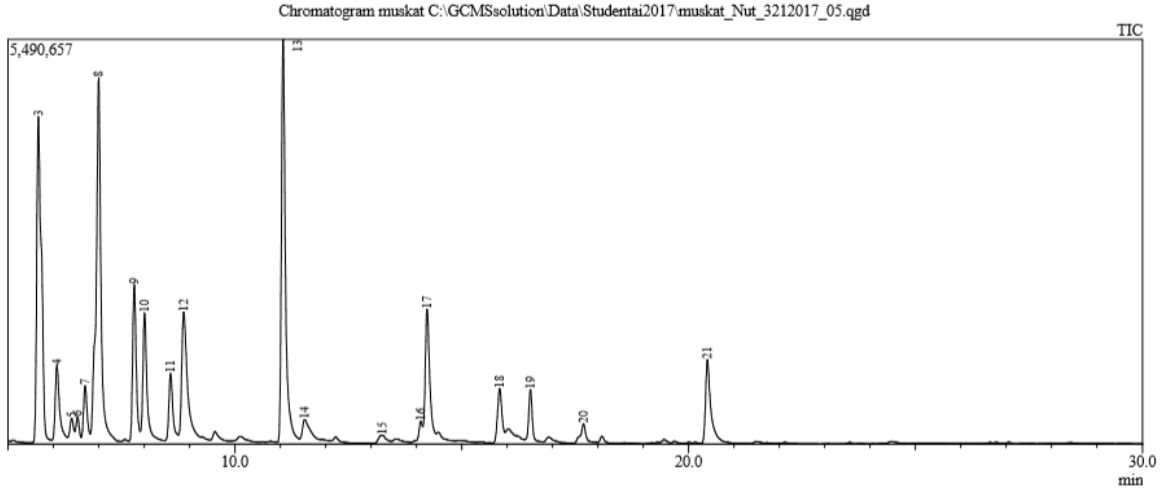
Chromatogram garlic C:\GCMSsolution\Data\Studentai2017\garlic_G_3212017_02.qgd



Peak Report TIC

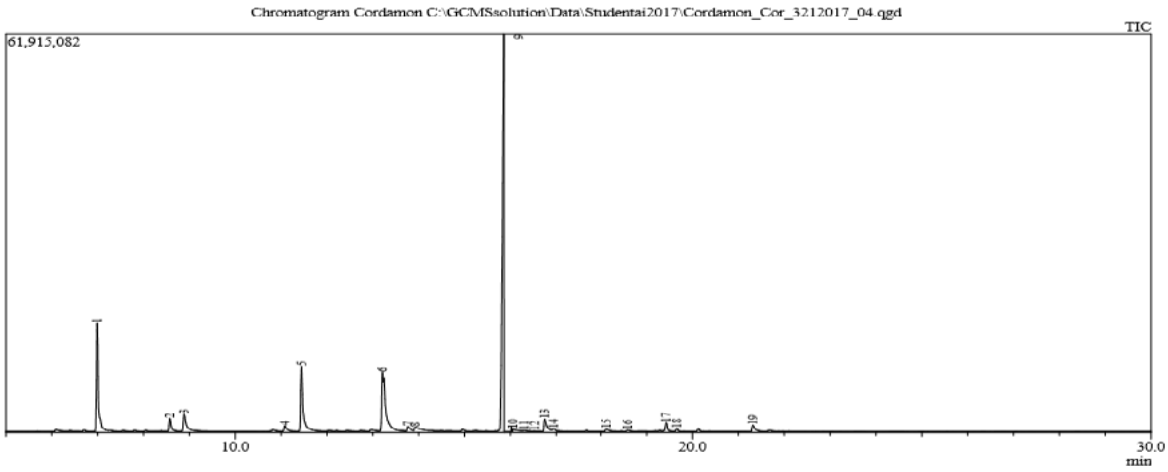
Peak#	R.Time	I.Time	F.Time	Area	Area%	Height	Height%	A/H	Mark	Name
1	4.559	4.425	4.975	1298834	3.88	145431	5.14	8.93		Thiophene, tetrahydro-, 1,1-dioxide
2	5.438	5.300	5.692	1116188	3.33	131537	4.65	8.49		Methoxymethyl isothiocyanate
3	8.411	8.142	8.717	4598482	13.73	442702	15.65	10.39		Tetrasulfide <diallyl->
4	8.983	8.717	9.475	5405495	16.14	386793	13.68	13.98	V	Propyl cyanide
5	10.113	9.975	10.383	410845	1.23	41428	1.46	9.92	V	Allyl methyl trisulfide
6	11.416	11.233	11.692	4993100	14.91	473500	16.74	10.55	V	Thiophene <2,5-dimethyl->
7	11.780	11.692	11.858	631148	1.88	68329	2.42	9.24	V	1,2,3-Thiadiazole, 5-methyl-
8	12.103	11.858	12.958	7604426	22.70	664035	23.48	11.45	V	Thiazole <2,4-dimethyl->
9	14.669	14.508	14.933	440232	1.31	39727	1.40	11.08		Disulfide <allyl->
10	15.380	15.150	15.575	568800	1.70	40675	1.44	13.98		Sulfide <diallyl->
11	15.995	15.825	17.050	4910601	14.66	270582	9.57	18.15	V	Menthalactone
12	24.440	24.125	24.808	494487	1.48	28380	1.00	17.42		Tridecane <n->
13	26.192	26.042	26.450	709979	2.12	67721	2.39	10.48		Pent-3-enoate <ethyl-, 2-methyl->
14	26.607	26.450	26.742	314844	0.94	27448	0.97	11.47	V	Butanoate <2-methyl-, hexyl->
				33497461	100.00	2828288	100.00			

Chromatogram muskat C:\GCMSsolution\Data\Studentai2017\muskat_Nut_3212017_05.qgd



Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height	Height%	A/H	Mark	Name
1	4.670	4.500	4.733	1833406	0.87	361355	1.18	5.07	V	Thujene <alpha->
2	4.807	4.733	5.042	5714065	2.72	1159973	3.78	4.93	V	Pinene <alpha->
3	5.669	5.492	5.958	31601666	15.03	4425074	14.43	7.14		Sabinene
4	6.079	5.958	6.308	7324531	3.48	1043178	3.40	7.02	V	Myrcene
5	6.404	6.308	6.467	1905997	0.91	324286	1.06	5.88	V	Phellandrene <alpha->
6	6.534	6.467	6.617	1851552	0.88	341820	1.11	5.42	V	Carene <delta-3->
7	6.701	6.617	6.817	4457955	2.12	765847	2.50	5.82	V	Terpinene <alpha->
8	6.998	6.817	7.483	36697117	17.45	4938706	16.11	7.43	V	Ocimene <(Z)-, beta->
9	7.783	7.667	7.917	11680683	5.55	2127088	6.94	5.49	V	Terpinene <gamma->
10	8.011	7.917	8.458	10983658	5.22	1740612	5.68	6.31	V	Sabinene hydrate <trans->
11	8.584	8.458	8.758	5554354	2.64	912601	2.98	6.09		Terpinolene
12	8.874	8.758	9.458	15769122	7.50	1743569	5.69	9.04	V	Sabinene hydrate <trans->
13	11.063	10.933	12.092	34527278	16.42	5461536	17.81	6.32	S	Terpinen-4-ol
14	11.541	11.425	11.875	2886701	1.37	267049	0.87	10.81	T	Terpineol <alpha->
15	13.246	12.975	13.442	1227436	0.58	106388	0.35	11.54		Linalyl formate
16	14.099	13.875	14.142	1560981	0.74	282837	0.92	5.52		Bornyl acetate
17	14.237	14.142	15.033	13691923	6.51	1800729	5.87	7.60	SV	Safrole
18	15.836	15.683	15.958	5160200	2.45	738314	2.41	6.99		Terpinyl acetate <alpha->
19	16.514	16.383	16.800	4003307	1.90	715058	2.33	5.60	V	Copaene <alpha->
20	17.685	17.458	17.992	2322632	1.10	263921	0.86	8.80	S	Caryophyllene <(E)->
21	20.414	20.258	21.058	9520209	4.53	1137427	3.71	8.37		Myristicin
				210274773	100.00	30657368	100.00			



Peak Report TIC

Peak#	R.Time	I.Time	F.Time	Area	Area%	Height	Height%	A/H	Mark	Name
1	6.991	6.942	7.508	46902004	12.76	16787084	14.97	2.79	SV	Limonene
2	8.577	8.500	8.825	6613478	1.80	2012173	1.79	3.29	V	Terpinolene
3	8.887	8.825	9.525	13437075	3.66	2626518	2.34	5.12	SV	Linalool
4	11.084	11.033	11.400	4180080	1.14	858520	0.77	4.87	SV	Terpinen-4-ol
5	11.448	11.400	12.367	36809438	10.02	10101382	9.01	3.64	SV	Terpineol <alpha->
6	13.212	13.150	14.783	55124860	15.00	9133333	8.14	6.04	SV	Linalyl acetate
7	13.770	13.725	13.892	3027429	0.82	665763	0.59	4.55	T	Geranial
8	13.926	13.892	14.267	5656590	1.54	544244	0.49	10.39	TV	Cinnamaldehyde <(E)->
9	15.855	15.700	15.875	146271624	39.80	61899836	55.19	2.36	V	Terpinyl acetate <alpha->
10	16.061	16.042	16.250	4678468	1.27	524189	0.47	8.93	V	Limonene oxide <cis->
11	16.303	16.250	16.492	3854842	1.05	324609	0.29	11.88	V	Lavandulyl acetate
12	16.532	16.492	16.708	2617361	0.71	233556	0.21	11.21	V	Copaene <alpha->
13	16.751	16.708	16.883	9586950	2.61	2064791	1.84	4.64	V	Geranyl acetate
14	16.948	16.883	17.200	5475994	1.49	535055	0.48	10.23	V	Elemene <beta->
15	18.124	18.050	18.417	3182517	1.41	523609	0.47	9.90	V	Terpinyl formate <alpha->
16	18.587	18.517	18.792	3174824	0.86	323535	0.29	9.81	V	Isopulegyl acetate
17	19.430	19.333	19.583	6290057	1.71	1447047	1.29	4.35	V	Selinene <beta->
18	19.663	19.583	19.792	2756114	0.75	517702	0.46	5.32	V	Selinene <alpha->
19	21.319	21.267	21.592	5858140	1.59	1037022	0.92	5.65	V	Nerolidol <(E)->
				367497845	100.00	112160168	100.00			

It is difficult to part for the analytical researcher to identify and quantify all the active chemical compounds which are present in spices, but we have successfully analysed this with the help of GC-MS. In an average more than ten active chemical compound, we have identified and quantified in each spice. The founded chemical compounds have pharmacological property, the garlic has 33 types of sulphur compounds and this sulphur compounds show anti-bacterial activity [5]. Meanwhile, in the future research purpose, we can consider this data.

4. CONCLUSION

The study shows that GC-MS is the efficient and very successful method for the identification and quantification of chemical active from the spice plant. From the identified compounds, we can easily target particular one or more compounds, which has the more and vital pharmacological effect.

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