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Investigation of Phytochemical Constituents of Tobacco (*Nicotiana Tobacum* L.) Methanol Extract

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Abstract

Tobacco is reported to be consists of more than 2000 compounds, and these constituents would change during the process of curing and combustion. The leaves and juice were much used for skin disorders in earlier generation. Since tobacco is the most important avoidable plant because of large number of premature death and diseases in the world due to cancer. The tobacco plant is more seen as destructive plant than as herb. Herein we interested in studying the phytochemical constituents present in the methanol extract of tobacco plant commonly available in Coimbatore district, Tamilnadu, India. About 36 compounds were obtained and most of them are found to be already reported carcinogens.

Introduction

India is the second largest producer of tobacco in the world. Different tobacco products are devised for the use of smoked tobacco. Smokeless tobacco is consumed without burning. So we interested in investigating the phytoconstituents using GC-MS analysis of the methanol extract of *Nicotiana Tobacum L.*

Materials and Methods

Reagent grade methanol is purchased from Merck and used without further purification. The tobacco material was identified and authenticated by the faculties of Post Graduate Department of Botany, Nallamuthu Gounder Mahalingam College, Pollachi, Coimbatore, Tamilnadu.

Methanol extraction

The dry shaded *Nicotianatabacum* leaves were collected and taken into the dry round bottom flask. Methanol was added into the round bottom flask [1,2]. The sample was heated for 3 hours at 40ºC in a mandle. The mother -liquid of the sample was collected in a beaker through what man filter paper. The sample was filtered and concentrated [3]. Then it is allowed to cool, storied in a container and kept in freezer for further investigation.

GC-MS analysis

Gas Chromatography Mass Spectrometry (GC/MS) is an instrumental technique, comprising a gas chromatograph (GC) coupled to a mass spectrometer (MS), by which complex mixtures of chemicals may be separated, identified and quantified [4,5]. GC-MS analysis of extract was carried out on Gas Chromatograph interfaced to a Mass Spectrometer equipped with a DB-5 MS capillary standard non-polar column of 30 m length, 0.25 µm thickness. Helium was used as a carrier gas at a constant flow of 0.1 ml/min and the temperature was 350ºC for 20 minutes. The sample of 100 ml was dissolved in 1ml of methanol and injected with split less mode. Mass spectra were recovered over 50-500 amu range with electron impact ionization energy 70eV, where injector and MS transfer line temperature were set at 230ºC and 280ºC respectively.

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Resultsand Discussion

GC-MS Analysis of Nicotiana Tabacum methanol extract

The methanol extract is subjected to GC-MS analysis. The components were identified by comparison of the retention time of the GC peaks with those obtained using NIFT library, analysis revealed that the presence of 36 compounds[6]. The major compounds are 1-Butyl(dimethyl)silyl oxy propane (1.187), Silane, diethoxy dimethyl (1.992), Ethanol, 2-(2- ethoxyethoxy) (1.083), 3(methylthio)-1-propanol (1.123), 2-propanol,1,1'-oxybis (0.760). The Minor compounds are 9,12,15- octadecatrienoic acid (z,z,z)(1.002), 11,13-dimethyl – 12-tetradecen-1-ol-acetate (0.759), 10,13-octadecadiynoic acid, methyl ester(0.519), Acetic acid(1,2,3,4,5,6,7,8-octahydro-3,8,8-trimethyl naphtha-2-yl)methyl ester (0.900), Hexanedioic acid, bis(2-ethylhexyl) ester (0.646) in Table 1 and Figure 1.

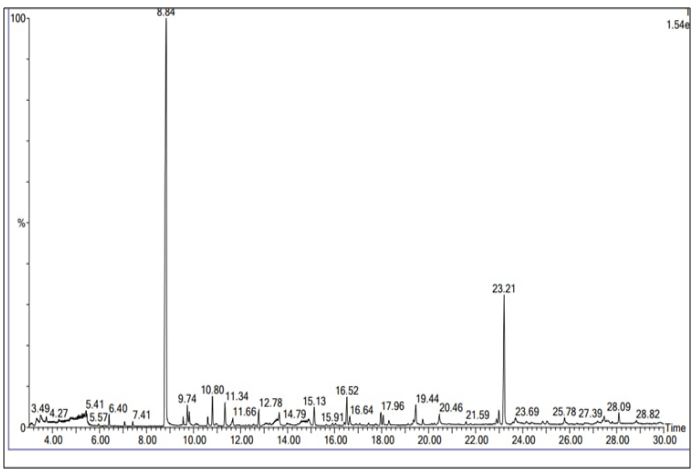

Figure 1: GC-MS Chromatogram of methanol extract of *Nicotiana tabacum.*

Table 1: Chemical composition of methanol extract of *Nicotiana tabacum*.

S.NO	NAME OF THE COMPOUND	MOLECULAR FORMULA	TOTAL PERCENTAGE	RETENTION TIME
1	1-Butyl (dimethyl)silyl oxy propane	$C_9H_{22}OSi$	1.187	3.334
2	Silane, diethoxy dimethyl	$C_6H_{16}O_2Si$	1.992	3.489
3	Ethanol, 2-(2- ethoxyethoxy)	$C_6H_{14}O_3$	1.083	3.744
4	3(methylthio)-1-propanol	$C_4H_{10}OS$	1.123	3.889
5	2-propanol,1,1'-oxybis	$C_6H_{14}O_3$	0.760	4.059
6	2,6-pyridine dicarboxaldehyde, 3-(phenyl methoxy)-bis[Methyl(2-pyridyl)hydrazone]	$C_{26}H_{25}N_7O$	0.803	4.274
7	D- Galactonic acid, ϒ-lactone	$C_6H_{10}O_6$	1.313	4.809
8	Methanamine, N-methoxy	C_2H_7NO	1.145	4.889
9	Methane, nitro	CH_3NO_2	1.104	5.104
10	1-propanol	C_3H_8O	0.599	5.139
11	Urea	CH_4N_2O	1.213	5.359
12	Phenylethyl alcohol	$C_8H_{10}O$	1.710	5.439
13	Pyridine,3-(1-methyl-2-pyrrolidinyl)-(s)	$C_{10}H_{14}N_2$	37.574	8.836
14	Anabasine	$C_{10}H_{14}N_2$	1.509	10.802
15	1,2,3,6-tetra hydro-2,3'-bipyridine	$C_{10}H_{14}N_2$	1.214	11.337
16	2,3'-dipyridyl	$C_{10}H_8N_2$	0.592	11.662
17	Diethyl phthalate	$C_{12}H_{14}O_4$	0.801	12.777
18	Phthalic acid,octyl 2- propyl pentyl ester	$C_{24}H_{38}O_4$	1.078	13.107
19	Phthalic acid, di (2-propyl pentyl) ester	$C_{24}H_{38}O_4$	1.440	13.568
20	Benzophenone	$C_{13}H_{10}O$	1.038	13.648
21	1,2-benzene dicarboxylic acid, dipropyl ester	$C_{14}H_{18}O_4$	0.710	14.713
22	Diisooctyl phthalate	$C_{24}H_{38}O_4$	0.856	14.893
23	Cotinine	$C_{10}H_{12}N_2O$	1.120	15.133
24	Benzyl benzoate	$C_{14}H_{12}O_2$	1.573	16.524
25	3-ethyl-3,4- dihydro 2(1H) quinoxalinone	$C_{10}H_{12}N_2O$	0.565	16.644
26	neophytadiene	$C_{20}H_{38}$	0.677	17.964
27	2' pentadecanone, 6,10,14 trimethyl	$C_{18}H_{36}O$	0.545	18.669
28	Bicyclo[4.4.0]dec 2 –ene-4-ol, 2-methyl-9(prop-1-en-3-ol-2-yl)	$C_{15}H_{24}O_2$	1.388	19.455
29	n-hexadecanoic acid	$C_{16}H_{32}O_2$	1.088	20.455
30	9,12,15- octadecatrienioc acid, methyl ester(z,z,z)	$C_{19}H_{32}O_2$	0.793	22.986
31	Phytol	$C_{20}H_{40}O$	8.488	23.211
32	9,12,15- octadecatrienoic acid (z,z,z)	$C_{18}H_{30}O_2$	1.002	23.687
33	11,13-dimethyl – 12-tetradecen-1-ol-acetate	$C_{18}H_{34}O_2$	0.759	25.778
34	10,13-octadecadiynoic acid, methyl ester	$C_{19}H_{30}O_2$	0.519	27.183
35	Acetic acid(1,2,3,4,5,6,7,8-octahydro-3,8,8-trimethyl naphtha-2-yl)methyl ester	$C_{16}H_{26}O_2$	0.900	27.463
36	Hexanedioic acid, bis(2-ethylhexyl) ester	$C_{22}H_{42}O_4$	0.646	28.093

Conclusion

The Phytoconstituents from methanol extract of *Nicotianatabacum* was analysed by GC-MS method. Results revealed that around 36 constituents were present in the extract, Most of the chemical constituents obtained were found to be known carcinogens. The product is toxic to the cells. So it is highly recommended for further cytotoxicity analysis.

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