STUDY OF NICOTIANA TABACUM L EXTRACTION, BY METHODS OF LIQUID AND SUPERCRITICAL FLUID EXTRACTION

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*The article presents a comparative analysis of various methods and conditions of extraction from the plant *Nicotiana tabacum* L, harvested in the Almaty region of the Republic of Kazakhstan in 2018. The study of extracts obtained by the traditional method (Extraction using non-polar organic solvents at atmospheric pressure) of solvent extraction in particular hexane, and the method of supercritical fluid extraction, carbon dioxide, under various conditions of the process. Three different supercritical extraction modes were selected, namely: at a relatively low pressure of 120 bar and a low temperature of 28°C, at an elevated temperature with the same pressure (60°C, 120 bar) and at an elevated temperature and pressure (80°C, 170 bar). As a result of studying the obtained extracts by GC-MS, their qualitative and quantitative composition was identified. It has been established that the largest number of various compounds is extracted by supercritical fluid extraction at a relatively low pressure of 150 bar for this method and a low temperature of 28°C, but the most popular compound in the studied raw material, nicotine, is extracted with approximately the efficiency similar to the classical method. To increase the yield of the target component, the extraction conditions were changed. As a result, a significant increase in the degree of extraction of the target product was achieved under the following conditions: pressure 120 bar, temperature 60°C. Under these conditions, the degree of extraction was 47.40%. Conducted to control and more fully study the laws of the extraction process (at elevated pressure) showed less efficiency, only 12.29%. This result is lower in efficiency than the classical extraction method. From the data obtained, it follows that for the extraction of nicotine, a substance belonging to the class of alkaloids and which is resistant to thermal destruction, preferred and most effective, is the mode of supercritical extraction at a temperature of 60°C and a pressure of 120 bar.*

Key words: *Nicotiana tabacum* L, liquid extraction, supercritical fluid extraction, GC-MS, nicotine

INTRODUCTION

It is known that the composition of plant extracts directly depends on many factors, such as the temperature of the extraction process, the extraction time, the ratio of plant raw material / extractant and, of course, the main factor is the nature of the solvent or solvent system used.

Depending on the tasks to be solved, a rich arsenal of organic solvents such as hexane, chloroform or benzene is used to extract biologically active substances.

However, most organic solvents are highly toxic compounds; exhibit an accumulation effect, which requires the introduction of additional stages of cleaning the drug substance and additional methods of quality control of such preparations. These measures lead to an inevitable rise in price of the final product. In addition, a significant part of organic solvents are in one way or another petrochemical products, their price will inevitably increase in the future.

In this regard, the last decades are actively studying and developing new methods for extracting complexes of biologically active substances from plant materials.

Supercritical extraction methods turned out to be very effective and have a number of advantages.

Supercritical fluids have been investigated since the last century, and at first the use of supercritical toluene in the processing of shale oil in the 1970s was of the greatest commercial interest. Supercritical water is also being investigated as a means of destroying toxic waste and as an exotic synthesis environment.

In the field of chemistry in the processing of plant materials, supercritical carbon dioxide, the critical temperature of which is 31°C, is of the greatest interest. Biological materials can be processed at 35°C, which contributes to their safety from thermal destruction. The density of supercritical CO₂ at a pressure of about 200 bar is similar in efficiency to hexane, and the solvation characteristics during extraction are also similar to hexane; thus, it acts as a non-polar solvent. [1]

The main advantage is that a slight decrease in temperature, or a slightly greater decrease in pressure, will lead to an almost complete precipitation of the target component.

Moreover, under normal conditions, carbon dioxide is a gas, so after the extraction process is completed; it becomes possible to obtain extracts with no solvent at all.

Examples of industrial products obtained using supercritical technologies include decaffeinated coffee,

cholesterol-free oil, lean meat, rose oil, etc. The solvation characteristics of supercritical CO₂ can be modified by adding a co-solvent such as ethanol, which can significantly increase the efficiency of extraction, however, the solvent residue in the product somewhat levels the main advantage of the process, which consists in the complete absence of impurities in the final extract [2,3].

As a plant raw material, we chose the plant *Nicotiana tabacum* l, due to the high content of biologically active substances in tobacco leaves, in particular, saturated and unsaturated fatty acids, fat-soluble vitamins, waxes, terpenes, terpenoids, pigments, alkaloids, essential oils and phytosterols, showing growth-regulating, fungicidal and insecticidal activity [4-15].

It should also be noted that due to a drop in demand for tobacco products, there is a release of a large amount of high-quality plant materials.

For comparison, two extraction methods were chosen, classical liquid extraction with hexane and supercritical extraction with carbon dioxide.

Objective: study of the qualitative and quantitative composition of the obtained extracts.

Materials and methods

The classical method of extraction (Extraction using non-polar organic solvents at atmospheric pressure) was carried out with hexane in the ratio of raw materials \ extractant 1:10 with infusion for 72 hours at a temperature of 28°C.

The supercritical extraction method was carried out on a Thar SFE-1000 CO₂-extraction unit under the following conditions: the temperature range in the reactor is 28-80°C, the pressure of CO₂ is 120-170 bar.

The obtained extract was investigated by gas chromatography on an Agilent Technologies 7890N / 5973N GC / MS gas chromatograph with a mass selective detector under the following conditions: a DB-35MS column (30 mx 250 mm x 0.25 mm) was used, the helium carrier gas velocity 1 ml/min The chromatographic temperature is programmed from 40°C (holding 0 min) to 300°C with a heating rate of 5°C / min (holding 5 min). Detection is carried out in the SCAN m / z mode 34-800. The software Agilent MSD ChemStation (version 1701EA) was used to control the gas chromatography system, record and process the obtained results and data. Data processing included determination of retention times, peak areas, as well as processing of spectral information obtained using a mass spectrometric detector. To decode the mass spectra obtained, the libraries Wiley 7th edition and NIST'02 were used (the total number of spectra in the libraries is more than 550 thousand) [16,17].

Results and discussion

In the course of the work, 7 extracts were obtained under various methods and conditions of extraction.

The data presented in tables 1-7

Table 1: The results of the study of hexane extract of the plant *Nicotiana tabacum* l by the method of chromatography-mass spectrometry

№	Retention time , min	Name of compounds	Content %
1	7,84	Sorbicacid	8,80
2	9,54	Benzoicacid	12,22
3	10,91	Tetradecane	0,73
4	11,54	Heptadecane, 4-methyl	0,14
5	11,81	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) - (E)	1,00
6	12,23	Pentadecane	1,04
7	12,44	Nicotine	19,52
8	12,91	1-dodecanol	0,27
9	13,09	5,9-indian-2-one, 6,10-dimethyl	0,35
10	13,49	Hexadecane	1,20
11	13,88	Quinoline, 1,2-dihydro-2,2,4-trimethyl	0,29
12	14,49	Pentadecane, 2,6,10,14-tetramethyl	0,39
13	14,68	Heptadecane	1,08
14	15,45	Megastigmasteron	0,83
15	15,70	2 (4H) -benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)	0,87
16	15,81	Octadecane	1,23
17	16,07	3-hydroxy-β-damascon	0,64
18	16,45	3,7,11,15-tetramethyl-2-hexadecen-1-ol	2,89
19	16,51	2-cyclohexen-1-one, 4- (3-hydroxy-1-butenyl) -3,5,5-trimethyl-	2,03
20	17,00	Nonadecane	1,49
21	17,45	2-cyclohexen-1-one, 4- (3-hydroxybutyl) -3,5,5-trimethyl	0,53
22	18,07	Bicyclo [3.1.1] hept-2-ene, 2,2'- (1,2-ethanediyl) bis [6,6-dimethyl]	0,26
23	18,48	Heptadecane, 9-hexyl-	0,46
24	18,58	Hexadecanoicacid, methylester	0,70

25	19,15	7-isopropenyl-1,4a-dimethyl-4,4a,5,6,7,8-hexahydro-3H-naphthalen-2-one	0,36
26	19,50	n-hexadecanoicacid	0,73
27	20,36	Cyclodecacyclotetradecene, 14,15-didehydro - 1, 4, 5, 8, 9, 10, 11, 12, 13, 16, 17, 18, 19, 20 - tetradecahydro	2,09
28	23,79	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12-(1-methylethyl)	14,49
29	24,71	Retinol	13,78
30	26,37	1-heptacantanol	7,17
31	26,59	3,3a-epoxydicyclopenta [a, d] cyclooctane-4 β -ol, 9,10a-dimethyl-6-methylene-3 β -isopropyl	2,42
Sum			100,00

From table 1 it can be seen that the hexane extract from the plant *Nicotiana tabacum* I revealed a large (19.52%) nicotine content of the natural alkaloid of the pyridine series, typical of the solanaceous family plants (*Solanaceae*) to which the object under study belongs. It should be noted that nicotine is often the target component in the processing of tobacco and the effectiveness of the extraction process is determined by its amount and completeness of extraction [18-23].

Found a high content of organic acids such as sorbic acid and benzoic acid, a significant content of hydrocarbons with a total content of 9.38%, a high content of vitamin A, retinol (13.78%), content of 1-heptatriacontanol (7.17%), which is a growth regulating agent.

Table 2: Results of chromatography-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* I, obtained by the method of supercritical fluid CO₂ extraction under conditions of 150 bar 28°C

№	Retention time , min	Name of compounds	Content %
1	5,22	1-butanol	0,19
2	5,40	Benzene, (1-methylethyl)	0,20
3	5,58	Benzene, 1,3-dimethyl	0,06
4	5,63	Propane, 1- (1-methylethoxy)	0,14
5	5,71	Methanesulfonylchloride	0,14
6	5,88	1-butanol, 3-methyl	4,39

7	6,00	Cyclopentane, 1,2,4-trimethyl	0,05
8	6,05	Furan, 2-Pentyl	0,05
9	6,11	Hexanoicacid, ethylester	0,07
10	6,61	Ethanone, 2- (formyloxy) -1-phenyl	0,66
11	7,14	2-propanone, 1-hydroxy	0,09
12	7,36	Pyrazine, 2,5-dimethyl	0,05
13	7,43	Pyrazine, 2,6-dimethyl	0,45
14	7,51	5-Hepten-2-one, 6-methyl	0,04
15	7,63	1-hexanol	0,03
16	8,33	Ethanol, 2-butoxy	0,42
17	8,68	Octanoicacid, ethylester	0,07
18	8,86	1-hexanol, 2-ethyl	0,30
19	8,91	Aceticacid	4,08
20	9,21	furfural	0,07
21	9,37	1-hexanol, 2-ethyl	0,16
22	9,70	Propanoicacid	0,53
23	9,99	Nenanoicacid, ethylester	0,14
24	10,48	Methoxyaceticacid, 3-tridecyl ether	0,33
25	10,68	Propyleneglycol	0,38
26	11,16	Butaneacid	0,39
27	11,35	biturolactone	0,09
28	11,55	2-furanmethanol	0,10
29	11,79	Butane acid, 4- (1,1-dimethylethoxy) -3-hydroxy-, methyl ester, (R) -	0,12
30	11,90	heptadecane	0,08
31	12,37	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) -, (E)	0,78
32	12,52	Oxime-, methoxyphenyl	0,19
33	12,93	1,3-propandiol	0,08
34	12,99	Ethanone, 1- (4-methylphenyl)	0,10
35	13,06	Pentanoicacid, 3-methyl	0,68
36	13,23	1,2-propandiol, 3-methoxy	0,94
37	13,60	Hexanoicacid	0,32
38	13,76	5,9-undecane-2-one, 6,10-dimethyl-, (Z)	0,19
39	14,08	Nicotine	19,34

40	14,43	3,7,11,15-tetramethyl-2-hexadecen-1-ol	2,34
41	14,57	Glycolaldehydedimethylacetal	0,21
42	14,70	Malonicacid, 2-butyldecyl ester	0,18
43	14,85	1-dodecanol	0,16
44	15,01	maltol	0,16
45	15,08	Ethanone, 1- (1H-pyrrol-2-yl) -	0,24
46	15,16	Dodecylacrylate	0,88
47	15,28	Diethylpimelate	0,13
48	15,41	Phenol	0,20
49	15,72	7-Methyl-Z-tetradecene-1-ol	0,27
50	15,85	Octanoicacid	0,37
51	16,25	7-methoxy-2,2,4,8-tetramethylcyclo [5.3.1.0 (4.11)] undecane	0,72
52	16,36	Diethylsuberate	0,21
53	16,69	Sorbicacid	1,77
54	16,97	10-methyldodec-2-en-4-olide	0,95
55	17,58	megastigmasterson	1,54
56	18,05	Hexadecanoicacid, ethylester	0,84
57	18,58	1,2,3-propantriol, 1-acetate	2,44
58	19,43	Glycerol	12,27
59	20,03	5,9,13-pentadecatrien-2-one, 6,10,14-trimethyl-, (E, E) -	0,21
60	21,31	Benzoicacid	16,67
61	21,90	trans-dehydroandrostosterone, pentafluoropropionate	0,41
62	22,42	2,3'-dipyridyl	0,56
63	22,61	transdehydroandrostosterone, trifluoroacetate	0,44
64	23,38	Bicyclo [3.1.1] hept-2-ene, 2,2' - (1,2-ethanediyl) bis [6,6-dimethyl-	1,13
65	23,86	3-hydroxy- β -damascon	0,66

66	26,00	4,8,13-Cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	3,06
67	27,04	2-cyclohexen-1-one, 4- (3-hydroxy-1-butenyl) -3,5,5-trimethyl	3,29
68	29,92	Tetradecanoicacid	2,65
69	30,63	triacontan	5,79
70	30,90	Cyclopenta [a, d] cyclooctene-5-one, 1,2,3,3a, 4,5,6,8,9,9a, 10,10a-dodecahydro-7- (1-methylethyl) -1,9a- dimethyl-4-methylene	2,76
Sum			100,00

In the supercritical extract, a high content of organic acids, in particular, sorbic acid and benzoic acid, is noted; a greater number of compounds are also isolated. 31 compounds were identified in the hexane extract, while in the carbon-dioxide supercritical - 70 compounds. The content of the main product of extraction, namely nicotine, was 19.34%, which is almost identical to the content in the extract obtained by the traditional method of solvent extraction [24-29].

To increase the yield of nicotine, the conditions for supercritical extraction were changed, namely: the process temperature was increased from 28 to 60°C.

The results of the study, obtained under these conditions, the extract is presented in table 3.

*Table 3: Results of chromato-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* l obtained by the method of supercritical fluid CO₂ extraction under conditions of 150 bar 60°C*

№	Retention time , min	Name of compounds	Content %
1	6,334	2,4-hexadienoic acid, methylester	0,21
2	6,99	Benzylalcohol	0,37
3	10,829	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) -, (E) -	0,99
4	11,942	Nicotine	23,70
5	12,09	2,2-dimethyl-1- (3-oxobut-1-enyl) -cyclopentanecarboxydehyde	1,13
6	13,249	Cyclohexanone, 2,3-dimethyl-2- (3-oxobutyl)	0,58
7	13,392	Pyridine, 3- (3,4-dihydro-2H-pyrrol-5-yl) -	0,40

8	13,596	heptadecane	0,17
9	13,999	Diethylsuberate	0,52
10	14,209	Tricyclo [6.3.0.0 (1.5)] undec-2-en-4-one, 5,9-dimethyl-	0,38
11	14,417	megastigmasteron	0,94
12	14,695	2 (4H) -benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R) -	1,15
13	14,851	2,3'-dipyridyl	0,35
14	15,039	3-hydroxy- β -damascon	0,81
15	15,335	Fitol, acetate	2,81
16	15,525	2-cyclohexen-1-one, 4- (3-hydroxy-1-butenyl) -3,5,5-trimethyl-	4,47
17	16,228	] 2-Cyclohexen-1-one, 4- (3-hydroxybutyl) -3,5,5-trimethyl-	1,22
18	16,653	DL-xylitol, 1-benzoate	2,85
19	16,867	Hexadecanoicacid, methylester	0,53
20	17,484	Hexadecanoicacid, ethylester	3,09
21	17,579	n-hexadecanoicacid	3,54
22	18,301	Bicyclo [3.1.1] hept-2-ene, 2,2' - (1,2-ethanediyl) bis [6,6-dimethyl-	0,96
23	18,573	Dibutylphthalate	0,91
24	18,959	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	2,47
25	19,327	2-pentenoic acid, 5- (decahydro-5,5,8a-trimethyl-2-methylene-1-naphthalenyl) -3-methyl	5,45
26	19,728	Cyclopenta [a, d] cyclooctene-5-one, 1,2,3,3a, 4,5,6,8,9,9a, 10,10a-decahydro-7- (1-methylethyl) -1,9a- dimethyl-4-methylene	3,25
27	19,774	metasterone	0,98
28	20,575	2-pentenoic acid, 5- (decahydro-5,5,8a-trimethyl-2-methylene-1-naphthalenyl) -3-methyl	1,20

29	21,185	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	5,44
30	22,984	heptacosan	4,97
31	23,114	Glycerin 1-palmitate	3,75
32	23,673	Octacosan	1,12
33	24,083	Hexacosan, 9 octyl	0,85
34	24,443	nonacosan	4,05
35	25,097	triacontan	0,96
36	25,903	gentriacontan	5,40
37	26,697	dotriocontan	1,81
38	27,223	hexatriacontan	1,69
39	27,691	tritriacontan	3,08
40	28,041	28-Nor-17 β (H) -gopan	0,43
41	28,47	Tetracosane, 12-decyl-12-nonyl	0,42
42	29,009	17. Alpha. 21 β -28,30-bis-norpan	0,60
Sum			100,00

In the data presented in Table 3, there is a significant increase in the efficiency of nicotine extraction of 23.70%, while in previous processes the yield was less than 20%. Compared with supercritical extraction at low temperatures, in this case there is a smaller number of identified compounds - 42, against 70, besides a large amount of compounds belonging to groups of alcohols and ethers were found in the extract obtained in the process with heating besides nicotine.

To determine the optimal mode of extraction, an extraction process was carried out at elevated pressure and temperature, namely: the extraction conditions of 60°C were established at a pressure of 170 bar, the data obtained after analyzing the total extract are presented in Table 4.

*Table 4: Results of chromatography-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* / obtained by the method of supercritical fluid CO₂ extraction under conditions of 170 bar 60°C*

№	Retention time, min	Name of compounds	Content %
1	6,25	Aceticacidbutylester	14,36
2	8,29	ethylbenzene	1,78
3	14,84	Sorbicacid	17,56
4	16,57	Benzoicacid	28,81
5	18,55	1-isopropyl-4-methyl-1,4-cyclohexadiene	0,46

6	19,20	Nicotine	12,29
7	22,18	Styron	0,99
8	23,14	9 octadecen	2,44
9	23,21	2-cyclohexen-1-one, 4- (3-hydroxy-1-butenyl) -3,5,5-trimethyl-, [R- [R *, R * - (E)]	0,82
10	24,68	Hexadecanoicacidmethylester	0,58
11	26,37	Dibutylphthalate	0,50
12	26,67	diethylbenzene	0,72
13	26,73	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	0,71
14	26,84	Methyl 8,11,14-heptadecathenoate	0,76
15	26,91	Cycloheptane, 4-methylene-1-methyl-2- (2-methyl-1-propen-1-yl) -1-vinyl-	1,40
16	27,08	Cyclohexanethanol, 4-methyl-β-methylene-	2,92
17	27,16	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	1,07
18	27,48	(R) - (-) - 14-methyl-8-hexadecen-1-ol	2,01
19	27,53	Ethanol, 1- (1-cyclohexenyl)	0,81
20	27,78	1-formyl-2,2,6-trimethyl-3- (3-methyl-but-2-enyl) -6-cyclohexane	1,05
21	27,94	3-carboxy-4-nitrophenyl disulfide	0,53
22	28,28	((2E, 4E) -3,7,11-trimethyldodecane-2,4,10-triene	0,34
23	28,33	4,8,13-Cyclotetradecatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	1,37
24	28,45	cyclopentanone	1,70
25	28,49	Cyclohexanethanol, 4-methyl-β-methylene-	1,71
26	28,89	2- (2-aminobutane-2-yl-diaz-enyl) -2-methylbutanethanitrile	1,63
27	31,64	Diisooctylphthalate	0,68
Sum			100,00

In the results of the analysis of the extract obtained at elevated pressure and temperature, we see a significantly smaller amount of extracted nicotine (12.29%), the total number of identified compounds is less, only 28, which can be explained by partial destruction or by a change in the dissolving ability of carbon dioxide.

Studied on the topic of literature the results indicated there indicate that in some cases the extraction proceeds more fully when the pressure is lowered and this leads to a change in the solubilizing indices of carbon dioxide. We decided to lower the pressure of the extraction process to 120 bar while maintaining the temperature at 60°C. The data presented in Table 5.

*Table 5: Results of chromatography-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* l obtained by the method of supercritical fluid CO₂ extraction under conditions of 120 bar 60°C*

№	Retention time , min	Name of compounds	Content %
1	6,25	1-butanol, 3-methyl-, acetate	0,02
2	6,49	3-heptanol	0,01
3	7,03	Ethanol, 2-butoxy-	0,17
4	8,53	Furan, 2-pentyl	0,01
5	8,83	Butaneacid, butylether	0,01
6	9,24	Cyclohexanol, 1-methyl-4- (1-methylethenyl) -, acetate	0,16
7	9,57	1-hexanol, 2-ethyl-	0,01
8	9,89	Benzaldehyde	0,01
9	9,99	Butanoicacid, 4-hydroxy-	0,01
10	11,77	Benzylalcohol	0,03
11	11,98	Pentanoicacid, 3-methyl-	0,37
12	12,36	Benzylacetatedigoxil	0,14
13	12,91	hexanoicanhydride	0,02
14	13,29	(R, S) -5-ethyl-6-methyl-3E-hepten-2-one	0,04
15	13,54	Hexanoicacid, anhydride	0,02
16	13,91	4-methyl-5H-furan-2-one	0,01
17	14,23	Octanoicacid, ethylester	0,01
18	14,51	1,1'-bicyclohexyl, 2- (2-methylpropyl) -, trans-	0,01
19	14,65	Phenylethylalcohol	0,06
20	14,99	3-Eicozen, (E) -	0,04
21	15,23	2-Hexadecene, 3,7,11,15-tetramethyl-, [R- [R *, R * - (E)]] -	0,03

22	15,45	1-decanol, 2-methyl-	0,02
23	15,64	4H-imidazol-4-one, 2-amino-1,5-dihydro	0,01
24	16,22	2-Isopropyl-5-oxohexanal	0,01
25	16,36	Ethanone, 1- (3-methyl-phenyl) -	0,01
26	17,72	tetradecane	0,06
27	18,27	2-buten-1-one, 1- (2,6,6-trimethyl-1-cyclohexen-1-yl)	0,01
28	19,73	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) -, (E) -	0,31
29	20,02	Propane, 2-isocyanato-2-methyl-	0,02
30	20,91	Nicotine	47,40
31	21,69	7-methoxy-2,2,4,8-tetramethyltricyclo [5.3.1.0 (4.11)] undecane	0,13
32	21,86	5,9-Undecadien-2-one, 6,10-dimethyl-, (E) -	0,02
33	22,72	hexadecane	0,19
34	22,87	1-dodecanol, 3,7,11-trimethyl-	0,01
35	23,14	Bottledhydroxytoluene	0,02
36	23,28	Phenol, 2,4-bis (1,1-dimethylethyl) -	0,02
37	23,89	Pyridine, 3- (3,4-dihydro-2H-pyrrol-5-yl) -	0,24
38	24,15	5-Isopropyl-6-methylhept-3,5-diene-2-ol	1,41
39	24,45	Pyridine, 3- (1-methyl-2-pyrrolidinyl) -, N-oxide, (2S) -	0,02
40	24,93	1-Heptatriacontanol	0,03
41	25,02	heptadecane	0,03
42	25,36	Nicotirin	0,32
43	25,45	trans-5-isopropyl-6,7-epoxy-8-hydroxy-8-methylnonan-2-one	0,01
44	25,51	tert-hexadecanethiol	0,02
45	25,62	Trichloroaceticacid, hexadecylether	0,03
46	25,70	1-hexadecansulfonyl	0,02
47	25,81	octacosanol	0,02
48	25,94	1-trikozen	0,04

49	26,12	5,6,6-trimethyl-5- (3-oxobut-1-enyl) -1-oxaspiro [2.5] octan-4-one	0,16
50	26,62	1-allyl-2-hydroxy-6-methylcyclohexanecarboxylic acid	0,07
51	26,88	2,3'-dipyridyl	0,03
52	27,24	Octadecane	0,23
53	27,33	Pentadecanal	0,14
54	27,59	4- (2,6,6-trimethyl-cyclohexen-1-enyl) butan-2-ol	0,03
55	27,82	6- (3-hydroxy-but-1-enyl) -1,5,5-trimethyl-7-oxabicyclo [4.1.0] heptan-2-ol	0,03
56	27,93	2,10-dodecadien-1-ol, 3,7,11-trimethyl-, (Z) -	0,11
57	28,61	phytol, acetate	10,87
58	29,05	Cedran, 8-propoxy-	0,07
59	29,13	Bicyclo [4.4.0] deca-5-ene-1-acetic acid	0,03
60	29,34	nonadecan	0,03
61	29,42	3-buten-2-one, 4- (4-hydroxy-2,2,6-trimethyl-7-oxabicyclo [4.1.0] hept-1-yl) -	0,02
62	29,63	2-pentadecanone, 6,10,14-trimethyl-	0,10
63	29,81	2-cyclohexen-1-one, 4- (3-hydroxybutyl) -3,5,5-trimethyl-	0,22
64	30,03	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	0,04
65	30,09	4a, 7,7,10a-tetramethyl-dodecabenzo [e] chromen-3-ol	0,05
66	31,09	Globulol	0,09
67	31,36	eicosane	0,38
68	31,49	2-butanone, 4- (2,6,6-trimethyl-1-cyclohexen-1-yl) -	0,18
69	31,67	3- (4,8,12-trimethyltridecyl) furan	0,08
70	31,78	1,4-bis (1'-hydroxycyclopentyl) -1,3-butadiyne	0,08
71	31,96	Cotinine	0,29
72	32,04	Scroleoxide	0,10

73	32,66	n-hexadecanoicacid	0,50
74	32,93	Cyclodecacyclotetradecene, 14,15- didehydro-1, 4,5,8,9,10,11,12,13,16,17 ,18,19,20-tetradecahydro	0,09
75	33,06	4a, 7,7,10a-tetramethyl-dodecahydro-benzo [e] chromen-3-one	0,10
76	33,84	12-Oxatetracyclo [5.2.1.1 (2,6) .1 (8,11)] dodecan-3-ol-10-one acetate	0,10
77	33,89	Bicyclo [3.1.1] hept-2-ene, 2,2 '- (1,2-ethanediyl) bis [6,6-dimethyl-	0,12
78	33,95	14-oxatricyclo [9..2.1.0 (1.10)] tetradecane, 2,6,6,10,11-pentamethyl-	0,06
79	34,62	1,2-benzenedicarboxylic acid, butyl-8-methylnonyl ether	0,15
80	35,06	sclareolid	1,42
81	35,15	Dokosan	0,22
82	35,57	1H-benzocyclohepten-7-ol, 2,3,4,4a, 5,6,7,8-octahydro-1,1,4a, 7-tetramethyl-, cis-	0,21
83	35,90	Isoaromadendrenepoxide	0,39
84	36,50	5- (7a-isopropenyl-4,5-dimethyl-octahydroinden-4-yl) -3-methyl-pent-2-en-1-ol	2,62
85	36,85	1-penten-3-one, 1- (2,6,6-trimethyl-1-cyclohexen-1-yl) -	2,89
86	37,46	8H-naphtho [1,2-b] pyran-8-one, 3-ethyl-dodecahydro-3,4a, 7,7,10a-pentamethyl-, [3R- (3α, 4aβ, 6aα, 10aβ, 10bα)] -	0,27
87	37,63	1-naphthalenepropanol, α-ethenyl decacidro-2-hydroxy-α, 2,5,5,8a-pentamethyl, [1R- [1α (R *), 2β, 4aβ, 8aα]]	0,29
88	37,90	5- (7a-isopropenyl-4,5-dimethyl-octahydroinden-4-yl) -3-methyl-penta-2,4-dien-1-ol	0,45

89	38,34	2,5,5,8a-tetramethyl-4-methylene-4a, 5,6,7,8,8a-hexahydro-4H-chromen	3,31
90	38,68	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	1,71
91	38,84	1-carbomethoxy-1,2,5,5-tetramethyl-cis-decalin (1R, 2S, 4aS, 8aS)	0,28
92	38,92	4a, 7,7,10a-Tctroamethyl-dodecabenzo [e] chromen-3-ol	0,23
93	39,08	isolonigifolmethylester	0,53
94	39,23	Hafilin	0,10
95	39,61	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	9,09
96	39,69	5- (1-Isopropenyl-4,5-dimethylbicyclo [4.3.0] nonan-5-yl) -3-methyl-2-pentenol acetate	0,86
97	39,93	1-carbomethoxy-1,2,5,5-tetramethyl-cis-decalin (1R, 2S, 4aS, 8aS)	0,85
98	40,24	Heptacosan	0,23
99	40,91	5,6,6-trimethyl-5- (3-oxobut-1-enyl) -1-ox-aspiro [2.5] octan-4-one	0,17
100	41,23	Tetracos-2,6,14,18,22-pentaene-10,11-diol, 2,6,10,15,19,23-hexamethyl-	0,10
101	41,55	Beganalcohol	0,78
102	41,87	2,5,5,8a-tetramethyl-4-methylene-4a, 5,6,7,8,8a-hexahydro-4H-chromen	0,26
103	41,98	1-methyl-3- (2,2,6-trimethylbicyclo [4.1.0] hept-1-yl) acetic acid ester	0,28
104	42,26	4a, 7,7,10a-tetramethyl-dodecahydro-benzo [e] chromen-3-one	0,39
105	43,04	1-Heptatriacontanol	0,28

106	43,55	1H-naphtho [2,1-b] pyran, 3-ethynyl-dodecahydro-3,4a, 7,7,10a-pentamethyl-, [3R- (3 α , 4 $\alpha\beta$, 6 $\alpha\alpha$, 10 $\alpha\beta$, 10 $\beta\alpha$)] -	0,22
107	43,82	Di (2-propylpentyl) phthalic acid ester	0,45
108	44,01	2- [4-methyl-6- (2,6,6-trimethyl-cyclohex-1-enyl) hex-1,3,5-trienyl] cyclohexeno-1-ene-1-carboxaldehyde	0,27
109	44,19	β -epi-shiobunol	0,21
110	44,37	(E) -5-isopropyl-8-hydroxy-8-methyl-non-6-en-2-one	0,23
111	44,53	5- (7 α -isopropenyl-4,5-dimethyl-octahydroinden-4-yl) -3-methyl-penta-2,4-dien-1-ol	0,07
112	45,64	Spiro [furan-2 (5H), 2' (1'H) -naphtho [2,1-b] furan] -5-one, 3'a, 4', 5', 5'a, 6', 7', 8', 9', 9'a, 9'b-decahydro-3,3'a, 6', 6', 9'a-pentamethyl-, [2'S- (2' α , 3'a α , 5 "Ap, 9'a α , 9'b β)] -	0,70
113	46,15	1-dimethyl (phenyl) siloxydecan	0,14
114	46,40	Squalene	0,42
115	47,13	Koprostan	0,04
116	47,23	tetratetracontan	0,14
117	48,12	5,8,9-endo-10-exo-tetramethyltricyclo [6.3.0.0 (5.11)] undecan-1-carboxylic acid	0,25
118	48,57	6,10,14,18,22- tetrakozapentaen-2-ol, 3-bromo-2,6,10, 15,19,23 - hexamethyl-, (all-E) -	0,13
119	51,30	28-Nor-17 β (H) -gopan	0,06
120	51,76	2,2,6,6-tetramethyl-4-piperidinyl ester decanedioic acid	0,85
121	52,06	DL- α -tocopherol	0,83
122	52,36	17. α ., 21 β -28,30-bisnorfan	0,11

123	52,60	17- (1,5-dimethylhex-yl) -10, 13-dimethyl-2,3,4,7, 8,9,10, 11,12,13,14,15,16,17 - tetradecahydro- 1H-cyclopenta [a] phenanthrene-3-ol	0,15
124	53,74	Lupan-3-he	0,06
125	54,67	stigmasterol	0,29
Sum			100,00

The data presented in Table 5 indicate that under conditions of supercritical fluid CO₂ extraction at a pressure of 120 bar and a temperature of 60°C, an extremely high yield of nicotine is achieved, which is 47.40%, and this is a very high figure for alkaloids.

Note that in addition to nicotine, its derivatives have been identified: cotinine and nicotirin (Fig.1).

Figure 1: Structural formulas of Cotinine and Nicotirin

In addition, in the extract obtained under these conditions, the largest number of compounds (125) was identified, but almost all of them, except for phytol acetate, are present in minor quantities.

These changes in composition are explained by the fact that in supercritical processes, pressure and temperature are of decisive importance not only on the completeness of extraction, but also on the properties of the extractant [30-34].

For a more detailed study of the influence of the temperature factor on the process of extracting biologically active substances from the aerial part of *Nicotiana tabacum* l, extraction was carried out at a pressure of 170 bar and a temperature of 80°C. The results are presented in table 6.

Table 6: Results of chromato-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* / obtained by the method of supercritical fluid CO₂ extraction under conditions of 170 bar 80°C

№	Retention time , min	Name of compounds	Content %
1	6,16	1-butanol, 3-methyl-, acetate	0,03
2	6,97	Ethanol, 2-butoxy-	0,29
3	7,22	Benzene, 1-ethyl-2-methyl-	0,02
4	7,57	ButylAcidHeptylEster	0,29
5	7,83	1,2-propandiol, 3-methoxy-	1,33
6	8,04	Pyrazine, 2,6-dimethyl-	0,10
7	8,31	4-cyclopenten-1,3-dione	0,07
8	8,49	Furan, 2-pentyl	0,02
9	8,79	butanoicacidbutylester	0,03
10	8,93	hexanoicacidethylester,	0,01
11	9,20	limonene	0,02
12	9,56	1-hexanol, 2-ethyl-	0,09
13	9,96	biturolactone	0,03
14	10,05	2 (5H) -furanone	0,02
15	11,73	Benzylalcohol	0,08
16	14,57	Sorbicacid	2,68
17	18,08	Benzoicacid	41,68
18	19,25	2H-pyran-2-one, 5,6-dihydro-6-pentyl-	0,19
19	19,45	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) -, (E) -	0,25
20	19,54	Butanoicacid 2,3-dihydroxypropyl ester	0,04
21	19,71	2-piperidinone	0,17
22	21,14	Nicotine	22,50
23	21,81	7-methoxy-2,2,4,8-tetramethyltricyclo [5.3.1.0 (4.11)] undecane	0,10
24	21,89	5,9-undecadien-2-one, 6,10-dimethyl-	0,06
25	22,38	Bicyclo [3.1.0] hexane-3-one	0,04
26	22,86	7-heptadecene, 1-chloro	0,05
27	24,13	Cyclohexanone, 2,3-dimethyl-2- (3-oxobutyl) -	0,25
28	24,73	2 pyridin-2-ylamino cyclohexyl ester of acetic acid	0,03

29	25,45	Nicotine	0,06
30	25,74	5,6-dimethyl-2-, benzimidazolion	0,05
31	26,66	2 (4H) -benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)	0,17
32	26,96	2,3'-dipyridyl	0,11
33	27,56	3-hydroxy-β-damascon	0,13
34	27,69	Megasigmastiron	0,42
35	27,83	Propyl 2,4-hexadiene carboxylate	0,55
36	28,40	2-cyclohexen-1-one, 4- (3-hydroxy-1-butenyl) -3,5,5-trimethyl-	2,19
37	29,53	1,2-benzenediol, 3,5-bis (1,1-dimethylethyl) -	0,04
38	30,04	Farnesylbromide	0,05
39	30,39	Phenol, 2,6-bis (1,1-dimethylethyl) -4-ethyl-	0,02
40	30,62	DL-xylitol, 1-benzoate	1,05
41	31,43	Hexadecanoicacid, methylester	0,19
42	31,66	Methyl ester of 3- (1-acetyl-2,2-dimethyl-5-oxocyclopentyl) acrylic acid	0,08
43	32,19	5,9,13-pentadecatrien-2-one, 6,10,14-trimethyl-, (E,E)-	0,23
44	32,34	2-cyclohexen-1-one, 4-hydroxy-3,5,5-trimethyl-4- (3-oxo-1-butenyl) -	0,09
45	32,72	n-hexadecanoicacid	1,74
46	33,23	Bayer 28,589	0,04
47	33,89	Bicyclo [3.1.1] hept-2-ene, 2,2' - (1,2-ethanediyl) bis [6,6-dimethyl-	0,44
48	34,62	hept-3-yl phthalic acid butyl ester,	0,15
49	35,24	Cyclopropanebutanoic acid, 2 - [[2 - [(2-pentylcyclopropyl) methyl] cyclopropyl] methyl] cyclopropyl] methyl] -, methyl ester	0,09
50	35,61	9,12,15-octadecataric acid, methyl ester, (Z, Z, Z) -	0,36
51	35,88	Resibufagenin	2,62
52	36,41	Oleicacid	0,33

53	36,66	1-Heptatriacontanol	1,41
54	36,75	Ethyl 6,9,12,15,18-genic-osapentaenoate	0,31
55	37,54	Card-20 (22) -enolide, 3,5,14,19-tetrahydroxy-, (3 β , 5 β)	0,40
56	38,05	Epiglobulol	0,33
57	38,64	3,3a-epoxydicyclopenta [a, d] cyclooctane-4 β -ol, 9,10a-dimethyl-6-methylene-3 β -isopropyl-	1,06
58	39,12	Pregn-4-ene-3,20-dione, 11-hydroxy-, (11 α) -	0,06
59	39,27	Dihydroartemisin	0,12
60	39,46	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12-(1-methylethyl) -	4,54
61	39,76	1-naphthalenecarboxylic acid, 5- (4-carboxy-3-methyl-3-butenyl) decahydro-1,4a-dimethyl-6-methylen, [1S- [1 α , 4 $\alpha\alpha$, 5 α (E), 8 $\alpha\beta$]] -	0,10
62	40,09	Fluoxymesterone	0,11
63	40,19	1b, 4a-Epoxy-2H-cyclopenta [3,4] cycloprop [8,9] cycoundec [1,2-b] oxyrene-5 (6H) -one, 7- (acetyloxy) decahydro-2,9,10- trihydroxy-3,6,8,8,10a-pentamethyl	0,23
64	40,62	Hexadeca-2,6,10,14-tetraen-1-ol, 3,7,11,16-tetramethyl-	0,09
65	41,52	17-pentatriaconten	0,21
66	42,20	(1S, 2E, 4S, 5R, 7E, 11E) -Cembra-2,7,11-triene-4,5-diol	0,32
67	43,34	Heptacosan	0,22
68	43,52	2-hydroxy-1- (hydroxymethyl) ethyl ester hexadecanoic acid	2,60
69	43,81	Di (2-propylpentyl) phthalic acid ester	0,64
70	44,04	Vidrolhydroxyether	0,31

71	45,59	Spiro [furan-2 (5H), 2 '(1'H) -naphto [2,1-b] furan] -5-one, 3'a, 4', 5', 5'a, 6', 7', 8', 9', 9'a, 9'b-decahydro-3,3'a, 6', 6', 9'a-pentamethyl-, [2'S- (2' α , 3' $\alpha\alpha$, 5 "Ap, 9' $\alpha\alpha$, 9'b β)] -	0,15
72	46,22	Hexacosan	0,18
73	46,38	Squalene	0,23
74	46,65	(Z) -, 2-hydroxy-1- (hydroxymethyl) ethyl ester of 9-octadecenoic acid	0,23
75	47,02	2-hydroxy-1- (hydroxymethyl) ethyl linolenic acid ester (Z, Z, Z) -	0,57
76	47,21	2-methyloctacosan	0,28
77	48,35	tetratetracontan	0,22
78	48,93	tetratriacontan	0,47
79	49,86	Triacontane, 1-bromine	0,26
80	51,47	tetratetracontan	0,23
81	52,05	Vitamin E	0,84
82	52,35	Phenanthrene, 9-dodecyltetradecahydro	0,15
83	52,61	17- (1,5-dimethylhexyl) -10,13-dimethyl-2,3,4,7,8,9, 10,11,12,13,14,15,16,17 -tetradecahydro- 1H-cyclopenta [a] phenanthrene-3-ol	0,34
84	54,69	stigmasterol	0,78
85	55,74	γ -sitosterol	0,34
Sum			100,00

The content of table 6 indicates that the analysis of the extract of the plant *Nicotiana tabacum* l by the method of supercritical fluid CO₂ extraction under conditions of 170 bar 80°C helps to identify a much larger number of compounds (85), to obtain benzoic acid with a high content (41.68%). However, the amount of nicotine produced under these conditions is 22.50%. This result for nicotine is comparable to the method of extraction with hexane and extraction at more, which is a factor of thermal destruction of low pressures.

Thus, we can conclude that these conditions are not optimal for the efficient extraction of nicotine. However, the selectivity of the process increases, namely: the dominance nicotine and benzoic acid with a large number of other substances found in minor or trace amounts.

At the end of this phase of the work, an extraction was carried out under a pressure of 120 bar at a temperature of 80°C in order to establish the effect of a temperature rise at lower pressures. The data presented in table 7.

Table 7: Results of chromato-mass spectrometric analysis of the extract of the plant *Nicotiana tabacum* / obtained by the method of supercritical fluid CO₂ extraction under conditions of 120 bar 80°C

№	Retention time , min	Name of compounds	Content %
1	6,89	Ethanol, 2-butoxy-	0,07
2	7,12	3-hexene-2-one, 5-methyl-	0,01
3	8,08	2-heptanone, 6-methyl-	0,01
4	8,76	Butanoicacidbutylester	0,01
5	9,17	Cyclohexene, 1-methyl-5- (1-methylethenyl) -, (R) -	0,01
6	9,24	exo-2-hydroxyenol	0,01
7	9,39	3-methyl butanoicacid	0,07
8	12,32	Pentanoicacid, 3-methyl-	0,26
9	12,91	Hexanoicacid, anhydride	0,04
10	13,27	(R, S) -5-ethyl-6-methyl-3E-hepten-2-one	0,09
11	14,72	Phenylethylalcohol	0,05
12	15,00	Cyclohexane, 1-ethyl-2-propyl	0,02
13	15,23	3-Eicozen, (E) -	0,02
14	15,46	2-Isopropyl-5-methyl-1-heptanol	0,01
15	17,72	tetradecane	0,05
16	19,74	6,8-nonadien-2-one, 8-methyl-5- (1-methylethyl) -, (E) -	0,21
17	20,92	Nicotine	25,87
18	21,73	7-methoxy-2,2,4,8-tetramethyltricyclo [5.3.1.0 (4.11)] undecane	0,26
19	22,73	hexadecane	0,14
20	23,16	Bottledhydroxytoluene	0,05
21	23,30	Phenol, 2,4-bis (1,1-dimethylethyl) -	0,03
22	23,93	Pyridine, 3- (3,4-dihydro-2H-pyrrol-5-yl) -	0,15
23	24,16	Cyclohexanone, 2,3-dimethyl-2- (3-oxobutyl) -	0,94
24	24,50	nicotine N-oxide	0,08
25	25,03	2 (1H) -naphthalenone, 3,4,4a, 5,6,7,8,8a-octa-hydro-5a-hydroxy-4a, 7,7-trimethyl-, acetate	0,13
26	25,38	nicotine	0,23

27	25,69	1-hexadecanol, 2-methyl-	0,06
28	25,93	1-dodecanol, 2-octyl-	0,09
29	26,13	5,6,6-trimethyl-5- (3-oxobut-1-enyl) -1-oxaspiro [2.5] octan-4-one	0,12
30	26,32	1-genikosyl formate	0,06
31	26,63	6-ethyl-3-octyl octanoicacid	0,08
32	26,91	2,3'-dipyridyl	0,06
33	27,25	Octadecane	0,30
34	27,60	Bicyclo [4.3.0] nonane, 2,2,6,7-tetramethyl-7-hydroxy-	0,07
35	27,94	n-Nonylsuccinicanhydride	0,19
36	28,60	Fitol, acetate	5,85
37	29,06	2-butenal, 2-methyl-4- (2,6,6-trimethyl-1-cyclohexen-1-yl) -	0,12
38	29,64	2-pentadecanone, 6,10,14-trimethyl-	0,26
39	29,83	2-cyclohexen-1-one, 4- (3-hydroxybutyl) -3,5,5-trimethyl-	0,20
40	30,09	Isoshiobunon	0,15
41	30,44	2H-pyran, 2- (7-heptadecyloxy) tetrahydro	0,25
42	30,69	5,6,6-trimethyl-5- (3-oxobut-1-enyl) -1-oxaspiro [2.5] octan-4-one	0,17
43	31,11	Epiglobulol	0,17
44	31,36	eicosane	0,26
45	31,52	6-epi-shibunol	0,20
46	31,68	3- (4,8,12-trimethyltridecyl) furan	0,24
47	31,99	Continin	0,34
48	32,47	1-penten-3-one, 1- (2,6,6-trimethyl-1-cyclohexen-1-yl) -	0,32
49	32,70	n-hexadecanoicacid	0,52
50	32,93	Cyclodecacyclotetradecene, 14,15- didehydro-1,4,5, 8,9,10, 11,12,13, 16,17,18, 19,20 - tetradecahydro	0,12
51	33,88	7H-indeno [5,6-b] furan-7-one, 4,4a, 5,6,7a, 8-hexahydro	0,43

52	34,12	7-methyl-Z-tetradecene-1-ol acetate	0,23
53	34,62	Dibutylphthalate	0,39
54	35,07	sclareolidlactol	1,54
55	36,52	5- (7a-isopropenyl-4,5-dimethyl-octagiroinden-4-yl) -3-methyl-pent-2-en-1-ol	2,67
56	36,85	1-penten-3-one, 1- (2,6,6-trimethyl-1-cyclohexen-1-yl) -	2,79
57	37,64	1-naphthalenepropanol, α -ethenyl decacidro-2-hydroxy- α , 2,5,5,8a-pentamethyl, [1R- [1 α (R *), 2 β , 4a β , 8a α]]	1,14
58	38,36	2,5,5,8a-tetramethyl-4-methylene-4a, 5,6,7,8,8a-hexahydro-4H-chromen	3,11
59	38,86	1-carbomethoxy-1,2,5,5-tetramethyl-cis-decalin (1R, 2S, 4aS, 8aS)	2,71
60	39,10	1-hexene, 2- (p-anisyl) -5-methyl-	1,27
61	39,66	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	8,78
62	40,40	Benz [e] azulene-3 (3aH) -one, 4,6a, 7,8,9,10,10a, 10b-octahydro-3a, 8,10a-trihydroxy-5- (hydroxymethyl) -2,10-dimethyl -, [3aR- (3a α , 6a α , 8 β , 10 β , 10a β , 10b β)] -	0,72
63	40,78	4a, 7,7,10a-tetramethyldodecabenzl [e] chromen-3-ol	1,76
64	40,94	5,6,6-trimethyl-5- (3-oxobut-1-enyl) -1-oxaspiro [2.5] octan-4-one	1,44
65	41,25	5- (7a-isopropenyl-4,5-dimethyl-octahydroinden-4-yl) -3-methyl-pent-2-enal	2,23
66	41,57	Beganalcohol	1,87
67	41,99	1-methyl-3- (2,2,6-trimethylbicyclo [4.1.0] hept-1-yl) acetic acid ester	2,04

68	42,28	4a, 7,7,10a-tetramethyl-dodecahydro-benzo [e] chromen-3-one	1,05
69	42,60	Tunbergol	1,67
70	42,85	4,8,13-cyclotetradecatatriene-1,3-diol, 1,5,9-trimethyl-12- (1-methylethyl) -	4,03
71	43,60	1H-naphtho [2,1-b] pyran, 3-ethynyl-dodecahydro-3,4a, 7,7,10a-pentamethyl-, [3R- (3 α , 4a β , 6a α , 10a β , 10b α)] -	1,51
72	43,85	Di (2-propylpentyl) phthalic acid ester	1,14
73	44,23	6-epi-shiobunol	0,65
74	44,40	Oxyrandodecanoicacid, 3-octyl-, cis-	0,95
75	44,57	2- Pentenoic acid, 5- (decahydro-5,5,8a-trimethyl-2-methylene-1-naphthalenyl) -3-methyl-, methyl ether, [1R- [1 α (E), 4a β , 8a α]]	1,41
76	45,17	Heptacosan, 1-chloro	1,25
77	45,43	1-naphthalene penta-noic acid, decahydro- β , 5,5,8a-tetramethyl-2-methylene, methyl ether, [1R- [1 α (S *), 4a β , 8a α]]	0,68
78	46,17	6 β , 17 β -dihydroxy-6 α -pentyl-5 α -androstan-3-one	0,85
79	46,40	Squalene	0,59
80	46,86	Spiro [furan-2 (5H), 2 '(1'H) -naphto [2,1-b] furan] -5-one, 3'a, 4', 5', 5'a, 6', 7', 8', 9', 9'a, 9'b-decahydro-3,3'a, 6', 6', 9'a-pentamethyl-, [2'S- (2' α , 3'a α , 5 "Ap, 9'a α , 9'b β)] -	1,12
81	47,70	A-nor-cholestan-2-one, (5 α) -	1,59
82	48,15	5,8,9-endo-10-exo-tetramethyltricyclo [6.3.0.0 (5.11)] undecan-1-carboxylic acid	0,96
83	48,35	Heptacosan, 1-chloro	0,38
84	48,58	Oxirane, 2,2-dimethyl-3- (3,7,12,16,20-pentamethyl-3,7,11,15,19-genikospentaenyl)	1,20

85	51,30	28-Nor-17 β (H) -gopan	0,90
86	51,75	Bis (2,2,6,6-tetramethyl-4-piperidiny) decanedi- oic ester	0,64
87	52,07	Vitamin E	1,00
88	52,36	18,19-Sekolupan-3-ol (3 β , 17.xi.) -	0,38
89	52,62	17- (1,5-dimethylhexyl) -10, 13-dimethyl-2,3,4, 7,8,9, 10,11, 12,13,14,15, 16,17 -tetradecahy- dro- 1H-cyclopenta [a] phenanthrene-3-ol	1,06
90	53,74	Loop-20 (29) -o-28-ol	0,14
91	54,27	5-cholesten-3-ol, 24-meth- yl-	0,22
92	54,69	stigmasterol	0,35
93	55,74	γ -sitosterol	0,14
Sum			100,00

The data presented in table 7 show that the temperature increase caused a significant decrease in the efficiency of nicotine extraction, compared with the result obtained at 120 bar and 60°C. In the extract, the nicotine content decreased by 21.53%, that is, almost twice, moreover, the content of phytolacetate fell almost twice, which confirms the pattern in the composition change. Also, despite the temperature increase in the extract, a large number of various compounds have also been identified (93).

CONCLUSION

In this paper, a comparative analysis of liquid and supercritical extraction of biologically active substances from the plant *Nicotiana tabacum* l, harvested in the Republic of Kazakhstan, was conducted for the first time.

For the first time, optimal values of pressure and temperature have been established for nicotine release by supercritical fluid extraction with carbon dioxide.

For the first time, the qualitative and quantitative composition of extracts obtained by supercritical fluid extraction with carbon dioxide at various pressures and temperatures has been established.

It should be noted that in all extracts the dominant compound is nicotine, which can be used as a standalone product, or oxidized to nicotinic acid.

Data on the degree of extraction of nicotine with different methods and modes of extraction are presented in Figure 2.

The carried out and analyzed modes of SCF extraction allow us to draw the following conclusions:

- An increase in temperature leads to a change in the properties of the solvent and does not cause ther-

Figure 2: Data on the degree of extraction of nicotine with different methods and modes of extraction

mal degradation processes. This is evidenced by the analysis of the chemical compositions of the obtained extracts: at a relatively high temperature for a BAS(Biologically Active Substances), substances of rather complex composition were identified. The reason for this may be the absence of an oxidizing agent in the system and a short heating time;

- degree of extraction of nicotine at a pressure of 170 bar and a temperature of 80°C was comparable with the extraction mode of 150 bar 28°C, 150 bar 60°C and 120 bar 80°C;
- as a preferred process, we propose a supercritical process with heating up to 60°C and a pressure of 120 bar, at which the recovery rate reached 47.40%. In this parameter, it significantly surpasses the classical liquid extraction with organic solvent and the supercritical process without heating.

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