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Bioactive Nanoparticles

Essential oil from Lamiaceae Family Plants / β- Cyclodextrin Supramolecular Systems

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This paper presents the molecular encapsulation of essential oils from the Lamiaceae family plants in β-cyclodextrin (β-CD). The complexation of the essential oils (basil-Ocimum basilicum L., thyme-Thymus vulgaris L., lavender-Lavandula angustifolia, and marjoram-Majorana hortensis) in β-cyclodextrin was achieved by the ethanol-water solution method, and the nanoparticles were analyzed by thermogravimetry (TG), differential scanning calorimetry (DSC), and scanning electron microscopy (SEM). The relative concentrations for the volatile compounds from the raw and encapsulated essential oils were obtained by gas chromatography - mass spectrometry (GC-MS). The biomolecule binding competitivities to the β-cyclodextrin were evaluated.

Keywords: nanoparticles, cyclodextrins, essential oils, Lamiaceae, thermogravimetry, gas chromatography-mass spectroscopy, scanning electron microscopy

The *Lamiaceae* (*Labitae*) family is particularly rich of biodiversity (over than 6500 species which are spreaded in mediterranean and subtropical regions) and since ancient times it was the subject of a great variety of uses by humans [1,2]. The main uses of the *Lamiaceae* plants are: human food and livestock feeding, aromatic, for liqueur production, as medicinal and veterinary plants, to combat pests and parasites, as cosmetic, for house decoration, as fuel or illumination, in the handicraft, to produce soap, to tan the leathers, to dye the cloths, in religious, magic or superstitious practices [2]. The main classes of compounds which belong to this family are: terpenoids (especially mono-, sesqui-, di- and triterpenes), phenolic compounds (phenolic acids, like rosmarinic and caffeic acids, flavonoids – including flavone 3- and 7-glucosides). Nitrogen containing compounds play a minor role, such as stachydrine and other simple alkaloids, and betaine [3-6]. The presence of the phenolic compounds (especially flavonoids) confers to the *Lamiaceae* plants extracts a strong antioxidant activity [68].

Some compounds, like mono- and sesquiterpenes, are susceptible to oxidation and can conduct to potential carcinogenic epoxy-derivatives (like limonene-dioxide, known with mutagenic activity) [9]. In order to reduce the oxidative process, these compounds must be protected against the action of the oxygen. The most appropriate host molecules which can complex the labile hydrophobic molecules are cyclodextrins. Cyclodextrins (CDs) are natural cyclic oligosaccharides, containing 6-8 (α-,β-,γCD) or more glucopyranose moieties, which have structures like a truncated cone with a hydrophobic inner cavity, that can interact with a hydrophobic organic molecule (geometrically compatible) and form a more stable, protective (to air, temperature, light), and controlled releasing supramolecular system [10-12].

A large number of studies were performed in the drug controlled release field [13-17], but very interesting studies were conducted in the food flavoring field [18-21]. Some studies were performed on the competitive binding of aroma compounds (alcohols, acids and esters) from natural strawberry by βCD [18], on the complexation and the protective capabilities of cyclodextrins for the odorant compounds [20], and on the complexation of some volatile oils [11,12,19, 22].

Studies on the nanoencapsulation of Lamiaceae volatile oils in cyclodextrins were realized [11,12,23], some of them with applications in aromatherapy [24,25], but the competitivity of compounds to the cyclodextrin cavity was not realized yet from this point of view. Our earlier studies [26-28] indicate important modifications on the composition of biosystems at the action of different degradative factors like air/oxygen, temperature, humidity, and the nanoencapsulation of these natural compounds in cyclodextrin-like matrices conduct to biosystems with higher stability and bioavailability.

This study investigates the capacity of encapsulation of essential oils from the *Lamiaceae* family plants (basil, thyme, lavender, and marjoram) in β-cyclodextrin (βCD) matrix, by ethanol solution method, using the thermogravimetry (TG-DTG), differential scanning calorimetry (DSC), and scanning electron microscopy (SEM) analyses. The competitivity of biocompounds to the encapsulation was evaluated from the composition of the raw volatile oils and the complexed ones by means of gas chromatography-mass spectrometry (GC-MS).

Materials and Method

Materials. Essential oils used for the molecular encapsulation were purchased from S.C. Natex S.A. Cluj Napoca (basil, thyme, and marjoram essential oils) and

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Table 1
QUANTITIES OF ESSENTIAL OILS FROM *LAMIACEAE* FAMILY PLANTS AND βCD USED FOR ENCAPSULATION, AND YIELDS FOR THE ENCAPSULATION PROCESS

No	Complex	m (essential oil) (mg)	m (βCD) (mg)	m (complex) (mg)	Yield (%)
1	Basil ess. oil/βCD	70.9	671.2	477.6	64.36
2	Thyme ess. oil/βCD	75.5	671.0	602.4	80.70
3	Lavender ess. oil/βCD	105.5	671.1	624.4	80.40
4	Marjoram ess. oil/βCD	75.4	671.3	492.5	65.96

from S.C. Fares S.A. Orãºtie (lavender essential oil). β-Cyclodextrin (βCD, >99%, reagent grade) were purchased from Merck&Co., Inc, New Jersey. Alkane Standard solution C_8-C_{20}, used for Kovats indices determination, was obtained from Fluka ChemieAG, Switzerland. Ethanol 96%, used for the obtaining of nanoparticles, was obtained from Chimopar SA – Bucureºti, Romania. The hexane used for recovering the bioactive compounds from nanoparticles has GC purity and was achieved from Merck&Co, Inc.

Complexation method. 0.5 mmole of β-cyclodextrin (table 1) were dissolved in 8 mL distilled water at 50±1°C in a thermocontrolled minireactor, equipped with reflux condenser, and then 5 mL ethanolic solution of essential oil (the weight corresponding to essential oil main compound : βCD molar ratio of 1 : 1, table 1) were slowly added under continuous stirring. The solution was then stirred another 15 min and slowly cooled at 20°C in about 4 h. The crystallization was perfected in refrigerator at 4°C for 24 h. The complex was filtered, washed with ethanol and dried in exicator.

Essential oil extraction from the complex. 100 mg Essential oil/βCD complex was dissolved in 4 mL distilled water in a thermocontrolled minireactor, equipped with reflux condenser and a magnetic stirrer; 2 mL hexane was then added and the emulsion was vigorously stirred for 20 min at 69°C. After cooling, the organic layer was separated and the aqueous layer was extracted for another three times with 3 × 2 mL hexane in the same manner. All organic layers were dried over anhydrous $CaCl_2$ and analyzed by GC-MS.

GC-MS analysis. For the analysis of the raw and recovered essential oils a Hewlett Packard HP 6890 Series gas chromatograph coupled with a Hewlett Packard 5973 mass spectroscopy detector (GC-MS) system was used. A HP-5 MS capillary column (30 m length, 0.25 mm i.d., 0.25 μm film thickness) was used for the GC system. The temperature program was set up from 50°C to 250°C with 4°C/min, both the injector and detector temperatures were 280°C and He was used as carrier gas. The injection volume was 2 μL. Ionization energy EI of 70eV was used for mass spectroscopy detector, with a source temperature of 150°C, scan range 50-300 amu, scan rate $1s^{-1}$. The mass spectra were compared with the NIST/EPA/NIH Mass Spectral Library 2.0.

TG-DTG analysis. The thermogravimetric analysis for the essential oil/βCD complexes was performed on a Netzsch TG-209 apparatus, using a temperature program of 20-200°C, with a heating rate of 4°C/min, and from 200 to 900°C with a heating rate of 10°C/min. All analyses were conducted under nitrogen atmosphere. Data acquisition was performed with the TG Netzsch 209-Acquisition Soft/2000 and the data analysis was realized with the Netzsch Proteus-Thermal Analysis ver. 4.0/2000 soft.

DSC analysis. Differential scanning calorimetry (DSC) was carrying out on a DSC 204 Netzsch apparatus, with a temperature program of -50÷200°C, the heating rate being 4°C/min. The acquisition and the processing of the data were performed with the DSC 204 Netzsch-Acquisition Soft/2000 and Netzsch Proteus-Thermal Analysis, ver. 4.0/2000, respectively.

SEM analysis. For morphological and dimensional evaluation of the *Lamiaceae* bioactive compounds/cyclodextrin nanoparticles scanning electron microscopy (SEM) technique was used. A JEOL JSM 5510-LV SEM apparatus was used, at different magnitude levels.

Results and discussion

The best yields were obtained for the complexation of the thyme and lavender essential oils of 80.7% and 80.4%, respectively, and the lower one was obtained in the case of basil essential oil (64.4%). The thermogravimetric analysis indicates that the concentration of the essential oils encapsulated was in the range of 7.2% (for the marjoram complex) to 9% (for basil / βCD complex) (fig. 1). The DTG analyses reveal that a part of the essential oil was not completely encapsulated (components were volatilized at a lower temperature) or an ethanol/βCD complex also can be formed in the encapsulation process. Very interesting is the DSC analysis of the basil essential oil/βCD complex, comparatively with the raw essential oil, that indicate an endothermic peak at 140°C, corresponding to α-bergamotene (in concentration of about 5.7%, obtained from the GC analysis), and which does not appear in the complex (concentration of bergamotene of about 0.1% in the complex extract). All complexes released the encapsulated essential oils at temperature above 100°C (the minimum of the endothermic peaks are 107.7°C for the basil essential oil/βCD complex, 105.6°C for thyme essential oil/βCD complex, 101°C for lavender essential oil/βCD complex and, 101.4°C for marjoram essential oil/βCD complex, fig. 2).

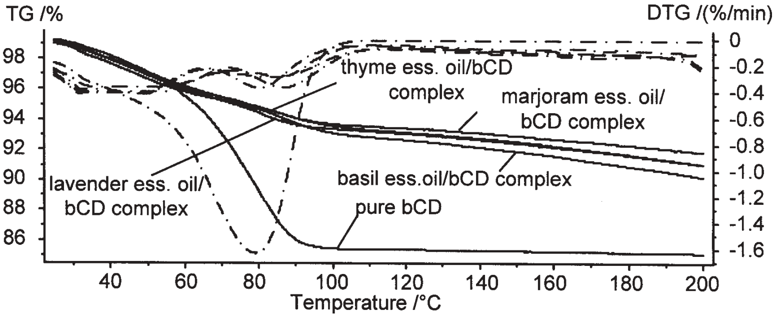

Fig. 1. TG-DTG analyses of the pure βCD and for the essential oil/βCD complexes

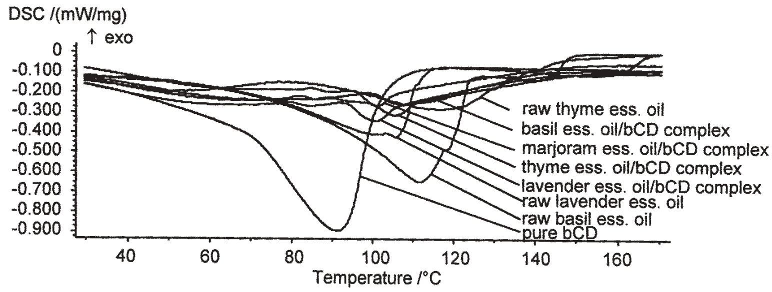

Fig. 2. DSC analyses of the pure βCD, raw essential oils and essential oil/βCD complexes

From the SEM analysis, *Lamiaceae* biocompounds/βCD nanoparticles seem to have a rhombohedral-parallelepipedic crystallization form, with dimensions between hundred of nanometers and two micrometers for the particle sides (fig. 3).

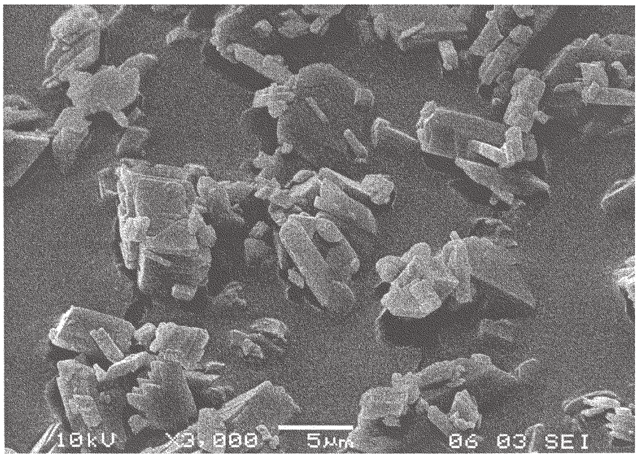

Fig. 3. SEM micrograph for the *Ocimum basilicum* L. essential oil/ βCD nanoparticles (×3 000 magnitude)

The main components from the essential oils from *Lamiaceae* family plants were the phenolic compounds and the tertiary terpenoid alcohols.

The GC-MS analysis of the basil essential oil revealed a number of 140 compounds (fig. 4), the main compound being methyl chavicol (66%); the minor components were eucalyptol, methyl eugenol, and linalool. The most concentrated sesquiterpene was α-bergamotene (5.7%),

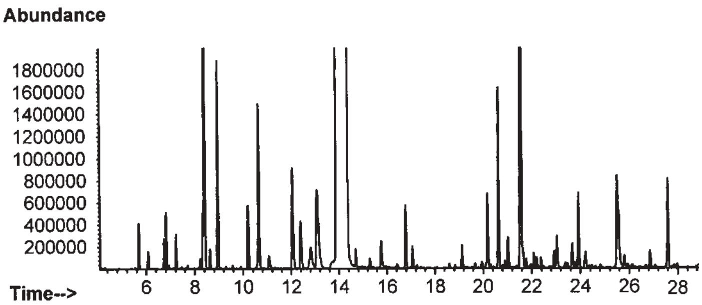

Fig. 4. The gas chromatogram from the GC-MS analysis of the basil essential oil

Table 2

THE RELATIVE CONCENTRATIONS OF THE RAW AND RECOVERED BASIL ESSENTIAL OIL, THE RATIOS BETWEEN THESE CONCENTRATIONS, AND THE KOVATS INDICES FOR THE MS IDENTIFIED COMPOUNDS

GC No	MS Identification	Kovats Indices (KI)	$c_{raw\ ess.\ oil}$ (%)	$c_{recov.ess.oikl}$ (%)	$c_{recovered}/c_{raw\ ess.\ oil}$
6	α-Pinene	936	0.36	0.25	0.69
7	Camphene	950	0.15	0.09	0.62
11	Sabinen	975	0.25	0.12	0.46
12	β-Pinene	978	0.45	0.28	0.62
15	β-Myrcene	993	0.29	0.15	0.54
24	Cymene	1026	0.09	0.11	1.20
25	Limonene	1030	0.54	0.41	0.76
26	Eucalyptol	1033	4.21	1.77	0.42
27	*trans*-β-Ocimene	1040	0.17	0.09	0.54
28	*cis*-β-Ocimene	1050	1.86	1.08	0.58
34	Fenchone	1089	0.65	0.31	0.48
35	Linalool	1103	1.90	0.57	0.30
43	Camphor	1145	1.30	0.38	0.29
44	Isomenthone	1155	0.70	0.29	0.42
45	Borneol	1168	0.54	0.05	0.10
46	Cyclohexanol, 1-methyl-4-(1-methylethyl)-	1176	2.03	0.26	0.13
47	Anisole, *p*-allyl-	1209	66.51	84.77	1.28
48	Fenchyl acetate	1224	0.16	0.10	0.63
49	Fenchyl acetate (isomer)	1232	0.01	0.33	
56	Bornyl acetate	1287	0.64	0.12	0.18
57	Menthol, acetate	1296	0.21	0.39	1.82
71	β-Elemen	1393	0.82	0.17	0.20
74	Methyleugenol	1408	1.98	0.17	0.09
77	Caryophyllene	1420	0.36	4.30	
80	α-Bergamotene	1438	5.69	0.13	0.02
81	α-Guaiene	1440	0.16	0.08	0.48
84	Humulene	1454	0.15	0.05	0.36
85	β-Farnesene	1458	0.11	0.08	0.71

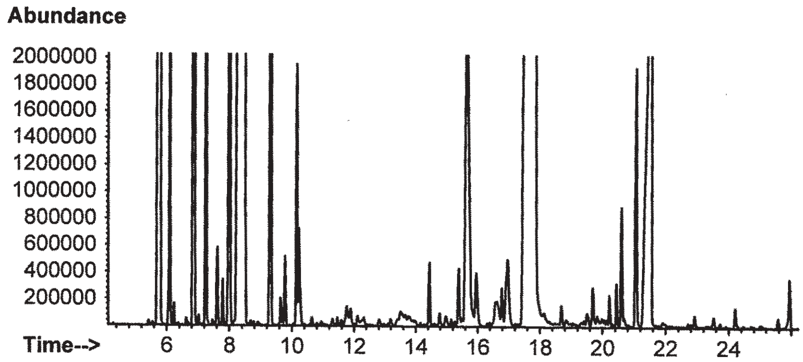

Fig. 5. The gas chromatogram from the GC-MS analysis of the thyme essential oil

Table 3

THE RELATIVE CONCENTRATIONS OF THE RAW AND RECOVERED THYME ESSENTIAL OIL, THE RATIOS BETWEEN THESE CONCENTRATIONS, AND THE KOVATS INDICES FOR THE MS IDENTIFIED COMPOUNDS

GC No	MS Identification	Kovats Indices (KI)	$c_{raw\ ess.\ oil}$ (%)	$c_{recov.ess.oikl}$ (%)	$c_{recovered}/c_{raw\ ess.\ oil}$
3	α-Pinene	939	11.08	30.19	2.72
4	Camphene	951	1.36	1.56	1.14
7	β-Pinene	980	5.88	10.13	1.72
9	β-Myrcene	994	2.03	2.13	1.05
11	α-Thujene	1006	0.38	0.34	0.92
12	3-Carene	1012	0.20	0.17	0.84
13	α-Terpinen	1019	2.92	2.75	0.94
14	β-Cymene	1029	7.28	6.67	0.92
15	Limonene	1035	12.51	22.10	1.77
19	γ-Terpinen	1062	3.37	3.58	1.06
21	Bicyclo[2.2.2]octan-1-ol, 2-chloromethyl-	1076	0.30	0.24	0.78
23	Terpinolen	1088	1.12	1.08	0.96
24	Terpinolen (or isomer)	1090	0.51	0.45	0.89
37	Verbenone	1189	0.08	1.67	
39	4-Acetyl-*m*-xylene	1216	0.31	0.15	0.48
45	Carvone	1245	0.30	1.35	4.46
50	*p*-Allylanisole	1287	0.16	0.34	2.10
53	Carvacrol	1320	31.45	11.03	0.35
59	α-Copaene	1377	0.21	0.15	0.71
63	Sativen	1391	0.02	0.11	4.70
66	Junipen	1406	0.60	0.54	0.89
67	Caryophyllene	1421	1.38	1.31	0.95

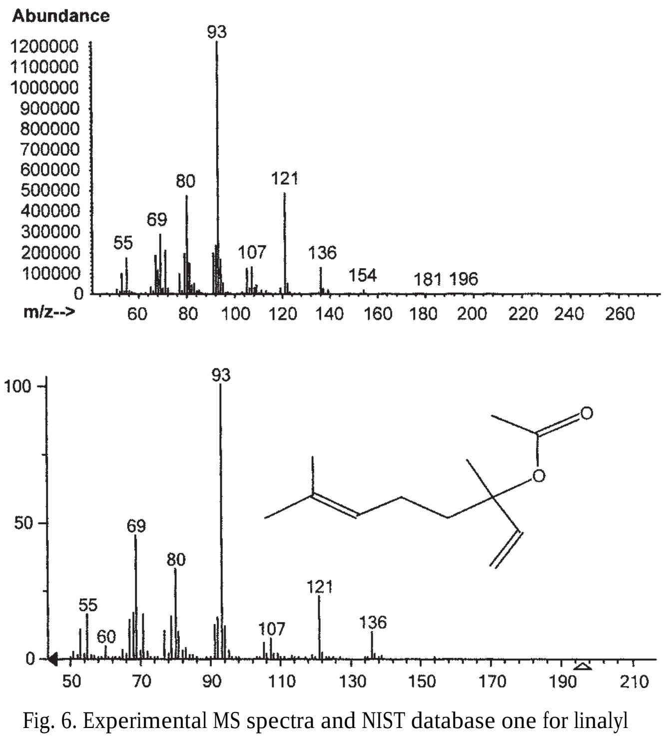

Fig. 6. Experimental MS spectra and NIST database one for linalyl acetate

all the rest of compounds having concentrations < 2%. The main compound, methyl chavicol, was encapsulated in the highest relative concentration of about 84% (the ratio between the recovered and raw concentrations being 1.28). All the rest of oxygenated compounds have these ratios < 1. The hydrocarbonate *p*-cymene was encapsulated in a higher concentration (a ratio of 1.2), and all the sesquiterpenoids (with exception of cubebene) were lower encapsulated (sub-unitary ratios) (table 2).

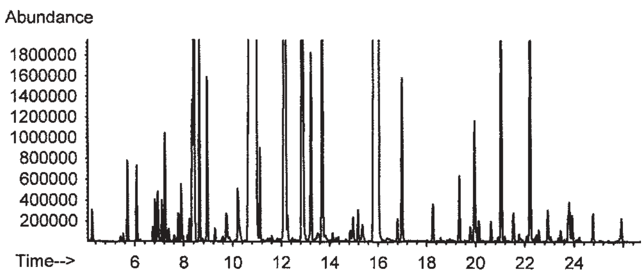

Fig. 7. The gas chromatogram from the GC-MS analysis of the lavender essential oil

Carvacrol was the most concentrated compound in the thyme essential oil (31.5%); concentrations above 5% were observed for limonene (12.5%), α –pinene (11.1%), and β–pinene (5.9%) (fig. 5). The monoterpenes α- and β –pinene were encapsulated in about double relative concentrations (the ratios between the concentrations of these terpenes in the recovered oil and in the raw one being 2.7 and 1.7, respectively), comparatively with carvacrol, that was encapsulated in lower relative concentration (a ratio of 0.35, table 3), probably due to the rigid structure

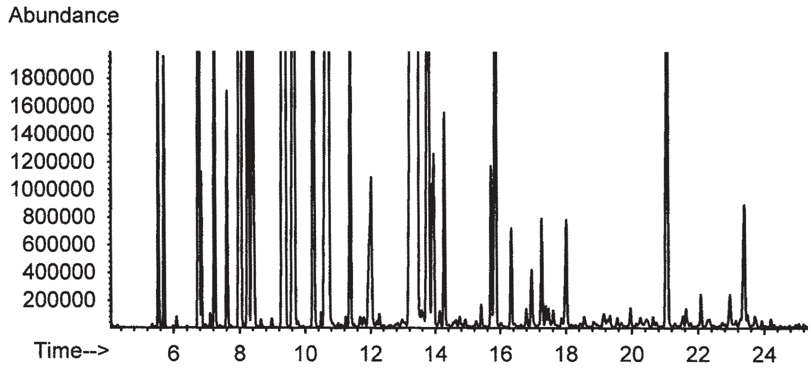

Fig. 8. The gas chromatogram from the GC-MS analysis of the marjoram essential oil

Table 4

THE RELATIVE CONCENTRATIONS OF THE RAW AND RECOVERED LAVENDER ESSENTIAL OIL, THE RATIOS BETWEEN THESE CONCENTRATIONS, AND THE KOVATS INDICES FOR THE MS IDENTIFIED COMPOUNDS

GC No	MS Identification	Kovats Indices (KI)	$c_{raw\ ess.\ oil}$ (%)	$c_{recov.ess.oikl}$ (%)	$c_{recovered}/c_{raw\ ess.\ oil}$
5	α-Pinene	936	0.50	0.35	0.70
6	Camphene	950	0.47	0.36	0.77
11	β-Pinene (or isomer)	978	0.28	0.18	0.64
14	β-Pinene	993	0.73	0.39	0.54
23	Limonene	1031	1.40	0.82	0.59
24	Eucalyptol	1034	3.96	3.67	0.93
25	*trans*-β-Ocimene	1040	1.65	0.99	0.60
27	*cis*-β-Ocimene	1050	1.16	0.63	0.54
33	Bicyclo[3.1.0]hexane, 6-isopropylidene-1-methyl-	1087	0.01	0.23	
35	Linalool	1113	30.40	34.80	1.15
43	Camphor	1149	8.84	6.06	0.69
47	Borneol	1171	5.49	1.34	0.24
48	Terpinen-4-ol	1181	1.65	0.80	0.49
52	α-Terpineol	1195	2.50	0.89	0.36
62	Linalyl acetate	1264	23.61	26.65	1.13
66	4-Hexen-1-ol, 5-methyl-2-(1-methylethenyl)-, acetate	1294	1.36	0.78	0.57
87	Caryophyllene	1421	1.86	1.53	0.82
92	(Z)-β-Farnesene	1459	2.21	1.20	0.54
101	Linalyl *iso*valerate	1511	0.33	0.24	0.71
119	α-Bisabolol	1686	0.60	0.21	0.35

Table 5

THE RELATIVE CONCENTRATIONS OF THE RAW AND RECOVERED MARJORAM ESSENTIAL OIL, THE RATIOS BETWEEN THESE CONCENTRATIONS, AND THE KOVATS INDICES FOR THE MS IDENTIFIED COMPOUNDS

GC No	MS Identification	Kovats Indices (KI)	$c_{raw\ ess.\ oil}$ (%)	$c_{recov.ess.oikl}$ (%)	$c_{recovered}/c_{raw\ ess.\ oil}$
6	α-Pinene	936	0.90	0.91	1.01
10	β-Phellandrene	976	3.73	4.11	1.10
11	β-Pinene	979	0.47	0.50	1.07
14	Myrcene	993	1.24	1.09	0.88
16	α-Phellandrene	1006	0.98	0.91	0.92
18	α-Terpinen	1021	7.84	11.23	1.43
20	Cymene	1028	4.60	6.16	1.34
21	Limonene	1032	4.23	4.72	1.12
24	γ-Terpinen	1064	11.03	19.05	1.73
25	Terpineol, *cis*-β-	1073	5.34	2.12	0.40
28	Terpinolen	1091	3.70	4.57	1.24
30	Terpineol, *cis*-β- (or isomer)	1105	8.93	3.39	0.38
34	2-Cyclohexen-1-ol, 1-methyl-4-(1-methylethyl)-, *trans*-	1125	1.58	0.71	0.45
46	Terpinen-4-ol	1187	19.28	25.01	1.30
48	α-Terpineol	1197	4.81	3.17	0.66
50	*p*-Allylanisole	1202	0.65	0.28	0.44
52	*trans*-Piperitol	1212	1.01	0.33	0.33
54	*cis*-Carveol	1222	0.04	0.13	3.50
61	Linalyl acetate	1259	2.96	2.76	0.93
68	Terpinene 4-acetate	1302	0.53	0.36	0.68
89	Caryophyllene	1421	2.99	3.06	1.02
95	α-Humulene	1454	0.16	0.11	0.69

with a higher secondary dimension and to the presence of a hydrophilic OH-phenolic group.

The raw lavender and marjoram oils show a great number of compounds (133 for the first and 181 for the second essential oil), revealed by the GC analysis. The main compounds were linalool and linalyl acetate (fig. 6 shows the experimental MS spectra and from the NIST database one) for the lavender essential oil (fig. 7) and terpinen-4-ol and γ-terpinene for the marjoram essential oil (fig. 8). In the case of linalool and the corresponding acetate, the relative concentrations in the recovered oils were closely to those from the raw oil, the ratios of these concentrations being 1.15 and 1.13, respectively (table 4). In the case of marjoram essential oil, γ-terpinene (a hydrophobic monoterpene) was encapsulated almost in a double relative concentration, comparatively with that from the raw oil (the ratio of 1.73); for the corresponding alcohol terpinen-4-ol this ratio was only 1.3 (table 5).

Conclusions

Four essential oils from the *Lamiaceae* family plants were encapsulated in β-cyclodextrin matrix with yields in the range of 64 and 81%, the best results being obtained for thyme and lavender oils. The DSC analysis revealed the formation of the complex, the concentration of the complexed oils being in the range of 7.2% to 9% (for the basil/βCD complex), as shows the TG-DTG analyses.

The main compounds from the essential oils used for the encapsulation were methyl chavicol for the basil essential oil, carvacrol for the thyme essential oil, linalool and linalyl acetate for the lavender essential oil, and terpinen-4-ol for the marjoram essential oil.

The ratios between the relative concentration of the tertiary terpenoid alcohols (linalool, terpinen-4-ol) and the phenolic ethers (methyl chavicol) in the recovered and in the raw essential oil were in the range of 1.27 and 1.6, and for the phenolic compounds (carvacrol) this ratio was sub-unitary.

Generally, the more hydrophobic compounds (especially monoterpenes) were encapsulated in higher concentrations comparatively with the hydrophilic ones (like terpene alcohols) in the *Lamiaceae* family essential oils.

References

1.RADU, A., ANDRONESCU, E., FÜZI, I., Botanicã farmaceuticã, Editura Didacticã ºi Pedagogicã, Bucureºti, 1981, p. 438

2.MULAS, M., Traditional Use of Labiatae in the Mediterranean Area; PISTELLI, L., Phytochemicals from Lamiaceae: from nutraceutics to hallucinogens. In: Proceedings of the International Symposium “The *Labiatae*: Advances in Production, Biotechnology and Utilization, Sanremo, 2006, p. 3

3.SILVA, D.B., VIEIRA, R.F., ALVES, R.B.N., MENDES, R.A., CARDOSO, L.D., QUEIROZ, L., SANTOS, I.R.I., Revista Brasileira de Plantas Medicinais, **8**, 2006, p. 27

4.SIDDIQUI, B.S., ASLAM, H., ALI, S.T., BEGUM, S., KHATOON, N., Chemical & Pharmaceutical Bulletin, **55**, 2007, p. 516

5.BILKE, S., MOSANDL, A., European Food Research and Technology, **214**, 2002, p. 1438

6.CULÁKOVÁ, V., MÁRIÁSSYOVÁ, M., HEILEROVÁ, L., Chemicke Listy, **99**, 2005, p. 277

7.KÄHKÖNEN, M.P., HOPIA, A.I., VUORELA, H.J., RAUHA, J.-P., PIHLAJA, K., KUJALA, T.S., HEINONEN, M., Journal of Agricultural and Food Chemistry, **47**, 1999, p. 3954

8.MATKOWSKI, A., PIOTROWSKA, M., Fitoterapia, **77**, 2006, p. 346

9.*** The Ullmann’s Encyclopedia of Industrial Chemistry, Wiley-VCH & AND CompLex Publ. Tech., Chichester, 2002, Electronic Release ver. 3.5

10.SZEJTLI, J., Pure and Applied Chemistry, **76**, 2004, p. 1825

11.SZEJTLI, J., Cyclodextrin Technology, Kluwer Academic Publishers, Dordrecht-Boston-London, 1988

12.SZENTE, L., SZEJTLI, J., Trends in Food Science & Technology, **15**, 2004, p. 137

13.BREWSTGER, M.E., LOFTSSON, T., Advanced Drug Delivery Reviews, **59**, 2007, p. 645

14.ZIDOVETZKI, R., LEVITAN, I., Biochimica et Biophysica Acta, **1768**, 2007, p. 1311

15.PUCADYIL, T.J., CHATTOPADHYAY, A., Progress in Lipid Research, **45**, 2006, p. 295

16.STELLA, V.J., RAO, V.M., ZANNOU, E.A., ZIA, V., Advanced Drug Delivery Reviews, **36**, 1999, p. 3

17.LOFTSSONA, T., JÄRVINEN, T., Advanced Drug Delivery Reviews, **36**, 1999, p. 59

18.GOUBERT, I., DAHOUT, C., SÉMON, E., GUICHARD, E., LE-QUÉRÉ, J.-L., VOILLEY, A., Journal of Agricultural and Food Chemistry, **49**, 2001, p. 5916

19.BHANDARI, B.R., D’ARCY, B., BICH, L.L.T., Journal of Agricultural and Food Chemistry, **46**, 1998, p. 1494

20. LIU, X.D., FURUTA, T., YOSHII, H., LINKO, P., COUMANS, W.J., Bioscience, Biotechnology and Biochemistry, **64**, 2000, p. 1608

21.YULIANI, S., BHANDARI, B., RUTGERS, R., D’ARCY, B., Food Reviews International, **20**, 2004, p. 163

22.ECKERT, M., ZFL, **47**, 1996, p. 63

23.MARTINS, A.D.P., CRAVEIRO, A.A., MACHADO, M.I.L., RAFFIN, F.N., MOURA, T.F., NOVAK, C., EHEN, Z., Journal of Thermal Analysis and Calorimetry, **88**, 2007, p. 363

24.WANG, C.X., CHEN, S.L., Journal of Industrial Textiles, **34**, 2005, p. 157

25.WANG, C.X., CHEN, S.L., Fibres and Textiles in Eastern Europe, **13**, 2005, p. 41

26.HÃDÃRUGÃ, N.G., HÃDÃRUGÃ, D.I., LUPEA, A.X., PÃUNESCU, V., TATU, C., ORDODI, V.L., BANDUR, G., Rev. Chim. (Bucureºti), **56**, 2005, p. 876

27.HÃDÃRUGÃ, N.G., HÃDÃRUGÃ, D.I., PÃUNESCU, V., TATU, C., ORDODI, V.L., BANDUR, G., LUPEA, A.X., Food Chemistry, **99**, 2006, p. 500

28. HÃDÃRUGÃ, D.I., HÃDÃRUGÃ, N.G., RESIGA, D., PODE, V., DUMBRAVÃ, D., LUPEA, A.X., Rev. Chim. (Bucureºti), **58**, 2007, p. 566

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