

Supplementary data

Essential Oils as Antimicrobial Agents – Myth or Real Alternative?

Katarzyna Wińska, Wanda Mączka, Jacek Łyczko, Małgorzata Grabarczyk, Anna Czubaszek, Antoni Szumny

Samples and materials:

Samples of essential oil were obtained from local distillery (Herbiness, Poland).

GC-MS Analysis

GC-MS analysis was performed on Shimadzu GCMS-QP2020 NX apparatus (Shimadzu, Kyoto, Japan) equipped with a ZB-5 (30 m x 0.25 mm x 1.40 μ m) column Phenomenex (US). Briefly, the GC oven temperature was programmed from 50 °C to 130°C at a rate of 4.0 °C and holding for 2 min, then to 280°C at a rate of 10.0°C and holding for 5 min. Samples were injected at 1:10 split ratio and helium was used as a carrier gas with flow rate of 1.0 ml/min. Scanning was performed from 35 to 500 m/z with an absolute detector voltage of 1 kV. Ionization was performed in electron impact (EI) mode at 70 eV.

Identification and quantification of Volatile Compounds

Identification of volatile compounds was based on the comparison of experimentally obtained mass spectra with mass spectra available in NIST 14 database. Moreover, obtained retention indices (RI) by Kovats were compared with RI available in literature sources. The data proceeding was performed using GCMS Postrun Analysis software (Shimadzu, Kyoto, Japan). Quantification was based on internal standard (2-undecanone), as well as *n*-hexanol calibration curves.

Figure S1. The chromatogram of lavender oil.

Figure S2. The chromatogram of thyme oil.

Figure S3. The chromatogram of peppermint oil.

Figure S4. The chromatogram of cajeput oil.

Figure S5. The chromatogram of cinnamon oil.

Figure S6. The chromatogram of eucalyptus oil.

Figure S7. The chromatogram of clove oil.

Figure S8. The chromatogram of sage oil.

Figure S9. The chromatogram of tea tree oil.

Table S1. Chemical composition of EOs.

Table S2. The antimicrobiological activity of EOs.

lawenda_11_1_Centroided Mass Spectrum_EI+

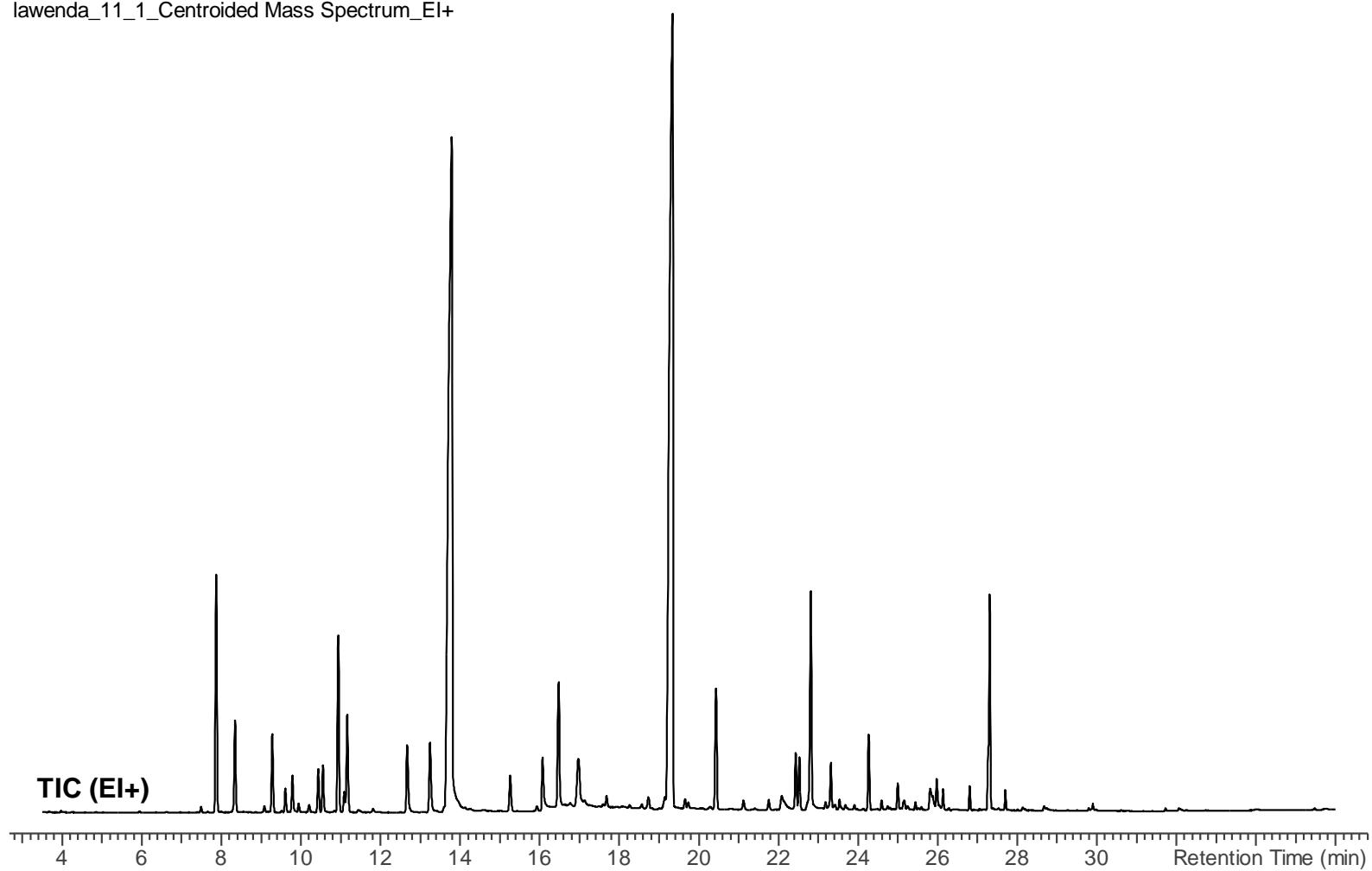


Figure S1. The chromatogram of lavender oil.

tymiankowy_4_1_Centroided Mass Spectrum_EI+

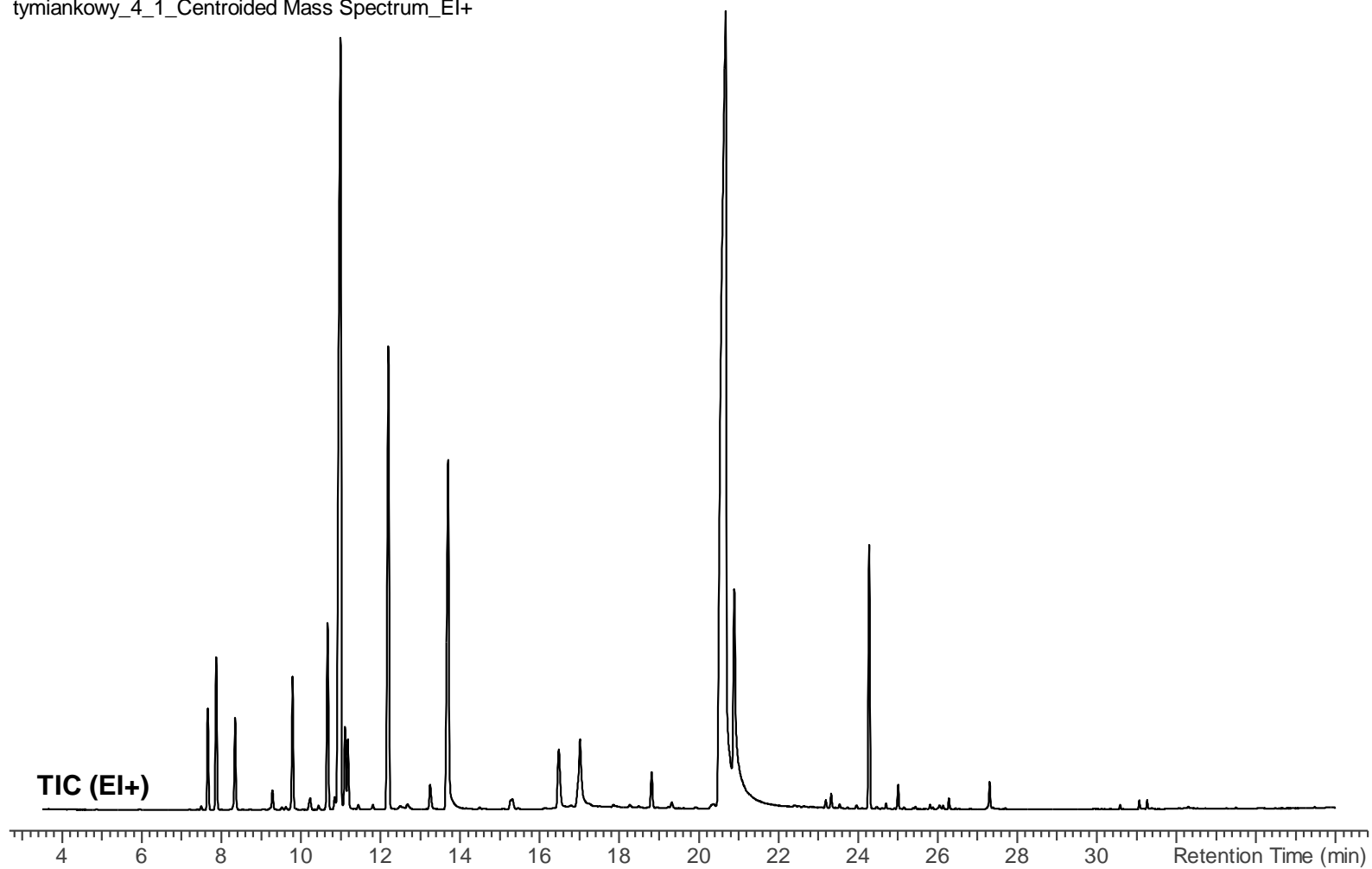


Figure S2. The chromatogram of thyme oil.

mieta_8_1_Centroided Mass Spectrum_EI+

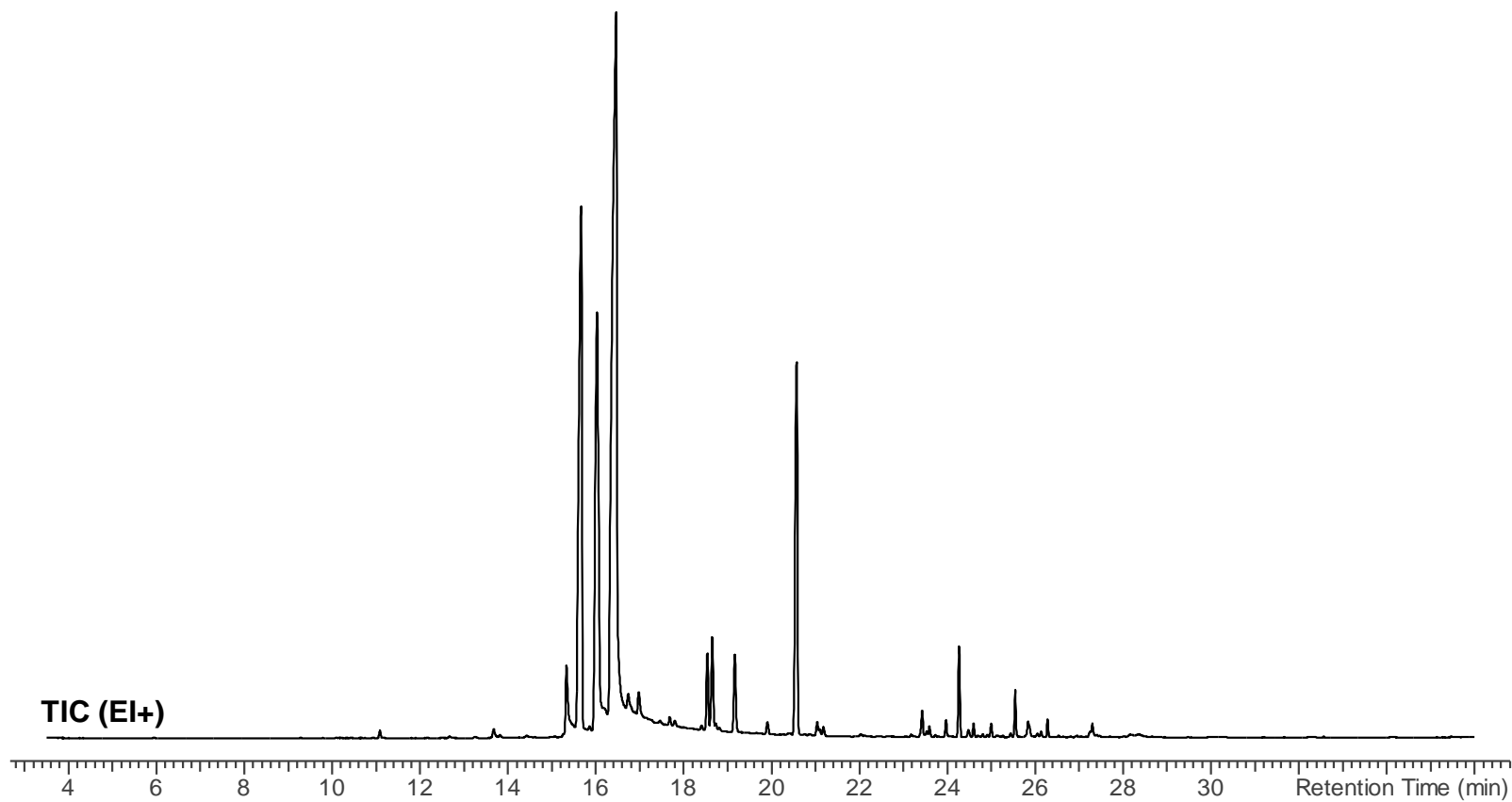


Figure S3. The chromatogram of peppermint oil.

kajeputowy_6_1_Centroided Mass Spectrum_EI+

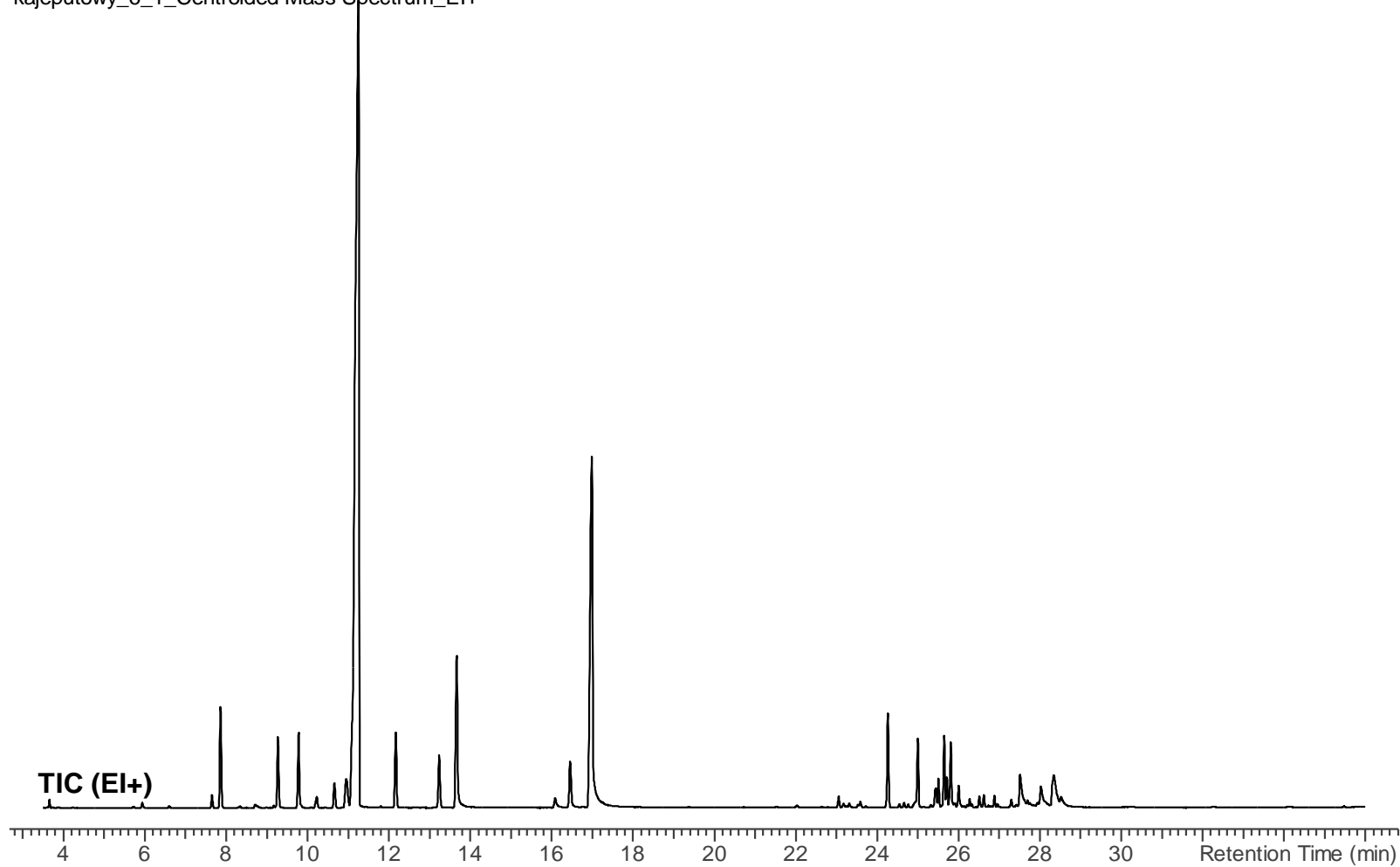


Figure S4. The chromatogram of cajeput oil.

cynamonowy_3_1_Centroided Mass Spectrum_EI+

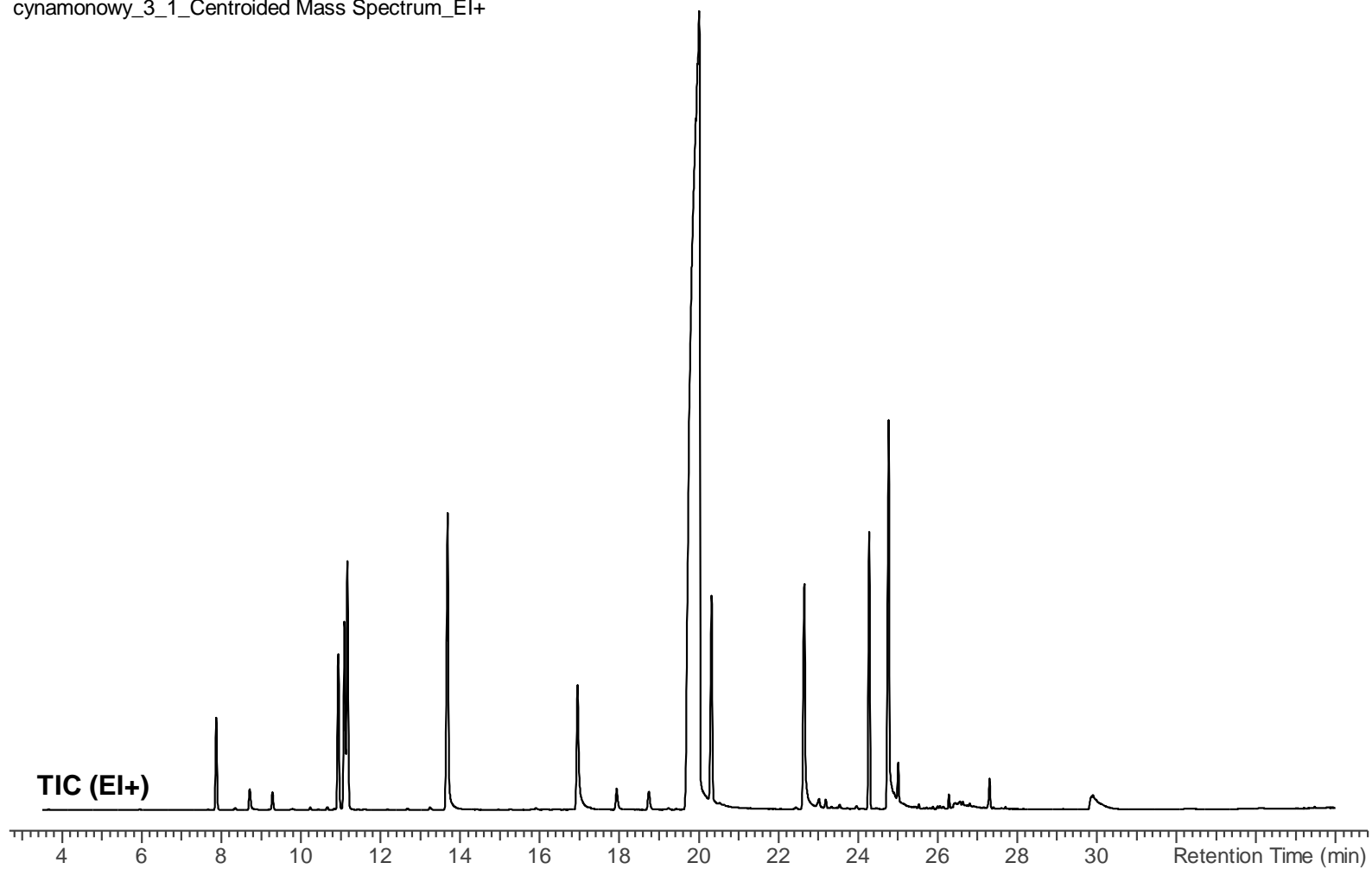


Figure S5. The chromatogram of cinnamon oil.

eukaliptusowy_5_1_Centroided Mass Spectrum_EI+

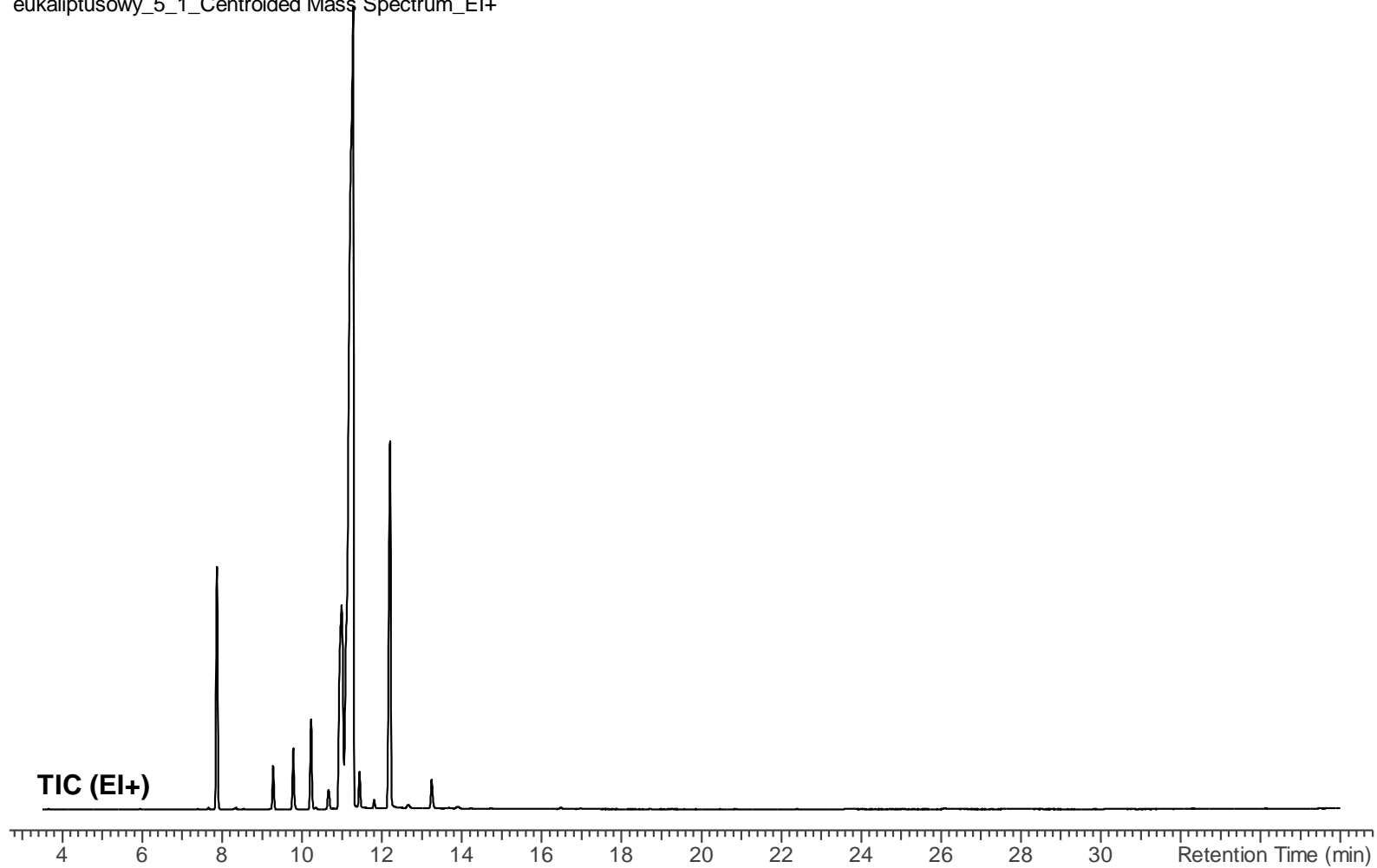


Figure S6. The chromatogram of eucalyptus oil.

gozdik_9_1_Centroided Mass Spectrum_EI+

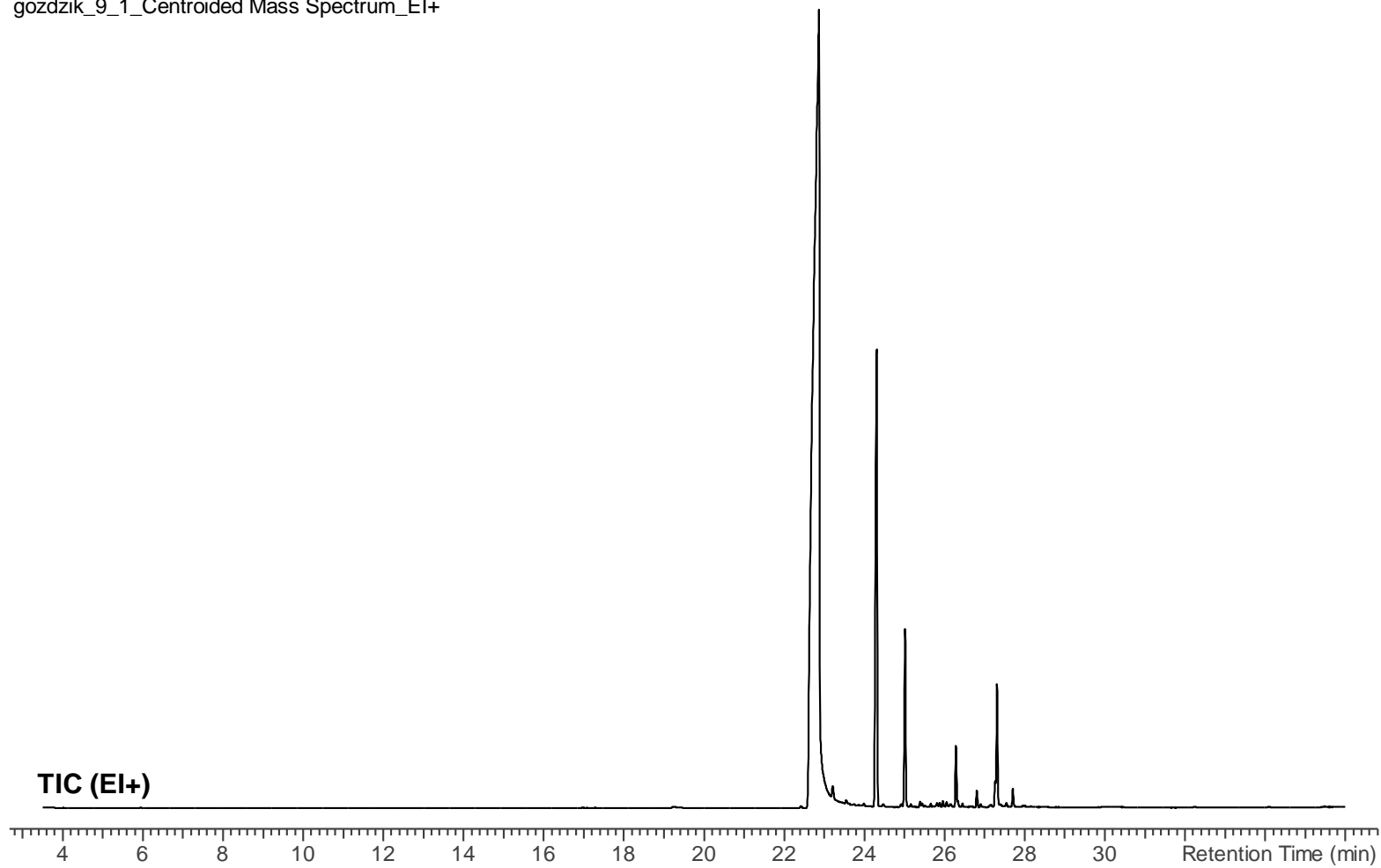


Figure S7. The chromatogram of clove oil.

Szalwia_2_1_Centroided Mass Spectrum_EI+

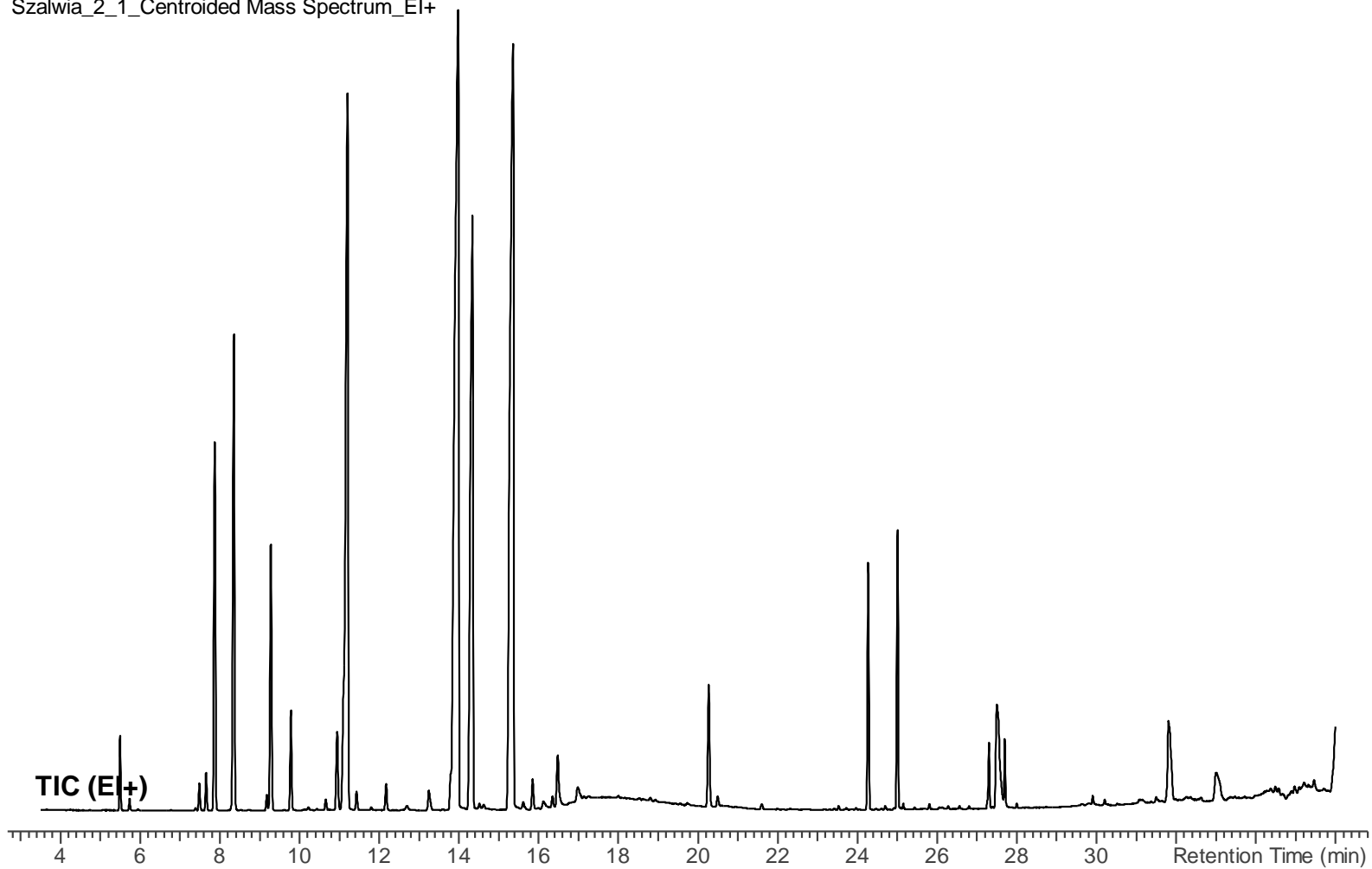


Figure S8. The chromatogram of sage oil.

dzrewo herbaciane_10_1_Centroided Mass Spectrum_EI+

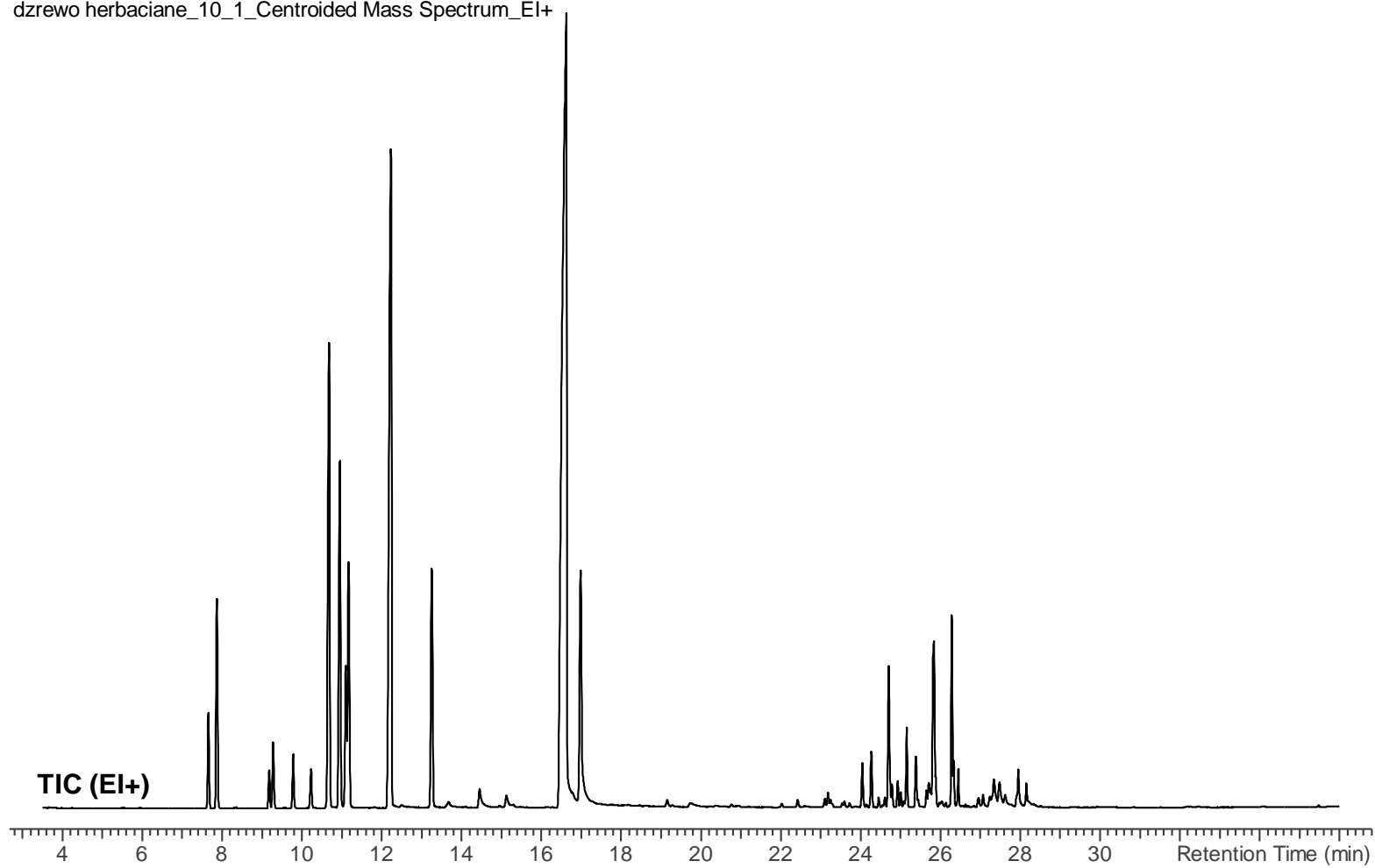


Figure S9. The chromatogram of tea tree oil.

Table S1. Chemical composition of EOs.

	Rt	Lavender oil	Thyme oil	Peppermint oil	Cajeput oil	Cinnamon oil	Clove oil	Eucalyptus oil	Sage oil	Tea tree oil
Z-Salvene	5.48								0.63	
E-Salvene	5.73								0.10	
β -Picoline	5.937				0.13			0.01		
Tricyclene	7.45								0.27	
α -Thujene	7.656		1.26		0.29			0.04		1.17
α -Pinene	7.868	3.71	2.07		2.39	1.18		6.24	4.22	2.80
α -Fenchene	8.285		1.17					0.02	5.85	
Camphene	8.343							0.04		
(2E)-Heptenal	8.343	1.42				0.02				
Thuja-2,4(10)-diene	8.531							0.01		
Benzaldehyde	8.713					0.31				
Sabinene	9.179								0.13	0.50
β -Pinene	9.277	1.27	0.24		1.76	0.23		1.08	3.10	0.90
Octan-3-one	9.608	0.36								
Myrcene	9.781	0.61	1.79		1.87			1.58	1.07	0.70
α -Phellandrene	10.227		0.21		0.34	0.03		2.44	0.03	0.53
Linalool oxide <dehydro-, cis->	10.345							0.05		
delta-3-Carene	10.435	0.69								
Hexyl acetate	10.552	0.73								
α -Terpinene	10.681		2.68		0.66	0.03		0.58	0.12	7.89
para-Menth-1-ene	10.850		0.18							
Cymene <para->	10.948	2.99	18.50		1.22	2.22		12.17	1.03	5.38

Limonene	11.089	0.34	1.15	0.13		2.79			14.15	
Sylvestrene	11.102									2.10
Eucalyptol	11.171	1.66	0.97		50.18	3.48		61.29		3.65
(Z)- β -Ocimene	11.440							0.89	0.20	
(E)- β -Ocimene	11.806							0.21	0.03	
γ -Terpinene	12.230		8.14		2.01			12.22	0.30	14.70
<i>cis</i> -Linalool oxide	12.670	1.28						0.13	0.06	
<i>cis</i> -Linalool oxide	13.244	1.37								
<i>para</i> -Cymenene	13.25								0.31	
Terpinolene	13.256		0.42		1.47			0.84		3.76
Linalool	13.673	28.45	7.14	0.21	4.61	5.15			23.30	0.12
Solusterol	13.870							0.04		
Hex-2-enal <2-isopropyl-, 5-methyl->	13.916							0.05		
β -Thujone	14.3								11.63	
<i>cis</i> - <i>para</i> -Menth-2-en-1-ol	14.452									0.43
α -Campholenal	14.51								0.05	
<i>allo</i> -Ocim-(4E,6Z)-ene	14.728							0.01		
<i>trans</i> - <i>para</i> -Menth-2-en-1ol	15.125									0.23
Camphor	15.258	0.68	0.31						20.32	
Isopulegol	15.335			2.34						
Menthone	15.667			18.20					0.09	
<i>trans</i> -Pinocamphone	15.857			0.15					0.36	
Menthone	16.027			15.69						
Borneol	16.071	0.96								
δ -Terpineol	16.085				0.32				0.15	
<i>cis</i> -Pinocamphone	16.35								0.12	
Menthol	16.461			40.24						
<i>Iso</i> Menthol	16.736			0.42						

Terpinen-4-ol	16.623	2.42				2.58		0.05	0.80	33.94
Terpineol <alpha->	16.984	1.38	1.84	0.55	15.41				0.28	4.65
<i>n</i> -Decanal	17.457			0.07						
<i>n</i> -octyl acetate	17.680	0.16								
<i>trans</i> -Oct-2-enyl acetate	17.797			0.11						
(<i>Z</i>)-Cinnamaldehyde	17.933					0.37				
3-Hexenyl 2-methylbutyrate	18.407			0.08						
<i>cis</i> -3-hexenyl-Isovalerate	18.539			1.54						
Pulegone	18.652			2.08						
hexyl-, 3-methyl butanoate < ->	18.735	0.21		0.13						
<i>o</i> -Anisaldehyde	18.742					0.35				
Carvacryl methyl ether	18.812		0.56							
Carvone	18.820			0.06						
Carvotanacetone	19.163			1.69						
<i>cis</i> -Piperitone oxide <	19.140	0.22								
Dec-(4 <i>Z</i>)-en-1-ol	19.153									0.11
Neobergamate	19.336	2.19	0.07							
Neomenthyl acetate	19.906			0.24						
(<i>E</i>)-Cinnamaldehyde	20.007					61.48				
isobornyl acetate	20.26								1.60	
(<i>E</i>)-Anethole	20.319					3.48				
Lavandulyl acetate	20.433	31.09								
Bornyl acetate	20.49								0.13	
Menthyl acetate	20.568			9.39						
Thymol	20.674		39.14							
Carvacrol	20.890		6.03							
Isomenthyl acetate	21.038			0.29						
Nonyl acetate	21.100			0.12						

cis- α Terpinyl acetate	21.124	0.18								
iso-Verbanol acetate	21.181			0.18						
Myrtenyl acetate	21.59								0.06	
n-hexyl-Tiglate	21.758	0.19								
Citronellyl acetate	22.087	0.64								
α -Cubebene	22.424									0.11
neoiso-Dihydro carveol acetate	22.438	1.03								
Carbonic acid <4-cycloocten-1-yl-, methyl-> ester	22.529	0.84								
Linalool isobutanoate	22.815	4.27								
Eugenol	22.649					4.08	78.84			
Propan-2-one <methoxy-3-phenyl->	23.019					0.15				
Longicyclene	23.055				0.10					
Isoledene	23.105									0.12
α -Copaene	23.185	0.23			0.27	0.12	0.25			0.20
2-epi-a- Funebrene	23.189		0.10							
β -Patchoulene	23.248									0.13
trans-Geranyl acetate	23.320	0.81			0.12					
But-(2E)-en-1-one <1-(2,4,4-trimethyl-, 2-cyclohexen-1-yl)->	23.325		0.20							
β Cubebene	23.538	0.18								
Tetradec-1-ene	23.548					0.04	0.06			
β -Elemene	23.596			0.52	0.13					0.08
Longifolene	23.957					0.05				
Decyl acetate	23.969			0.27						
α -Gurjunene	24.042									0.59
(E)- β -Caryophyllene	24.268	1.18	3.57	1.55	2.22	3.61	11.93		2.48	0.71

β -Copaene	24.479			0.15						
γ -Maaliene	24.452									0.12
α -Guaiene	24.545				0.07					
<i>n</i> -octyl-Isovalerate	24.598			0.20						
Maaliene <alpha->	24.609									0.12
Himachalene <alpha->	24.703		0.07						0.04	1.99
(<i>E</i>)-Cinnamyl acetate	24.770					6.73				
Selina-5,11-diene	24.786									0.26
<i>trans</i> -Muuroala-3,5-diene	24.929									0.30
<i>trans</i> -Prenyl limonene	24.999		0.29		1.74					
α -Humulene	25.003	0.39		0.22		0.58	3.62		2.78	0.17
9-epi-(<i>E</i>)-Caryophyllene	25.154						0.06			0.95
<i>trans</i> -Dodec-2-en-1-ol	25.165	0.17								
Cadina-1(6),4-diene	25.385						0.10			0.62
γ -Muurolene	25.440			0.05						0.07
γ -Gurjunene	25.440				0.64					
Germacrene D	25.450			0.71			0.06			
α -Amorphene	25.507				0.59					
α -Curcumene	25.526					0.04				
β -Selinene	25.650				1.58					0.19
δ -Selinene	25.710				0.60					0.38
γ -Amorphene	25.755									0.18
α -Selinene	25.814				1.62		0.06			
1-Methyl-3-(4-methylpent-3-enyl)cyclohex-3-ene-1-carboxaldehyde	25.815	0.78								
Bicyclogermacrene	25.836			0.37						3.33
α -Muurolene	25.883						0.06			
β -Dihydroagarofuran	25.910				0.09					

(<i>E,E</i>)- α -Farnesene	25.960						0.10			
Lavandulyl <2-methyl-> butyrate	25.979	0.60								
β -Bisabolene	25.995					0.02				
δ -Amorphene	26.005				0.52					
δ -Cadinene	26.135			0.08						
α -Bulnesene	26.035									0.12
γ -Cadinene	26.137	0.28								
Z-nerolidol	26.286		0.11	0.25	0.17	0.15	1.20			2.12
γ -Cuprenene	26.334									0.44
Cadina-1,4-diene <trans->	26.448						0.06			0.41
Selina-4(15),7(11)-diene	26.510				0.26					
Selina-3,7(11)-diene	26.622				0.23					
α -Calacorene	26.631					0.03				
<i>epi</i> -Longipinanol <	26.808	0.30				0.03	0.30			
Longipinanol	26.883				0.21					
Guaiol	26.954									0.16
Ledol	27.068									0.18
Fokienol	27.236			0.11						0.15
Caryophyllene oxide	27.311	3.50		0.22	0.16	0.37	2.69		0.63	
Viridiflorol	27.339		0.31						2.62	0.67
Guaiol	27.480									0.52
Cubenol <1,10-di- <i>epi</i> ->	27.512					1.37				
Eremoligenol	27.623									0.21
allo-Aromandendrene epoxide	27.7								0.48	
Bulnesol	27.951									0.62
Patchoulyl acetate	28.027				0.62					
Caryophyllene acetate	28.154									0.22
Guaiac acetate	28.341				2.00					

Guaiac acetate	28.520				0.41					
β -Bisabolenol	29.904					0.30				
Manool	30.0								0.08	
(5Z,9E)-Farnesyl acetone	31.067		0.09							

Table S2. The antimicrobiological activity of EOs.

Essential oil	Main constituents	Microorganism	Reference	
<i>Lavandula angustifolia</i> Mill.	linalyl acetate linalool	<i>Herpes simplex</i>	[15]	
		<i>Staphylococcus aureus</i> (MRSA)	[17]	
		<i>S. aureus</i> ATCC 25923	[13]	
		<i>S. aureus</i> MRSA / ORSA		
		<i>Escherichia coli</i> [enro (-)]		
		<i>Pseudomonas aeruginosa</i>		
		<i>Salmonella typhimurium</i> ATCC 14028		
		<i>Candida albicans</i> ATCC 10231	[18]	
		<i>Enterococcus</i> sp. (VRE)		
		<i>Shigella flexneri</i>		[20]
		<i>Haemophilus influenzae</i>		[23]
		<i>Aureobasidium pullulans</i>		[25]
		<i>Penicillium citrinum</i>		
		<i>Penicillium simplicissimum</i>		
		<i>Botrytis cinerea</i>		[26]
<i>Fusarium solani</i> var. <i>coeruleum</i>	[27]			
<i>Aspergillus niger</i>				
<i>Aspergillus tubingensis</i>				
<i>Thymus vulgaris</i> L.	thymol p-cymene γ -terpinene	<i>Herpes simplex</i>	[32]	
		influenza virus A1/Denver/1/57 (H1N1)	[33]	
		<i>Streptococcus mutans</i>	[38]	
		<i>Streptococcus pyogenes</i>	[39]	

		<i>S. aureus</i> ATCC 25923	[28]
		<i>K. pneumoniae</i> ATCC 13882	
		<i>Brachyspira hyodysenteriae</i>	
		<i>Malassezia furfur</i> (drożdże)	[40]
		<i>Fusarium graminearum</i> Fg 06-17	[41]
		<i>Aspergillus flavus</i>	[43]
		<i>Aspergillus niger</i>	
		<i>Botrytis cinerea</i> ATCC12481	[44]
		<i>Penicillium expansum</i>	
		<i>Herpes simplex</i>	[47]
		<i>Escherichia coli</i> WDCM 00013	[49]
		<i>Listeria monocytogenes</i> WDCM 00020	
		<i>Pseudomonas aeruginosa</i> WDCM 00024	
		<i>Salmonella enterica</i> WDCM 00030	
		<i>Staphylococcus aureus</i> WDCM 00032	
		<i>Candida albicans</i>	[7, 50, 52]
		<i>Candida tropicalis</i>	[50]
		<i>Pichia anomala</i>	
		<i>Saccharomyces cerevisiae</i>	
		<i>Streptococcus mutans</i>	[56]
		<i>Alternaria alternata</i>	[57]
		<i>Aspergillus flavus</i>	
		<i>Aspergillus niger</i>	
		<i>Colletotrichum gloeosporioides</i>	
		<i>Fusarium solani</i>	
		<i>Macrophomina phaseol</i>	
		<i>Bacillus cereus</i>	[63]
		<i>Bacillus subtilis</i>	
		<i>Corynebacterium diphtheriae</i>	
		<i>Corynebacterium minutissimus</i>	
		<i>Enterococcus faecium</i>	
		<i>Listeria monocytogenes</i>	
<i>Mentha piperita</i> (L.) Hudson	menthol menthone cineol menthyl acetate isomenthone		
<i>Melaleuca leucadendron</i> L.	1,8-cyneole γ -terpinene α -pinene viridiflorol		

		<i>Staphylococcus epidermidis</i>	
		<i>Alcaligenes faecalis</i> ATCC 356551	[65]
		<i>Bacillus cereus</i> ATCC 14579	
		<i>Enterobacter cloacae</i> ATCC 23355	
		<i>Escherichia coli</i> ATCC 25922	
		<i>Streptococcus faecalis</i> ATCC 19433	
		<i>Staphylococcus aureus</i> ATCC 259231	
		<i>Micrococcus luteus</i> ATCC 4698	
		<i>Candida albicans</i> ATCC 14053	
		<i>Acinetobacter</i> spp.	[59]
		<i>Klebsiella</i> spp.	
		<i>Pseudomonas aeruginosa</i>	
		<i>Aspergillus niger</i>	
<i>Cinnamomum zeylanicum</i>		HSV1	[72]
		H1N1	[33, 72]
		<i>Acinobacter baumannii</i>	[73]
		<i>Staphylococcus aureus</i>	[73, 77]
		<i>Escherichia coli</i>	[73, 77, 81]
		<i>Pseudomonas aeruginosa</i>	[73,77]
		<i>Borrelia burgdorferi</i>	[74]
		<i>Enterococcus faecalis</i>	[77]
		<i>Salmonella typhimurium</i>	[6]
		<i>Listeria monocytogenes</i>	
		<i>Escherichia coli</i>	[81]
		<i>Candida albicans</i>	
		<i>Escherichia coli</i> PTCC 1163	[79]
		<i>Staphylococcus aureus</i> PTTC 25923	
		<i>Aspergillus niger</i>	
		<i>Cladosporium herbarum</i>	[83]
		<i>Fusarium culmorum</i>	
		<i>Fusarium oxysporum</i>	
		<i>Fusarium solani</i>	
	<i>trans</i> -cinnamaldehyde cinnamyl aldehyde benzaldehyde borneol eugenol cinnamic acid		

		<i>Fusarium verticillioides</i>	
		<i>Fusarium poae</i>	
<i>Cinnamomum cassia</i> (L.) J. Presl		<i>Streptococcus mutans</i> ATCC 35668	[80]
<i>Eugenia caryophyllata</i> Thunb	eugenol eugenol acetate anethol benzyl salicylate β-caryophyllene	<i>Herpes simplex</i>	[32, 55, 87]
		<i>Bacillus cereus</i> WU10	[89]
		<i>Escherichia coli</i> WU40, W1485, K12	
		<i>Listeria innocua</i> WU 507	
		<i>Salmonella typhimurium</i> WU73	[90]
		<i>Aeromonas hydrophila</i> ATCC 7966	
		<i>Candida albicans</i> ATCC 10231	
		<i>Klebsiella pneumoniae</i> ATCC 13883	
		<i>Proteus mirabilis</i> ATCC 10005	
		<i>Pseudomonas aeruginosa</i> ATCC 27853	
		<i>Staphylococcus aureus</i> ATCC 6538,	[10]
		<i>Staphylococcus epidermidis</i> ATCC 14990	
		<i>Escherichia coli</i> ATCC 8739	
		<i>Streptococcus pyogenes</i>	
		<i>Campylobacter jejuni</i>	
		<i>Haemophilus influenzae</i>	
		<i>Streptococcus agalactiae</i>	
		<i>Staphylococcus aureus</i>	
		<i>Klebsiella pneumoniae</i>	
		<i>Salmonella enteritidis</i>	
		<i>Bacillus subtilis</i>	
		<i>Morganella morganii</i>	
		<i>Yersinia enterocolitica</i>	[10, 92]
		<i>Listeria monocytogenes</i>	[91]
	<i>Candida albicans</i>	[86]	
	<i>Aspergillus niger</i>		
	<i>Penicillium christopherum</i>		
		<i>Mycobacterium phlei</i>	
<i>Eucalyptus globulus</i> Labill	1,8-cineol	<i>Herpes simplex</i>	[32]

	limonene α -pinene γ -terpinene α -terpineol	mumps virus	[100]
		Coxsackievirus B3 Nancy	[9,98]
		<i>Acinetobacter baumannii</i>	[101]
		<i>Porphyromonas gingivalis</i>	
		<i>Streptococcus mutans</i>	
		<i>Edwardsiella tarda</i> FP 5060	[102]
		<i>Lactococcus garviae</i> FP 5245	
		<i>Photobacterium damsela</i> FP 4101	
		<i>Streptococcus iniae</i> FP 5228	
		<i>Streptococcus parauberis</i> FP 3287	
		<i>Vibrio harveyi</i> FP 8370	
		<i>Vibrio ichthyoenteri</i> FP 4004	[103]
		<i>Staphylococcus aureus</i>	
		<i>Escherichia coli</i>	[95]
		<i>Aureobasidium pullulans</i> L6F	
		<i>Candida diversa</i> T6D	
		<i>Hansenula polymorpha</i> CBS 4732	
		<i>Pichia fermentans</i> T2A1	
		<i>Pichia kluyveri</i> T1A	
		<i>Pichia anomala</i>	
<i>Saccharomyces cerevisiae</i> SPA			
<i>Zygosaccharomyces bailii</i>			
<i>Alternaria alternata</i>	[104]		
<i>Fusarium roseum</i>			
<i>Mucor hiemalis</i>			
<i>Penicillium glabrum</i>			
<i>Salvia officinalis</i> L.	tamphor α -thujone 1,8-cineol β -thujone;	coronavirus SARS-CoV	[114]
		<i>Aeromonas hydrophila</i> ,	[122]
		<i>Klebsiella oxytoca</i>	
		<i>Aeromonas sobria</i>	
		<i>Bacillus megatherium</i>	[9, 122]
<i>Bacillus cereus</i>			

		<i>Bacillus subtilis</i>	[9, 115, 122]
		<i>Escherichia coli,</i>	[115]
		<i>Salmonella anatum</i>	[120]
		<i>Salmonella typhi</i>	[9, 116]
		<i>Shigella sonei</i>	
		<i>Salmonella enteritidis</i>	[116]
		<i>Staphylococcus aureus</i>	[9, 117]
		<i>Candida albicans</i>	
		<i>Staphylococcus epidermidis</i>	[118]
		<i>Streptococcus mutans</i>	[105]
		<i>Candida glabrata</i>	
		<i>Candida krusei</i>	
		<i>Candida parapsilosis</i>	
		<i>Aspergillus carbonarius</i>	[123, 124]
		<i>Aspergillus niger</i>	[9, 125]
		<i>Ashbiya gossypii</i>	[126]
		<i>Trichoderma reesei</i>	[126]
		<i>Rhizopus oryzae</i>	[126]
		<i>Botrytis cinerea</i>	[127]
		<i>Alternaria solani</i>	
		<i>Ascochyta rabies</i>	
		<i>Monilia laxa</i>	
		<i>Penicillum italicum</i>	
		<i>Rhizoctonia solani</i>	
<i>Melaluleca alternifoli</i> Tea tree Tea tree cd	terpine-4-ol teripene α -terpinene q-cymene	<i>Herpes simplex virus type 1 (HSV-1)</i>	[8, 32, 131]
		<i>Escherichia coli</i>	[132]
		<i>Staphylococcus aureus</i>	
		<i>Streptococcus mutans</i>	[129]
		<i>Porphyromonas endodontalis</i>	
		<i>Porphyromonas gingivalis</i>	[132, 134]
		<i>Candida albicans</i>	
		<i>Candida glabrata</i>	[8]

		<i>Aspergillus niger</i>	[135]
		<i>Penicillium expansum</i>	[136]
		<i>Botrytis cinerea</i>	[137]