Chiral Separation and Optimization on Substituted ß-Cyclodextrin Columns

Jana Hepner, Jaap de Zeeuw, Whitney Dudek-Salisbury, Kristi Sellers, Chris English; Restek Corporation

Basics of chiral chromatography

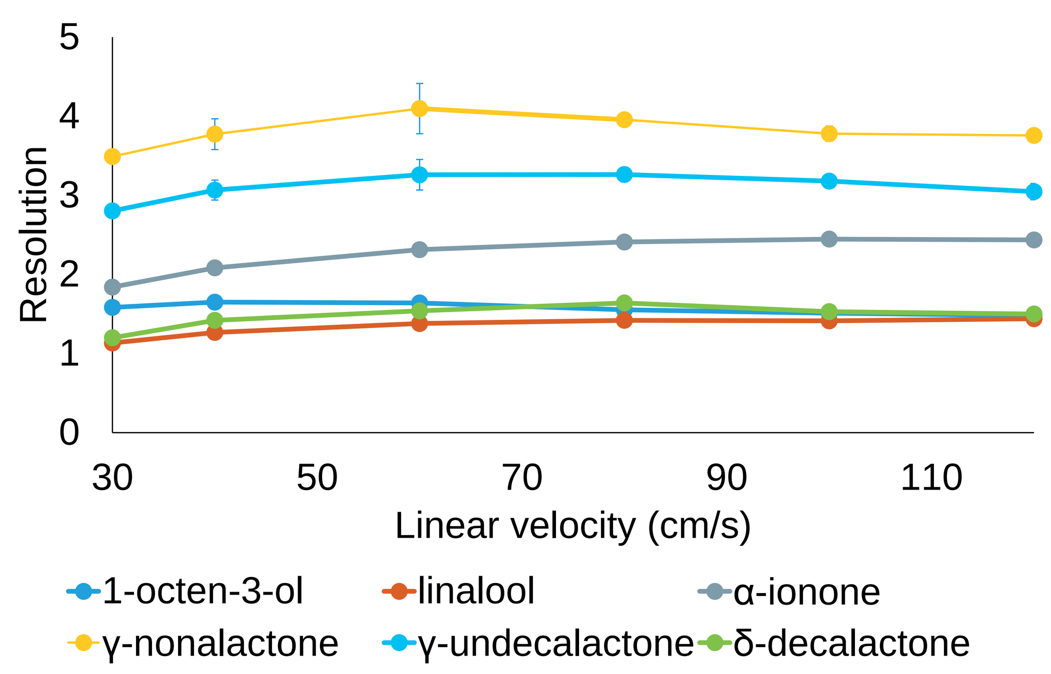
Chiral chromatography is the separation of enantiomeric compounds. Common liquid stationary phases used in gas chromatography resolve components from one another, but they do not possess adequate selectivity for enantiomeric separation. Addition of derivatized cyclodextrin macromolecules to common stationary phases creates capillary columns with the ability to separate enantiomers as well. The common chiral phase (permethylated derivative of betacyclodextrin in cyanopropyl-dimethylpolysiloxane liquid stationary phase) is used for such stereochemical separations, but it exhibits limited applications on its own. Beta-cyclodextrins derivatized with alkyl substituents can enhance the enantiomeric resolution of various compound classes. Restek's five capillary columns incorporate various combinations of alkylated beta-cyclodextrins into a cyanopropyl-dimethyl polysiloxane liquid stationary phase to achieve significant separation.

Optimization of chiral separation

Variation in linear velocity and temperature ramp rate can greatly affect the resolution of enantiomers. Depending on the type of chiral column, initial GC oven temperature can affect peak width. Column sample capacity varies with different compounds, and overloading results in broad tailing peaks and reduced enantiomeric separation.

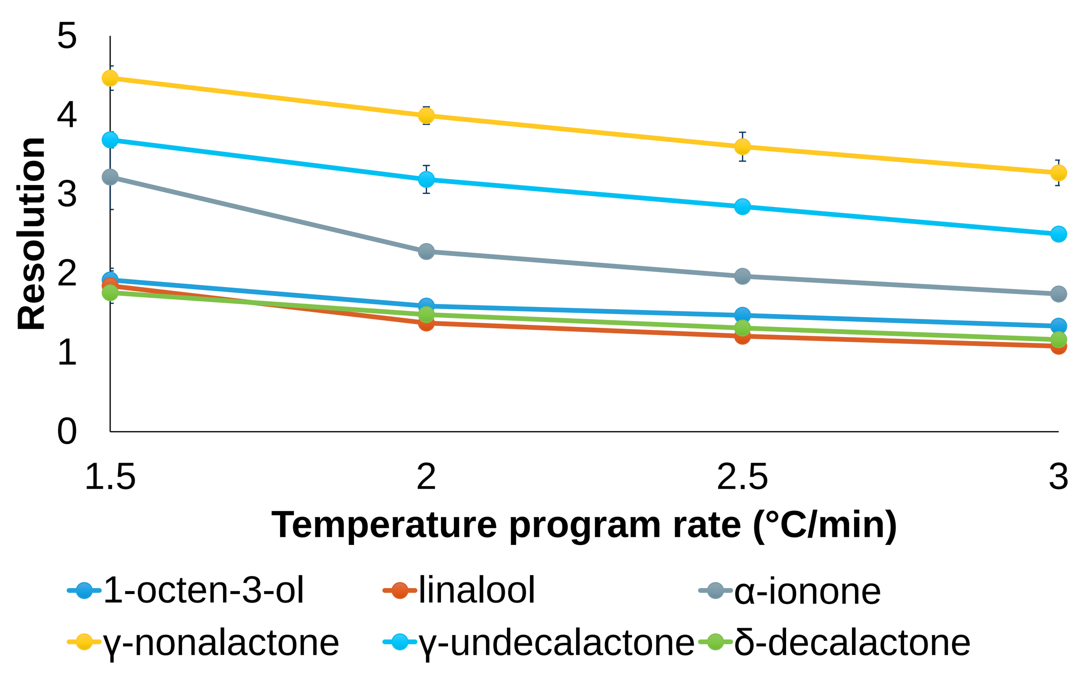
Linear velocity/column flow

Hydrogen's optimal linear velocity is around 40 cm/s at 40°C. However; better resolutions were achieved with higher flows/linear velocity (60-80 cm/s).

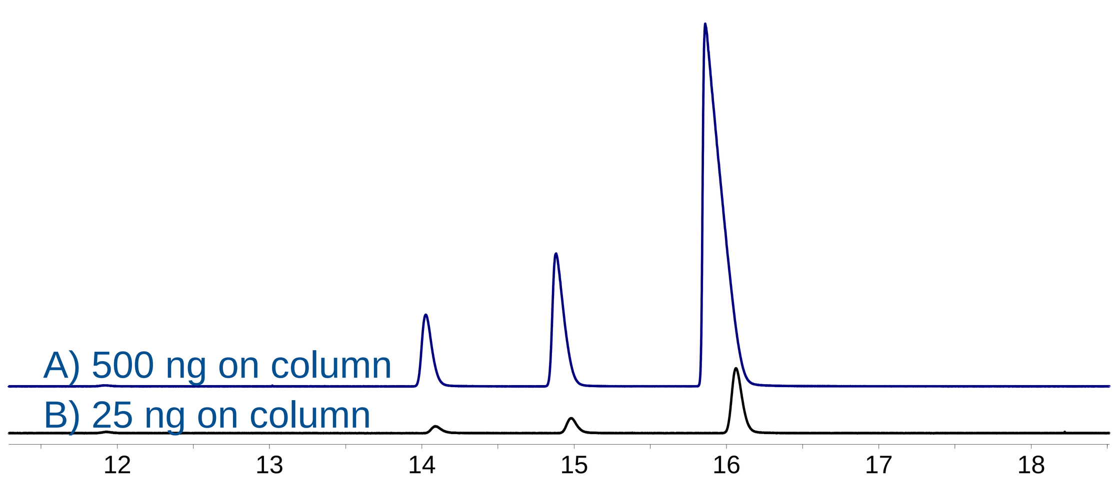


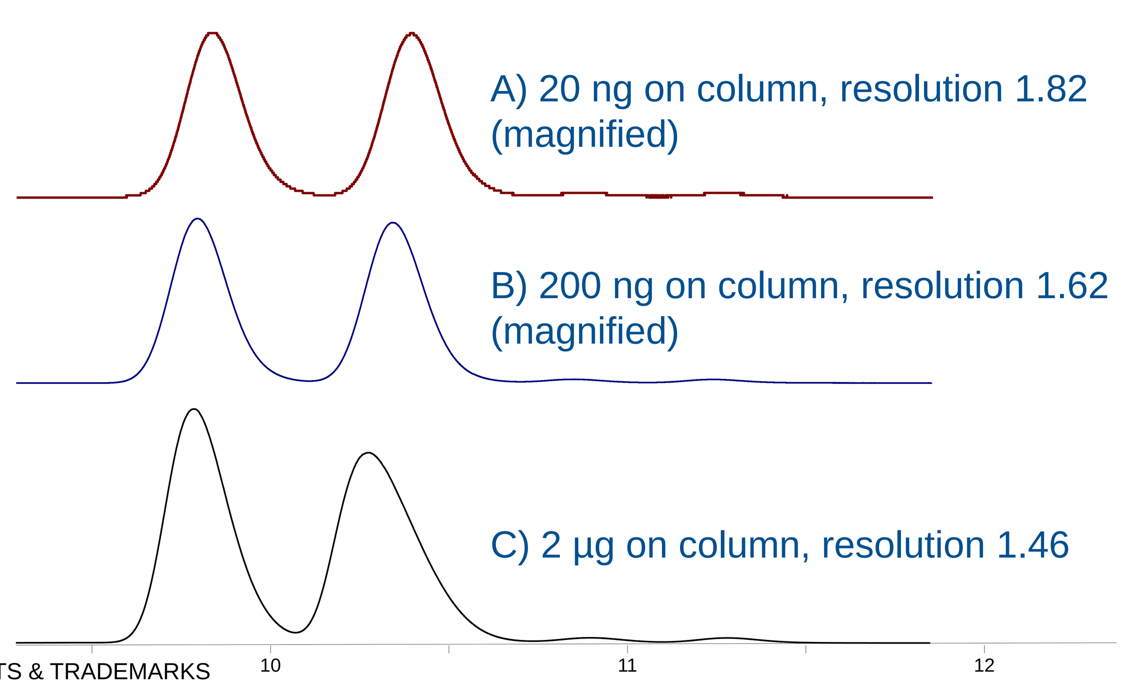
Temperature ramp

The chiral resolution can improve with decrease in the temperature ramp speed. The best temp programs are 1-2 °C/min



Overloading and tailing





Chiral separation on Restek's cyclodextrin-based columns

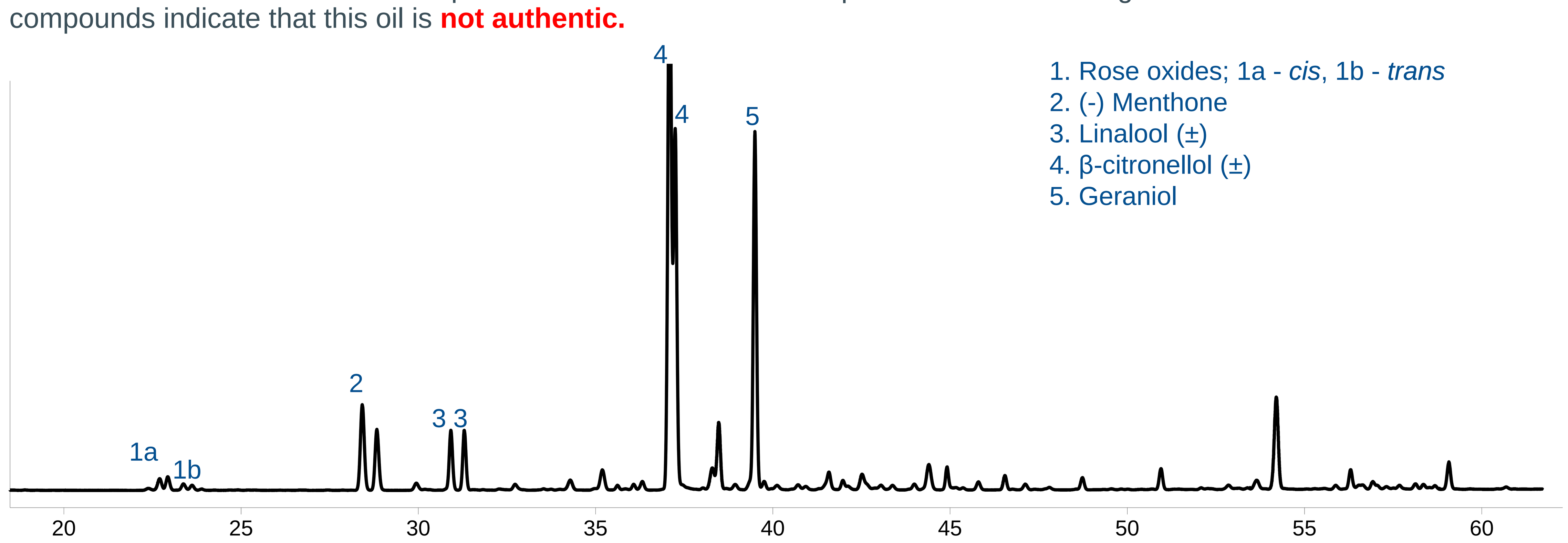
Compounds			Column Resolution Factors					
			Rt-βDEX sm	Rt-βDEX se	Rt-βDEX sp	Rt-βDEX sa	Rt-βDEX cst	Rt-βDEX m
Terpenes	1	α-pinene	3.14	0.82	ns	ns	0.82	3.19
	2	limonene	5.60	8.05	1.22	ns	2.04	1.41
Alcohols	3	1-octen-3-ol	1.08	ns	ns	2.00	ns	ns
	4	linalool	3.10	5.96	1.79	1.60	2.25	1.01
	5	α-terpineol	4.72	5.20	1.29	2.82	2.69	1.54
	6	terpinen-4-ol	1.90	1.92	ns	2.47	ns	1.41
	7	isoborneol	3.76	3.35	ns	0.79	1.87t	2.15
	8	β-citronellol	0.80	0.89	ns	0.98	ns	ns
	9	menthol	1.24	1.24	1.07	0.93	0.89	1.58
	10	2,3-butanediol	6.44	7.10	1.46	2.61	1.27	2.43
	11	1-phenylethanol	7.52	6.52	ns	6.43	4.87	5.88
Ketones	12	camphor	1.70	2.13	0.50	4.20	2.22t	ns
	13	α-ionone	5.67	3.31	0.80	4.69	1.37	2.81
	14	menthone	0.59	5.76	ns	1.16	2.60	4.11
Lactones	15	γ-nonalactone	4.19	5.07	2.11	4.00	3.82	1.03
	16	γ-undecalactone	2.39	3.24	1.20	3.65	3.18	ns
	17	δ-decalactone	0.80	ns	ns	1.91	1.69	ns
	18	ethyl-2-methylbutyrate	3.94	4.66	ns	ns	ns	0.92
Esters	19	linalyl acetate	ns	2.36	ns	ns	ns	ns
	20	styrene oxide	4.53	10.77	2.86	2.26	1.16t	3.03
Epoxides	21	trans-linalool oxide	9.71	2.96	ns	1.20	3.02t	7.44
	21	cis-linalool oxide	6.06	4.28	ns	0.91	2.19t	4.38

Rose oil analysis on Rt-ßDEXsa

The Rt-βDEXsa has a significantly different selectivity than the other chiral columns. It provides the best separation of 1-octen-3-ol, carvone, camphor, β-citronellol, and rose oxides, which makes it a good fit for certain essential oils' analysis.

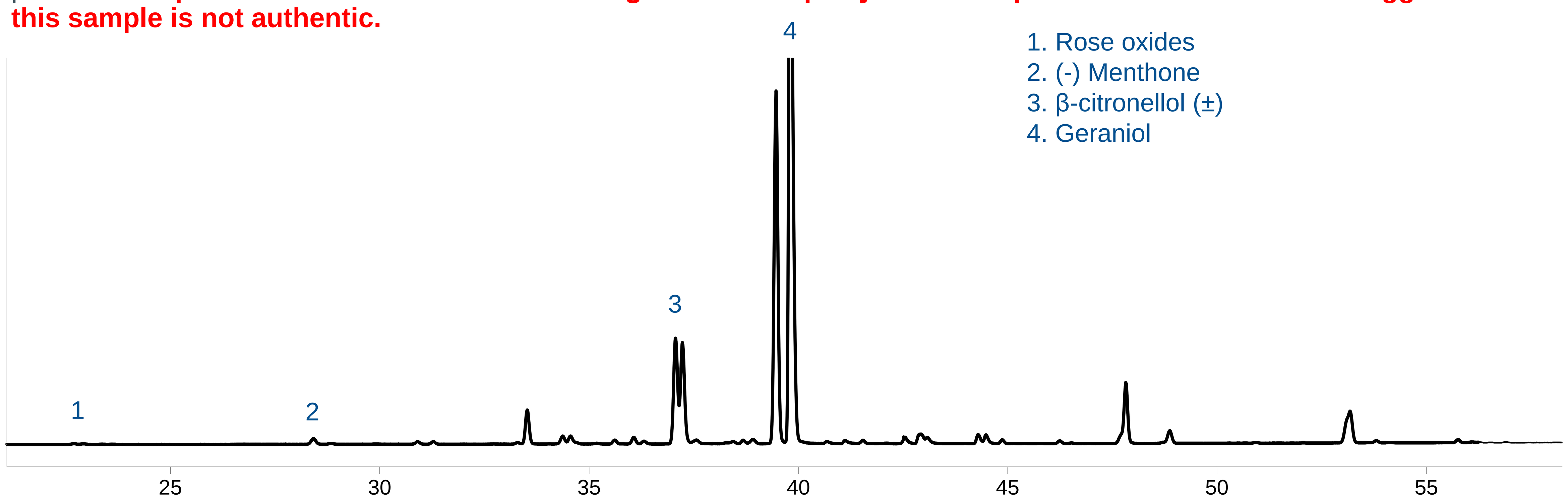
Geranium oil

Geranium is a plant from Rosids clade, thus its oil's composition is similar to the rose oil. Chiral constituents in geranium oils include cis and trans rose oxides, menthone, linalool, and β-citronellol. The Rt- βDEXsa column provides chiral resolution for all of these compounds. In authentic samples of geranium oil, (-)-(4R)-configured diastereomers of *cis-* and *trans-*rose oxides predominate over their (+)-enantiomers. **The (-)-(S) form of ß-citronellol should be 74-80% of the enantiomeric ratio.** Note that *cis-* and *trans-*rose oxides and β-citronellol are racemic in this particular commercial geranium oil. These racemic compounds indicate that this oil is **not authentic.**



Rose oil

As geranium oil, rose oil also contains rose oxides and β-citronellol. Menthone is present as well, however in a lower amount. In this sample of rose oil, the rose oxides were also in much lower amount than in the geranium and all four isomers were present. **The presence of all rose oxides with together with equally abundant β-citronellol enantiomers suggest that this sample is not authentic.**



Rose Fragrance

The rose fragrance had some of the typical components of the rose oil, however, the rose oxides were completely missing