Essential oil of peppermint (*Mentha* × *piperita* L.) from fields in Eastern Slovakia

A. SÚSTRIKOVÁ[1], I. ŠALAMON[2]

[1]*Research Institute of Agroecology, Michalovce, Slovak Republic*
[2]*Department of Ecology, Prešov University, Prešov, Slovak Republic*

ABSTRACT: The peppermint, *Mentha* × *piperita* L., is a plant that represents the oldest and traditional medicinal herbs used in both Eastern and Western traditions until recent time. The peppermint has a history of use in herbal medicine dating back to the ancient Egyptian, Greek and Roman times (MURRAY 1995) although it need not have been used for the same reasons. The large therapeutic effects of peppermint dry drug as well as essential oil in human medicine caused the peppermint to be appreciated by the pharmaceutical industry. The world peppermint production is realized by large-scale cultivation using suitable intensive practices. Studies of the qualitative and quantitative characteristics of peppermint essential oil produced under the agroecological conditions of Eastern Slovakia confirmed its high composition quality considering the heavy metal contamination. At the same time, the determination of peppermint essential oil composition in comparison with the analysis of peppermint oil produced in the other parts of the world suggested its competitive quality parameters in the world market. A suitable menthol content of peppermint of Slovakian provenience predestinates this peppermint gene material for the breeding of new cultivars opposite to foreign ones and its introduction into agricultural conditions.

Keywords: peppermint; *Mentha* × *piperita* L.; qualitative and quantitative composition of essential oil; Eastern Slovakia

In the world markets there are many medicinal and aromatic plants from different parts of the world characterised by various therapeutic effects. Slovakia represents traditional producers and growers of medicinal and aromatic plants. Sufficient herb composition with a maximum of effective substances is the main condition for their further cultivation and processing by the Slovak pharmaceutical industry as well as their export abroad. This contribution deals with national project *Protection of Plant Gene Pool in the Slovak Republic*. Research Institute of Agroecology in Michalovce is engaged in partial tasks of this project: collecting expeditions of genetic resources of selected plants in the East-Slovakian Lowland, determination by GC and GC-MS of medicinal and aromatic plant chemotypes that were collected in this part of Slovakia.

Generally, the reconnoitring of the autochthonous gene pool of medicinal and aromatic plant species as well as of the cultural gene pool of field crops provides the most suitable chemotypes that are necessary to be introduced into further plant breeding programmes and also large-scale production.

Peppermint genetic resources could be recommended for a breeding process to obtain new varieties. The technology of peppermint production was successfully tested in the East-Slovakian Lowland and the results confirmed satisfactory ecological quality of peppermint drug.

In recent time the peppermint has belonged to economically important aromatic and medicinal plants commercially grown in many areas of the world. The three biggest world producers and exporters of peppermint raw material and essential oil in the world market are the U.S.A., Japan and Great Britain. Germany, Russia, Italy, Bulgaria, Greece, Norway, etc. produce lower volumes of the mentioned plant material.

The peppermint (*Mentha* × *piperita* L.), as a representative of the large family Lamiaceae, belongs to the oldest and traditional medicinal and culinary herbs.

Peppermint is a perennial plant with procumbent, ascending, branched, reddish stems, quite smooth or fringed, with a few spreading hairs, furrowed and quadrangular, and 2 or 3 feet in height. The leaves are ovate-oblong or somewhat lanceolate, rounded at the base, deep-green, smooth or hairy on the underside, serrate, and borne on ciliated petioles. The flowers are in whorls, small, and purplish; upper floral leaves are small, lanceolate-subulate, shorter than the flowers. The whorls are few, lax, uppermost in a short, oblong, obtuse, reddish spike; lowermost remote, with the cymes shortly stalked. Bracts subulate, outer ones as long as the calyx. Pedicels are quite smooth. Calyx 5-toothed, hispid teeth, subulate, erect. The corolla is 4-cleft, tubular, with the broadest segment emarginated (DOSTÁL 1989).

Peppermint is indigenous to England, and has been extensively cultivated in various parts of Europe and throughout the United States. It grows wild in damp places. The whole plant has a peculiar, aromatic, diffusive odour, and an agreeable, warm, burning, bitter taste, followed by a feeling of coolness during inhalation. These properties are more marked in the fresh than in the dried plant and both the odour and taste may be reserved for a long time.

The entire herb is medicinal (peppermint dry leaves and leafed shoot tops are the pharmacopoeial material) usually used as a component of herb teas and mixtures, but much more frequently the peppermint essential oil (*Menthae piperitae aetheroleum*) is used in cosmetics, pharmaceutical and food industry.

Peppermint is a powerful diffusible stimulant. Medicines made of it may be applied both externally and internally. From the phytotherapeutic aspect the peppermint drug is allowed to be a specially important stomachic, cholagogue, antispasmodic, carminative, anodyne, antiseptic, tonic, refrigerant and fancied ingredient modifying the action and masking the flavour – olfactory properties of remedies. Peppermint tea or essential oil can be taken for nervousness, insomnia, cramps, coughs, migraine, poor digestion, heartburn, nausea, abdominal pains, and various problems such as headache and vomiting due to nervous causes (LUST 1974).

The major active component of peppermint dry herb and leaves is essential oil whose content ranges from 0.5 to 4%. Other compounds found in the peppermint are: flavonoids (12%), polymerised polyphenols (19%), carotenes, tocopherols, betaine and choline (MURRAY 1995).

Distillation of peppermint oil in the United States began in 1816. Until 1846 distillation was carried out by means of simple copper stills heated by direct fire; since that date distillation by steam has become the rule.

Essential oil is contained in little vesicles existing throughout the plant, and visible in the leaves. Peppermint essential oil has unique therapeutic properties that have been documented by use throughout history as one of the most well known essential oils.

Peppermint oil is a very grateful agent to allay nausea and vomiting, indigestion, fevers, flatulence, headaches, migraine, liver problems and arthritis. It is stimulating to the nervous system, cooling to the body for fevers or in hot weather. A strong digestive aid and breathe freshener. Peppermint's strong antispasmodic action makes it useful in massage for sports injuries. Its anti-inflammatory action helps sciatica, neuralgia, and arthritis. The oil of peppermint, on account of the menthol present in it, is a local anaesthetic, and may be employed to relieve local pain such as in the inflamed joints of rheumatism, as a spray in painful inflammation of the throat and fauces, and in any painful condition where a direct application of the anaesthetic can be made. Its stimulating ability helps mental concentration and memory. It is very stimulating to the mind, relieves mental fatigue and depression.

Menthol, a monoterpene (10 carbons) isolated from various mints, is a topical pain reliever and antipuretic. Plants in the mint family have been used for medicinal purposes since before 2000 BC, but menthol was not isolated until 1771. The first knowledge explaining the menthol chemical structure and properties was provided by SIMONSEN (1947) scientific research. Fig. 1 illustrates the chemical structures of natural forms of menthol.

Menthol activates coolness feeling on the skin by the specific irritation of nervous axons with the desensitisation in the locality of application (WATSON 1978). It has anaesthetic, antiseptic, antibacterial effects, removes the itch and reduces gland secretion. It is a suitable carminative and antiemeticum.

This paper is aimed at the study of qualitative and quantitative characteristics of peppermint essential oil cultivated under the agroecological conditions in Eastern Slovakia and isolated by distillation.

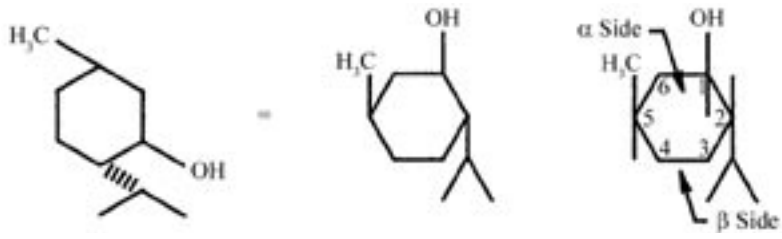
Qualitative and quantitative analysis of peppermint essential oil presents the quality composition and character of the peppermint gene pool habit cultivated in the East-Slovakian Lowland.

MATERIAL AND METHODS

The plant material, peppermint dry leaves and leafed shoot tops, was obtained from *Mentha piperita* L., an interesting chemotype with increased content of menthol, from large-scale cultivation in various parts of the East-Slovakian Lowland. In regard to varying soil and climatic conditions, the East-Slovakian Lowland provides heterogeneous of facilities to the occurrence of various plant species that are characterised by biodiversity and abundant gene pool material. On the other hand, this plain is one of the most productive regions of the Slovak agriculture.

Essential oil from peppermint was isolated by distillation with water. Hydrodistillation lasted for 2 hours, sample weights were 2 g of drug dry matter. A modified distillation apparatus manufactured by Coocking & Middleton was used.

The identification and evaluation of peppermint essential oil were realised by means of some analytic methods. Main compounds of essential oil were identified and determined by gas chromatography under experimental conditions.



(–) - Menthol (–) - Menthol (–) - Menthol
1R (1α, 2ß, 5α) - Menthol

Fig. 1. The chemical structures of natural forms of menthol

Table 1. Selected physical and chemical parameters of essential oil cultivated in the East-Slovakian Lowland (2002)

Physical and chemical parameters	Technical parameters	Determined parameters
Consistency (Ph. Eur. 1997)	0.900–0.916 g/cm^3	0.899 ± 0.001 g/cm^3
Optical versatility (Ph. Eur. 2001)	–10 to –30°	–24 ± 1°
Refractive index (Ph. Eur. 2001)	1.457–1.467	1.464 ± 0.001
Acidity number (Ph. Eur. 1997)	max. 1.4 mg KOH/g	0.6 ± 0.1 mg KOH/g

Total composition of peppermint essential oil was carried out using an apparatus HEWLETT – PACKARD 5890/5970 GC/MSD system, with split-splitless system for injection, MSD detector, column BPX – 5 (SGE Ltd., Melbourne), fused capillary column, 50 m long × 0.25 mm i.d., film thickness 0.25 mm. The following experimental conditions were observed: carrier gas UHP helium, column pressure 21 p.s.i. (flow velocity 250 mm/s), injection temperature 240°C, detector temperature 290°C, oven temperature was programmed to 50–250°C at 3°C/min, and then maintained for 15 minutes. Sample sizes were 1.0 µl and manual type of injection was used.

Another type of possible GC analysis used for the determination of peppermint essential oil is HEW-LETT – PACKARD 5890 Series II system, with capillary column HP – 5, 50 m long × 0.25 mm i.d., film thickness 0.25 mm, FID detector, split-splitless system for injection and automatic injector HP 7673. The following operating conditions were used: injection temperature 150°C, detector temperature 250°C, carrier gas nitrogen (flow velocity 274 mm/s), auxiliary gases nitrogen (30 ml/min), hydrogen (30 ml/min), air (400 ml/min). The following temperature programme was used: 90°C (0 min), 10°C/min at 150°C (5 min), then 5°C/min at 180°C (maintained for 3 min), then 7°C/min to finally isothermal 280°C maintained for 25 min. Sample sizes were 1.0 µl and manual type of injection was used.

Determination of main components of essential oil was realised on the basis of the use of standard compounds and knowledge acquired by studying technical literature. The following authentic compounds were purchased for subsequent identification: (–) – menthol, menthone, menthofuran, menthylacetate, carvone, isomenthone, limonene and linalool. Merck, Fulka, Sigma and Roth companies delivered standard compounds.

Qualitative determination of selected components was realised by a comparison of retention times of all detected components with retention time of standard compounds. Peak areas and retention times were measured by electronic integration.

Selected physical and chemical characteristics (consistency, optical versatility, refractive index and acidity number) of peppermint essential oil were determined by the methods defined in Slovak Dispensatory I (Pharmacopoea Slovaca I. 1997).

RESULTS AND DISCUSSION

Peppermint oil can be described as a colourless or yellowish or greenish-yellow liquid, becoming darker and thicker with the age and exposure to the air, having the characteristic strong odour of peppermint and a strongly aromatic pungent taste, followed by a sensation of cold when the air is drawn into the mouth. It forms a clear solution with an equal volume of alcohol, becoming turbid when somewhat further diluted, and is soluble in all proportions, in carbon disulphide and in glacial acetic acid. The alcoholic solution of the oil is neutral to litmus paper.

In accordance with the distillation process selected physical and chemical characteristics of peppermint essential oil were determined (Table 1).

The obtained results of the parameters – consistency, optical versatility, refractive index and acidity number confirmed the fact that the observed parameters were in compliance with pharmacopoeial limits.

The optical laevorotatory versatility of essential oil ranges in American oil from –25° to –33° (GILDEMEISTER, HOFFMANN 1961).

The composition of essential oil from peppermint determines its therapeutic effects. The most relevant

Table 2. Qualitative and quantitative parameters of peppermint essential oils cultivated in the different parts of Eastern Slovakia (1999–2001)

Peppermint essential oil	Percentage contents of major components					
	menthol	menthone	menthyl acetate	iso-menthone	linalool	limonene
(1)	38.3	30.1	–	5.7	–	6.70
(2)	49.6	16.3	–	4.1	3.1	–
(3)	44.1	20.9	–	8.3	5.1	–
(4)	46.0	0.4	–	8.8	–	2.62
(5)	43.0	14.0	3.5	7.0	–	2.50
(6)	69.1	3.5	4.5	0.8	0.6	–

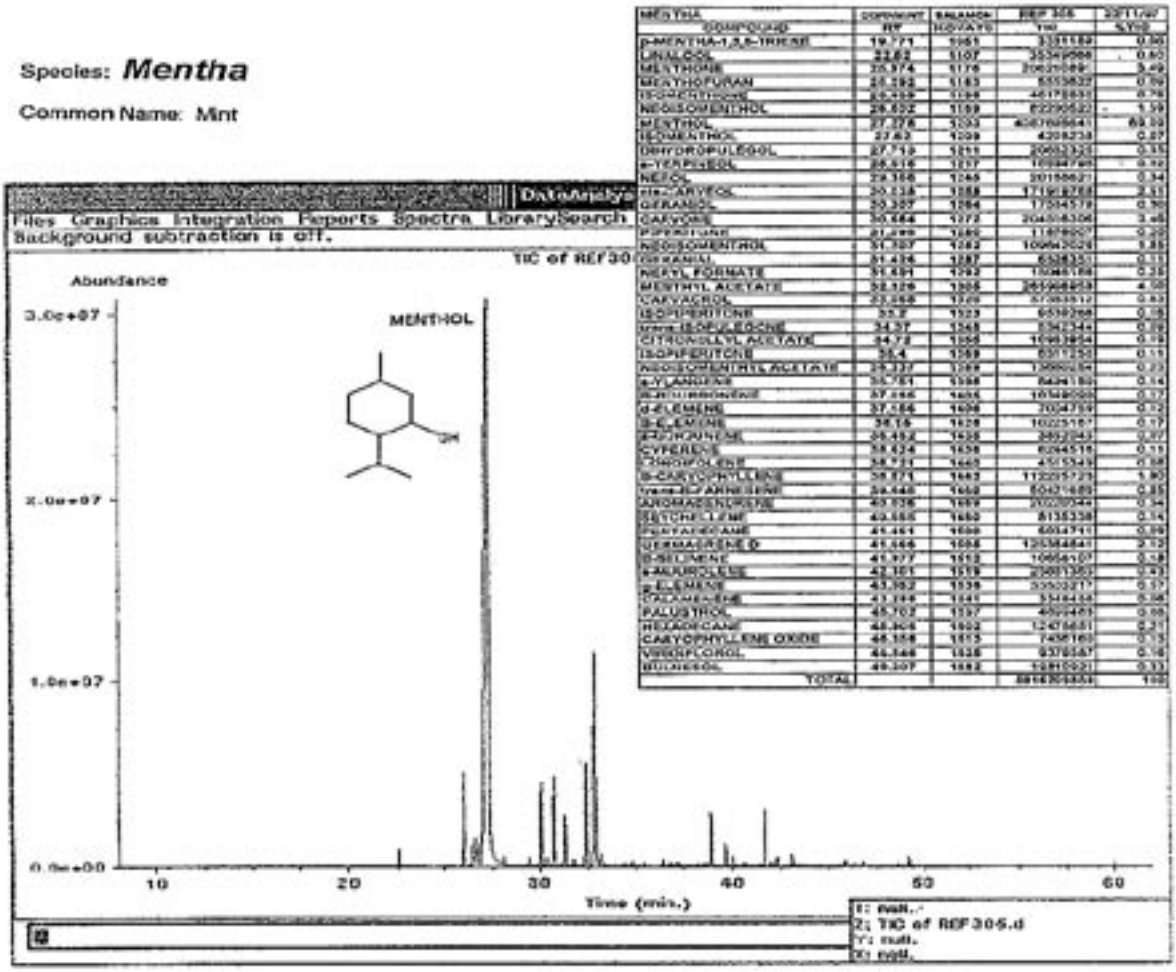


Fig. 2. Total GC – MSD identification of the peppermint essential oil composition, *Mentha* × *piperita* L., 2000

component is menthol whose content depends on the soil and climatic conditions of agricultural region, plant origin and date of harvest. Peppermint essential oil can consist of up to 40–70% of menthol. Menthol is a solid constituent of peppermint oil forming colourless, acicular or prismatic crystals; its characteristics are strong and pure odour and warm, aromatic taste followed by a sensation of cold when the air is drawn into the mouth. It was formerly known as peppermint camphor.

Other active constituents are menthone (7–25%), menthofuran (2.5–5%), menthylacetate (3.5%), carvone (3.5%) and less than 1% content of iso-menthone, 1.8-cineole, linalool, limonene, etc. Essential oil compounds of several peppermint cultivars have the same composition without expressive differences.

Slovak Dispensatory I (Pharmacopoea Slovaca 1997, 2001) demands the minimum content of essential oil 0.8–1.2%. LINDAUEROVÁ et al. (1991) reported the oil content of 1.5% in the variety Perpeta, which is one of the most frequently cultivated varieties in Slovakia.

TOMKO et al. (1989) determined the following contents of the peppermint essential oil constituents: esteric-bound (–) – menthol (50–80%), menthone (7–25%), menthofuran (2.5–5%), piperitone (menthenone, 3%), menthylacetate (3.5%), carvone (3.5%), iso-menthone (0.35%), linalool (0.6%), cineole, limonene and others.

Other active constituents are menthone (7–25%), menthofuran (2.5–5%), menthylacetate (3.5%), carvone (3.5%) and less than 1% content of iso-menthone, 1.8-cineole, linalool, limonene, etc. Essential oil compounds of several peppermint cultivars have the same composition without expressive differences.

Results of the qualitative and quantitative analysis of peppermint essential oil cultivated under different growing conditions in the various parts of Eastern Slovakia are illustrated in Table 2. According to the qualitative analysis peppermint essential oils cultivated in the East Slovakian region have a sufficient composition of the chief active constituents. The content of menthol ranged from 38 to 69%. The most important chemotype of peppermint with the highest content of menthol was introduced into cultivation under experimental conditions in Vysoká nad Uhom (Experimental Research Station of the Research Institute of Agroecology in Michalovce). 47 components of essential oil were identified in this chemotype. The active compounds with highest proportions were

menthol 69.1%, menthone 3.5%, menthylacetate 4.5%, carvone 3.5%, iso-menthone 0.8% and linalool 0.6%. Fig. 1 shows the chromatographic record of qualitative and quantitative determination of one analysed essential oil.

The qualitative and quantitative analyses of peppermint essential oils of Slovakian provenience carried out in the last years confirmed the same satisfactory parameters. ŠALAMON and SVOBODA (1996) reported the menthol content of 44–49% and menthone content of 16–21% in peppermint oil.

In regard to the comparison of qualitative characteristics of peppermint oil cultivated in Eastern Slovakia with peppermint oil originating from different countries of the world some results of foreign authors were published.

In Europe, Bulgaria is a major exporter of mint essential oil. The mentioned volatile oil has these qualitative parameters: menthol 26.7%, menthone 31.6%, iso-menthone 6.86% and linalool 0.4% (LAWRENCE 1987).

In Montana (U.S.A.) the most famous cultivar grown there is cv. *Black* Mitcham, whose composition of peppermint oil can be characterised by the following compound content: menthol 36%, menthone 21.0%, menthofuran 2.63%, menthylacetate 3.3% (WELTY et al. 2000). New York oil contains 50 to 60% of total menthol, of which 40 to 45% are free and the rest is combined in the form of ester. It also contains 12% of menthone. Michigan oil contains about 48 to 58% of total menthol, of which 43 to 50% are free menthol.

American oil of peppermint is subjected to cold, crystals of menthol fall out. The Japanese oil is semisolid at an ordinary temperature, owing to the large quantity of menthol it contains.

According to UMNEY (1896) English oil of peppermint contained 63 to 66% of total menthol, 3 to 14% of menthol in the form of ester, 9 to 11% of menthone.

The peppermint essential oil produced in Tasmania (Australia) has the following average qualitative parameters: menthol 29%, menthone 23–30%, menthylacetate 3–10% (PETERSON, BIENVENU 1999).

The peppermint oil composition depends on taxon, origin, soil and climatic growing conditions, used harvest and isolation methods and effective weed protection.

Peppermint is currently one of the most economically important aromatic and medicinal crops. The world production of peppermint oil is about 8,000 tons and the increase in demand for this raw material actually moves around 5% yearly (PETERSON, BIENVENU 1999).

The total production of peppermint essential oil has achieved enormous quantities, the United States alone producing 251,000 pounds in 1987. Lower quantities of peppermint oil are produced in Russia, Germany, Italy, Norway, etc. The yield from German herb (fresh) is reported to be 0.1 to 0.25%, from dried herb 0.7%.

The consumption of this natural product in the U.S.A. increases on average by about 15% per year. Natural products made from the peppermint essential oil were sold for 12 mil. US$ in 2000. The financial value of the world peppermint drug production is higher than 96 mil. US$. The sale price of peppermint oil ranges from 12 to 15 US$/kg.

Other mint species cultivated in the world for peppermint oil production are as follows: *Mentha pulegium* (creeping habit, occurs across the whole Europe, the main component – pulegone), *M. spicata* (spearmint, cultivated mainly across the U.S.A., the main component – carvone), *M. citrata* (eau de cologne, derived from spearmint, main components – linalool and linylacetate).

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Received for publication July 30, 2003
Accepted after corrections September 17, 2003

Silica mäty piepornej (*Mentha* × *piperita* L.) pestovanej na východnom Slovensku

ABSTRAKT: Štúdium kvalitatívno-kvantitatívnych charakteristík silice mäty piepornej produkovanej v agroekologických podmienkach východoslovenského regiónu potvrdilo jej vysoké kvalitatívno-kvantitatívne parametre. Znamená to, že silica mäty piepornej je kvalitou porovnateľná až lepšia ako silica veľkokapacitne izolovaná v iných štátoch sveta. Vykonané porovnania výsledkov analýz mätovej silice tak naznačujú možnosť vysokej konkurencieschopnosti na svetovom trhu s liečivými rastlinami a prírodnými produktmi. Podstatne vyšší obsah mentolu oproti kultivarom pestovaným v Severnej Amerike, Austrálii a Bulharsku predurčuje tento rastlinný materiál pre vyšľachtenie novej odrody a jej introdukciu do pestovateľských podmienok. Faktormi, ktoré môžu ovplyvniť kvalitu silice, získanej z veľkoplošného pestovania mäty, sú výber vhodného agroklimatického regiónu, použitie správneho pestovateľského postupu a ochranných prostriedkov, vytvorenie konkurenčne silného porastu, spôsoby zberu a izolácie silice z rastlinného materiálu. V konečnom dôsledku majú na kvalitatívno-kvantitatívne charakteristiky silice vplyv všetky faktory environmentálneho prostredia.

Kľúčové slová: mäta pieporná; *Mentha* × *piperita* L.; kvalitatívno-kvantitatívne zloženie silice; východné Slovensko

Corresponding author:

Ing. ANDREA SÚSTRIKOVÁ, Oblastný výskumný ústav agroekológie, Špitálska 1273, 071 01 Michalovce, Slovenská republika
tel.: + 421 56 644 38 88, fax: + 421 56 642 02 05, e-mail: ovua_marketing@minet.sk