

Variation of Essential Oils in Selected Species of the Genus *Echinacea* Moench During Vegetation

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Abstract: We have investigated the variation of essential oils in selected *Echinacea* species during the ontogenetic life of plants cultivated in three different locations. The contents of essential oil in the aerial and underground parts varied. The leaves and stems of *E. purpurea* were richer in essential oil, while *E. sanguinea* and *E. species* had relatively high essential oil contents in roots in the stage of biological maturity or in the prebud stage. With respect to climatic conditions, no significant differences in essential oil contents have been observed between particular locations.

Keywords: *Echinacea purpurea*, *Echinacea sanguinea*, *Echinacea species*, aerial and underground parts, essential oils.

Introduction

Healing herbs and their constituents, because of their natural origin, have been considered as the most natural medicines, in spite of scepticism about them in the recent past, when the beliefs in “almighty” synthetic drugs influenced also the expert opinions of their healing capacity. The comeback of medicinal plants started at the beginning of this century. Since then, this trend has taken on new dimensions and the interest in natural healing has increased worldwide. This development has been supported by the latest knowledge about the substances contained in medicinal herbs, which opened up prospects for production of new drugs.

A particularly important group of medicinal plants is represented by the species of the genus *Echinacea*, originating from North America. Among the ones processed by pharmaceutical industry have been *Echinacea purpurea*, *Echinacea angustifolia*, *Echinacea leavigata* and *Echinacea atrorubens*. They are perennial, 70 – 120 cm tall herbs with characteristic single, large flower heads terminating the stem. The *Asteraceae* are characterized by production of essential oils, balsams, latex and terpene-based substances.

From phenolic substances *Echinacea* plants contain in particular caffeic acid derivatives and flavonoids, which act as anti-inflammatory and antibacterial agents (PENGELLY 1996, SCHARR 1998). Cichoric acid, a polar derivative of caffeic acid, is concentrated in roots and flowers of *E. purpurea*, and in smaller amounts also in other species (BECKER & HSIEH 1985, BAUER et al. 1988, AWANG & KINDACK 1991). It stimulates cell-mediated immunity, has antioxidant and virostatic effects, inhibits the hyaluronidase enzyme (PAAS & GRANT 2002) has a stimulant effect on phagocytes (BONE 1998) and an inhibitive effect on the HIV infection (REINKE et al. 2004). Echinacoside is a glycoside with minor bactericide, hypotensive and analgesic effects, which is used for standardization of drugs (BECKER 1982a, BECKER & HSIEH 1985).

Important from the therapeutic point of view are polysaccharides, characterized by strong immunostimulant (WAGNER 1985, BAUER & WAGNER 1991, ROESLER et al. 1991, BODINET et al. 1993) and anti-inflammatory (TUBARO et al. 1987) effects. They have been shown to stimulate macrophage activity and several functions related to cytokine production (RININGER et al. 2000, GOEL et al 2002, RANDOLPH et al 2003). The composition of polysaccharides differs among species of *Echinacea*, among organs of the same species, among provenance, and among extracts based on the solvent used (BERGERON & GAFNER 2007).

An important constituent of the *Echinacea* species is essential oil. There is a variation in the amounts of essential oil, while its quality depends on particular plant organs and the cultivar used (BAUER & WAGNER 1991). MAZZA & COTRELL (1999) have analysed and identified over 70 volatile components in the plant parts of Canadian-grown *E. purpurea*, *E. angustifolia* and *E. pallida*. The main essential oils in all plant tissues, irrespective of species, were camphene, β -pinene, and limonene, together with other volatiles such as acetaldehyde, dimethylsulphide, and hexanal. SCHULTHESS et al. (1991) have analysed the essential oil content in achenes of the above species. Essential oils present in the achenes of *Echinacea* spp. have an important role as phytochemical markers in differentiating the three species.

Our work has focused on a qualitative evaluation of the content and the quality of essential oils in selected species of *Echinacea Moench.* with respect to various climatic conditions.

Material and Methods

The plant material for experiments included *Echinacea purpurea* /L./, *Echinacea sanguinea* /Nutt./ and its cultivar referred to as *Echinacea species*. The plants were grown from seed (source Seva Seed spol. s.r.o. Valtice) and

cultivated at the standard agrochemical conditions in qualitatively different locations (I. Stará Ľubovňa, II. Nitra and III. Oponice). Detailed agro-ecological characteristics of individual localities are archived at research workplace.

The plant material was collected in the main development phases: a) prebud, b) stem growth, c) pre-blooming, d) blooming and e) biological maturity. Three replicate samples were analysed. The analysed samples included underground and aerial parts – roots, leaves and stems, and flower heads. The samples were dried at 32-34°C. The results of analysis were recalculated to the dry matter.

Determination of the oil content in the drug

20 g of the drug was subjected to hydrodistillation for 3.5 hours according to the European Pharmacopoea (2000). Isolated oil was diluted in n-hexane and dried over anhydrous sodium sulphate.

Statistical analysis

Data were shown as means \pm standard errors of the mean (SEM). Statistical significance of the differences between parameters was evaluated by means of Student's test. $P < 0.05$ was chosen as a criterion of statistical significance.

Results and Discussion

Essential oil is a major constituent present in nearly all organs of the *Echinacea Moench.* species. Its content and quality varies during the ontogenesis. The variation depends on the species, climatic conditions and location. Of course, there are also different essential oil contents in particular organs of the mother plant (BECKER 1982b).

The three sorts of *Echinacea* genus under study in various stages of their ontogenetic growth contained from 0.05 to 2.92% of the oil in the roots, and from 0.03 to 0.87% in the aerial parts, respectively. The content of essential oil in *E. sanguinea* and *E. species* roots was greater than in *E. purpurea* roots. The root system of *E. sanguinea* contained the highest oil amount at the stage of biological maturity, while that of *E. species* reached the maximum value at the prebud stage. The aerial parts of *E. sanguinea* and *E. species* contained only trace amounts of essential oil. The highest oil content in *E. purpurea* was observed in the pre-blooming and blooming stages (Fig. 1, 2). This variability may be caused by the origin of the plant, as well as by the locality of its cultivation, environmental conditions and developmental stage of the plant (HOLLA et al. 2005). BAUER (1999) mentions that the roots of *E. purpurea* have been reported to contain up to 0.2% of essential oil and the flowering aerial parts less than 0.1% of essential oil, which corresponds with our results. The results of GRAY et al. (2003) indicate that plant developmental stage and growing conditions can significantly influence the components that determine raw material quality.

Dependence of the oil content upon a cultivation locality has been most evident in the flower heads of *E. purpurea*. For *E. sanguinea* and *E. species*, it

has been found that the greatest sensitivity in this respect is exerted by their root systems. The quality and content of essential oil depended on climatic conditions and location. However, the total essential oil content in the aerial and underground parts of particular *Echinacea* species has not changed significantly in response to actual existing conditions (Fig. 3).

Our results correspond with those of HOLLA et al. (2005), who noted that the content of essential oil obtained from the flower heads of *Echinacea purpurea* was 1.85% of the weight of dry material. VAVERKOVA et al. (2007) have studied the variability of essential oil from *E. purpurea*, *E. pallida* and *E. atropurpurea* in the main ontogenetic stages. The results obtained from *E. purpurea* correspond with our results.

The problem of the influence of various climatic conditions along with the quality of individual plant organs during the plant growth has been examined by several authors (PERRY et al. 1997, MAZZA & COTRELL 1999, THAPPA et al. 2004). MAZZA & COTRELL (1999) observed that the content of germacrene-D in oil from the flower heads of *E. purpurea* from diverse sources is different. The results of THAPPA et al. (2004) indicate that the various components of the essential oil of *E. purpurea* are specific to climatic factors, which influence their concentration. The variations in their concentrations under subtropical conditions may be due to the expression of these characters under altered agroclimatic conditions, as *Echinacea* is primarily a plant grown commercially in countries having a temperate agroclimate.

References

- AWANG D. V. & KINDACK D. G. (1991): Herbal CheMedicine: *Echinacea*. Can. Pharm. J. 124: 512-516.
- BAUER R. (1999): Chemistry, analysis and immunological investigations of *Echinacea* phytopharmaceutics, in Wagner, H., Ed. Immunomodulatory Agents from Plants, Birkhauser Verlag, Basel, p. 212.
- BAUER R., REMIGER P. & WAGNER H. (1988): *Echinacea*: Verhleichende DC- und HPLC analyse der herba drogen *Echinacea purpurea*, *E. pallida*, und *E. angustifolia*. Deutsche Apoth. Z. 128: 174-180
- BAUER R. & WAGNER H. (1991): *Echinacea* species as potential immunostimulatory drugs. In: WAGNER H. & FARNSWORT N. R. (eds.). Econ. and med. plants res., London, Acad. Press 5: 253-321.
- BECKER H. (1982a): Structure of Echinadoside. Z. Naturforsch. 37: 351-353.
- BECKER H. (1982b): Verwendung und Inhaltstoffe von *Echinacea angustifolia* und *Echinacea purpurea*. Deutsch. Apoth. Ztg. 122: 2320-2323.
- BECKER H. & HSIEH W. (1985): Cichoric acid and its derivatives from *Echinacea* species. Z. Naturforsch. 40c: 585-587.
- BERGERON CH. & GAFNER S. (2007): Quantitative Analysis of the Polysaccharides and Glycoprotein Fractions in *Echinacea purpurea* and *Echinacea angustifolia* by HPLC – ELSD for Quality Control of Raw Material. Pharm. Biol. 45 (2): 98-105.
- BODINET C., WILLIGMANN I. & BEUSCHER N. (1993): Host-resistance increasing activity of root extracts from *Echinacea* species. Planta Med. 59 (Suppl.): A 672.
- BONE K. (1998): *Echinacea*: what makes it work? British J. Phytoter. 5 (1): 5.
- European Pharmacopea (Ph. Eur.) 4-nd.ed. Maissonneuve AS, France 2000, p. 375-376.

- GOEL V., CHANG C., SLAMA J. V., BARTON R. & BAUER R. (2002): Alkylamides of *Echinacea purpurea* stimulate alveolar macrophage function in normal rats. *Inter. Immunopharm.* 2 (2-3): 381-387.
- GRAY D.E., PALLARDY S.G., GARRETT H.E., ROTTINGHAUS G.E. (2003): Acute Drought Stress and Plant Age Effects on Alkamide and Phenolic Acid Content in Purple Coneflower Roots. *Planta Med.* 69: 50-55.
- HOLLA M., VAVERKOVA S., FARKAS P., TEKEL J. (2005): Content of essential oil obtained from flower heads of *Echinacea purpurea* L. and identification of selected components. *Herba Pol.* 51: 26-30.
- MAZZA G. & COTRELL T. (1999). Volatile components of roots, stems, leaves and flowers of *Echinacea* species. *J. Agric.Food Chem.* 47: 3081-3085.
- PAAS E. & GRANT P. (2002): *Echinacea*; National centre for Agri-food research in medicine, <http://www.sbrc.ca/ncarm/>.
- PENGELLY A. (1996): The constituents of medicinal plants. USA, New South Whales:sunflower herbals, p. 121-189.
- PERRY N.B., VAN KLING J.W., BURGESS E.J., PARMENTER G. (1997): Alkamide levels in *Echinacea purpurea*: a rapid analytical method revealing differences among roots, rhizomes, stems, leaves and flowers. *Planta Med.* 63: 58-62.
- RANDOLPH R.K., GELLENBECK K., STONEBROOK K., BROVELLI E., OUIAN Y., BANKAITIS-DAVIS D. & CHERONIS J. (2003): Regulation of human gene expression an influenced by a commercial blended *Echinacea* product: Preliminary Studies, *Exp. Biol. Med.* 228: 1051-1056.
- RININGER J. A., KICKNER S., CHIGURUPATI P., MCLEAN A. & FRANCK Z. (2000): Immunopharmacological activity of *Echinacea* preparations following simulated digestion on murine macrophages and human peripheral blood mononuclear cells. *J. Leucocyte Biol.* 68(4): 503-510.
- REINKE R.A., LEE D.J., MCDUGALL B.R., KING P.J., VICTORIA J., MAO Y., LEI X., REINECKE M.G., ROBINSON W.E. (2004): L-cichoric acid inhibits human immunodeficiency virus type 1 integration in vivo and is a noncompetitive but reversible inhibitor of HIV-integrase in vitro. *Virology*, Sep. 1 (2): 326-332.
- ROESLER J., EMMENDORFER A., STEINMÜLLER C., LUETTIG B., WAGNER H., LOHMANN-MATTHES M. L. (1991): Application of purified Polysaccharides from Cell Cultures of the Plant *Echinacea purpurea* to test Subjects medites Activation of the Phycocyte System. *Int. J.Immunopharm.* 13: 931-941.
- SCHARR D.D. (1998): *Echinacea*: the plant that boosts your immune system. UK, London, p.382.
- SCHULTHESS H, GIGER E. & BAUMANN T.W. (1991): *Echinacea*: Anatomy, Phytochemical Pattern, and Germination of the Achene. *Planta Med.* 57: 384-388.
- THAPPA R.K., BAKSHI S.K., DHAR P.L., AGARWAL S.G., KITCHLU S., KAUL M.K., SURI K.A. (2004): Significance of changed climatic factors on essential oil composition of *Echinacea purpurea* under subtropical conditions. *Fla. Fragr. J.* 19 (5): 452-454.
- TUBARO A., TRAGNI E., DEL NEGRO P., GALLI C.L. & DELLA LOGGIA R. (1987): Anti-inflammatory activity of polysaccharide fraction of *Echinacea angustifolia*. *J. Pharm. Pharmacol.* 39: 567-569.
- VAVERKOVA S., MIKULASOVA M., HABAN M., TEKEL J., HOLLA M., OTEPKA P. (2007): Variability of the essential oil from three sorts of *Echinacea* Moench genus during ontogenesis. *Ces. Slov. Farm.* (56): 212-124.
- WAGNER H. (1985): Immunostimulating polysaccharides /heteroglycans/ of higher plants. *ArzneimForsch.* 35: 1069-1075.

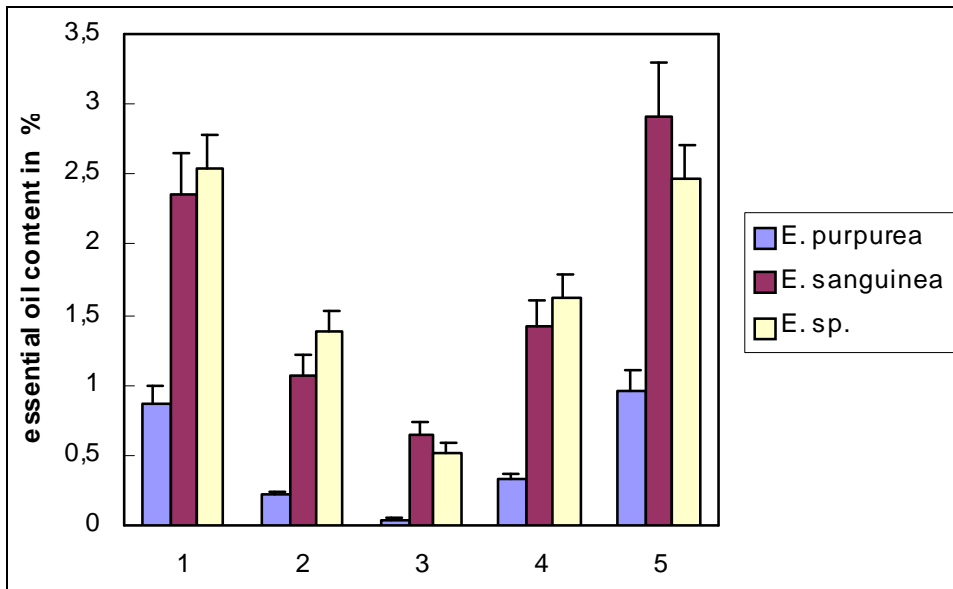


Fig. 1. Essential oil content (% v/w) in roots of three different *Echinacea* Moench. species during their ontogenesis. 1 - prebud stage, 2 - stem growth, 3 - pre-blooming, 4- blooming, 5 - biological maturity.

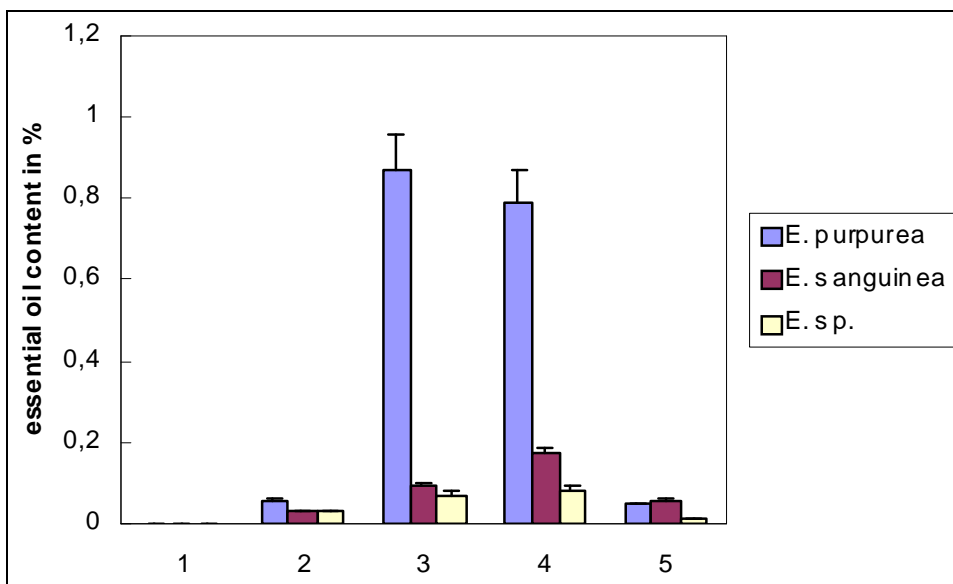


Fig. 2. Essential oil content (% v/w) in the aerial parts of three different *Echinacea* Moench. species during their ontogenesis. 1 - prebud stage, 2 - stem growth, 3 - pre-blooming, 4- blooming, 5 - biological maturity.

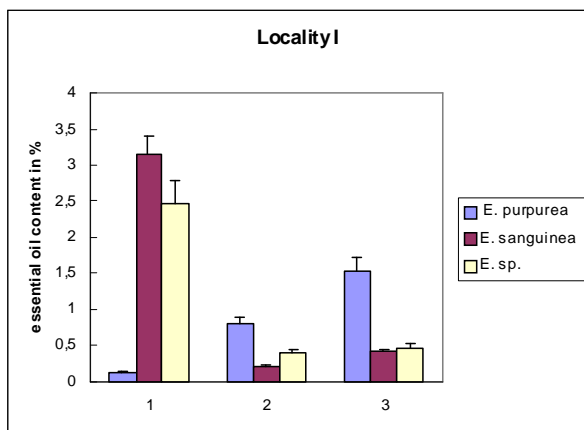
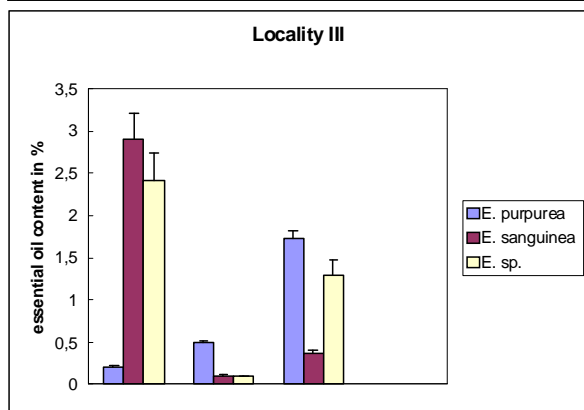
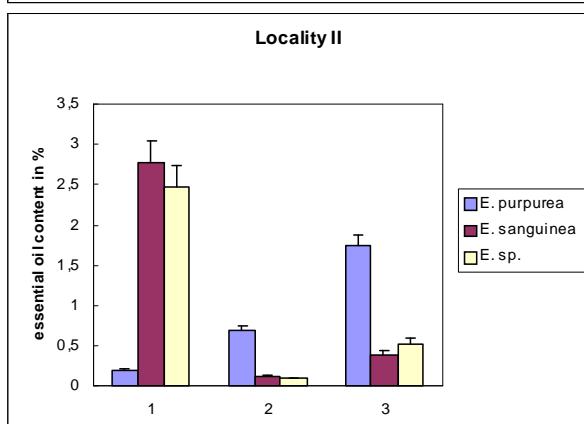


Fig. 3. Essential oil content (% v/w) depending on location in three different *Echinacea* Moench species. 1 - root, 2 - aerial part, 3 - flower head



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