ACTA UNIVERSITATIS AGRICULTURAE ET SILVICULTURAE MENDELIANAE BRUNENSIS

Volume LXI 86 Number 3, 2013
http://dx.doi.org/10.11118/actaun201361030777

# THE CONTENT OF VITAMIN E IN HOP CONES OF THE SAAZ VARIETY

Helena Pluháčková, Karolína Benešová, Jaroslava Ehrenbergerová, Blanka Kocourková

**Received: February 22, 2013**

## Abstract

PLUHÁČKOVÁ HELENA, BENEŠOVÁ KAROLÍNA, EHRENBERGEROVÁ JAROSLAVA, KOCOURKOVÁ BLANKA: *The content of vitamine E in hop cones of the Saaz variety.* Acta Universitatis Agriculturae et Silviculturae Mendelianae Brunensis, 2013, LXI, No. 3, pp. 777–785

The activity of vitamin E, total content of tocols and the content of individual isomers: α-tocopherols, β-tocopherols, γ-tocopherols and δ-tocopherols was monitored in samples of hop cones of the world-important Saaz variety. Hop cone samples originated from hop-breeding area Tršice, Czech Republic. The method used for the determination of vitamin E in barley was modified and used for this quantitative analysis. The results indicate that monitored characteristics are influenced by the year of harvest (2010 or 2011) but also by the age of hop-gardens (hop bucks). High values of vitamin E activity (up to 67.79 $mg.kg^{-1}$) and total content of tocols (up to 76.31 $mg.kg^{-1}$) in hop cones are worth further attention from the viewpoint of alternative use of hops.

hop, vitamin E, antioxidant, tocopherol, Saaz

Antioxidants are important mediators of energy transfer, immune defense factors and signal molecules of cell regulation (Chrpová, Kouřimská, Pánek, 2008). Food quality is given not only by nutrients and nutritionally important substances, but also by components that enhance the immunity of the organism and its defensive ability against diseases, reduce the risk of disease, act against pathogenic microorganisms or otherwise protect human health. These components slow unwanted processes in human body and contribute to the protection against negative effects of lifestyle changes and aging (Kopec, 1998; Štípek, 2000).

Antioxidants are divided according to their function (Benešová, 2000; Velíšek, 2002) to primary antioxidants that end the chain reaction of free radicals by supplying hydrogen or electrons. They work also with lipid radicals, creating complexes lipid-antioxidant. Tocopherols belong to this group together with many other phenolic compounds found in natural samples etc. According to Benešová (2000), natural antioxidants are compounds naturally occurring in plants, animals and microorganisms. Antioxidation activity of these compounds varies in a wide range from very mild to very high.

Vitamins are non-energetical low-molecular organic compounds that the organisms need unconditionally in some minimum concentration, but are unable to synthesize them itself or can do it only in insufficient quantities and thus the compounds must be contained in food. The complex of vitamins contained in fruits and vegetables protects human organism in many ways. Some vitamins have other protective effects, for example against tumor diseases.

Eight basic structurally-related derivatives of chroman (3,4-dihydro-2h-1-benzopyran) called tocols show the activity of vitamin E. The structural base is common for all the compounds exhibiting the vitamin E activity (so-called vitagenum E), contains chroman cycle with saturated or unsaturated isoprenoid side chain with 16 carbon atoms. This cycle is derived from diterpenoid alcohol phytol, but it contains also additional methyl group in the position C-2 and hydroxy group in the position C-6 of the chroman cycle. The presence of these functional groups is necessary for biological activity of all eight vitamers. Four forms of the vitamin E with saturated terpenoid side chain derived from tocol are tocopherols, four forms with unsaturated side chain are called tocotrienols. Individual tocopherols

and tocotrienols differ in the position and number of methyl groups in the chroman cycle. The vitamin E, especially α-tocopherol, is the most important lipophilic antioxidant that is the part of eukaryotic cells. Appropriate intake of vitamin E is considered to help preventing the oxidation of biomembrane lipids. Vitamin E is necessary for the cell division, for the right function of nerves, muscles, brain, kidneys and liver. It enhances also the life duration of red blood cells (Velíšek, 1999; Kopec, 1998). Vitamin E, group of substances synthesized by all higher plants, belong to the most important natural antioxidants. In the human organism they help to slow down the ageing process and play a role in prevention of cancer and heart and vascular diseases (Weber and Rimbach, 2001; Zingg, 2007).

The content of antioxidants, including vitamin E, is most often studied in vegetables. Ensminger (1995) states that the most studied are following species: cabbage, onion, tomatoes, carrot, melon, bell pepper, cucumber and cauliflower. Antioxidants are the focus of interest also in other plant species, especially in cereals (Prýma, 2007). The content of antioxidants hasn't been studied in detail yet in hops, where there would be a good idea to look for alternative ways of use due to the world overproduction of hops.

Olas *et al.* (2011) dealt with the use of extract from hop cones (*Humulus lupulus*) as an oxidation stress modulator in blood cells and according to them there is little known about the antioxidation activity of hop cones. In the comparative study they found out that hop extracts aren't more efficient antioxidant than pure commercial preparation. Cleemput *et al.* (2009) found out that bitter acids from hops are effective against inflammation and metabolic disorder. They pointed out there is a possibility of using hop extracts for the treatment of diabetes, cardiovascular diseases and metabolic syndromes.

In the Czech Republic the most widespread cultivated variety is the Saaz hops. As for its quality, it belongs to the top quality hops in the world. It is an original population that was developed and naturally selected at the conditions of current hop-breeding regions. From the beginning of nineties the clones are propagated from virus-free planting material. International degustation held since 1998 by the Hops Institute Ltd., Žatec, clearly demonstrate the high quality of this variety. (Nesvadba, 2012). Hops is a crop that is being grown at the site for more than 20 years; hop varieties became more yielding thanks to the plant breeders, which is one of the factors contributing to the worldwide overproduction of hops in recent years. Hops are irreplaceable raw material for brewing, but it is necessary to use the overproduction of hops also in other areas, for example in pharmaceutics, cosmetics, but also in animal nutrition and breeding (treatment of inflammation diseases).

The aim of our work was the determination of the content of vitamin E antioxidant. Previous scientific research indicate that this antioxidant hasn't been determined in hops before.

## MATERIALS AND METHODS

The samples of Saaz variety hops were used for the determination of individual isomers of vitamin E and its activity. Hop cone samples from hop-breeding region Tršice were obtained from the harvests 2010 and 2011 and were taken from 9 hop-gardens of different age (hop bucks) (Tab. I).

I: *Places where hop cones samples were taken*

| Planting year | Cadastral area | Track name |
|---|---|---|
| 1993 | Dolní Újezd | U lípy |
| 1995 | Prosenice | Rol |
| 1996(1) | Lazníky | Nad česačkou |
| 1996(2) | Prosenice | Kouta |
| 1996(3) | Dolní Újezd | Pod cestou |
| 1997(4) | Dolní Újezd | Pod cestou |
| 1997(5) | Lazníky | Zabitá I. |
| 1998 | Lazníky | Zabitá II. |
| 2000 | Dolní Újezd | Pod cestou |

Original method according to EN 12 822:2000 and according to Mc Laughlin and Weihrauch (1979) adapted for hop cone samples was used for the determination of vitamin E. Vitamin E activity is given in milligrams of α-tocopherol equivalent which represents the sum of the individual tocopherols and tocotrienols with respect to their biological activity (calculation according to McLaughlin and Weihrauch 1979).

### Saponification:

The sample of hop cones was ground in laboratory mill. 2 g of the sample were weighted, 100 mg of ascorbic acid and 50 ml of ethanol were added. Then the sample was shaken in the shaker machine for 1 minute, kept at rest for 10 minutes in the dark and after that 10 ml of 50% KOH were added. The prepared sample was left to stay overnight in the dark in nitrogen atmosphere at the laboratory temperature to let the saponification occur.

### Extraction:

Saponified sample was transferred quantitatively to the separating funnel, the flask was rinsed with 100 ml of distilled water. The extraction was performed with 3 × 50 ml of diethylether. Combined etheric phases were washed with water to the neutral reaction; water was added to the etheric phase (3 × 100 ml) and shaken gently not to form emulsion. Washed etheric phase was dried by anhydrous sodium sulfate. The solvent was evaporated to dryness at the vacuum evaporator and the residue was immediately dissolved in 4 ml of methanol and transferred quantitatively to the vial.

### Analytical determination:

Chromatographic analyses were preformed employing high-performance liquid chromatograph (HPLC) Ultimate 3000 (Dionex, USA). The system is equipped with quaternary gradient pump with integrated vacuum degasser, autosampler with injection block Rheodyne, programmable termostated column and two programmable detectors, diode-array detector on-line coupled to four-channel fluorescence detector. The software Chromeleon was used for data acquisition and evaluation. The column Ascentis® Express RP-Amide, size 150 × 3 mm, particle size 2.7 µm was used for the analyses, the mobile phase was 100% acetonitrile at the flow rate 0.75 ml.min$^{-1}$. Column temperature was 30 °C, injection volume 1 µl and both tocopherols and tocotrienols were determined by a fluorescence detector with $\lambda_{ex}$ 290 and $\lambda_{em}$ 330 nm, resp. Statistical program STATISTICA (data analysis software system), version 10 (StatSoft Inc., 2011) was used for data evaluation.

Results were evaluated by multi-factor analysis of variance, followed by testing the differences in average values by the means of LSD test.

## RESULTS AND DISCUSSION

Only the content of tocopherols was determined in the monitored set of samples, tocotrienols were not detected. This is in good accordance with the analysis of barley green mass; there were also only tocopherols determined (Ehrenbergerová *et al.*, 2009).

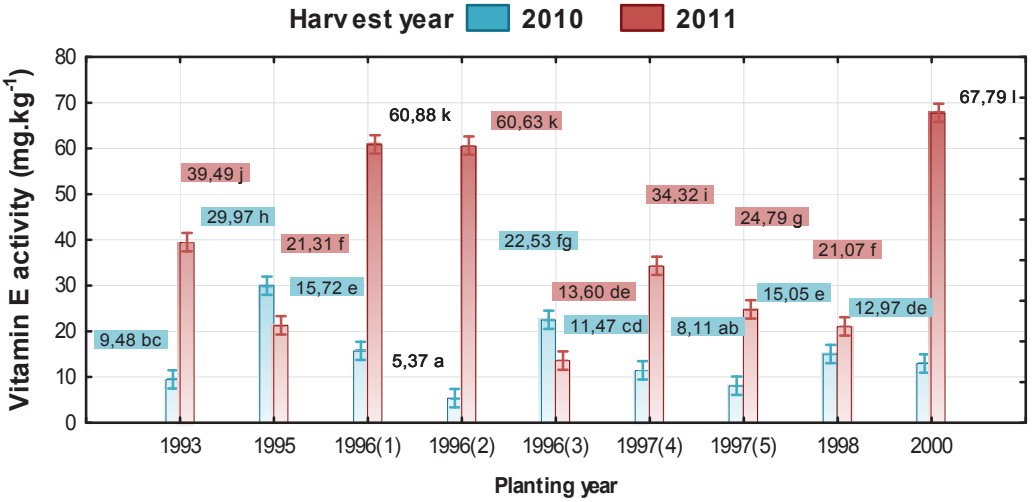
Based on the evaluation of obtained experimental results by the means of the multi-factor analysis of variance (see Tab. II), statistically highly significant effects were found for the planting year (age of hop bucks) and the harvest year (2010, 2011). Also the interaction of these factors (planting year x harvest year) influenced monitored parameters in a statistically highly significant way. Subsequent testing using Fisher's LSD test found many significant differences in the average activity values and the content of individual vitamin E isomers (see Fig. 1–Fig. 6) caused by the above given factors.

As for the average activity of vitamin E (see Fig. 1, see Tab. II) it was found out for the Saaz variety and individual years of the hop-gardens planting during the two-year monitoring that statistically significantly higher activity was determined for the

II: *Analysis of variance for the activity of vitamin E and content of its isomers in hop cones of the Saaz variety for the years 2010 and 2011*

| SOURCE OF VARIANCE | d.f. | Vitamin E activity | Total tocols | α-T | β-T | γ-T | δ-T |
|---|---|---|---|---|---|---|---|
| | | MS | | | | | |
| **Planting year** | 8 | 635,17*** | 772,11*** | 615,23*** | 0,08*** | 0,24*** | 7,93*** |
| **Harvest year** | 1 | 10100,99*** | 11966,80*** | 9801,34*** | 0,34*** | 2,95*** | 65,39*** |
| Interactions: **Planting year* Harvest year** | 8 | 1234,65*** | 1613,26*** | 1175,56*** | 0,19*** | 1,21*** | 20,82*** |
| **Error** | 54 | 3,98 | 4,94 | 3,79 | 0,00 | 0,03 | 0,10 |

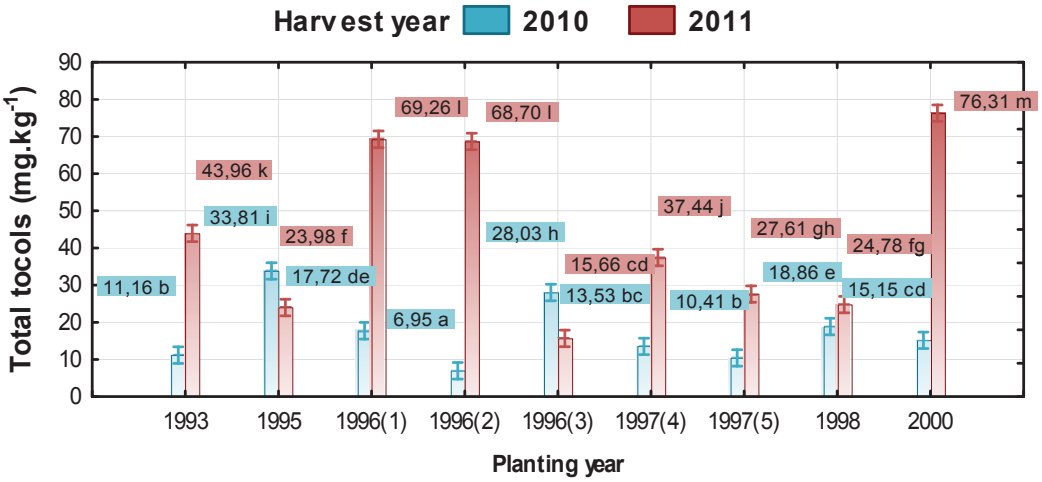
Note: * - p ≤ 0.05; ** - p ≤ 0.01; *** - p ≤ 0.001; T = tocopherol




1: *Average values of vitamin E activity in hop cones of the Saaz variety in harvest years 2010 and 2011*
Note: Average values marked with different letters showed statistically significant differences from each other at P = 0.05; (1) Lazníky „Nad česačkou", (2) Prosenice „Kouta", (3) Dolní Újezd „Pod cestou", (4) Dolní Újezd „Pod cestou", (5) Lazníky „Zabitá I."

harvest year 2011 (38.21 mg.kg$^{-1}$) in comparison with the year 2010 (14.52 mg.kg$^{-1}$). This finding could lead to the conclusion that the harvest year 2011 was a more favourable for the activity of vitamin E, but in some samples higher values were found in the harvest year 2010 (planting years 1995 – 29.97 mg.kg$^{-1}$ and 1996 – 22.53 mg.kg$^{-1}$). The highest activity was found in the samples from the youngest hop-garden (the planting year 2000 – 67.79 mg.kg$^{-1}$) in the harvest year 2011, on the other hand the lowest activity of vitamin E was found in samples from the planting year 1996 (5.37 mg.kg$^{-1}$), which is not significantly statistically different from the samples from the planting year 1997 (8.11 mg.kg$^{-1}$).

Similar to the vitamin E activity, the highest average content of total tocols (see Fig. 2, see Tab. III) in the two-year period was determined in the harvest year 2011 in samples from the youngest hop-garden (harvest year 2000 – 76.31 mg.kg$^{-1}$). High content of total tocols was found also in samples from the same harvest year 2011 and planting year 1996 (69.26 a 68.70 mg.kg$^{-1}$) which were not significantly




2: *Average values of the total tocols content in hop cones of the Saaz variety in harvest years 2010 and 2011*

III: *Average values of the vitamin E activity, total tocols content and vitamin E isomers (mg.kg$^{-1}$) in hop cones of the Saaz variety*

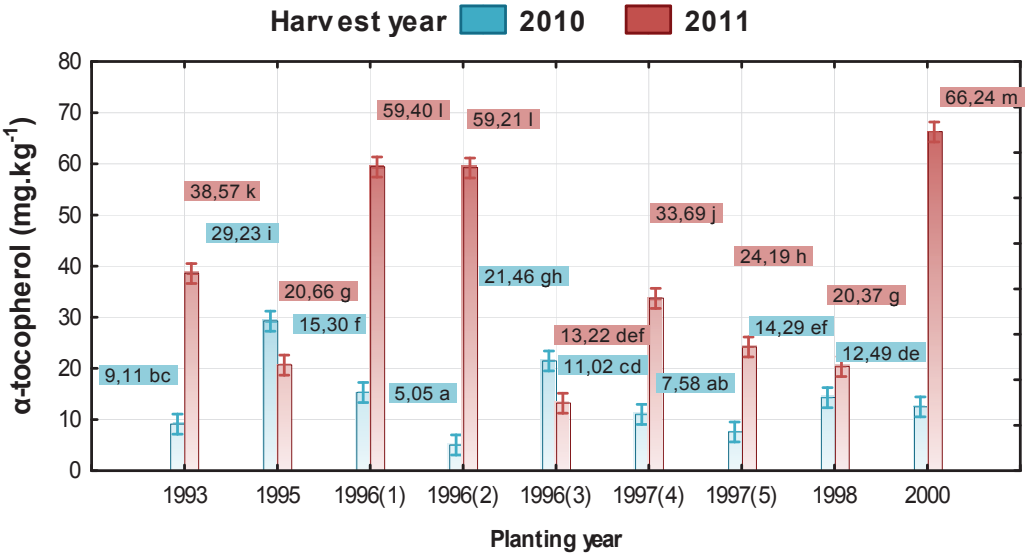
| Harvest year | Planting year | Cadastral area | Track name | Area (ha) | Vitamin E activity | Total tocols | α-T | β-T | γ-T | δ-T |
|---|---|---|---|---|---|---|---|---|---|---|
| 2010 | 1993 | Dolní Újezd | U lípy | 2,6 | 9,48 $^{bc}$ | 11,16 $^{b}$ | 9,11 $^{bc}$ | 0,58 $^{ab}$ | 1,34 $^{a}$ | 0,14 $^{a}$ |
| | 1995 | Prosenice | Rol | 12 | 29,97 $^{h}$ | 33,81 $^{i}$ | 29,23 $^{i}$ | 1,03 $^{ef}$ | 3,26 $^{e}$ | 0,30 $^{de}$ |
| | 1996(1) | Lazníky | Nad česačkou | 5,2 | 15,72 $^{e}$ | 17,72 $^{de}$ | 15,30 $^{f}$ | 0,64 $^{abc}$ | 1,57 $^{ab}$ | 0,21 $^{abc}$ |
| | 1996(2) | Prosenice | Kouta | 3,6 | 5,37 $^{a}$ | 6,95 $^{a}$ | 5,05 $^{a}$ | 0,50 $^{a}$ | 1,26 $^{a}$ | 0,15 $^{a}$ |
| | 1996(3) | Dolní Újezd | Pod cestou | 9,5 | 22,53 $^{fg}$ | 28,03 $^{h}$ | 21,46 $^{gh}$ | 1,56 $^{hi}$ | 4,38 $^{g}$ | 0,63 $^{h}$ |
| | 1997(4) | Dolní Újezd | Pod cestou | 7,4 | 11,47 $^{cd}$ | 13,53 $^{bc}$ | 11,02 $^{cd}$ | 0,72 $^{abcd}$ | 1,60 $^{ab}$ | 0,19 $^{ab}$ |
| | 1997(5) | Lazníky | Zabitá I. | 7 | 8,11 $^{ab}$ | 10,41 $^{b}$ | 7,58 $^{ab}$ | 0,87 $^{cde}$ | 1,80 $^{bc}$ | 0,17 $^{ab}$ |
| | 1998 | Lazníky | Zabitá II. | 7 | 15,05 $^{e}$ | 18,86 $^{e}$ | 14,29 $^{ef}$ | 1,13 $^{fg}$ | 3,04 $^{e}$ | 0,40 $^{fg}$ |
| | 2000 | Dolní Újezd | Pod cestou | 6 | 12,97 $^{de}$ | 15,15 $^{cd}$ | 12,49 $^{de}$ | 0,78 $^{bcde}$ | 1,69 $^{ab}$ | 0,19 $^{abc}$ |
| **Mean** | | | | | **14,52** | **17,29** | **13,95** | **0,87** | **2,22** | **0,26** |
| 2011 | 1993 | Dolní Újezd | U lípy | 2,6 | 39,49 $^{j}$ | 43,96 $^{k}$ | 38,57 $^{k}$ | 1,37 $^{gh}$ | 3,70 $^{f}$ | 0,33 $^{def}$ |
| | 1995 | Prosenice | Rol | 12 | 21,31 $^{f}$ | 23,98 $^{f}$ | 20,66 $^{g}$ | 1,15 $^{fg}$ | 1,86 $^{bc}$ | 0,31 $^{de}$ |
| | 1996(1) | Lazníky | Nad česačkou | 5,2 | 60,88 $^{k}$ | 69,26 $^{l}$ | 59,40 $^{l}$ | 1,86 $^{jk}$ | 7,26 $^{h}$ | 0,74 $^{i}$ |
| | 1996(2) | Prosenice | Kouta | 3,6 | 60,63 $^{k}$ | 68,70 $^{l}$ | 59,21 $^{l}$ | 1,72 $^{ij}$ | 7,29 $^{h}$ | 0,48 $^{g}$ |
| | 1996(3) | Dolní Újezd | Pod cestou | 9,5 | 13,60 $^{de}$ | 15,66 $^{cd}$ | 13,22 $^{def}$ | 0,50 $^{a}$ | 1,78 $^{b}$ | 0,17 $^{ab}$ |
| | 1997(4) | Dolní Újezd | Pod cestou | 7,4 | 34,32 $^{i}$ | 37,44 $^{j}$ | 33,69 $^{j}$ | 0,94 $^{def}$ | 2,54 $^{d}$ | 0,28 $^{cde}$ |
| | 1997(5) | Lazníky | Zabitá I. | 7 | 24,79 $^{g}$ | 27,61 $^{gh}$ | 24,19 $^{h}$ | 0,93 $^{def}$ | 2,23 $^{cd}$ | 0,25 $^{bcd}$ |
| | 1998 | Lazníky | Zabitá II. | 7 | 21,07 $^{f}$ | 24,78 $^{fg}$ | 20,37 $^{g}$ | 0,97 $^{def}$ | 3,09 $^{e}$ | 0,35 $^{ef}$ |
| | 2000 | Dolní Újezd | Pod cestou | 6 | 67,79 $^{l}$ | 76,31 $^{m}$ | 66,24 $^{m}$ | 2,01 $^{k}$ | 7,34 $^{h}$ | 0,72 $^{hi}$ |
| **Mean** | | | | | **38,21** | **43,08** | **37,28** | **1,27** | **4,12** | **0,40** |

Note: Average values marked with different letters showed statistically significant differences from each other at P = 0.05

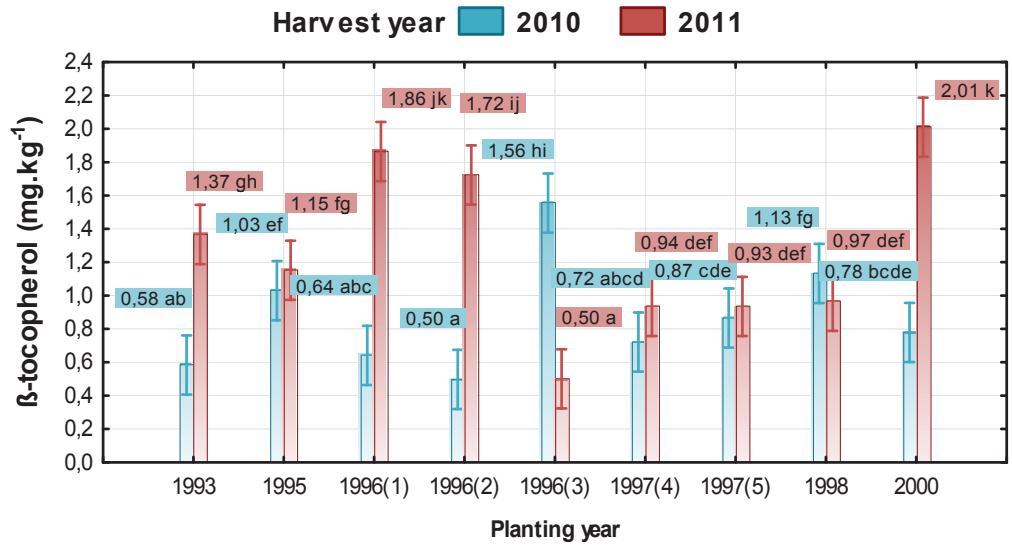
statistically different from each other. Interesting results were obtained for three hop-gardens from the planting year 1996 of different areas from different cadastral districts (hereinafter referred to as cadastral). The results can point out the influence of the hop-garden habitat. In the samples from the hop-garden from planting year 1996 (cadastral Dolní Újezd „Pod cestou"), higher values (28.03 mg.kg$^{-1}$) were found in the harvest year 2010 in comparison with the harvest year 2011 (15.66 mg.kg$^{-1}$).

α-tocopherol (see Fig. 3, see Tab. III) is generally one of the isomers most involved in the total content of tocols and the vitamin E activity. Its high percentage was confirmed also in the hop cone samples. Thus, similar to the vitamin E activity and the total content of tocols, also the highest values of α-tocopherol were found in samples from the youngest hop-garden (planting year 2000 – 66.24 mg.kg$^{-1}$) in the harvest year 2011. In the harvest year 2010, several times lower values (12.49 mg.kg$^{-1}$) were found in samples from the same hop-garden planted in the year 2000. However, the results were not significantly statistically different either from the samples from the planting year 1998 and harvest year 2010 (14.29 mg.kg$^{-1}$) or from the samples from the planting year 1996 and harvest year 2011 (13.2 mg.kg$^{-1}$).

Statistically significantly higher average content of β-tocopherol isomer (see Fig. 4, see Tab. III) during the two-year period was determined in hop cone samples from the planting year 2000 and harvest year 2011 (2.01 mg.kg$^{-1}$) compared to other samples in the set. Statistically significantly lower content was found in samples from the planting year 1996




3: *Average values of the α-tocopherol content in hop cones of the Saaz variety in harvest years 2010 and 2011*




4: *Average values of the β-tocopherol content in hop cones of the Saaz variety in harvest years 2010 and 2011*

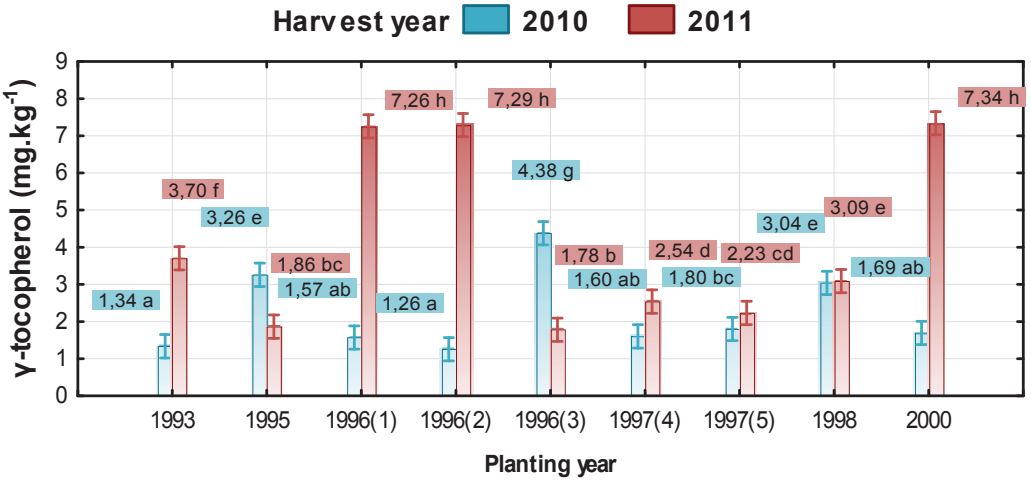
and harvest year 2011 (0.50 mg.kg$^{-1}$, cadastral Dolní Újezd „Pod cestou“), but it wasn't significantly statistically different from samples from the planting year 1996 (cadastral Prosenice „Kouta“ – 0.50 mg.kg$^{-1}$), the planting year 1993 (cadastral Dolní Újezd „U lípy“ – 0.58 mg.kg$^{-1}$), the planting year 1996 (cadastral Lazníky „Nad česačkou“ – 0.64 mg.kg$^{-1}$) and the planting year 1997 (cadastral Dolní Újezd „Pod cestou“ – 0.72 mg.kg$^{-1}$), all in the harvest year 2010.

The γ-tocoferol isomer (see Fig. 5, see Tab. III) was found in monitored years (2010, 2011) to be in the range of 1.26–7.34 mg.kg$^{-1}$. This isomer is the second highest represented one in the total content of tocols after α-tocopherol.

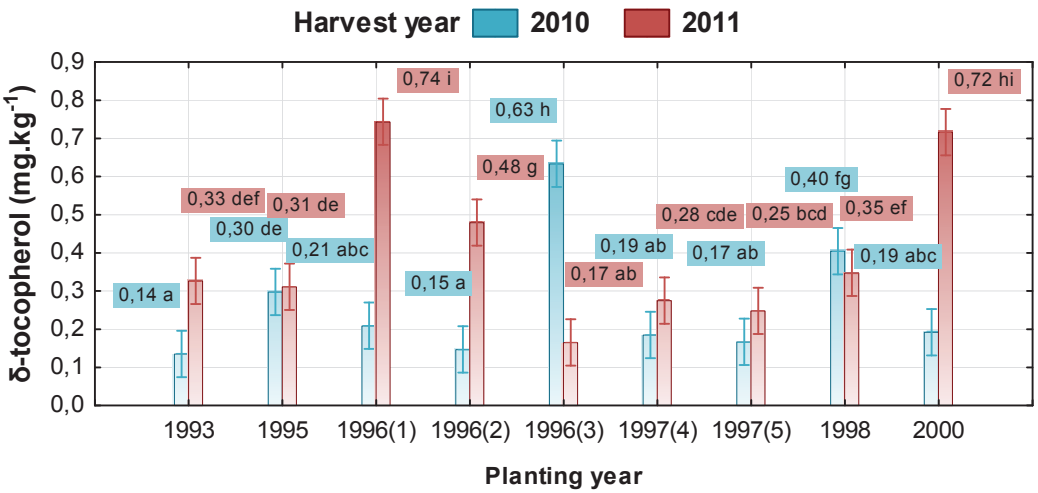
The highest values of δ-tocopherol (0.74 mg.kg$^{-1}$) (see Fig. 6, see Tab. III) were found in the samples from the planting year 1996 and harvest year 2011 (cadastral Lazníky „Nad česačkou“). However, the results weren't significantly statistically different from samples from the youngest hop-garden from the planting year 2000 (0.72 mg.kg$^{-1}$).

The lowest content of δ-tocopherol was found in the samples from the planting year 1993 (0.14 mg.kg$^{-1}$), but this was significantly statistically different neither from samples from the planting year 1996 (0,15 mg.kg$^{-1}$), from the planting year 1997 and cadastral Lazníky „Zabitá I.“ (0.17 mg.kg$^{-1}$), from the planting year 1997 and cadastral Dolní Újezd „Pod cestou“ (0.19 mg.kg$^{-1}$) and from the planting year 2000 (0.19 mg.kg$^{-1}$), all in the harvest year 2010, nor from the planting year 1996 and cadastral Dolní Újezd „Pod cestou“ 1996 in the harvest year 2011 (0.17 mg.kg$^{-1}$).

Lachman (2000) states that the vitamin E, mostly α-tocopherol, is the most important lipophilic antioxidant contained in eukaryotic cells as the protection of unsaturated lipids against damage caused by free radicals. In our research the content of α-tocopherol in hop cones of investigated Saaz variety varied in the range between 5.05 mg.kg$^{-1}$–




5: *Average values of the γ-tocopherol content in hop cones of the Saaz variety in harvest years 2010 and 2011*




6: *Average values of the δ-tocopherol content in hop cones of the Saaz variety in harvest years 2010 and 2011*

29.23 mg.kg$^{-1}$ and 13.22–66.24 mg.kg$^{-1}$ in the harvest years 2010 and 2011, resp., given in average values from all sites. These quantities are much higher than values given for the selected vegetables, because according to Kopec (1998) the amount of vitamin E varies from 1.3 mg.kg$^{-1}$ in cauliflower and 2 mg.kg$^{-1}$ in cabbage to 7.2 mg.kg$^{-1}$ in tomatoes and 10.5 mg.kg$^{-1}$ in green bell pepper and 11.9 mg.kg$^{-1}$ in red bell pepper. The content of tocols found in genotypes of spring barley by Ehrenbergerová *et al.* (2006) was highest for *waxy* varieties (60.32–67.55 mg.kg$^{-1}$) and reached the level of 49.93–53.56 mg.kg$^{-1}$ in standard malting type varieties; it means that some samples of hop cones in our research are also of higher content than these values.

The authors Goupy *et al.* (1999) and Cavallero *et al.* (2004) state that the range of total tocol content in barley grain is from 9.7 to 61.4 mg.kg$^{-1}$; it is in good accordance with the range of our results in hop cones that was from 6.95 to 76.31 mg.kg$^{-1}$. Peterson and Qureshi (1993) also studied the concentration of vitamin E in barley grain and they state that the content of vitamin E is influenced by the environmental conditions during the growth, ripening, harvest and storage, which is confirmed also by the authors of more recent works (Březinová Belcredi *et al.*, 2008). The results of this research were confronted primarily with those of authors who dealt with vitamin E in barley, because hops are reported to have similarly high levels of vitamin E as above-mentioned barley.

Average activity of vitamin E in hop cones was 14.52 mg.kg$^{-1}$ in the year 2010 and 38.21 mg.kg$^{-1}$ in the year 2011, average values of total tocols and α-tocopherol in the samples were adequately higher. It means that the weather course conditions during individual hop-planting years influences significantly the activity of vitamin E and the content of its isomers, as was also statistically verified by analysis of variance. However, due to significant interaction of the years of cultivation with the years of hop-garden planting both lower amount of total tocols and lower activity of vitamin E was determined in the hop cone samples from the hop-gardens Prosenice "Rol" a Dolní Újezd "Pod cestou" (1) in the harvest year 2011 compared to the year 2010 when vast majority of samples showed higher values.

The evaluation of the results achieved in the hop cones research indicates that the content of tocopherols and vitamin E activity was influenced by the planting year (age of the hop-garden). Also the soil and climatic conditions of the habitat and weather course conditions in the harvest years of hops influenced levels of vitamin E, which is confirmed e.g. by different values obtained for samples from different cadastral areas in hop-growing region Tršice.

Prýma *et al.* (2007) confirmed the dependence of the content of vitamin E on the variety of barley; thus it is likely that the situation will be similar for hops and our current research work will brings interesting results of the study of more hop varieties.

The activity of vitamin E in hop cones was monitored for the purpose of alternative use. Due to the overproduction of hops not only in the Czech Republic but also in the whole world it is important to look for other use of hops, for example in food additives that aren't exposed to high temperatures during the processing.

## CONCLUSIONS

The obtained results show that the content of tocopherols and vitamin E activity was influenced by the year of planting (age of hop bucks), but also by soil and climatic conditions of the site and the harvest year.

From the temperature viewpoint the harvest year 2010 can be characterised as slightly above the average with the average temperature 8.17 °C. The rainfall during the year 2010 was 768.40 mm, that is 215 mm above the long-term average. In the harvest year 2011 the average temperature was even higher (9.02 °C) in comparison to the year 2010, but as for the rainfall the year 2011 was closer to the long term average with 551.2 mm rainfall. From the different weather conditions of these two years we can assume that the weather conditions of the year 2011 influenced favourably higher vitamin E activity.

It is evident from the two-years results that the hop cones contain significant amount of vitamin E and isomers of tocopherols. Tocotrienols were not detected in the samples due to their low level.

Further research of the use of vitamin E from hop cones is needed. In the harvest year 2011 the highest activity of vitamin E and its isomers was found in samples from the youngest hop-garden, planted in the year 2000. Unfortunately, it was not the same in the year 2010; thus, the result show that the impact of harvest year is very important. According to overall results we can say that the harvest year 2011 was more favourable for the synthesis of vitamin E compared to the year 2010.

## SUMMARY

This work deals with the methodology of determination of vitamin E activity in hop cones of the most important Czech hop variety Saaz. The method proved and validated to be used on barley grain (Ehrenbergerová *et al.*, 2006) was used for the determination; it has been partially adapted for determination from hop cone samples. The samples were saponified, extracted and chromatographic analyses were performed. The results were evaluated by multi-factor analysis of variance, followed by testing the differences among the average values via LSD test.

Four tocopherol isomers (α, β, γ and δ) were detected in the samples of Saaz variety hop cones from hop-growing region Tršice in the harvest years 2010 and 2011. Hop samples originated from the hop-gardens of different planting age (age of the hop bucks).
The evaluation of the results achieved in the hop cones monitoring indicates that the content of tocopherols and vitamin E activity was influenced by the planting year (age of the hop-garden) and also by the soil and climatic conditions of the site and weather course conditions during the harvest year.
The vitamin E activity found in the samples of the Saaz variety varied in the range of 5.37–67.79 $mg.kg^{-1}$, total tocols content varied in the range of 6.95–76.31 $mg.kg^{-1}$. The α-tocopherol, β-tocopherol, γ-tocopherol and δ-tocopherol isomers were present in hop cone samples in the ranges of 5.05–66.24 $mg.kg^{-1}$, 0.5–2.01 $mg.kg^{-1}$, 1.26–7.34 $mg.kg^{-1}$ and 0.14–0.74 $mg.kg^{-1}$, respectively. It is evident from the two-years results that the hop cones contain significant amount of vitamin E and its isomers.

Acknowledgement

The study was supported by the Ministry of Educations, Youth and Sports of the Czech Republic, Project CR No. 1M0570 and the project No. QJ1210301, National Agency of Agricultural Research, Ministry of Agriculture of the Czech Republic.

## REFERENCES

BAMFORTH CH. W., 2004: *Beer, Health and Nutrition.* Oxford OX4 2DQ, UK: Blackwell Science Ltd a Blackwell Publishing company, 179 p., ISBN 0-632-06446-3.

BENEŠOVÁ, L. a kol., 2000: *Potravinářství VI.* Ústav zemědělských a potravinářských informací, Praha: ÚZPI Praha, 39–58. ISBN 80-7271-003-6.

BŘEZINOVÁ BELCREDI, N., MARKOVÁ, J., EHRENBERGEROVÁ, J., PRÝMA J., VACULOVÁ, K., LANCOVÁ, K., HAJŠLOVÁ, J., 2008: Antioxidant and Mycotoxins in Barley Grain. In: UGARČIČ-HARDI, Ž. (ed.) *Proceedings of 4th International Congress FLOUR – BREAD'07 and 6th Croatian Congress of Cereal Technologists*. Osijek: Faculty of Food Technology Osijek University of Osijek, Croatia, p. 248–253. ISBN 978-953-7005-15-3.

CAVALLERO, A., GIANINETTI, A., FRANCA, F., DELOGU, G., STANCA, A. M., 2004: Tocols in hull-less and hulled barley genotypes grown in contrasting environments. *J Cereal Sci*, 39, 2: 175–180. ISSN 0733-5210.

CLEEMPUT, M., VAN CATTOOR, K., BOSSCHER, K., DE HAEGEMAN, G., KEUKELEIRE, D., DE HEYERICK, A., 2009: Hop (Humulus lupulus)-derived bitter acids as multipotent bioactive compounds. *J Nat. Prod.*, 72, 6: 1220–1230. ISSN 0163-3864.

ENSMINGER, A. H., 1995: *The Concise Encyclopedia of Foods and Nutrition*, Boca Raton: CRC Press, 458–469. ISBN 0-8493-4455-7.

EHRENBERGEROVÁ, J., BELCREDIOVÁ, N., PRÝMA, J., VACULOVÁ, K., NEWMAN, C. W., 2006: Effect of Cultivar, Year Grown, and Cropping System on the Content of Tocopherols and Tocotrienols in Grains of Hulled and Hulless Barley. *Plant Food Hum. Nutr.*, 61, 3: 145–150. ISSN 0921-9668.

EHRENBERGEROVÁ, J., BŘEZINOVÁ BELCREDI, N., KOPÁČEK, J., MELIŠOVÁ, L., HRSTKOVÁ, P., MACUCHOVÁ, S., VACULOVÁ, K., PAULÍČKOVÁ, I., 2009: Antioxidant enzymes in barley green biomass. *Plant Food Hum. Nutr.,* 64, 2: 122–128. ISSN 0921-9668.

GOUPY, P., HUGUES, M., BOIVIN, J. P., and AMIOT, M. J., 1999: Antioxidant composition and activity of barley (Hordeum vulgare) and malt extracts and of isolated phenolic compounds. *J Sci Food Agri*, 79, 12: 1625–1634. ISBN 0022-5142.

CHRPOVÁ, D., KOUŘIMSKÁ, L., PÁNEK, J., 2008: Antioxidační působení vybraných druhu koření používaných v šetřících dietách: Antioxidanty. In: NEUGEBAUEROVÁ, J., VÁBKOVÁ, J., *Aktuální otázky pěstování léčivých, aromatických a kořeninových rostlin*. 1. vyd. Brno: MZLU v Brně, 151 s. ISBN 978-80-7375-245-3.

KOPEC, K., 1998: *Tabulky nutričních hodnot ovoce a zeleniny*. 1. vyd. Praha: ÚZPI Praha, 72 s. ISBN 80-86153-64-9.

LACHMAN, J., ORSÁK, M., PIVEC, V., 2000: Antioxidant Contents and Composition in Some Vegetables and Their Role in Human Nutrition. *Hort. Sci.*, 27, 2: 65–78. ISSN 0861-867X.

Mc LAUGHLIN and WEIHRAUCH, J. L., 1979: Vitamin E content of foods. *J. Am. Dietet. Assoc.*, 75, 6: 647–651. ISSN 0002-8223.

NESVADBA V., POLONČÍKOVÁ Z., HENYCHOVÁ A., 2012: Hodnocení Žateckého poloraného červeňáku z pivovarského hlediska. *Kvasny Prum*. 58, 7–8: 209–214. ISSN 0023-5830.

OLAS, B., KOLODZIEJCZYK, J., WACHOWICZ, B., JEDREJEK, D., STOCHMAL, A., OLESZEK, W., 2011: The extract from hop cones (Humulus lupulus) as a modulator of oxidative stress in blood platelets. *Platelets*, 22, 5: 345–352. ISSN 0953-7104.

PETERSON, D. M., QURESHI, A. A., 1993: Genotype and Environment Effects on Tocols of Barley and Oats. *Cereal Chem.*, 70, 2: 157–162. ISSN 0009-0352.

PRÝMA, J., EHRENBERGEROVÁ, J., BELCREDIOVÁ, N., VACULOVÁ, K., 2007: Tocol content in barley. *Acta Chim. Slov.*, 54, 1: 102–105. ISSN 1318-0207.

ŠTÍPEK, S. a kol., 2000: *Antioxidanty a volné radikály ve zdraví a v nemoci*. Praha: Grada Publishing, 314 s. ISBN 80-7169-704-4.
VELÍŠEK, J., 1999: *Chemie potravin, II.,* Tábor: OSSIS, 51–55. ISBN 80-902391-4-5.
WEBER, S., U., RIMBACH, G., 2002: *Handbook of antioxidants*. 2nd edition. Switzerland: Marcel Dekker. Biological Activity of Tocotrienols, 109–115. ISBN 978-0-8247-0547-3.
ZINGG, J.-M., 2007: Vitamin E: An overview of major research directions. Review. *Mol Aspects Med,* 28, 5–6: 400–422. ISSN 0098-2997.

Address

Ing. Helena Pluháčková, prof. Ing. Jaroslava Ehrenbergerová, CSc., Ing. Blanka Kocourková, CSc., Department of Crop Science, Breeding and Plant Medicine, Mendel University in Brno, Zemědělská 1, 613 00 Brno, Czech Republic, Ing. Karolína Benešová, Ph.D., Research Institute of Brewing and Malting, Mostecká 7, 614 00 Brno, Czech Republic, e-mail: schella@centrum.cz, benesova@beerresearch.cz