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# Antioxidant activity and selected nutritional values of plums (*Prunus domestica* L.) typical of the White Carpathian Mountains

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ABSTRACT

The paper presents a description of chemical characteristics of selected cultivars and subspecies of plum trees (*Prunus domestica* L.). Traditional commercial cultivars were compared with typical regional cultivars originating from the same locality of the White Carpathian Mountains. In this region, growing and use of specific local cultivars, and processing of fruit for alimentary purposes are a traditional activity which has been performed there for centuries. Regional cultivars showed outstanding nutritional properties, especially as far as the total content of phenolic substances was concerned (3.48–4.95 mg GAE $g^{-1}$ FM); this parameter was highly correlated with the total antioxidant capacity of the fruit ($r^2$ = 0.893). A higher content of minerals and pectins in some local cultivars was also of interest. This paper demonstrates beneficial properties of some less known but regionally typical European cultivars of plums and contributes to their wider use in breeding practice and also as a potential source of nutrients for human diet.



## 1. Introduction

The White Carpathians is a mountain range situated along the south-east border of the Czech Republic with Slovakia. Today this mountain range belongs to a protected landscape area with the total acreage of approximately 1150 $km^2$; this enables to maintain and develop its natural abundance and ecological fund. There are unambiguous proofs that the territory was inhabited already ten thousand years ago and the first proofs of growing of wild *Prunus* species stretch back to the 8th century A.D. (Kuca et al., 1992).

Already in the Middle ages, the growing of plums had a long tradition there and this has persisted till the present time. In the course of centuries, many different taxa arose there and they were used when selecting today's more frequent hybrids that are dealt with in this paper. Besides the direct consumption, plums were used also for drying, plum jam making and (above all after the year 1835) production of distillates. This tradition has survived until now and it can be said that plums are the most characteristic fruit tree species in the region of the White Carpathians (Tetera, 2006).

Plums represent an excellent source of nutrients and contribute significantly to human nutrition (Cao et al., 1997). They are also an important source of compounds influencing human health and preventing the occurrence of many diseases (Stacewicz-Sapuntzakis et al., 2001). In this context it is necessary to mention above all their contents of flavonoids, anthocyans, carotenes and polyphenolic acids, which contribute to a strong antioxidant capacity of their fruit (Vinson et al., 2001). Since plants contain many different classes and types of antioxidants, knowledge of their total antioxidant capacity (TAC), which is the cumulative capacity of food components to scavenge free radicals, would be useful for epidemiology purposes (Gey, 1998). TAC is thought to be one of the basic measures of foodstuffs biological value (Wang et al., 1996).

In the current pomological system, plum cultivars are classified as members of the species *Prunus domestica* L. From the pomological point of view, plums (*P. domestica* L.) species are classified as follows (Kutina, 1991):

- *P. domestica* L., subsp. *insititia* – that involves yellow plums (var. *pomariorum*);
- *P. domestica* L., subsp. *syriaca* – that involves mirabelles (var. *cerea*);
- *P. domestica* L., subsp. *italica* – that involves rounded renclode (var. *claudiana*) and egg-shaped (oval) renclodes (var. *ovoidea*) and
- *P. domestica* L., subsp. *oeconomica* – that involves varieties *pruneauliana* and *mammilaris*.

From the viewpoint of their occurrence and (also) commercial utilisation, the most important are plums (*P. domestica* L., subsp. *oeconomica* – that involves varieties) in the study territory (Tetera, 2006) and for that reason we decided to study six typical regional

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cultivars of plums. For comparison, the most frequent cultivars cultivated also in other European regions were harvested in the study area as well. The main objective of this study was to point out those significant properties of these specific plum cultivars that were related to their effects on human health. The aim was to determine particularly the correlation between polyphenols and antioxidant activity and also find out the differences in contents of nutritional components such as pectins, crude protein, potassium, calcium, magnesium and sodium. The described cultivars could also become a potential source of breeding material for selection and cultivation in other parts of Europe or even other parts of the temperate zone worldwide.

## 2. Materials and methods

### 2.1. Locality description and collection of samples

Fruit were harvested in experimental orchards of Tomas Bata University Zlin within the period of 2006 – 2008. These orchards are situated in the south-western part of the White Carpathians near Zlin, the Czech Republic. The average altitude is 340 m above sea level, and the mean annual temperature and precipitation are 7.9 °C and 760 mm, respectively. Soil type was classified as the Mesotrophic Cambisol.

Fruit were harvested in consume ripeness from five trees of each cultivar (thus each year in five replications) under study in the course of September. 20 randomly chosen fruit from each tree were used for analyses (i.e. altogether 100 per each cultivar). The age of experimental trees ranged from 8 to 15 years.

### 2.2. Sample processing

Fruit of individual cultivars were processed immediately after the harvest (not later than within two days). Harvested plums were puréed in a mixer and the average sample was obtained by means of quartation. Each parameter was measured in five replications.

Fresh samples were used for the measurement of the total content of phenolic substances and of overall antioxidant activity. To characterise the nutritional value of plums, these basic parameters were supplemented with data about contents of some mineral elements (phosphorus, potassium, calcium, magnesium and sodium), crude protein value and pectins.

To our best knowledge the following cultivars were analysed – regional plum cultivars 'Bluma', 'Durancia', 'Kulovacka', 'Pavluvka', 'Svestka domaci', and 'Wangenheimova' and European plum cultivars 'Augustinka', 'Bryska', 'Hamanova svestka', 'Kirkeho', 'Stanley' and 'Vlaska'.

### 2.3. Extraction procedure

Extraction was performed according to the method described by Kim et al. (2003a) (in Vasantha Rupasinghe et al., 2006) using the following procedure: 10 g of fresh sample were homogenised for 10 s in an extraction mixture (hydrochloric acid: methanol: water in the ratio 2: 80: 18). The resulting paste was placed into Erlenmeyer flasks (120 ml) and let to stand in a water bath with the temperature of +50 °C for a period of 2 h.

Thereafter the suspension was centrifuged for 10 min (at 3000 r.p.m) and the supernatant was filtered under vacuum through the glass frit; 30 ml of sample were vacuum evaporated to a final volume of approximately 5 ml and the total solids were quantitatively transferred into Eppendorf beakers (10 ml) and diluted up to the mark with water.

To measure total contents of phenolic substances, 0.5 ml of the sample was taken and diluted with water in a 50-ml volumetric flask. Thereafter, 2.5 ml of Folin-Ciocalteau reagent and 7.5 ml of a 20% solution of sodium carbonate were added. The resulting absorbance was measured in the spectrophotometer LIBRA S6 at the wavelength of 765 nm against a blind sample, which was used as reference. Results were expressed as mg of gallic acid $g^{-1}$ of fresh mass (FM).

### 2.4. Antioxidant activity assay

The antioxidant activity was measured using the method described by Sulc et al. (2007). This test is based on monitoring of the course of inactivation of the cation $ABTS^{+}$, which is produced during the oxidation of 2,2′-azinobis(3-ethylbenzothiazoline-6-sulphonate). $ABTS^{+}$ shows a strong absorbance in the visible region of the electromagnetic spectrum (600–750 nm); this solution is green and its antioxidant activity can be easily measured by means of spectrophotometry. This method is standardly used when measuring antioxidant activity and its great advantage consists in reproducibility and a good fit with other methods also used for measuring of antioxidant activity (Thaipong et al., 2006).

Altogether 54.9 mg of ABTS were dissolved in 20 ml of phosphate buffer (pH 7.0; 5 mM) and activated on cation radical of $ABTS^{+}$ by means of an addition of 1 g of $MnO_2^{+}$; the resulting solution was intermittently stirred for an activation period of 30 min. Thereafter, the solution was centrifuged for 5 min and at 7000 r.p.m and filtered through a syringe filter (0.25 μm) and 2 ml of the filtrate were diluted with phosphate buffer to the absorbance ($t_0$) of $0.500 \pm 0.01$, which was measured at the wavelength of 734 nm. After absorbance measured in time $t_0$, 0.5 ml of the sample was added and the new absorbance value was measured in time $t_{20}$, i.e. after 20 min. The antioxidant activity was calculated as a decrease in absorbance value using the formula:

$$\% = 100 - \left[\frac{A_{t_{20}}}{A_{t_0}}\right] \times 100$$

The calculated activity was converted using a calibration curve of the standard and expressed in ascorbic acid equivalents (AAE) (Vasantha Rupasinghe et al., 2006).

### 2.5. Mineral content assay

The sample was dried to a constant weight in a drier at $105 \pm 2$ °C; thereafter, 1 g of homogenised dry matter (with the size of particles 1 mm) was further mineralised in a mixture of concentrated sulphuric acid with 30% hydrogen peroxide. After the mineralization, the obtained samples were quantitatively transferred into a 250-ml volumetric flask and filled to the volume with re-distilled water. The resulting mineralisate was measured in an atomic absorption spectrometer (PHILIPS PU 9200X). The amount of crude protein was estimated on the base of total nitrogen in the Kjeldahl apparatus KJELTEC TM 2300 and the result was multiplied by the coefficient 6.25 (Novotny, 2000).

### 2.6. Content of pectin assay

The content of pectins was measured using the modified method described by Rop et al. (2008). 10 g of crushed fruit material were extracted with hydrochloric acid ($c = 1$ mol $dm^{-3}$) in a shaker at 80 °C for 90 min. The hydrolyzate was quantitatively transferred into a 250-ml volumetric flask and filled to the volume with water. Pectins were thereafter measured by photometry as a coloured complex consisting of the product of thermal decomposition of galacturonic acid with *m*-hydroxybiphenyl in a medium containing concentrated $H_2SO_4$. Thereafter, samples of 5 ml were gradually taken off, put into 50 ml flasks and supplemented always with 6 ml of the solution of sodium tetraborate ($c = 0.013$ mol $dm^{-3}$) and 1.5 mg of *m*-hydroxybiphenyl in concentrated sulphuric acid,