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INFLUENCE OF RIPENING ON THE ETHYLENE AND CARBON DIOXIDE PRODUCTION DURING STORAGE OF PLUM FRUITS

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Abstract

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The fruits of 13 plum cultivars were analysed at two different stages of maturity: first when they were ready for picking according to conventional commercial criteria, and again after seven days of maturation at 20 °C in a normal oxygen atmosphere during shelf life. Firmness, soluble solids concentration (SSC), respiration rate and the production of ethylene were measured. In the period of over-ripening SSC increased, and no differences were found in relation to the cultivar. The fruits in this period were physiologically in a phase of reduced intensity of respiration, while production of ethylene increased and was associated with the earliness of the cultivar. Cultivars with a short vegetation period produce more ethylene while late-maturing cultivars have a low potential for ethylene production. Based on post-harvest changes in the tested quality factors, the late-ripening plum cultivars (cv.) 'Jojo', 'Topend', 'President', 'Tophit' and 'Elena' have higher storage potential. Changes in ethylene production in response to shelf life seem to play a regulatory role in fruit firmness. A high significant negative correlation was detected between levels of skin firmness and ethylene production.

plum fruits, cultivar, ripening, ethylene, respiration, firmness

Plums (*Prunus domestica* L.) are members of the *Rosaceae* family, genus *Prunus* along with other stone fruits such as peaches, apricots, nectarines, cherries. Based on a survey done by Blažek (2007) the author commented that plums constitute the most numerous and diverse group of fruit tree species traded internationally. Plums rank as the fourth most important fruit species of temperate area.

The plum has been categorised as a climacteric fruit, with a high rate of respiration between the end of the fruit's development and the start of the ripening process. The increase in respiration is concomitant with an enhancement of ethylene synthesis (KAYS and PAULL, 2004), which is cultivar-dependent. Ethylene is thought to function as a plant hormone responsible for coordinating and initiating ripening events in climacteric fruit (BAPAT *et al.*, 2010; ALEXANDER and GRIERSON, 2002). It triggers the processes of ripening and senescence. The main consequences of this behaviour are a reduced shelf life and a decrease in the quality parameters, such as skin and mesocarp colour changes, autocatalytic ethylene production, chlorophyll degradation, sugar accumulation, occurrence of decay and fruit softening (TRAINOTTI *et al.*, 2007; GIOVANNONI, 2001). The latter is critical for the plum fruit industry, as it directly dictates fruit shelf life and quality (VALERO *et al.*, 2007). There is a lack of reference data for different varieties, particularly for European plums (USENIK *et al.*, 2008).

The plum fruit is highly perishable. Ripening, softening and deterioration of the plum fruit after harvest, which results in their short shelf life at ambient temperature, occur very quickly (MENNITI *et al.*, 2004). Therefore, they require special care during handling and storage (ABDI *et al.*, 1998). As part of the senescence process, the over-ripe stage of a climacteric fruit is considered to be when the fruit loses part of its marketable characteristics, such as texture, colour and flavour. According to CRISOSTO *et al.* (2004), plums with flesh firmness below 9 and 13 N are considered to be in the over-ripe stage, hence commercially unmarketable. The change in fruit firmness has been a reliable parameter for measuring ripening changes (KADER, 2002), since it correlates to ethylene production (KHAN and SINGH, 2007).

The management and control of fruit ripening is important for the successful transport and marketing of fresh fruit. An increased shelf life period could be useful for improving distribution and could favour long-distance transport. Today, the extended availability of seasonal fruit in the marketplace is commonplace thanks to both post-harvest technologies and fruit varieties that allow for longer periods of storage. Each season, consumers increase their requirements and demand higher quality products. Flavour is remarkably important among the attributes consumers use to assess fruit quality.

The purpose of this study was to evaluate the physicochemical changes in 13 plum cultivars during their ripening and over-ripening stages. The fruit were harvested at the ripe stage and ripened at ambient atmosphere and room temperature for a period of 7 days. From a traditional fresh market point of view, studying the over-ripening stage may not be of practical interest since the fruit is commercially unmarketable. However, the focus is mainly on the amount of ethylene being synthesized. The aim of this study is to analyze the influence of maturation on measured quality changes in European (*Prunus domestica* L.) plums and to find any correlations between the measured parameters. This research also helps to reveal aspects that may help to explain some of the physiological differences among the varieties being evaluated. For this purpose, experiments were carried out on 13 plum fruit cultivars with differing climacteric behaviour.

MATERIALS AND METHODS

Plant material

A total of 13 plum fruit cultivars (*Prunus domestica* L.) were harvested at commercial maturity stage between third week of July and first week of September from two orchards during the growing season in 2011. Eight plum fruit varieties ('Topfirst', 'Čačanska lepotica', 'Stanley', 'Valjevka', 'Čačanska najbolja', ' Topend', 'President', Tophit') were obtained from a commercial orchard in Agro Stošíkovice, Ltd., South Moravia, Czech Republic. Five cultivars ('Gabrovska', 'Hanita', 'Valor', 'Jojo', 'Elena') were from the orchards of the Faculty of Horticulturae in Lednice (Mendel University in Brno). Fruit maturity was judged on the basis of soluble solids concentration (SSC) and fruit firmness. Plum fruits were harvested manually and transported within several hours to the technological laboratory of the Institute of Postharvest Technology of Mendel University in Brno, Lednice, Czech Republic. Immediately before storage plums were sorted in order to remove mechanically damaged fruits and fruits without stems. Subsequently they were allowed to ripen at 20 °C for up to 7 days, in order to simulate the period of commercial shelf life. Samples were taken immediately after harvest and seven days of shelf life for the analyses of ethylene and respiration rate.

Ethylene and CO_2 measurement

Ethylene production and respiration rate were measured of each variety initially and after seven days of storage in ambient atmosphere at 20 °C. Ethylene and CO_2 production were measured by placing three samples of 5 fruits each in a 1 litre glass jar hermetically sealed with a rubber stopper for one hour. Ethylene and CO_2 were monitored by injecting 1 ml of a headspace gas sample into an Agilent 4890D gas chromatograph (Agilent Technologies, Inc., Wilmington, DE, USA). Both gases were determined simultaneously on dual columns. A HP-Plot/Q column 30 m, I.D. 0.53 mm, film 40 µm was used for ethylene and detected on FID, and a HP-AL/KCL column 30 m, I.D. 0.53 mm, film 15 µm was used for CO_2 and detected on TCD. Helium at 1.2 ml/min was used as a carrier gas. Ovens were programmed to rise from 80 to 120 °C at a rate of 10 °C/minute. The concentration of ethylene are expressed as microliters per kilogram per hour, those for CO_2 are expressed as mg per kilogram per hour.

Firmness measurement and soluble solids

Fruit quality parameters (firmness, weight, total soluble solids) were determined using conventional destructive methods. A set of 15 fruit from each variety was used to measure firmness, soluble solids content (SSC) initially and after seven days of shelf life. The technique of pushing a plunger into the intact fruit under a defined loading rate was used for evaluating the firmness of the skin and flesh and for determination of the fruit's toughness. Firmness was measured on two opposite sides of each fruit using a universal testing machine TEXAN 2000 instrument (constructed at Mendel University, Brno, Czech Republic) which recorded compression and rate of loading. A steel plunger with 5 mm diameter was pressed into the fruit at a rate of 8 $mm.min^{-1}$ and the resulting force deformation curve with two turns was plotted. The break in the curve indicates the puncture point when the plunger breaks the skin (a measure of skin firmness) and the sudden decrease in force observed provides a measure of flesh firmness. The area under the deformation curve measures toughness. For each fruit two deformation curves were generated. The measurements were expressed in units of MPa. For each sample and after firmness determination, plums were homogenized in a blender. Samples were then immediately frozen and stored at –21 °C prior to analysis.

The soluble solids content (SSC) was determine in the juice obtained from each subsamples of flesh tissue and measured in replicate using a digital refractometer giving readings as °Brix.

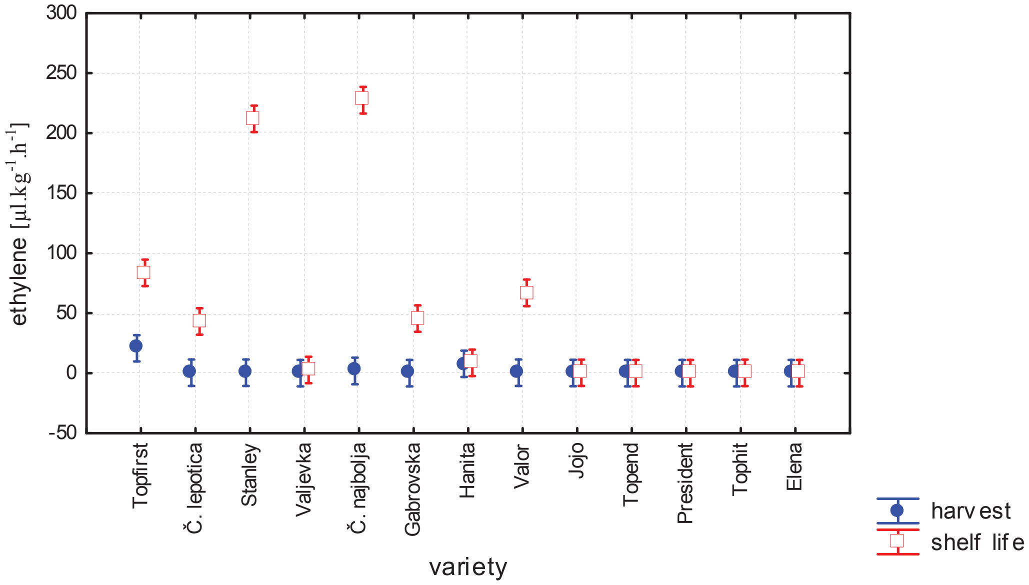
Statistical analysis

Analysis of variance was performed and means were compared with the Tukey's multiple range test of significant difference ($p < 0.05$). Pearson correlations were performed between the ethylene production and fruit firmness. Statistical analyses were performed using Statistics (version 9.0 for Windows).

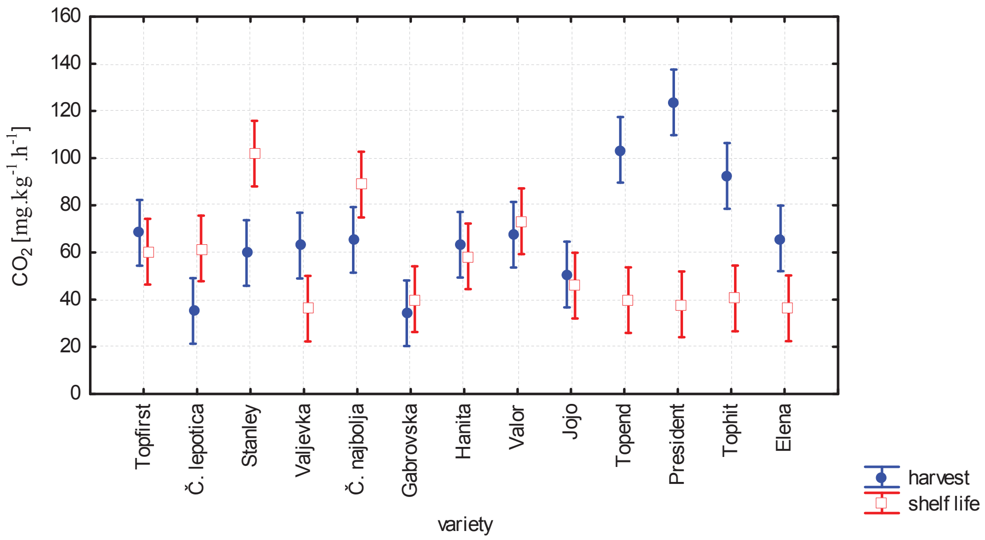
RESULTS AND DISCUSSION

Respiration and ethylene production

The plum is a climacteric fruit which significantly increases ethylene production during ripening (BAPAT *et al.*, 2010). In climacteric fruit, ripening events such as fruit softening, chlorophyll degradation, colouring and sugar accumulation are considered to be induced by ethylene (ALEXANDER and GRIERSON, 2002). These effects lead to an acceleration of the ripening process, and in turn short periods of shelf life lasting no longer than several days even during cold storage (GOLIÁŠ *et al.*, 2010). However, ethylene production in plums depends on the cultivar (ABDI *et al.*, 1998).



1: *Production of ethylene in 13 plum fruit varieties at harvest and after seven days of the shelf life period at 20 °C in normal oxygen atmosphere. Plum varieties are sorted by harvest date. Vertical bars denote standard error of the mean (n = 15).*



2: *Respiration rate in 13 plum fruit varieties at harvest and after seven days of the shelf life period at 20 °C in normal oxygen atmosphere. Plum varieties are sorted by harvest date. Vertical bars denote standard error of the mean (n = 15).*

Fruits judged to be at the optimal degree of maturity for commercial harvest were completely blue (purple) over the whole surface. Ethylene was present in very low concentrations in fruits at harvest. Ethylene production in plum fruits varied from 0.04 to 1.89 μl.kg^{-1} per hour (Fig. 1) with the exception of the cv. 'Hanita' and 'Topfirst', in which the production of ethylene was significantly higher in the mature fruits (7.89 and 20.76 μl.kg^{-1}.h^{-1}). This was explained by the fact that plums, at the initial phase of ripening, normally demonstrate very low or undetectable levels of ethylene production (VALERO *et al*., 2003, WU *et al*., 2011).

Respiration was measured as CO_2 production. The range of CO_2 production was from 34.31 mg.kg^{-1} to 68.35 mg.kg^{-1} per hour (Fig. 2). Significantly higher production of carbon dioxide was detected in the cv. 'Tophit', 'Topend' and 'President' (92.51 ± 6.72 mg. kg^{-1} per hour, 103.56 ± 3,12 and 123.66 ± 12.83 mg. kg^{-1} per hour, respectively).

After the fruits were incubated at ambient atmosphere and room temperature of 20 °C, plum fruits increased their ethylene production. After seven days of the shelf life simulation, a statistically significant increase in ethylene production was observed in early-maturing cultivars ('Topfirst', 'Čačanska lepotica', 'Stanley', Čačanska najbolja', 'Gabrovská' and 'Valor'), with the exception of 'Hanita' and 'Valjevka' (Fig. 1). The production of ethylene in late-maturing cultivars after seven days of shelf life varied from 0.035 μl.kg^{-1} to 0.397 μl. kg^{-1} per hour. The cultivar 'President' exhibited the lowest rate of ethylene production in shelf life (0.035 μl.kg^{-1} per hour). Production of ethylene is in general lowest in the late-maturing cultivars than in the early-maturing fruits. This trend was observed in harvested fruits and after incubation of fruits at ambient conditions, too. According to Goliáš *et al.* (2011), this is also seen in apricots, where the fruits of late-maturing cultivars produce lower levels of ethylene than early-maturing cultivars. Typically, early-maturing fruits have a shorter storage life (GOLIÁŠ *et al*., 2011).

When the fruits were transferred to 20 °C, the respiration rate increased greatly only in cultivar 'Stanley', reaching a peak of 101.9 ± 3.8 mg.kg^{-1} per hour, which was double the level measured in the mature fruits. There was a statistically significant decrease in the respiration rate in the cultivars 'Topend', 'President', ' Tophit' and 'Elena'.

Fruit quality parameters

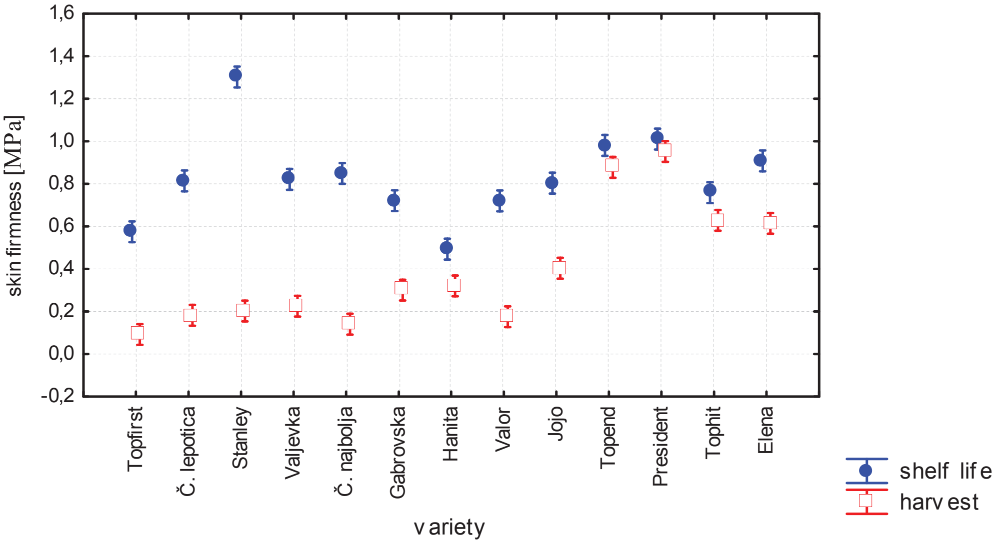
Fruit quality is generally measured in terms of colour, shape, texture, total soluble solids, titratable acidity and particularly for the consumer, aroma-related volatiles (ZHANG, 2011). However, large variations in these parameters can be found depending on the cultivar, production area, climatic conditions and harvest season (KADER and MITCHELL, 1998).

Deformation after breaking of the skin was taken as the criterion for plum firmness. The parameter of skin firmness provides more reliable values than flesh firmness (GOLIÁŠ *et al*., 2010). Initial values for skin firmness (Fig. 3) varied from 0.72 MPa to 0.98 MPa, with the exception of cv. 'Hanita' and 'Topfist' (0.49 MPa and 0.58 MPa, respectively), although a higher value was recorded in the cv. 'Stanley' (1.30 MPa).

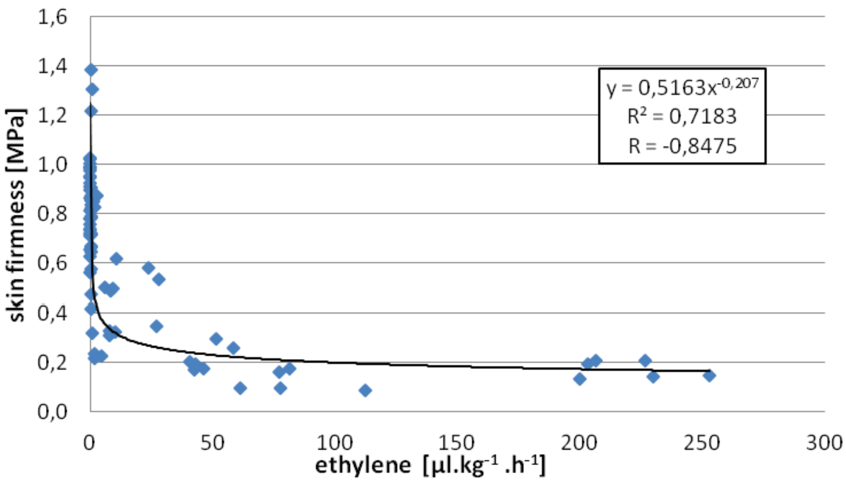
During shelf life simulation at 20 °C a decline in skin and flesh firmness was observed for all of the analysed varieties. After a seven-day period of shelf life, the fruits softened further at a rate ranging from 0.03 MPa/day to 0.1 MPa/day but still within the bounds of acceptability. Values higher than this range were recorded for the early-season cv. 'Stanley' (0.16 MPa per day). The rate of softening of skin and flesh was lower in the late-season plum fruit varieties during shelf life. Thus, a significant effect of higher temperature and ambient conditions on early-season plum varieties was observed. Early season plum varieties are usually less firm at the minimum maturity time than late season varieties (CRISOSTO, 1994). The influence of the season of harvest and stage of maturation was significantly ($P \leq 0.05$) high in all the subjected cultivars with the exception of the cv. 'President'. The main problem during the marketing of plums is excessive softening (CRISOSTO *et al*., 2004). During ripening, tissue softening related to the reduced shelf life occurs, especially at 20 °C (VALERO *et al*., 2003). Fruit softening is therefore a suitable predictor of the potential shelf life for plums (VALERO *et al*., 2007). Fruit softening is associated with cell wall loosening. Pectin degradation plays an important role in fruit softening, which leads to the disassembly of the cellulose and hemicellulose network and a decrease in fruit firmness (LIU *et al*., 2009, PRASANNA *et al*., 2007). Fruit firmness measurement has been a reliable parameter for monitoring the changes in ripening (KADER, 2002) and is a good way to monitor fruit softening and to predict bruising damage during harvest and post-harvest handling (VALERO *et al*., 2007).

Fruit softening is a ripening process that is sensitive to ethylene, but little is known of the role ethylene plays in the softening of these fruits. To check the effect of ethylene production on fruit quality parameters during maturation, nonlinear regressions were performed taking into account the data for all cultivars. Skin firmness, flesh firmness and toughness were negatively correlated to ethylene production for both ripening stages. Results from nonlinear regressions revealed that skin firmness was ethylene dependent during shelf life for 13 plum fruit cultivars (Fig. 4).

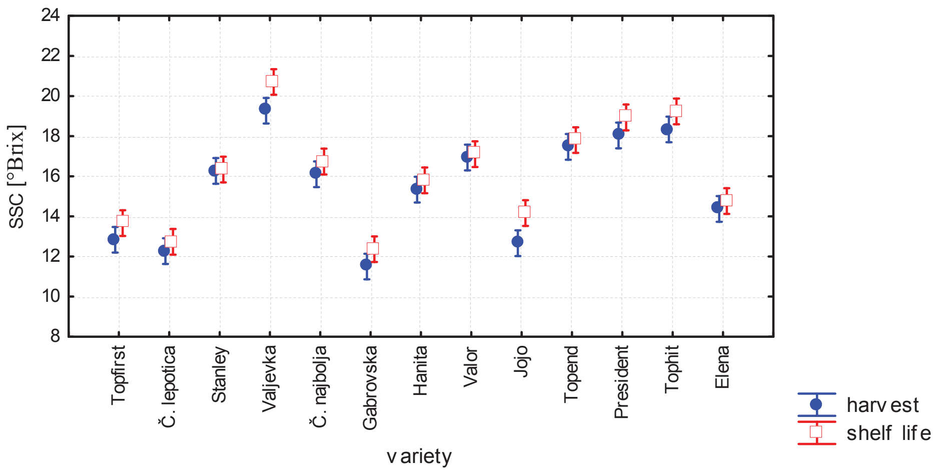
A high significant negative correlation was detected between levels of skin firmness and the production of ethylene ($y = 0.5163x^{-0.207}$, $r = -0.8475$). This finding is in agreement with those provided by Khan and Singh (2007). They found a significant negative correlation ($r = -0.737$) between ethylene production and fruit firmness (KHAN and SINGH, 2007). Thus, a maximum peak in ethylene



3: *Changes over time in skin firmness of 13 plum fruit cultivars at harvest and after seven days of the shelf life period at 20 °C in normal oxygen atmosphere. Plum varieties are sorted by harvest date. Vertical bars denote standard error of the mean (n = 15).*



4: *Nonlinear regression between ethylene production and skin firmness*



5: *Soluble solids content in 13 plum fruit varieties at harvest and after seven days of the shelf life period at 20 °C in normal oxygen atmosphere. Plum varieties are sorted by harvest date. Vertical bars denote standard error of the mean (n = 15).*

production will correspond to a lower firmness value (ABDI *et al.*, 1998, KHAN and SINGH, 2007). Firmness of the plum fruit negatively correlated with increased production of ethylene in fruit pulp and its subsequent release from the intact fruit.

The soluble solids concentration (SSC) in stone fruit is an essential quality index which is highly associated with sensory quality and thus plays a crucial role in consumer acceptance (CRISOSTO *et al.*, 2004). Furthermore, SSC is one of the quality parameters most influenced by environment and rootstock. Changes in the SSC of plum fruit cultivars during post-harvest ripening at 20 °C are shown in Fig. 5.

SSC increases during fruit ripening depending on the cultivar (USENIK *et al.*, 2008). In general, early-season plum cultivars have a lower SSC than late-season plum cultivars (MANGANARIS *et al.*, 2008; CRISOSTO *et al.*, 2007). Significant changes in soluble solids content were not observed in any of the cultivars tested, which confirms the work on other plum cultivars by Vangdal et al. (2007). The increase in SSC was significantly higher only in the cv. 'Jojo' (12.7 °Bx and 14.2 °Bx).

CONCLUSIONS

Plums, as a typical climacteric type of fruit, have the potential to produce ethylene. In the monitored cultivars, which ripened over a broad time period, it was shown that early-ripening cultivars have a higher production of ethylene, and in late-ripening cultivars ethylene production is clearly reduced. Cultivars which are stored at up to 20 °C (shelf-life) respond with a high production of ethylene and softening. The production of ethylene is significantly associated with softening. Fruits should be stored in a period when the production of ethylene is in the climacteric phase. The determined values of ethylene corresponding to the degree of ripeness labelled as "ripe" correspond to the possible distribution of fruits in the commercial sphere. An increased shelf life period could be useful to improve distribution and favour long-distance transport. Changes in ethylene production in response to shelf life have a regulatory role in fruit firmness.

SUMMARY

The aim of this study is to analyze the influence of maturation on measured quality changes in European (*Prunus domestica* L.) plums and to find any correlations between the measured parameters. This research also helps to reveal aspects that may help to explain some of the physiological differences among the varieties being evaluated. The fruits of 13 plum cultivars were analysed at two different stages of maturity: first when they were ready for picking according to conventional commercial criteria, and again after seven days of maturation at 20 °C in a normal oxygen atmosphere during shelf life. Firmness, soluble solids concentration (SSC), respiration rate and the production of ethylene were measured. The monitored textural parameters are of importance for determining the durability of plum fruits and their quality. It was determined that softening was minimal with the storage of the fruits of late cultivars of plums in a normal oxygenated atmosphere at 20 °C. At room temperature the firmness of the flesh and skin rapidly decreases in early ripening plum cultivars. The speed of softening of the fruits is higher in plums with a higher production of ethylene. Late-ripening cultivars are characterised after harvesting by production of ethylene in a range of values from 0.04 to 0.21 $\mu l.kg^{-1}.h^{-1}$. Likewise, in the course of their shelf life only a negligible surge of ethylene was found in late-ripening cultivars. The late-ripening plum cultivars 'Jojo', 'Topend', 'President', 'Tophit' and 'Elena' have a higher storage potential. They can be stored at 20 °C without a significant growth in ethylene and only a moderate loss of firmness. The fruits of these cultivars produce a lower content of ethylene upon harvesting and in the course of their shelf life. A highly conclusive indirect association was found between ethylene content and firmness of plum fruits. Ethylene production could serve as criterion for estimation of varietal differences.

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