

EFFECT OF TEMPERATURE AND 1-METHYLCYCLOPROPENE ON THE STORABILITY OF 'ALEXANDER LUCAS' PEAR¹

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ABSTRACT - The storage life of 'Alexander Lucas' pear is limited by the occurrence of internal storage disorders during controlled atmosphere (CA). 1-Methylcyclopropene (1-MCP) can be used to delay pear ripening, however, its interaction with CA conditions is not well understood. 'Alexander Lucas' fruit were treated with 1-MCP at 300 nL L⁻¹ for 24 h in air at -0.5 or 1.0 °C before being stored. Fruit were kept at -0.5 or 1.0 °C in normal air or CA (2.0 kPa O₂ plus <0.7 kPa CO₂). After six months of storage, superficial scald did not develop in fruit. The highest incidence of flesh browning (72.2 %) was found in air-stored fruit at -0.5 °C without 1-MCP treatment. Storage in air at 1.0 °C plus 300 nL L⁻¹ 1-MCP produced 88.8 % of healthy fruit. In contrast, 1-MCP increased the incidence of flesh and core browning under CA conditions. 1-MCP and CA maintained greener skin color and higher titratable acidity. No significant differences were found for fruit firmness, total soluble solids and ascorbic acid content between the treatments. In conclusion, the quality of 'Alexander Lucas' pear was best maintained during six months storage under normal air at 1.0 °C combined with 300 nL L⁻¹ 1-MCP treatment.

Keywords: Ethylene action inhibition. Internal storage disorders. *Pyrus communis*. Controlled atmosphere.

EFEITO DA TEMPERATURA E 1-METILCICLOPROPENO NA CONSERVAÇÃO DE PERA 'ALEXANDER LUCAS'

RESUMO - O tempo de armazenamento da pera 'Alexander Lucas' é limitado pela ocorrência de distúrbios fisiológicos, principalmente quando em atmosfera controlada (AC). O 1-metilciclopropeno (1-MCP) pode ser usado para retardar o amadurecimento das peras, no entanto, sua interação quando usado em AC não está bem compreendida, podendo por um lado, retardar o processo de amadurecimento e preservação da qualidade do fruto, mas por outro lado, podendo agravar a ocorrência de desordens fisiológicas em AC. Frutos da cultivar 'Alexander Lucas' foram tratados com 300 nL L⁻¹ de 1-MCP por 24 h em armazenamento refrigerado (AR) nas temperaturas de -0,5 ou 1,0 °C, antes do armazenamento. Os frutos foram mantidos a -0,5 ou 1,0 °C em AR ou AC (2,0 kPa O₂ + <0,7 kPa CO₂). Após seis meses de armazenamento, não ocorreu escaldadura superficial. A maior incidência de escurecimento da polpa (72,2 %) foi observada em frutos armazenados em AR a -0,5 °C sem tratamento com 1-MCP. O armazenamento em AR a 1,0 °C mais 300 nL L⁻¹ 1-MCP manteve 88,8 % dos frutos sadios. No entanto, o uso de 1-MCP aumentou a incidência de escurecimento da polpa e do miolo quando em condições de AC. 1-MCP e AC mantiveram a coloração da casca mais verde e maior acidez titulável. Não foram encontradas diferenças na firmeza de polpa dos frutos, nos sólidos solúveis totais e na concentração de ácido ascórbico. Concluiu-se que, a qualidade da pera 'Alexander Lucas' se conservou melhor durante seis meses de armazenamento em AR a 1,0 °C com a aplicação de 300 nL L⁻¹ de 1-MCP.

Palavras-chave: Inibição da ação do etileno. Distúrbios internos de armazenamento. *Pyrus communis*. Atmosfera controlada.

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INTRODUCTION

'Alexander Lucas' pear is considered a high-quality cultivar with a good acceptance rate by consumers in Germany, which in 2014 represented close to 29 % of total pear production (DIEREND; BIER-KAMOTZKE, 2014). 'Alexander Lucas' is usually stored under normal air and not under controlled atmosphere (CA) because of its sensitivity to high CO₂ partial pressure, and thus has a limited storage life, even at -1 °C (GARCIA; STREIF, 1993). CA storage can induce the development of internal physiological disorders in 'Alexander Lucas', with the frequent formation of cavities occurring in fruit flesh when O₂ partial pressures (pO_2) are kept below 3.0 kPa or carbon dioxide partial pressures (pCO_2) are above 0.7 kPa (GARCIA; STREIF, 1993; STREIF; SAQUET, 2006).

Application of 1-Methylcyclopropene (1-MCP) can prolong the storage life of pears (EKMAN et al., 2004). However, depending on the 1-MCP concentration, maturity stage at harvest and storage conditions, some pear cultivars have difficulty to ripen and develop satisfactory skin color and organoleptic characteristics such as juiciness and adequate softening after 1-MCP treatment (CHIRIBOGA et al., 2011). Streif and Saquet (2006) investigated the effect of 1-MCP treatment on the storability and quality maintenance of 'Conference' and 'Alexander Lucas' pears and concluded that 1-MCP concentrations need to be much lower than those used on apples. However, 1-MCP could maintain satisfactory fruit quality under both air and CA storage, particularly for late harvested pears. 'D'Anjou' pears treated with 1-MCP showed low ethylene production and respiration rates (ARGENTA; FAN; MATTHEIS, 2003). 'La France' pears had higher firmness and lower respiration rates in comparison to the untreated control fruit (KUBO et al., 2003). Alpalhão et al. (2006) showed that less

mature 'Rocha' pear stored for seven months in air maintained good fruit quality when treated with 1-MCP, but there were no benefits of 1-MCP application on later harvested pears.

The challenge for the long-term CA storage of 'Alexander Lucas' pears is to prevent the development of superficial scald and internal physiological disorders while maintaining acceptable fruit quality for further marketing and consumption. Therefore, this research evaluated the effect of 1-MCP application on the storability of 'Alexander Lucas' pear under different storage conditions.

MATERIAL AND METHODS

Plant material and sampling

'Alexander Lucas' pears were harvested from the Competence Centre for Fruit Growing at Lake Constance (KOB Bavendorf) in Ravensburg, Germany. Fruit maturity parameters at harvest were: flesh firmness (46.2 N), titratable acidity (4.44 % malic acid), total soluble solids (11.8 %), starch index 6 (at a scale of 0 to 10; 0 = high starch content and the flesh surface is totally stained, 10 = no stained flesh), and skin color (116.8 ° hue angle). Pears were harvested at a Streif-Index of 0.07, which is slightly below the optimal harvest window for 'Alexander Lucas' pears, which is 0.08 to 0.18 (STREIF, 1996). Samples for storage treatments (Table 1) and analyses were composed of 3 replicates. For quality analysis, each replicate comprised of 8 fruit and around 20 fruit per replicate were used to assess diseases and physiological disorders.

Storage conditions, procedure and 1-MCP treatment

Storage conditions

Table 1. Storage conditions and 1-MCP treatment for 'Alexander Lucas' pear.

Storage system	Temperature (°C)	pO_2 (kPa)	pCO_2 (kPa)	1-MCP (nL L ⁻¹)
Air	1.0	20.9	< 0.7	0
Air	1.0	20.9	< 0.7	300
CA storage	1.0	2.0	< 0.7	0
CA storage	1.0	2.0	< 0.7	300
Air	-0.5	20.9	< 0.7	0
Air	-0.5	20.9	< 0.7	300
CA storage	-0.5	2.0	< 0.7	0
CA storage	-0.5	2.0	< 0.7	300

Storage procedure

For air storage, fruit were kept in storage

rooms with temperature and relative humidity control. The relative humidity was between 92 and 94 %. Since 'Alexander Lucas' is very sensitive to

CO₂, normal air storage rooms were periodically ventilated to avoid an accumulation of CO₂ or hypoxic conditions. The CO₂ was absorbed with lime in a relation of 3% of fruit volume. The conventional CA storage (Table 1) procedure was carried out according to KOB (2021).

The temperature inside all storage rooms was automatically controlled by electronic thermostats and by temperature sensors inserted in the fruit flesh.

1-MCP treatment

One day after harvest, fruit were cooled and put into gas-tight plastic boxes (650 L) for 1-MCP treatment. Each treatment was performed in a separate box. Fruit were exposed to 1-MCP for 24 h at -0.5 or 1 °C according to the respective treatments (Table 1). The 1-MCP used was SmartFresh™ for research. CA storage conditions for each treatment was established within 24 h after 1-MCP treatment.

Fruit analyses

a) Skin color: Changes in fruit skin color were measured with a Minolta CR-300 (Konica Minolta, Tokyo, Japan) colorimeter and expressed with hue angle.

b) Flesh firmness: Firmness was determined with a motorized penetrometer (Fruit Texture Analyser GÜSS, South Africa) fitted with 8 mm tip. One measurement was made on the medial diameter region of each fruit, where the skin was previously peeled. Firmness values were expressed in N.

c) Total soluble solids (TSS): From the same samples used for firmness determination, juice of the whole replicate was extracted, and the value was measured with a hand refractometer (Atago Palette PR 32a, Tokyo, Japan). The total soluble solids were expressed in percentage.

d) Titratable acidity: The same fruit samples used for firmness and soluble solids were used. 10 mL of juice was diluted with 50 mL distilled water and titrated with NaOH (0.1 N) until pH 8.1 was reached. The titratable acidity was analyzed using a Metrohm robotic 702 SM Titrino and an autosampler (815 robotic USB sample Processor XL, Metrohm, USA) and was expressed in % malic acid.

e) Ascorbic acid (AA) determination: All fruit in each replicate were cut transversely through the equatorial region, and a similar slice from the sun exposed side without core was sampled, immediately frozen in liquid nitrogen and then stored in the dark at -28 °C until analysis. The frozen samples were finely crushed and homogenized with 3 % HPO₃ solution (ratio of 10 g sample : 15 mL of 3 % HPO₃ solution) for 2 min and centrifuged for 15 min at 14,000 g at 4 °C. The supernatant was filtered (0.45 µm), and a 20 µL of sample was injected into a high-

performance liquid chromatograph (HPLC, Bischof). All steps were performed with the sample in the dark and at a temperature below 5 °C. The HPLC analysis was performed according to Veltman et al. (2000): ProntoSil column 60-5-C18 (5 µm; 125 x 4 mm); eluent was 2.5 g tetra butyl ammonium hydrogen sulphate and 55 mL methanol in 1 L Milli-Q water; flow: Isocratic, 0.8 mL min; UV detector (Lambda 1010) at 254 nm. Ascorbic acid was measured and quantified using a standard and expressed as µg g⁻¹ fresh weight (FW).

f) Incidence of storage disorders: Fruit were assessed immediately after removal from storage and after 7 days shelf life at 20 °C. Assessments made immediately after storage were performed using 3 repetitions of 70 fruit for superficial physiological disorders, and for internal disorders, only the 24 fruit from quality analysis were used. Three repetitions of 60 fruit were used for assessments after shelf life. Results of scored external and internal physiological disorders are presented as the percentage of affected fruit in a sample.

Statistical analysis

All data were subjected to analysis of variance (ANOVA), and the means of the treatments were compared using Tukey's HSD test ($p < 0.05$) using the statistical software R version 4.0.2.

RESULTS AND DISCUSSION

Superficial scald

After six months of storage, superficial scald was not observed in fruit from any storage condition. 'Alexander Lucas' is susceptible to superficial scald, and the lack of symptoms may be explained by seasonal variability or attributed to the slightly advanced maturity of the fruit at harvest. Late harvested pears are known to be less susceptible to superficial scald during long-term storage under air or CA storage (LURIE; WATKINS, 2012).

Internal storage disorders

Flesh browning was the internal disorder that most affected 'Alexander Lucas' pear during storage (Table 2). Affected fruit were found in all storage conditions, with the highest incidence of 72.2 % under normal air conditions at -0.5 °C without 1-MCP, followed by 61.1 % in air at 1 °C without 1-MCP (Table 2). In normal air storage, treatment with 300 nL L⁻¹ 1-MCP significantly reduced the incidence of flesh browning when fruit were stored at -0.5 °C (44.4 %) and 1 °C (11.1 %) compared to untreated fruit (Table 2). The higher damage may be

related to the advanced state of ripening as late harvested pears are known to be most susceptible to internal disorders during long-term storage (STREIF, 2008; DEUCHANDE et al., 2012). Normal air plus 1-MCP likely better regulated fruit metabolism during storage, resulting in the lowest flesh browning incidence (Table 2). In contrast, treatments that

strongly inhibited fruit ripening during storage increased the incidence of flesh browning as found in fruit stored under CA with 1-MCP at $-0.5\text{ }^{\circ}\text{C}$. The greater stress caused by the combination of CA plus 1-MCP at $-0.5\text{ }^{\circ}\text{C}$ was clearly detrimental for maintaining healthy 'Alexander Lucas' pears during storage.

Table 2. Incidence of storage disorders and healthy fruit of 'Alexander Lucas' pear stored under various conditions plus a 7 day shelf life at $20\text{ }^{\circ}\text{C}$.*

Treatment	Temperature ($^{\circ}\text{C}$)	Flesh browning (%)	Core browning (%)	Cavities (%)	Healthy (%)
Air	1.0	61.1 b	0	0	38.8 c
Air plus 1-MCP	1.0	11.1 e	0	0	88.8 a
CA	1.0	50.0 c d	22.2 b	0	50.0 b
CA plus 1-MCP	1.0	55.5 c	44.4 a	0	38.8 c
Air	-0.5	72.2 a	0	0	27.7 d
Air plus 1-MCP	-0.5	44.4 d	0	0	55.5 b
CA	-0.5	44.4 d	0	5.5	55.5 b
CA plus 1-MCP	-0.5	55.5 c	22.2 b	0	44.4 b c

*Means were compared by the Tukey HSD Test at $\alpha=0.05$. Values within a column followed by the same letter are not significantly different.

The incidence of core browning was higher in fruit stored under CA at $1.0\text{ }^{\circ}\text{C}$ than fruit stored under normal air (Table 2). The application of 1-MCP in CA-stored fruit further increased core browning incidence at both temperatures. Similar behavior was observed by HENDGES et al. (2015, 2018), who reported increased browning disorders in 'Alexander Lucas' pear when fruit were treated with 300 nL L^{-1} 1-MCP and stored under CA (2.0 kPa O_2 plus $<0.7\text{ kPa CO}_2$). Saquet, Streif, and Almeida (2017) observed an increase in core browning in 'Rocha' pear treated with 300 nL L^{-1} 1-MCP after 8.5 months storage.

Cavity formation in fruit flesh was only observed in fruit stored under CA without 1-MCP treatment (Table 2). These results suggest that cavity formation in 'Alexander Lucas' pear may not be related to 1-MCP as was the case for internal browning as previously mentioned (Table 2). The cavity formation in this investigation was not a limiting factor to long-term storage. Other pear cultivars such as 'Conference' (SAQUET; STREIF; BANGERTH, 2000, 2003) and 'Rocha' (SAQUET; ALMEIDA, 2017a, b; SAQUET; STREIF; ALMEIDA, 2017) frequently develop cavities during long-term CA storage.

The highest healthy fruit percentage (88.8 %) was found in fruit stored under air with 1-MCP at $1\text{ }^{\circ}\text{C}$ (Table 2). All the other storage treatments showed a maximum of 55.5 % healthy fruit. 'Alexander Lucas' does not appear to tolerate the high stress caused by either the low storage temperature or CA storage combined with 1-MCP

treatment.

Quality attributes of 'Alexander Lucas' pear

After storage, the flesh firmness of 'Alexander Lucas' did not differ among treatments (Table 3). Our results indicate that 'Alexander Lucas' harvested at the maturity stage of this study (firmness 46.2 N , TSS 11.8% and skin color $116.8\text{ }^{\circ}\text{hue}$) may be stored for six months under normal air with 1-MCP treatment at a slightly higher storage temperature without significant quality losses. Thus, storage facilities will receive some energy saving advantages and have fewer maintenance costs for cooling systems.

The titratable acidity was higher in CA-stored fruit than air without 1-MCP at both storage temperatures but did not differ from fruit under air with 1-MCP treatment (Table 3). Fruit stored under air at $-0.5\text{ }^{\circ}\text{C}$ without 1-MCP application had the lowest acidity (Table 3). This result may be related to the higher incidence of browning disorders in these samples, which could negatively affect the fruit acidity. Alternatively, these results could be due to the advanced ripening stage of fruit following storage, as indicated by the low hue angle (Table 3).

The total soluble solids (TSS) content of fruit juice varied from 11.7 to 12.8% but did not statistically differ among storage treatments (Table 3). Even after long-term storage, TSS in pear did not change drastically under different gas combinations which is a common behavior (STREIF; SAQUET, 2006; SAQUET; STREIF; ALMEIDA, 2017).

Table 3. Fruit quality of 'Alexander Lucas' pear stored under various conditions plus a 7 day shelf life at 20 °C.*

Treatment	Temperature (°C)	Firmness (N)	TA (%)	TSS (%)	Color (°hue)	AA ($\mu\text{g g}^{-1}$ FW)
Values at harvest		46.2	4.4	12.1	117.7	-
Air	1.0	32.3 a	1.7 b	12.7 a	104.6 c	28 a
Air plus 1-MCP	1.0	33.3 a	1.7 a b	12.4 a	108.6 b	29 a
CA	1.0	37.2 a	2.1 a	11.7 a	111.9 b	28 a
CA plus 1-MCP	1.0	37.2 a	2.2 a	12.4 a	113.6 a b	31 a
Air	-0.5	36.2 a	1.5 c	12.4 a	111.7 b	31 a
Air plus 1-MCP	-0.5	38.2 a	1.7 a b	12.8 a	114.6 a	35 a
CA	-0.5	36.2 a	2.1 a	11.9 a	114.6 a	27 a
CA plus 1-MCP	-0.5	36.2 a	2.1 a	11.9 a	117.1 a	26 a

*Means were compared with the Tukey HSD Test at $\alpha=0.05$. Values within a column followed by the same letter are not significantly different.

Changes in skin color are shown in Table 3. Fruit stored in air at 1 °C without 1-MCP had a lower ° hue than the other treatments, which indicates these fruit were yellower. Fruit stored at -0.5 °C under CA or air with 1-MCP had the highest hue angle values. These results demonstrate the beneficial effects of storage under low temperatures, CA conditions and 1-MCP application. The effect of CA or 1-MCP treatment on maintaining green skin color during storage in pears is frequently reported (CHIRIBOGA et al., 2011; SAQUET; ALMEIDA, 2017a, b; SAQUET; STREIF; ALMEIDA, 2017). As chlorophyllase is ethylene dependent (LELIÈVRE et al., 1997), the inhibition of ethylene action results in greener skin color in many fruits (ARGENTA; MATTHEIS, 2003; SAQUET; ALMEIDA, 2017a, b).

The ascorbic acid (AA) values varied between 25 and 38 $\mu\text{g g}^{-1}$, but there were no significant differences among the storage treatments (Table 3). Veltman et al. (2000) reported higher losses in ascorbic acid in pears when kept under long-term CA storage. Franck et al. (2003) noticed that browning inducing CA storage conditions in 'Conference' pear resulted in more than 4-fold faster decrease in ascorbic acid concentration. Cocetta, Mignani, and Spinardi (2016) found that treatment of 'Passe Crassane' pear with 400 nL L^{-1} 1-MCP maintained a higher ascorbic acid during air storage at 0 °C and during shelf life. The effect of 1-MCP on pear seems to be similar to apple. 1-MCP at 625 nL L^{-1} , and AVG at 125 $\mu\text{L L}^{-1}$, alone or in combination, reduced ascorbic acid losses in apples after 12 weeks storage at 1 °C (NEUWALD; STREIF, 2010).

Apples treated with AVG plus 1-MCP or treated with only 1-MCP showed the highest postharvest ascorbic acid content (NEUWALD; STREIF, 2010). However, ascorbic acid concentrations could not be associated with the incidence of storage disorders in 'Alexander Lucas' pear in this study.

Practical implications about the storability of 'Alexander Lucas'

This study evaluated the effect of various storage conditions on 'Alexander Lucas' pear. Storage type, temperature and 1-MCP application did not significantly affect the quality attributes, fruit firmness, acidity, TSS, skin color and ascorbic acid.

However, regarding storage disorders, the most acceptable levels of healthy 'Alexander Lucas' fruit were found with 1-MCP application (300 nL L^{-1}) under normal air at 1 °C, which produced 88.8 % of healthy fruit after six months of storage. Storage under slightly higher temperatures could help storage facilities reduce their energy costs and avoid the use of expensive CA storage systems. McCormick, Neuwald, and Streif (2010) reported that treating 'Gala' apple with 1-MCP allowed the storage temperature to increase from 1.5 to 4.5 °C during 5.5 months CA storage, saving 35 % of the energy costs. Furthermore, after storage, a tasting panel slightly preferred 1-MCP-treated apple fruit over untreated fruit. 'Alexander Lucas' pears are not easy to keep under long-term storage, which necessitates further investigations to better understand the storage behavior of this pear cultivar.

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