

Quality and Chemical Changes Associated with Flavor of ‘Camarosa’ Strawberries in Response to a CO₂-enriched Atmosphere

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Abstract. Quality and chemical changes associated with flavor were evaluated in ‘Camarosa’ strawberries (*Fragaria ×ananassa*) that had been kept at 5 °C in air or in air + 20 kPa CO₂ for 3 and 6 days to elucidate possible factors contributing to the loss of flavor during storage. The elevated CO₂ treatment did not affect flesh firmness, total soluble solids, pH, or titratable acidity. In contrast, decreases in color (as indicated by a higher hue angle value) and in concentrations of sucrose, reducing sugars, and citric acid were detected in fruits exposed to elevated CO₂. Fermentative metabolites were present in strawberries stored in air and in higher concentration in those kept in air + 20 kPa CO₂. Also, strawberries kept in air + 20 kPa CO₂ had higher levels of ethyl esters and a major reduction in the level of methyl esters. Thus, clear differences in the aroma profile of strawberries at harvest and after 3 and 6 days of storage at 5 °C in air or air + 20 kPa CO₂ were observed. This change in the volatile aroma profile is probably the primary factor contributing to the loss of strawberry flavor during storage.

The quality of fresh fruits can be defined in terms of factors such as appearance, firmness, color, flavor, and nutritional value. Modified atmospheres (MA) containing 10–20 kPa CO₂ have been applied commercially for many years to reduce decay incidence and to preserve quality attributes of strawberries (Harvey, 1982). However, not all quality characteristics can be preserved to the same extent. Flavor tends to decline before changes in appearance signal the end of acceptable postharvest life (Pelayo et al., 2003).

Sugars and acids together with aroma compounds are the primary constituents determining strawberry (*Fragaria ×ananassa* Dutch.) flavor. Strawberries stored in CO₂-

enriched controlled atmospheres (CAs) (Holcroft and Kader, 1999a) and those stored in MAs (Sanz et al., 1999) had reduced sucrose concentration with a concomitant increase in the levels of glucose and fructose over time. Also, decreased levels of citric and malic acids in ‘Selva’ strawberries stored at 5 °C in air were greater in strawberries stored in CO₂-enriched atmospheres (Holcroft and Kader, 1999a). Thus, pH increases during storage were greater in CO₂-stored strawberries than in those kept in air, and a corresponding change was observed in titratable acidity (TA). Similarly, Fernandez-Trujillo et al. (1999) found that malate concentrations were 22% lower in 20 kPa CO₂- stored than in air-stored fruit of seven strawberry cultivars, but the level of citric acid was unaffected by the CO₂ treatment.

Among aroma compounds, esters are apparently the volatiles most affected by CO₂-enriched atmospheres. The levels of ethyl acetate and ethyl butyrate increased over isopropyl, propyl, and butyl acetates in ‘Chandler’ strawberries at 5 °C in 50 kPa CO₂-

enriched atmospheres (Ke et al., 1994). The authors hypothesized that, under these conditions, the synthesis of ethyl esters predominates over other alkyl esters and the synthesis of other acyl esters predominates over acetates. Larsen and Watkins (1995a) reported an increase in ethyl butyrate and ethyl hexanoate in ‘Pajaro’ strawberries stored at 0 °C in 20 kPa CO₂. Watkins et al. (1999) found that the increase in acetaldehyde, ethanol, and ethyl acetate in strawberries stored at 2 °C in 20 kPa CO₂ was cultivar dependent. However, no information is available about the effect of CA on branched esters, which apparently are important contributors to the aroma of strawberries (Schieberle and Hofmann, 1997).

In addition to flavor components, CO₂-enriched atmospheres slow the softening rate of strawberries (Kader, 1986; Ke et al., 1991) or increase the fruit flesh firmness (Harker et al., 2000). Strawberries stored in CO₂-enriched atmospheres were lighter and less red than air-stored fruit (Gil et al., 1997; Holcroft and Kader, 1999b; Watkins et al., 1999). Strawberries contain 580 to 2100 mg·kg⁻¹ of total phenolic compounds (Kader, 1991), and these compounds play a role in the astringency perception of strawberries (Perkins-Veazie, 1995). The total content of phenolic compounds increased with time in storage at 5 °C but was unaffected by storage atmosphere (in air or in air + 10–20 kPa CO₂) in ‘Selva’ strawberries (Holcroft and Kader, 1999b).

The objectives of the present work were to evaluate changes of the primary flavor components of ‘Camarosa’ strawberries in response to a 20 kPa CO₂-enriched atmospheres and to elucidate the possible factors contributing to the loss of flavor during storage.

Materials and Methods

Experimental procedure. Strawberries (*Fragaria ×ananassa* Dutch. cv. Camarosa) were harvested in ripe condition (red color on more than 95% of the surface) from Watsonville, Calif., transported to the University of California, Davis, on the same day, and stored at 0 °C until the following morning. The strawberries were sorted to eliminate damaged, overripe, and poor-quality fruit and to obtain samples of uniform color. Twenty-five fruits were selected randomly and placed in a 10-L jar as one replicate. Three replicates were used per treatment. The jars were stored at 5 °C and ventilated with a continuous humidified flow of either air or air enriched with 20 kPa CO₂, at a rate of 150 mL·min⁻¹ using flow boards and capillary tubing as flow meters. The composition of the atmosphere was verified daily with an IR gas analyzer (Horiba 2000R; Horiba Instruments, Irvine, Calif.) and maintained within ± 10% of the required partial pressure. The RH was 95% to 100%, and the fruit weight loss was less than 1% in every jar. Three 20-berry samples at harvest and 20 fruits from each jar after 3 and 6 d of storage at 5 °C were randomly selected and analyzed for color

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and firmness. Same fruits were then cut in small pieces, wrapped in cheesecloth, and squeezed with a hand press, and the clear juice used for analysis. Total soluble solids (TSS), pH, and TA were determined using standard procedures and fermentative metabolites and aroma compounds by the analytical methods described below. In addition, three five-berry samples at harvest and five fruits from every jar, randomly selected, were directly frozen in liquid nitrogen and stored at -25°C for subsequent analysis of sugars, organic acids, and phenolics.

Sugars, organic acids, and phenolics. Frozen strawberries were thawed and homogenized in a blender. From the homogenate, three samples of 10 g were used for the analysis of sugars and organic acids, and 10 g for phenolic quantification. Glucose, fructose, sucrose, and citric and malic acids were analyzed according to Pérez et al. (1997) with HPLC (Hewlett-Packard, Palo Alto, Calif.) using a photodiode array detector (DAD model 1040 M) (Hewlett-Packard) in series with a refractive index detector. Organic acids were detected at 210 nm, and sugars were identified by the retention times of the reference compounds and quantified by standard curves. Phenolics were analyzed by using the Folin-Ciocalteu spectrophotometric method (Singleton and Rossi, 1965). The quantification was based on a *p*-coumaric acid standard curve. Results are reported on a fresh weight basis.

Fermentative metabolites. Samples of 5 mL of fresh strawberry juice were placed in crimp-seal 10-mL vials containing 2 g of NaCl, sealed and frozen at -25°C until the analysis of acetaldehyde, ethanol, and ethyl acetate was conducted. The frozen samples were thawed, and the vials were incubated at 30°C for 15 min. After 10 s of agitation, a sample of 1 mL was withdrawn from the headspace and injected into a HP 5890 GC equipped with a flame ionization detector (FID) and analyzed using a 60/80 Carbowax B/5% Carbowax 20 M, $1.8\text{ m} \times 2\text{ mm}$ ID column (Supelco, Bellefonte, Pa.). Injector and detector temperatures were 115 and 200°C , respectively, and the oven temperature started at 80°C , increased to 130°C at $10^{\circ}\text{C}/\text{min}$ and held for 6 min. Fermentative metabolites were identified by the retention times of reference compounds and concentrations calculated by using standard aqueous solutions of every analyte and by preparing the corresponding standard curves under the same conditions as those used for the strawberry samples.

Aroma compounds. Fresh strawberry juice was immediately frozen in liquid N_2 and kept at -25°C until volatiles were analyzed. After 10–20 min, the frozen samples were at room temperature, a NaOH + sodium salt of the ethylenediaminetetraacetic acid (EDTA) solution was added to the juice to obtain a final pH of 6.2–6.5 and a 50 mM concentration of the chelating agent ($\approx 10\%$ of the juice volume). EDTA was added to limit enzymatic and chemical reactions and therefore to prevent the generation of aroma

artifacts. At a pH of 6–7, EDTA sequesters important cations acting as cofactors of enzymes and catalyzers of oxidative reactions. Five-milliliter samples of this juice were placed in a crimp-sealed 16-mL vial containing 2 g of NaCl to facilitate the release of aroma compounds, sealed with a black Viton septum and 20-mm crimp caps, agitated for 30 s, and analyzed by a headspace-solid phase micro-extraction technique (SPME) using an HP 5890 GC apparatus (Hewlett Packard) coupled to a mass spectrometer (MS) (HP 5971 with an electronic upgrade to a model 5972) and a Varian 8200 cx autosampler (Varian, Walnut Creek, Calif.). A $60\text{ m} \times 0.32\text{ mm}$ ID, $1\text{ }\mu\text{m}$ film thickness DB-WAXETR capillary column (J & W Scientific, Folsom, Calif.) and a temperature program (50°C for 1 min, increased to 110°C at $5^{\circ}\text{C}/\text{min}$, then to 180°C at $20^{\circ}\text{C}/\text{min}$, and held for 10 min) were used to separate the analytes. Injector and detector temperatures were 200 and 280°C , respectively. The autosampler was fitted with a $65\text{ }\mu\text{m}$ Carbowax divinyl benzene SPME fiber (Supelco) and programmed for an 11-min cycle: 10-min adsorption time for sampling the headspace and 1 min for desorption in the GC injector. The headspace samplings were done at $25\text{--}30^{\circ}\text{C}$. The identification of aroma compounds was initially accomplished by matching mass spectra with library values. Identities of the major volatiles were confirmed by injecting standard aqueous solutions of each compound directly into the GC-MS and also by trapping the volatiles from the headspace by the SPME fiber under the same condition as those used for the strawberry juice samples. Quantification was carried out by comparing peak areas of analyte to that of 2,6-dimethyl-5-heptenal added at 280 mL^{-1} as the internal standard to the strawberry samples (Ulrich et al., 1995).

Statistical analysis. SAS (version 7.0, SAS Institute, Cary, N.C.) was used to perform analysis of variance (ANOVA) and to obtain LSD (5%) values of each of the main effects. Data presented are the means of three replicates.

Results and Discussion

Quality attributes. Lightness and chroma in ‘Camarosa’ strawberries decreased after 6 d in air storage as has been previously reported for other cultivars (Gil et al., 1997; Holcroft and Kader, 1999b; Pérez et al., 1996; Watkins et al., 1999). However, in contrast with these reports an increase in hue value was observed in the air stored ‘Camarosa’ fruits (Table 1). Because anthocyanins continue to be synthesized in strawberries kept at 5°C in air (Holcroft and Kader, 1999b; Perkins-Veazie, 1995), this unexpected increase in the *h* value could be attributed to the fruit being picked in a more advanced stage of ripeness. The only color variable that was significantly different between elevated CO_2 - and air-stored fruit was hue value after 6 d. In agreement with previous reports (Gil et al., 1997; Holcroft and Kader, 1999b), the hue value of straw-

berries kept in CO_2 -enriched atmospheres was higher, and consequently the fruits were orange red compared with those stored in air for 6 d at 5°C , which were red.

Firmness increased with storage time, but was unaffected by the elevated CO_2 -atmosphere (Table 1). Low temperature increases the flesh firmness of strawberries (Larsen and Watkins, 1995b; Watkins et al., 1999), and an increase in fruit firmness by high levels of CO_2 has been reported (Goto et al., 1996; Larsen and Watkins, 1995a; Smith and Skog, 1992; Ueda and Bai, 1993). The degree of increase in flesh firmness depended on cultivar and days in storage (Watkins et al., 1999). Lack of effect of CO_2 on firmness of ‘Camarosa’ strawberries may be due to cultivar differences or the use of riper fruit than in other studies.

Reduced TSS was observed with time in both air- and CO_2 -stored strawberries, but there was no clear difference between the two storage treatments (Table 1). Similarly, the concentration of sucrose decreased during storage and to a greater extent after 6 d under CO_2 enrichment (Table 2). Sucrose is hydrolyzed during ripening (Woodward, 1972), and this hydrolysis continues during storage (Holcroft and Kader, 1999a; Pelayo et al., 2003). Sucrose decreased by 35% in air- and by 56% in air + CO_2 -stored fruits after 6 d at 5°C . Also, a clear decrease on the level of glucose, fructose and total sugars was observed by the effect of air + 20 kPa CO_2 after 6 d. Thus, sugars were more actively metabolized in CO_2 - than in air-stored strawberries. Under stress conditions, such as those imposed by high levels of CO_2 , the rate of glycolysis increases and it has been hypothesized that this change may be mediated by the regulatory metabolite fructose 2,6-biphosphate (Purvis, 1997). The level of this compound regulates the adaptive pathway of glycolysis and its concentration changes with environmental perturbations. With respect to the proportion of individual sugars, it remained essentially the same in strawberries stored in either air or air + 20 kPa CO_2 (sucrose 4% to 7%, glucose 41% to 44%, and fructose 52% to 53%).

Fruits under elevated CO_2 did not show changes in TA or pH (Table 1) but they had higher losses of citric and total organic acids than strawberries stored in air after 6 d of storage (Table 2). Holcroft and Kader (1999a) reported a greater increase in pH with parallel decreases in TA and in the level of organic acids in ‘Selva’ strawberries stored at 5°C in 10 or 20 kPa CO_2 than in those stored in air. It appears that smaller changes in juice acidity took place in ‘Camarosa’ strawberries harvested in a more ripe condition. Thus, changes in pH and TA were not observed over time, and the reduction in the level of citric acid, the dominant organic acid in strawberries, was small.

An increase in the concentration of total phenolic compounds from 1850 to 2120 $\text{mg}\cdot\text{kg}^{-1}$ was observed after 6 d of storage in air-stored but not in CO_2 -stored fruits (1850–1945 $\text{mg}\cdot\text{kg}^{-1}$). Similarly, Holcroft and Kader

Table 1. L*, C*, and hue angle color measurements, pH, titratable acidity, total soluble solids and firmness of 'Camarosa' strawberries at harvest and after storage for 3 and 6 days in air or air + 20 kPa CO₂ at 5 °C (means ± sd).

Storage		Color			Firmness (N)	TSS (%)	pH	TA (%)
Days	Atmosphere	L*	C*	H (degrees)				
0		34.9 ± 0.5	38.1 ± 0.7	25.3 ± 0.5	3.0 ± 0.1	9.5 ± 0.1	3.67 ± 0.06	0.94 ± 0.05
3	Air	32.5 ± 0.4	38.8 ± 0.5	25.6 ± 0.2	3.3 ± 0.2	9.2 ± 0.3	3.67 ± 0.03	0.91 ± 0.02
	Air + 20 kPa CO ₂	31.7 ± 0.6	36.8 ± 0.6	25.3 ± 1.5	3.2 ± 0.1	9.3 ± 0.1	3.69 ± 0.06	0.92 ± 0.03
6	Air	32.9 ± 1.2	35.3 ± 0.2	28.4 ± 1.5	3.8 ± 0.2	8.3 ± 0.1	3.69 ± 0.05	0.90 ± 0.04
	Air + 20 kPa CO ₂	34.1 ± 0.9	36.2 ± 1.0	30.5 ± 1.1	3.8 ± 0.1	8.5 ± 0.1	3.67 ± 0.06	0.93 ± 0.04
LSD (5%)								
Atmosphere		1.4	1.1	2.0	0.3	0.20	0.09	0.06
Days		1.3	1.6	1.4	0.1	0.23	0.03	0.02

Table 2. Sugars and organic acids of 'Camarosa' strawberries at harvest and after 3 and 6 d of storage at 5 °C in air or air + 20 kPa CO₂ (means ± sd).

Storage		Sugars (mg·g ⁻¹)				Organic acids (mg·g ⁻¹)		
Days	Atmosphere	Sucrose	Glucose	Fructose	Total	Citric acid	Malic acid	Total
0		3.4 ± 0.7	21.7 ± 0.3	27.1 ± 0.7	52.2 ± 0.5	9.0 ± 0.8	2.7 ± 0.3	11.7 ± 0.6
3	Air	2.6 ± 0.1	18.3 ± 0.7	22.2 ± 2.1	43.1 ± 1.0	6.0 ± 0.3	3.1 ± 0.4	9.1 ± 0.3
	Air + 20 kPa CO ₂	2.3 ± 0.2	19.3 ± 0.4	24.3 ± 0.9	46.0 ± 0.5	6.3 ± 0.2	2.0 ± 0.2	8.3 ± 0.2
6	Air	2.2 ± 0.1	20.7 ± 0.8	26.3 ± 0.9	49.2 ± 0.6	9.5 ± 0.7	2.4 ± 1.4	11.9 ± 1.1
	Air + 20 kPa CO ₂	1.5 ± 0.2	16.5 ± 0.4	19.9 ± 0.8	37.9 ± 0.4	7.2 ± 0.6	1.8 ± 0.2	9.0 ± 0.4
LSD (5%)								
Atmosphere		0.6	1.8	3.0	5.2	1.1	1.2	1.6
Days		0.8	5.6	7.6	14.6	1.4	1.4	2.3

(1999b) found in 'Selva' strawberries that the level of these compounds increased with time at 5 °C, but it was unaffected by 10 or 20 kPa CO₂-enriched atmospheres. However, in the same work the authors found that concentrations of some individual phenolic compounds were usually lower in the CO₂-than in the air-stored fruits, suggesting that the rate of phenolic degradation may increase in strawberries subjected to elevated CO₂ atmospheres after 10 d of storage. The mechanism by which CO₂ interacts with phenolic compounds is not clearly understood. It is known that acetaldehyde, a fermentative metabolite that can accumulate during exposure to high levels of CO₂, binds phenolic compounds such as tannins and promotes their polymerization (Es-Safi et al., 1999; Pesis and Ben-Arie, 1986).

Fermentative metabolites. Ethyl acetate was the only fermentative metabolite detected in freshly harvested strawberries (Fig. 1A), but during storage acetaldehyde and ethanol were also detected in both air- and air + CO₂-stored fruits. The concentration of these compounds increased dramatically with time and storage atmosphere, dominating the aroma profiles of 'Camarosa' strawberries during storage (Fig. 2). After only 3 d at 5 °C, they represented 57% and 63% of the total volatiles quantified in the air- and CO₂ stored fruit, respectively. Some symptoms of over-ripeness such as calyx dehydration and external tissue breakdown were detected in fruit stored in air, but not in those kept under air + 20 kPa CO₂. Thus, the accumulation of these volatiles seemed to be an indicator of overripeness in the air-stored fruit and a manifestation of physiological stress in the CO₂-stored fruit. Our results indicate that 'Camarosa' strawberries belong to the category of cultivars accumulating

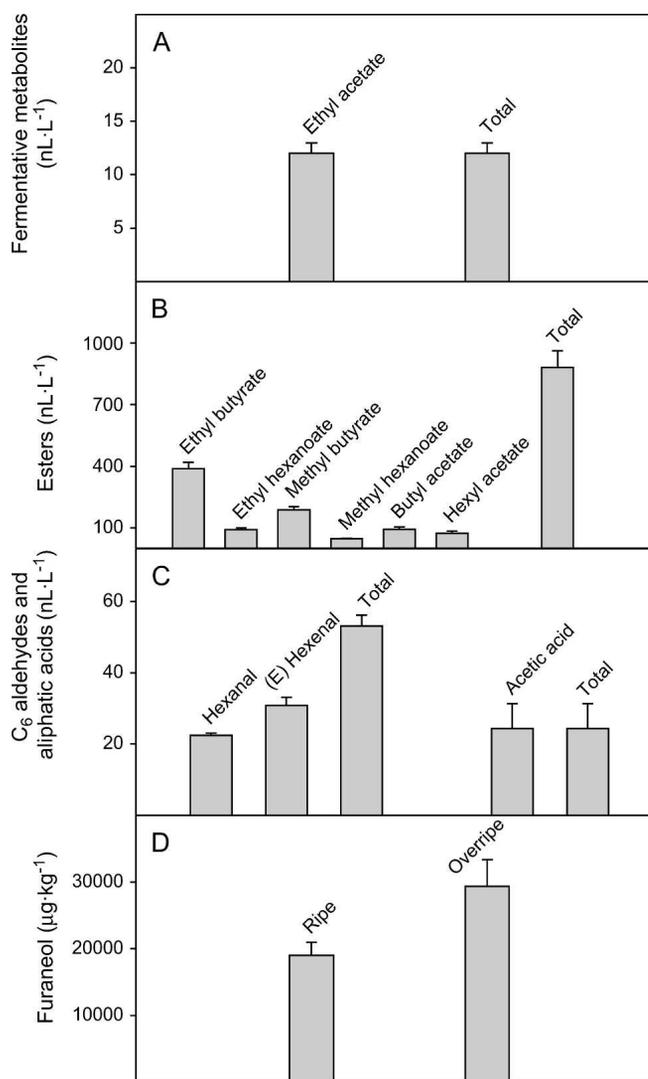


Fig. 1. Aroma composition of 'Camarosa' strawberries harvested in ripe condition, except for furaneol for which overripe fruits were also analyzed (means ± sd).

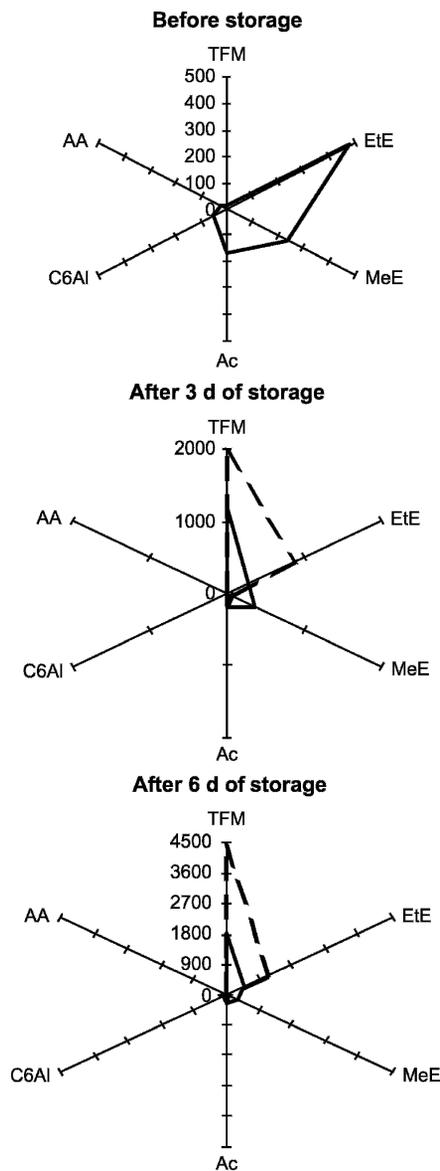


Fig. 2. Aroma profiles of 'Camarosa' strawberries before storage and after 3 and 6 d at 5 °C in air (continuous line) or air + 20 kPa CO₂ (dotted line) (means ± SD) (TFM, total fermentative metabolites; EtE, ethyl esters; MeE, methyl esters; Ac, acetates; C₆Al, C₆ aldehydes; AA, acetic acid, in nL·L⁻¹).

fermentative metabolites in response to elevated CO₂ (Watkins et al., 1999).

Aroma compounds. The most abundant aroma compounds present in ripe 'Camarosa' strawberries were esters and furaneol. Esters comprised 91% of the total aroma compounds (971 nL·L⁻¹) quantified by GC-MS, followed by C₆ aldehydes (5%) and acetic acid (3%) (Fig. 1B and C). Furaneol, which was quantified by HPLC because of its thermoinstability, was present at 19,000 µg·kg⁻¹ in fruit harvested ripe and at 29,000 µg·kg⁻¹ in fruit harvested in an overripe condition (Fig. 1D). These data confirm previous reports indicating that the synthesis of furaneol, and other furanones, continues after strawberries have attained full ripeness on the plant (Pérez et al., 1996).

Table 3. Levels (nM) of alkyl and acyl fractions required to synthesize the observed amount of esters in 'Camarosa' strawberries stored at 5 °C in air or air + 20 kPa CO₂.

Days	Storage	Alkyl and acyl straight chain fractions (nM)						Acyl branched chain fractions (nM)	
		C ₁ (methyl)	C ₂ (ethyl)	C ₂ (acetate)	C ₄ (butyl, butyrate)	C ₆ (hexyl, hexanoate)	C ₄ (methyl-2 propanoate)	C ₅ (methyl-2- butyrate)	
0		2.0	3.5	1.1	5.3	1.3	0	0	
3	Air	2.9	1.9	1.3	4.7	1.5	0	0	
	Air + 20 kPa CO ₂	0.6	6.1	1.0	4.7	2.6	0.1	0.3	
6	Air	2.5	3.5	1.8	5.5	2.4	0.0	0.07	
	Air + 20 kPa CO ₂	0.2	7.9	0.9	5.5	2.7	0.2	0.6	

Ethyl esters, which were already abundant in strawberries at the beginning of storage (Fig. 1B), were also present in air-stored fruits after 3 and 6 d, but a remarkable increase in the level of these compounds was observed in the strawberries stored under air + 20 kPa CO₂ (Fig. 2). Levels of methyl esters and butyl and hexyl acetates tended to increase in air-stored fruit (from 236 and 166 to 314 and 264 nL·L⁻¹, respectively, after 6 d of storage), but to decrease in strawberries stored under air + 20 kPa CO₂ (28 and 132 nL·L⁻¹, respectively, after 6 d of storage). These results support the findings of Ke et al. (1994), who reported the predominant synthesis of ethyl esters over methyl esters in CO₂-stored fruit, as well as the reduction in the level of acetates. Notably, the branched esters, ethyl 2-methyl propanoate and ethyl 2-methyl butyrate, considered important aroma compounds of strawberries (Schieberle, 1994; Schieberle and Hofmann, 1997), were not present before storage. In air-stored fruits, only ethyl 2-methyl butyrate was detected in trace amounts while both ethyl 2-methyl propanoate and ethyl 2-methyl butyrate were present in the air + CO₂-stored fruits in more than trace amounts (32 and 88 nL·L⁻¹, respectively, after 6 d of storage). Because valine and isoleucine are precursors of these branched esters (Drawert, 1975; Lindsay, 1996; Tressl et al., 1975), their presence may be an indication of enhanced protein degradation in the overripe, air-stored fruits and in those subjected to the physiological stress incited by elevated CO₂.

The concentration of total aroma compounds increased throughout the storage period in air and to a greater extent in CO₂-stored fruit mainly due to the enhanced production of fermentative metabolites and ethyl esters (Fig. 2).

In the end, although quantification of aroma compounds with SPME using only one internal standard is not very accurate, in this study, that method was able to show differences between treatments for many peaks.

To analyze the contribution of metabolic pathways to the synthesis of esters other than ethyl acetate during the storage of 'Camarosa' fruit, the average concentration of the three replicates of esters (in nL·L⁻¹) of each sample were converted into nanomoles. Considering that every ester molecule comes from an alcohol molecule (alkyl fraction) and an aliphatic acid molecule (acyl fraction), the number of nanomoles of alkyl and acyl

fractions required to synthesize the observed amount of every ester was calculated. Finally, fractions of the same carbon number coming from the same metabolic pathway were added (Table 3).

The amount of methanol, an alcohol coming from the hydrolysis of pectins, used for the synthesis of methyl esters increased in air and decreased in air + CO₂-stored fruit (Table 3). However, the concentration of this alcohol, measured by GC in the juice of strawberries (data not shown), remained the same throughout the 6 d in both storage atmospheres. Thus, apparently it was not the availability of methanol, but the higher levels of ethanol present in the CO₂-stressed fruit, that resulted in the preferential synthesis of ethyl esters over methyl esters in the CO₂-stored fruit.

The nanomoles of fractions C₄ (butyl and butyrate), reported to come from β-oxidation and synthesis of fatty acids (Sanz et al., 1997), were the same in strawberries kept under both storage atmospheres. However, the C₆ fractions (hexyl and hexanoate) increased after 6 d in air and to a higher level after 3 and 6 d in air + 20 kPa CO₂. Because C₆ fractions can be generated by both β-oxidation and the lipoxygenase (LOX) pathways, the observed increase in C₆ fractions may be due to an alteration of LOX activity by over-ripeness and by the exposure of the fruit to elevated CO₂.

Finally, the nanomoles of C₄ and C₅ acyl branched chains indicated a greater contribution of the amino acids valine and isoleucine, respectively, to the synthesis of esters in CO₂- than in air-stored fruits.

Conclusions

The accumulation of fermentative metabolites in both air- and air + 20 kPa CO₂-stored 'Camarosa' strawberries and the shift in the synthesis of methyl to ethyl esters in fruit exposed to air + 20 kPa CO₂ created a new profile of aroma compounds that may affect fruit aroma perception. This change in the volatile aroma profile is probably the primary factor contributing to the loss of strawberry flavor during storage. Simultaneous changes in other nonvolatile flavor components, including sugars and citric acid, may also contribute to the loss of strawberry flavor during storage. Additionally, changes in phenolic compounds and the increase in firmness by the effect of low temperature may have an

influence on the release of volatiles and therefore, on the flavor perception of strawberries. Sensory evaluation is required to confirm these results.

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