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Ali BATU

A. K. THOMPSON

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Ali BATU
Gaziosmanpaşa University, Agriculture Faculty, Food Engineering Department, 60100, Tokat-TURKEY

A. K. THOMPSON
Postharvest Technology Department, Silsoe College, Cranfield University, Silsoe, Bedford, MK45 4DT ENGLAND

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Abstract: Tomato (*Lycopersicon esculentum* Mill.) fruits (Cv Criterium) were harvested at the mature green stage and stored at 13°C in controlled atmosphere (CA) conditions for 1, 3 and 5 days. The CA conditions were 5, 10, 20, 40 and 60% CO_2 all with 5.5%O_2 plus air as control. The tomatoes were then stored at 20°C in air until they were fully ripe. At that time colour, firmness, titratable acidity, total soluble solids (TSS) and days to ripening were measured.

Fruit exposed to CO_2 for 5 days subsequently ripened more slowly than those exposed only one day. The controlled atmosphere stored fruit took 11 to 12 days to ripen compared to only 8 days for fruits stored in air. The ripening time of the fruits exposed to 60% CO_2 for only one day was 18 days without CO_2 injury, whereas it was 14 and 15 days for fruits exposed 20% CO_2 for 5 days or fruits exposed to 40% CO_2 either for 1 or 3 days. Fruits treated with 5, 10, and 20% CO_2 did not show any harmful effects on colour development and fruit softening. Treatments with 40 and 60% CO_2 for 1 day also did not cause any harmful effects on colour development while there was only a slightly inhibition of colour development after 3 days and completely inhibited it by 5 days exposure. There was also considerable CO_2 injury on tomatoes exposed to to 40% to 60% CO_2 for 5 days. Fruits exposed to 40% and 60% CO_2 for 1 to 5 days were found to be softer than the fruits from other treatments. It was observed that titratable acidity and TSS values of fruits stored in CA for 1 and 3 days were similar to each others. But both acidity and TSS values of 40 and 60% CO_2 treated tomatoes for 5 days were found to be lower than the 5, 10 and 20% CO_2 exposed fruits.

Kısa Süreli Yüksek Karbondioksit Uygulamasının Domates Olgunlaşması Üzerine Etkisi

Özet: Criterium çeşidi domates meyveleri yeşil olum döneminde hasadı yapılarak 13°C de kontrollü atmosfer (KA) koşullarında 1, 3 ve 5 gün süre tutularak bu süre zarfında %5, 10, 20, 40 ve 60 oranında CO_2 (hepsi % 5.5 O_2 ile) uygulanmıştır. Ayrıca diğer bir muameleyede 'kontrol' olarak hava verilmiştir. Daha sonra domatesler kırmızı oluma ulaşıncaya kadar 20°C de tutulmuştur. Kırmızı oluma ulaştıklarında renk, sertlik, asitlik, suda çözünür toplam katı madde (SÇKM) ve olgunlaşma süresi belirlenmiştir.

5 gün süre ile CO_2 uygulanmış domatesler 1 ve 3 gün süre ile uygulananlardan daha geç olgunlaşmışlardır. KA koşullarında CO_2 uygulanarak olgunlaştırılan domatesler 11-12 gün sonra olgunlaşırken kontrol amacı ile normal hava ortamında tutulan domatesler 11-12 gün sonra olgunlaşmıştır. 1 gün süre ile %60 CO_2 uygulanan domatesler CO_2 zararlanması görülmeksizin 18 gün sonra kırmızı oluma ulaşırken hem 5 gün %20 CO_2 uygulanan hemde 1 veya 3 gün %40 CO_2 uygulanan domatesler ise 14-15 gün sonunda kırmızı oluma ulaşmıştır. Karbondioksidin %5, 10 ve 20 oranlarında uygulanmasının domateslerde renk olgunlaşması üzerine herhangi bir zararlı etki yapmamıştır. 1 gün %40 ve %60 CO_2 uygulamasının renk üzerinde herhangi bir olumsuz etkisi olmazken aynı oranda CO_2 3 gün uygulaması durumunda domateslerin renklenmesinde kısmen gecikme olurken uygulama süresinin 5 güne çıkması ile renklenme tamamen durmuştur. 5 gün %40 ve %60 CO_2 uygulanan domateslerin hepsinde önemli derecede CO_2 zararlanmasının olduğu belirlenmiştir. Bu meyvelerin diğerlerine göre oldukça yumuşak oldukları saptanmıştır. 1 ve 3 gün KA de depolanmış meyvelerin asitlik ve SÇKM değerleri yaklaşık aynı olurken 5 gün %40 ve %60 CO_2 uygulanmış domateslerin asitlik ve SÇKM değerleri %5, 10 ve 20 CO_2 uygulanan meyvelerinkinden daha düşük oldukları belirlenmiştir.

Introduction

Losses often occurred from excessive deterioration during holding and marketing of tomatoes. This problem is especially acute with tomatoes harvested when at the breaker or more advanced stages of ripeness. Atmospheres containing elevated levels of CO_2 are known to inhibit fruit ripening. The application of CO_2 to delay ripening of tomatoes could be performed easily (1; 2) but several reports have indicated that tomatoes are susceptible to CO_2 injury (3). If the level of CO_2 in the storage is increased this will increase its levels within the crops tissue. Physiological disorders in fruit associated with excess CO_2 levels may be associated with this disruption of the respiratory pathway leading to an accumulation in the crop cells of alcohol and acetaldehyde (4).

Controlled atmosphere storage (CAS) is usually successful in controlling physiological disorders and in maintaining good appearance and suitable acid and sugar levels of tomatoes (5). Since O_2 and CO_2 are important components of the respiratory process, it is usually assumed that the beneficial and detrimental effects observed with different gas compositions are related to aerobic and anaerobic oxidations. The evidence for beneficial results from CAS is clear enought to understand, causing the decreasing O_2 and increasing CO_2 concentration in storage environment. Those O_2 and CO_2 concentrations ranging from 3-9% for O_2 and from 2-12% for CO_2 were reported for apples (6). High CO_2 delayed the onset of the climacteric rise in tomatoes, and therefore postponed the ripening of tomatoes. The effects of CO_2 in extending the storage life of crops appears to reduce respiration of the crop. Suppression of the O_2 uptake rate during high CO_2 exposure, accompanied with a decrease of ethylene evaluation, was reported in ripening tomatoes (7). They also reported that under the 60% CO_2, O_2 uptake rates of ripening tomatoes at pink and red stages declined and reached about 12-13 ml/kgh which was equal to that at the mature green stage. The idea of respiratory depression by CO_2 has been supported by the factors that CO_2 could have a strong controlling effect on mitochondrial activity. Additionally, higher concentrations of CO_2, especially above 40%, inhibited the NAD-cytochrome c oxidise (7) and high CO_2 also inhibits breakdown of pectic substance so that a firmer texture is retained for a longer period (8). Another way of application of CO_2 is pre-storage high CO_2 treatment and it has been tested on some apple varieties in USA and Europe in order to reduce the ripening rate using less CO_2 in the short term (9). Preliminary tests at several experiment stations, indicated that CO_2 pre-treatments could reduce 'McIntosh' softening in CAS. It was reported that increasing the length of pre-treatment with 12% CO_2 to as much as 6 weeks, slightly increased its effectiveness in delaying softening, but external CO_2 injury was also increased as treatment time increased (10). In the present study, therefore, the effects of high CO_2 exposure in a short time on ripening period and colour development of mature green tomatoes were determined. Additionally, deterioration and softening of those tomatoes was evaluated.

Material and Methods

Tomatoes (cv 'Criterium') were harvested at the mature green stage of maturity and sorted for uniformity of size and colour. Only undamaged fruits, free of disease, were selected for experiment. Two replications of ten fruits were placed into 3 litre jars, and exposed to ambient, 5, 10, 20, 40 and 60% CO_2 in CA conditions at 13°C for, 1, 3 and 5 days. Oxygen levels were maintained at 5.5(± 0.5)%. The controlled atmospheres were obtained by mixing CO_2 and O_2. All gas mixtures were analysed using an Oxysat 2 gas analyser type 770 produced by David Bishop in UK.

Skin colour values were measured using a Minolta Chromometer Model CR 200 and average readings at three pre-determined points on the circumference of the fruits were recorded. The instrument was calibrated against standard while colour Plate (Y=93.9, x=0.313, y=0.321) (11).

A destructive deformation test was used to evaluate fruit firmness by applying a constant 50 N force using with an Instron Universal Testing Machine, model 1122. In the firmness measurements, a 6 mm diameter round stainless stell probe with a flat end, with a chart speed of 20 mm $minute^{-1}$. The force (N) which was required to penetrate through the skin to the tomato flesh and deformation (mm) values, during that penetration, were recorded (12). Firmness (N mm^{-1}) was defined as the average slope of the force/deformation curve (13).

Titratable acidity wass determined by titrating juice to pH 8.1 with 0.1 N NaOH using a Jenway Digital pH meter (model 3020). Total soluble solids (TSS) content of fruits were determined using a banch top Atago digital refroctometer model PRI (14).

Results and Discussion

Maturation time of tomatoes were expanded by CO_2 exposure. For one day exposure, extension of maturation time was 3 days longer than untreated fruits for 5, 10 and 20% CO_2 treatments, whereas it was 6 and 9 days longer for 40% and 60% CO_2 treatments respectively. Maturation time was increased to 5, 10 and 20% CO_2 treatments with inreased exposure time (Figure 1). The tomato fruits which were treated with 60% CO_2 for 3 days and 40 to 60% CO_2 for 5 days were not ripe even after 18 days by keeping at 20°C in air, and their maturation was significantly P=0.01 longer than the others. Furthermore, unacceptable incidence of high CO_2 injury was found in those fruits.

Short term high CO_2 treatment to mature green tomatoes had beneficial effects on the retardation of fruit ripening. Red colour development of tomatoes was delayed by increasing both CO_2 concentrations and exposure time. Colour development of tomatoes was inhibited while exposed to all levels of CO_2 (Figures 2).

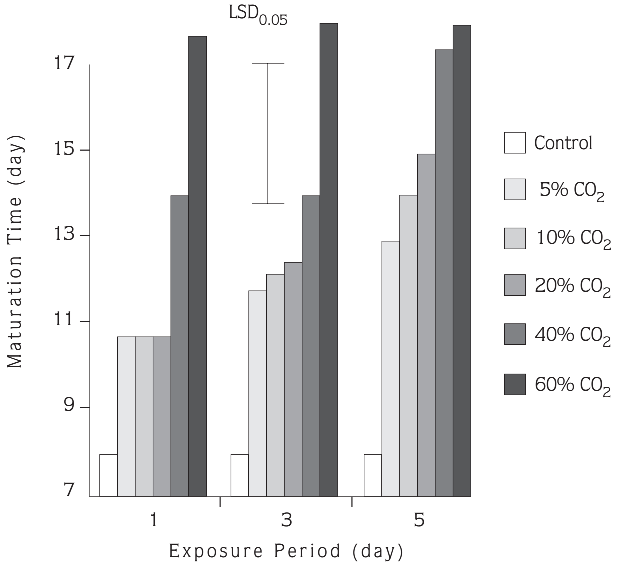

Figure 1. Time taken for tomatoes to be fully ripe when fruits were fully ripe after being exposed various CO_2 levels all with 5.5% O_2 for 1,3 and 5 days.

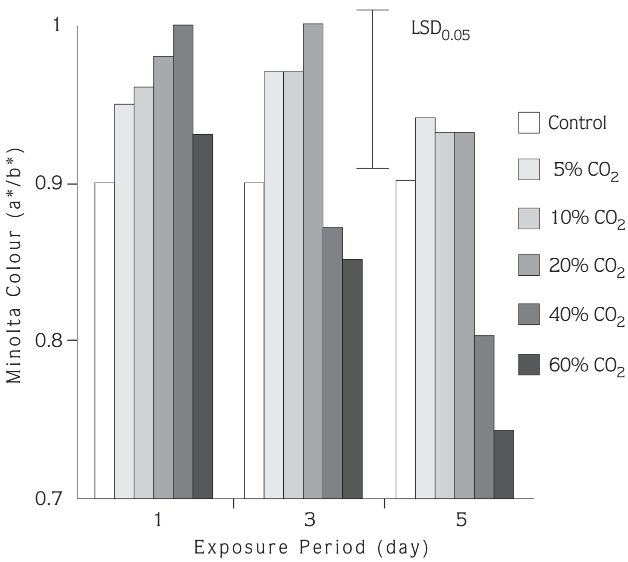

Figure 2. Minolta colour values when fruits were fully ripe after being exposed various CO_2 levels all with 5.5% O_2 for 1,3 and 5 days.

There was a reduction in Minolta a*/b* values of tomatoes exposed to 40 and 60% CO_2 for 5 days, possibly due to their exposure to high CO_2 and continued synthesis of carotenoids, but not lycopene (15). After transfer to air, colour development of fruits was advanced in all treatments except the fruits exposed to 40 and 60% CO_2 for 3 for 5 days. Colour values of those tomatoes were significantly (P=0.01) lower than the other treatments. This was could be due to higher acetaldehyde and ethanol accumulation in the fruit tissues. It was reported that strawberries treated with 50% CO_2 for 8 days at 5°C resulted in highest accumulation of acetaldehyde, and ethanol while 20% CO_2 treatment only slightly increased concentration of the anaerobic volatiles (16). Exposure of fruits to more than 40% CO_2 for 3 and 5 days reduced the number of saleable fruits. The severity of injury to fruits by CO_2 increased with increasing concentration of CO_2 and duration of exposure. These observations are in agreement with Ke *et al.*, (17). Mould growth, water soaked areas and uneven pigmentation were the primary symptoms of injury observed, as mentioned by Buescher (18). Fruits exposed to 5, 10 and 20% CO_2 for even 5 days were not apparently injured.

High CO_2 delayed onset of the climacteris rise in tomatoes, and therefore postponed the ripening of fruits. Tomatoes stored at high CO_2 atmosphere over the 40% levels for 1 or 3 days had no obvious benefits, although increasing CO_2 up to 40% for 1 day exposure was found to be beneficial for fruit ripening, higher than 40% CO_2 level caused unacceptable fruit texture (Figure 3). Keeping the fruits in high CO_2 atmosphere for 5 days caused significant CO_2 injury. When tomatoes were kept in 5, 10 and 20% CO_2 atmospheres for 1 to 5 days and then stored in normal ripening room at 20°C for 18 days, the levels of CO_2 concentrations did not significantly P=0.01 affect the fruit firmness. Porritt and Meheriuk (19) reported that 20-35% CO_2 for 15 days reduced softening of Newton apples at 0°C and CO_2 injury was not observed during that storage period. It was also reported that 'The Golden Delicious' apples have been successfully treated with CO_2 immediately after harvest and placed in CA storage and those fruits were firmer and had a longer storage life than the untreated fruits (20). Lau *et al.*, (20) also reported that treatments of 10 to 20% CO_2 for 10-14 days, reduced the softening of 'Golden Delicious' apples but caused CO_2 injury. Increasing the length of pre-treatment to 12% CO_2 for as long as 6 weeks, slightly increased its effectiveness in delaying softening, but CO_2 injury also increased when treatment time increased (10). It was also reported by Bramlage, (10) that increasing the CO_2 concentration during treatment to 10, 15 or 20% increased the firmness of the fruit after storage. However, the firmness of the 20% CO_2 pre-treatment for 2 weeks injured 60% of the fruits, while a 10% treatment produced no injury but produced a small delay in softening. CO_2 injury occurs in tomatoes if mature-green fruits are subjected to levels above 2% or if particularly ripe fruits are exposed to levels above 5% for more than 7 days at 20°C or 10 days at 12.5°C (4). The injury due to high concentration of CO_2 appears in various forms. Surface blemishes, increased softening, and uneven ripening after removal from the elevated CO_2 atmosphere were among the symptoms related to CO_2 injury (3). The common CO_2 injury is brown heart on apples. High CO_2 at higher temperatures increases the amount and severity of brown heart. The supply of O_2 as well as the amount of CO_2 in the tissue appears to influence the incidence of brown heart (21).

Significant differences were not found in acidity levels between the treatments of 1 day exposure although acidity values of fruits were decreased when CO_2 concentration was applied higher than 10% (Figure 4). It was also very similar for 3 days exposure, except acidity values of the fruits in 60% CO_2. It was significantly lower than the acidity values of other treatments. Acidity value of the fruit was increased up to 20% CO_2 concentration but it was decreased when the applied CO_2 concentration increased higher than 20%. Lau *et al.,* (20) reported that a pre-storage 10 days exposure of 'Golden Delicious' apples to CO_2 levels of about 20% delayed loss of titratable acidity during subsequent CA storage. Changes in titratable acidity were very similar to the TSS values during storage period (22).

Couvey and Olsen also (22) reported that acidity was slightly affected by CO_2. Bramlage (10) reported that CO_2 pretreatment did not significantly influence soluble solids and titratable acidity quality factors of apple fruits. Acidity is related to maturation levels of tomatoes. Maturation also directly correlated with ethylene levels in the storage environment (23). Respiration suppression, observed in ripening climateric fruits, might be mainly involved in the effects of CO_2 via its suppression of ethylene production and action rather than a direct effect of CO_2 on respiratory metabolism (7). Experiments carried out showed that treatmends displayed decreasing acidity. This observation is in agreement with Hall (24) and Hobson and Davies (25). Titratable acidity was affected by CO_2 treatments. This is also in agreement with Parsons *et al.,* (26) and Goodenough and Thomas (27). It would seem logical that fruits stored in higher CO_2 environments would be more acid because CO_2 is an acid gas which should be dissolved in the cell sap in proportion to its concentration in the surrounding atmosphere, but results from this current work disagreed with this. There is little information on acidity changes during CAS of tomatoes. Parsons *et al.,* (26) found that titratable acidity increased with increasing CO_2 concentration from zero to 5% CO_2 during controlled atmosphere storage and there was also no information about comparison of control tomatoes at that time. It is difficult to find information about the relationship between acidity changes and CO_2 concentration during controlled atmosphere storage but there are conflicting reports on some other fruits. In a recent review paper, Riquelme *et al.,* (28) reported that storage of strawberries in low O_2 and high CO_2 concentrations does not affect titratable acidity. It was also reported that in 60% CO_2 there was no affect on titratable acidity in Valencia oranges, while storage of lemons under high CO_2 leads to accumulation of organic acids (29). Salunkhe and

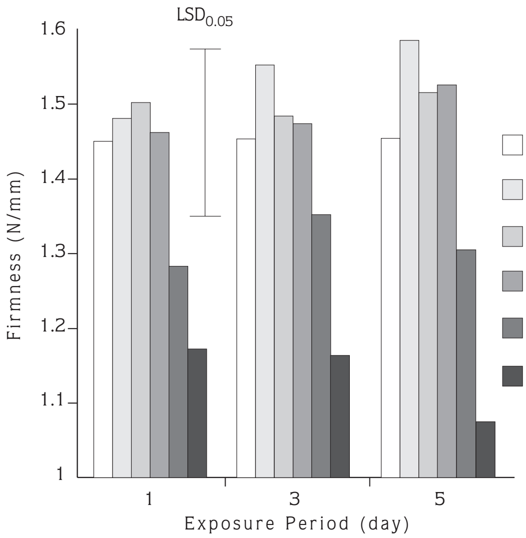

Figure 3. Firmness values of tomatoes exposed various CO_2 levels all with 5.5% O_2 for 1,3 and 5 days.

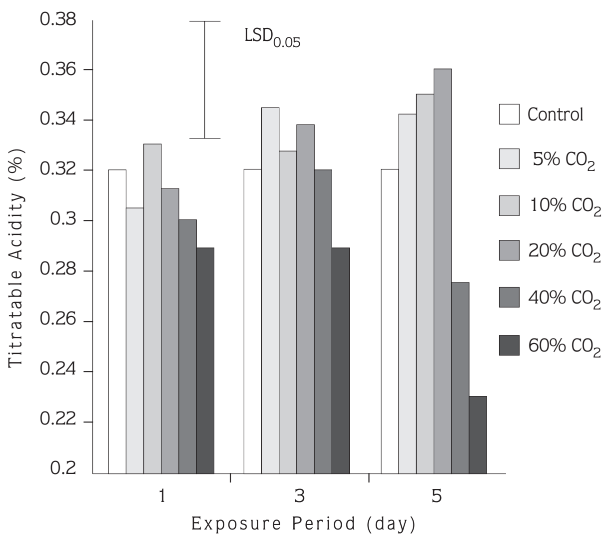

Figure 4. Titratable acidity values of tomatoes exposed various CO_2 levels all with 5.5% O_2 for 1,3.

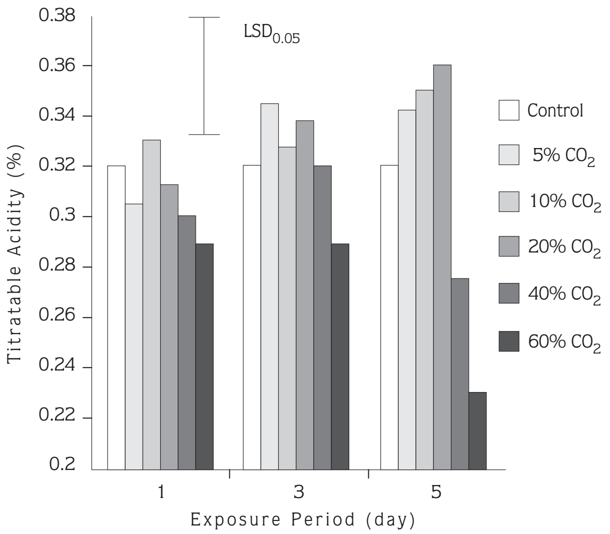

Figure 5. Total soluble solids values of tomatoes exposed various CO_2 levels all with 5.5% O_2 for 1,3.

Wu (30) reported that the titratable acidity of green beans increased during air storage, but decreased slightly in controlled atmosphere storage. They also reported that the titratable acidity of broccoli and asparagus decreased progressively with increasing concentration of CO_2 in the controlled atmosphere storage. If the CO_2 level is higher than 9% in the storage environment these CO_2 levels cause a decrease in the titratable acidity levels of the tomatoes (5; 14). It was more effective on ripe fruits.

The relation between exposure time and treatments for TSS levels was very similar to titratable acidity. TSS values of tomatoes exposed to ambient, 5, 10, or 20% CO_2 did not differ significantly (P=0.01) between the 3 and 5 days exposure period (Figure 5). TSS levels of the fruits exposed to over 40% CO_2 tended to decrease with the increasing exposure period and it caused considerably lower TSS values than the treatments lower than 40% CO_2 at both 3 and 5 days exposure. It was reported that 10 to 20% CO_2 treatment for 2 weeks had no effect on soluble solids or acid levels of McIntosh apples (19). Couvey and Olsen (21) reported that TSS was unaffected by CO_2 level or time in storage. Generally TSS values of tomatoes stored in higher CO_2 environments are lower compared to those stored in lower CO_2, because the ripening rate is inhibited by high CO_2 concentration. Therefore production of sugars, organic acids and other substances which contributes to TSS values of tomatoes, were inhibited. Hobson and Davies (25) reported that higher CO_2 prevented the production of sugars, organic acids and other chemicals which are the main substance of TSS. Herner (4) also reported that the accumulation of reducing sugars in potato tubers is prevented by concentration of 5% CO_2 or more. He also reported that the conversion of sugar in peas and sweet corn can be inhibited by high CO_2 levels. but there was also the potential for considerable CO_2 injury, both internal and external from the treatment.

In conclusion, it is possible to extend the shelf life of tomatos for 6 days, by 1 day exposure as compared with a shelf life of control fruits. The most suitable treatment to produce the best colour with acceptable texture and firmness improvement, was observed with the treatment of 40% CO_2 for 1 or 3 days exposure. Those tomatoes ripen after 14 days. This effect of CO_2 would be applicable economically in commercial storage with using less amount of CO_2 by exposing short term in comparison with continuos CA storage particularly when it would like to store for two weeks time.

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