Cold storage temperature and 1-MCP postharvest treatment on 'Starking Delicious' apple quality

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Abstract

It was investigated the effect of 1-methylcyclopropene (1-MCP) application, cold storage temperature (0, 2 or 5 °C) and 7 d shelf life on fruit quality characteristics, superficial scald incidence, total phenolic content (TPC) and total antioxidant activity (TAA) of 'Starking Delicious' apples. Apples treated with 1-MCP and stored at 0 °C had better flesh firmness (FF) and organoleptic quality and retained their external appearance (reduced scald) compared to untreated with 1-MCP fruit that stored at 0 °C throughout the storage period and after 7 d shelf life. After 2.5 months of cold storage, apples treated with 1-MCP and stored at 2 or 5 °C adequately retained FF compared to untreated with 1-MCP and stored at 0 °C even after 7 d shelf life, while all other quality parameters remained similar to apples treated with 1-MCP and stored at 0 °C. The application of 1-MCP, storage at 0, 2 or 5 °C for extended period of time plus shelf life had minor effects on TPC and TAA of 'Starking Delicious' apples.

Keywords — *Malus x domestica, 1-methylcyclopropene, storage temperature, shelf life, nutritional quality.*

I. INTRODUCTION

Apple fruit acceptance at consumer level is determined by external fruit quality characteristics such as size, shape, color and absence of defects and of internal quality characteristics such as texture, the presence of or the potential to develop acceptable varietal flavor and nutritional value [1]. Apple fruit ripening includes many physiological and biochemical changes and some of them contribute to fruit acceptability such as conversion of starch to sugars, the characteristic color development, loss of acidity, formation of cuticular waxes and synthesis of aromatic compounds ([2], [3]). Other changes such as apple fruit softening is generally considered an undesirable ripening process that is negatively perceived by consumers ([4], [5]).

Optimal storage conditions for apples aim to slow down degradation and maintain freshness, texture, flavor and nutritional value for extended periods close to levels at harvest time, in order to satisfy market and the consumer demands. Apples are commonly stored at 0 °C in air for up to 3-4 months or in an ultra-low oxygen controlled atmosphere (CA) at 0 °C for up to 12 months, depending on cultivar [3]. Postharvest treatment of apple fruits with 1-MCP is widespread as it is well documented that it retains fruit quality even after prolonged cold storage in an acceptable level for the consumer. In addition, 1-MCP could be an alternative to CA which requires significant installation costs.

1-MCP inhibits ethylene perception of plant tissues by blocking the ethylene binding sites [6]. Thus, 1-MCP is a postharvest tool to extend storage and shelf life of climacteric fruit [3]. Several studies on different apple cultivars have shown that postharvest treatment of apples with 1-MCP and thereafter storage at 0 °C prevented fruit softening even after shelf-life for 7 to 14 d at 20 °C, suppressed the incidence of superficial scald, maintained titratable acidity, but reduced aroma volatiles production, inhibited the loss of greenness of the background or ground color of the fruit skin, inhibited ethylene production and action, reduced respiration rate and decay, and gave variable on soluble solids content ([7], [8], [9], [10], [11], [12], [13], [14], [15]).

Flavor is a composite of taste and odor, and volatile production can be greatly affected by ethylene. Therefore, decreased and/or altered volatile production in 1-MCP treated fruit compared to untreated fruit may impact consumer acceptability ([9], [13], [15]). For many climacteric fruit, success of 1-MCP application should be based on delaying rather than preventing ripening, in order to provide a highly acceptable product for the consumer. Therefore, for apple fruits the inhibition of ethylene-mediated responses should be partially reversible so that desirable fruit ripening should proceed, resulting in a partially-ripened apple acceptable to the consumer [15]. This partial ripening could be achieved with higher than 0 °C storage temperatures.

Recently many authors have focused on the effect of various postharvest technologies on apple phenolic compounds and antioxidants because of their significant benefit on human health, contributing to the prevention of several diseases. Reports on the

effect of 1-MCP application on the content of phenolic compounds and antioxidants and their maintenance after prolonged storage are limited and the results vary according to the cultivar, pre-harvest conditions, maturity, storage conditions and duration, shelf life, evaluated at the peel, flesh or whole fruit, and the analytical method used ([16], [17], [18], [19], [20]).

The aim of this study was to investigate whether higher than 0 ^{o}C storage temperature might interfere with the fruit's ability to ripen normally after storage and/or develop storage disorder symptoms. Storing fruit at higher temperatures after 1-MCP application could provide significant energy cost savings if adequate post-storage apple quality is maintained. For this purpose, apple fruit quality attributes and nutritional characteristics, i.e. TPC and TAA, were evaluated during cold storage in air at different storage temperatures after treatment or not with 1-MCP.

II. MATERIALS AND METHODS

A. Fruit sampling and 1-MCP treatments

Apples cv. 'Starking Delicious' were harvested at commercial maturity from 10-year-old mature apple trees in an orchard at Zagora, central Greece, at an elevation of 700 m. Apples free of defects were randomly packed in plastic boxes (28 boxes and 90 fruit per box) by a commercial sorter and stored at the storage facilities of the Agricultural Cooperative of Zagora. Within a day of harvest, 18 boxes were treated with 625 nL L^{-1} 1-MCP at 10 °C and immediately thereafter six boxes were stored at 0 °C in air (1-MCP 0), six boxes were stored at 2 °C in air (1-MCP 2) and six boxes were stored at 5 °C in air (1-MCP 5). From the untreated with 1-MCP fruit six boxes were stored at 0 °C in air (no 1-MCP 0) while four boxes of fruit were used to evaluate initial fruit quality (one box per treatment).

B. Fruit quality evaluation

Fruit quality was evaluated just after harvest and after 2.5 (75 d after harvest-DAH), 5 (150 DAH) and 7.5 months (225 DAH) cold storage, immediately after cold storage and after 7 d shelf life (20-22 °C, 50-60% RH) in three replications of 10-fruit each per treatment except of scald that three replications of 30-fruit per treatment were used. Quality evaluation included skin color, flesh firmness (FF), soluble solids content (SSC), titratable acidity (TA) and superficial scald.

Skin color was measured by Minolta chroma meter (Model CR-400, Minolta Ltd, Osaka, Japan), which provided CIE L*, a* and b* values. These values were used to calculate hue angle (Hue) [hue^{o}=arctan(b/a)] and Chroma (C*) [C*=$(a^2+b^2)^{1/2}$] [21].

FF was measured on the equator and two opposite sides of each fruit after peel removal with a digital penetrometer (model 53205, Turoni Srl, Forli, Italy) equipped with 11-mm plunger. Data were calculated as the mean of the two measurements from each fruit of the 10-fruit per replication and expressed in Newtons (N).

SSC, expressed as %, was measured to the juice extracted from one slice of each fruit of the 10-fruit per replication by using an Atago Refractometer (Model PAL-1, Atago, Tokyo, Japan). TA was measured to the juice extracted from the ten slices of the 10-fruit replication after titration with 0.1 N NaOH to pH 8.2 and expressed in g malic acid per 100 mL apple juice.

C. Superficial scald evaluation

Three 30-fruit replications per treatment were used for scald evaluation per time period. Scald incidence was evaluated on a 0 to 3 scale with 0, no scald; 1, <25% of fruit surface with scald; 2, 25-75% of the surface with scald; and 3, >75% of the surface with scald. Also, the % of fruit with intense scald was calculated (% of fruit with scald values of 2 or 3, scald obvious to the consumer in red apples).

D. Extract preparation and TPC and TAA measurement

To determine the total phenolic content (TPC) and total antioxidant activity (TAA) of the fruit, three replications of 10-fruit each per treatment were used. Ten slices of the 10 fruit were grounded with liquid nitrogen with an electric grinder and a sample of 5 g pulp plus peel was extracted with 25 mL methanol. The extract was centrifuged at 4000*g* for 10 min and the supernatant was analyzed for TPC and TAA.

TPC was measured to the methanolic solution at 760 nm with a spectrophotometer (Milton Roy Spectronic 301, Ivyland, USA) using the Folin-Ciocalteu reagent and expressed as g of equivalent gallic acid per kg of fresh weight [22].

TAA of the methanolic solution of the fruit was evaluated by using the ferric-reducing antioxidant power (FRAP) assay with minor modifications [23]. The TAA of the samples was expressed as mmol L-ascorbic acid equivalents per kg fresh weight.

E. Statistical analysis

Analysis of variance was conducted over time of storage and treatment with the SPSS statistical package (SPSS Statistics for Windows, Version 20.0, IBM Corporation, Armonk, NY, USA). The difference among treatments was evaluated by using the least significant difference (LSD) for P≤0.05 level of significance. Correlation analysis was performed with the same statistical package using Pearson correlation coefficient (r).

III. RESULTS AND DISCUSSION

A. Skin color

1-MCP treatment of apples 'Starking Delicious' independently of cold storage temperature

resulted in a darker red skin color at 2.5 months measured as L*, C* and Hue, compared to no 1-MCP 0 even after 7 d shelf life and thereafter became progressively lighter red until 7.5 months plus 7 d shelf life (Fig. 1). Similar results were obtained from 'Red Chief' apples treated with 1-MCP and stored at 0 ^{o}C [11]. This finding coincided with superficial scald appearance (Fig. 4) in untreated fruit (no 1-MCP 0) that progressively obtained a less bright and lighter red skin color, and it is clearly demonstrated by the strong positive correlation between Hue and scald incidence (r=0.727, P<0.01). The above color changes were also macroscopically observed with the untreated with 1-MCP fruit having the characteristic brown color in the skin when scald occurred [24]. This means that cold storage of untreated apples at 0 ^{o}C in air even after 2.5 months plus 7 d shelf life caused fruit quality degradation and seriously reduced the percent of marketable fruit which after 7 months reached 50% of fruit immediately after cold storage and 56% of fruit after 7 d shelf life (Fig. 4).

In addition a strong negative correlation was found between Hue and FF (r=-0.614, P<0.01) and between Hue and TA (r=-0.707, P<0.01) showing that fruit color is also associated with fruit ripening during cold storage or plus 7 d shelf life. In general, 1-MCP treated fruit stored at 0 ^{o}C exhibited delayed ripening compared to untreated fruit stored at 0 ^{o}C or treated with 1-MCP and stored at 2 ^{o}C or 5 ^{o}C (Fig. 3).

B. SSC and TA

SSC initially increased in storage due to starch hydrolysis and remained relatively stable thereafter in all treatments with no significant differences among them (Fig. 2). Seven d shelf life did not affect or slightly increased fruit SSC, which ranged from 11.7 to 13.9% (Fig. 2). TA only slightly decreased mostly during shelf-life reaching from 0.3 g at harvest to around 0.2 g after 7.5 months storage plus 7 d shelf-life (Fig. 2). In addition 1-MCP treated fruit stored at 0, 2 or 5 ^{o}C had constantly slightly increased TA, especially the fruit stored at 0 ^{o}C, compared to untreated fruit (Fig. 2). Differences have been reported in quality characteristics between apple cultivars treated with 1-MCP that were stored at 0.5 ^{o}C with 'Red Delicious' apples having higher SSC than untreated fruit [14]. SSC in 1-MCP treated fruit might be expected to be higher than in untreated fruit due to lower respiration rates, but may vary depending on the product and the storage conditions ([9], [14]). In addition, our results are in agreement with other researchers which reported that apple treatment with 1-MCP and storage at 0 ^{o}C delayed fruit ripening, maintained fruit acidity and reduced ethylene production and respiration rates compared to untreated fruit, while SSC did not differ between treated or untreated fruit [9]. In our experiment the apples that were treated with 1-MCP and stored at 2 or 5 ^{o}C presented similar ripening behavior with those that were treated with 1-MCP and stored at 0 ^{o}C. In plums, 1-MCP did not affect SSC, but delayed the metabolism of TA during fruit ripening [25].

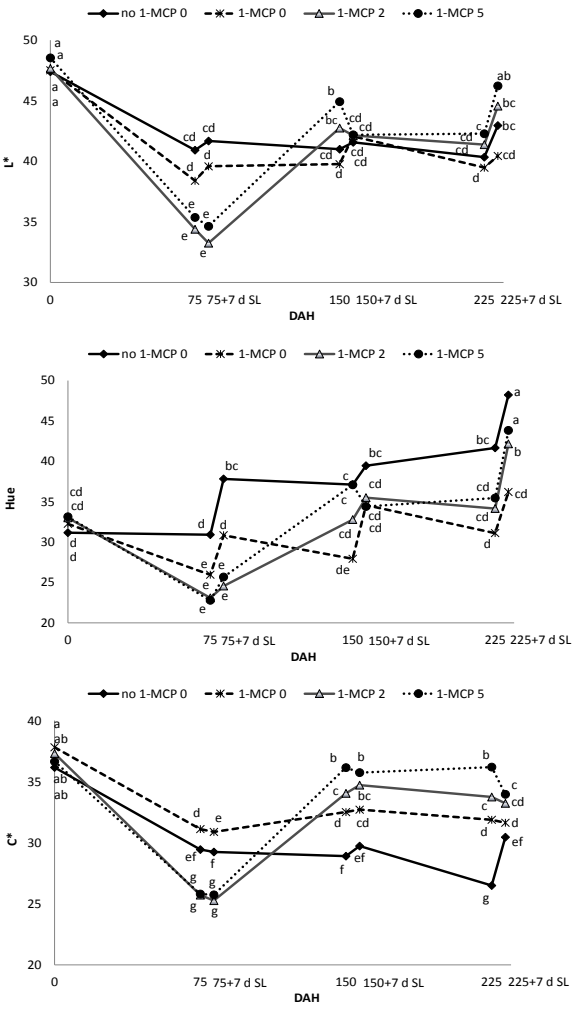

Fig. 1: Fruit skin color parameters L*, Hue and C* at harvest, at 75, 150 and 225 days after harvest (DAH) and after 7 d shelf-life (7 d SL) for untreated with 1-MCP and stored at 0 ^{o}C (no 1-MCP 0), 1-MCP treated and stored at 0 ^{o}C (1-MCP 0), 1-MCP treated and stored at 2 ^{o}C (1-MCP 2) and 1-MCP treated and stored at 5 ^{o}C (1-MCP 5) apples cv. 'Starking Delicious'. The values represent the mean of three 10-fruit replications (n=3) and values with the same letter are not significantly different, according to LSD at P≤0.05.

C. Flesh firmness

In this apple cultivar 'Starking Delicious' fruit softening with storage followed a similar pattern for all treatments (Fig. 3). 1-MCP treated apples stored at 0 ^{o}C showed delayed fruit softening and overall retained better FF during the whole storage

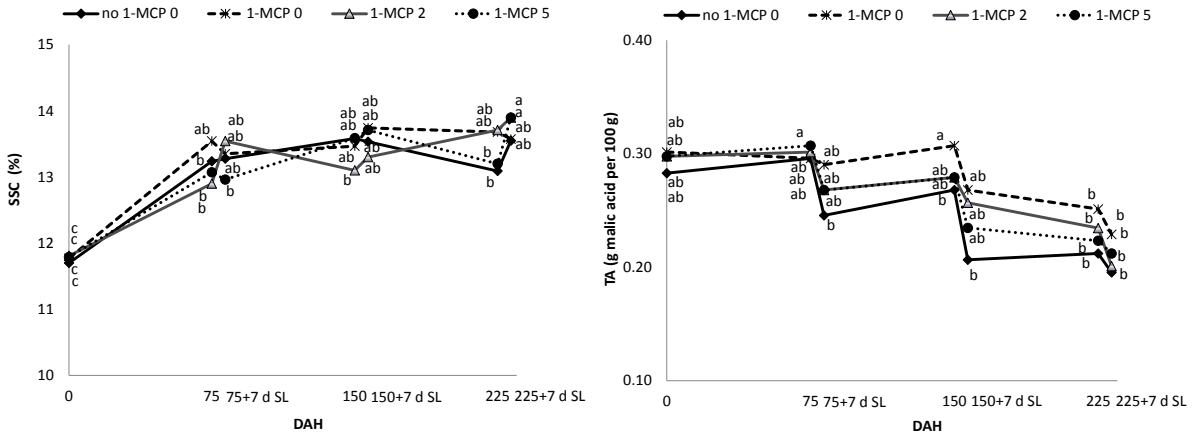

Fig. 2: SSC (%) and TA (g malic acid per 100 g) at harvest, at 75, 150 and 225 days after harvest (DAH) and after 7 d shelf-life (7 d SL) for untreated with 1-MCP and stored at 0 °C (no 1-MCP 0), 1-MCP treated and stored at 0 °C (1-MCP 0), 1-MCP treated and stored at 2 °C (1-MCP 2) and 1-MCP treated and stored at 5 °C (1-MCP 5) apples cv. 'Starking Delicious'. The values represent the mean of three 10-fruit replications (n=3) and values with the same letter are not significantly different, according to LSD at P≤0.05.

period compared with apples treated with 1-MCP and stored at 2 °C or 5 °C and especially with the untreated with 1-MCP fruit (Fig. 3). Seven d shelf life resulted in further fruit softening, which was significant in apples untreated and stored at 0 °C and apples 1-MCP treated and stored at 5 °C especially after 5 months cold storage and thereafter (Fig. 3).

One of the most important findings of this experiment is that the higher storage temperature overcame the 1-MCP effect and played a crucial role in fruit softening (Fig. 3). However, after 2.5 months of cold storage, apples treated with 1-MCP and stored at 2 °C or 5 °C retained their firmness to acceptable levels compared to the untreated with 1-MCP and stored at 0 °C even after 7 d shelf life, a significant characteristic for the consumer, with only slightly lower values compared to apples treated with 1-MCP and stored at 0 °C (Fig. 3).

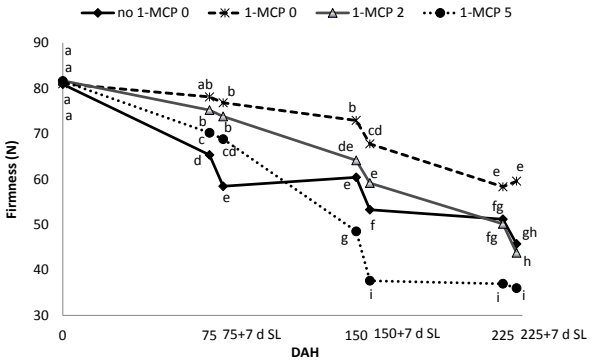

Fig. 3: Flesh firmness (N) at harvest, at 75, 150 and 225 days after harvest (DAH) and after 7 d shelf-life (7 d SL) for untreated with 1-MCP and stored at 0 °C (no 1-MCP 0), 1-MCP treated and stored at 0 °C (1-MCP 0), 1-MCP treated and stored at 2 °C (1-MCP 2) and 1-MCP treated and stored at 5 °C (1-MCP 5) apples cv. 'Starking Delicious'. The values represent the mean of three 10-fruit replications (n=3) and values with the same letter are not significantly different, according to LSD at P≤0.05.

Thereafter, at 5 months cold storage, apples treated with 1-MCP and stored at 2 °C retained their FF similar to untreated or higher after 7 d shelf-life showing slightly delayed ripening behavior compared to untreated stored at 0 °C in air, while scald incidence was still minor compared to untreated plus 7 d shelf life (Fig. 3, Fig. 4). Apples treated with 1-MCP and stored at 5 °C for 5 months softened significantly resulting in unacceptable texture for the consumer (Fig. 3), which is connected to a mealy texture in apples [26]. Thus, after 5 months cold storage at 5 °C 1-MCP action was diminished as far as flesh softening was concerned, but still scald incidence was negligible compared to untreated. At 7.5 months cold storage, 1-MCP treated and stored at 2 °C and the untreated with 1-MCP apples stored at 0 °C had similar flesh firmness immediately after storage and after 7 d shelf life, while 1-MCP treated and stored at 5 °C apples softened extensively. Other researchers found that apples maintained their firmness during cold storage at around 0 °C after 1-MCP treatment, with data presented on 'Delicious', 'Granny Smith', 'Fuji', 'Ginger Gold', 'Gala', 'Idared', 'Jonagold', 'Empire' and 'McIntosh' ([8], [9], [13], [14], [27]).

D. Superficial scald

Scald was not present in apples stored for 2.5 months immediately after removal from storage (Fig. 4). In the untreated apples with 1-MCP (no 1-MCP 0) fruit scald increased with storage time and especially during shelf life and was always higher than at the 1-MCP treated fruit. In 1-MCP treated fruit that were stored at 0, 2 or 5 °C, scald incidence remained negligible even after 7.5 months immediately after removal from cold storage at all storage temperatures (Fig. 4). After 7 d shelf life, in 1-MCP treated fruit scald appeared only after 5 months storage in apples stored

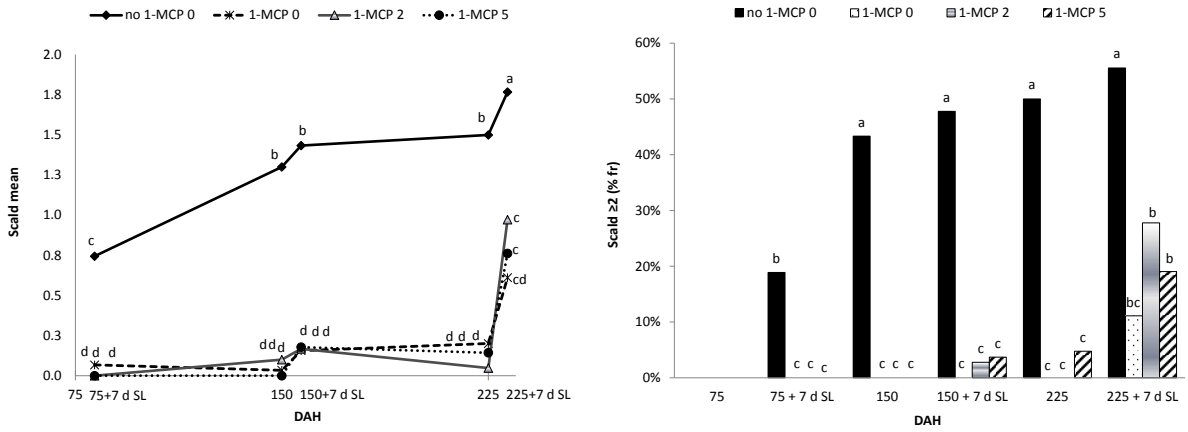

Fig. 4: Apple fruit scald incidence and scald intensity (Scald ≥2, % of fruit with scald at ≥25% of skin surface) stored for 75, 150 and 225 days after harvest (DAH) and after 7 d shelf-life (7 d SL) for untreated with 1-MCP and stored at 0 °C (no 1-MCP 0), 1-MCP treated and stored at 0 °C (1-MCP 0), 1-MCP treated and stored at 2 °C (1-MCP 2) and 1-MCP treated and stored at 5 °C (1-MCP 5) apples cv. 'Starking Delicious'. The values represent the mean of three 10-fruit replications (n=3) and values with the same letter are not significantly different, according to LSD at P≤0.05.

at 2 or 5 °C but not at 0 °C, while after 7.5 months plus 7 d shelf life scald incidence and intensity increased mainly in apples stored at 2 or 5 °C.

Apple 1-MCP treatment and storage at any temperature effectively suppressed the incidence and intensity of superficial scald compared to untreated apples (Fig. 4). Scald usually develops after cold stored fruit has been removed to ambient temperatures, although after extended storage it may be visible on fruit at cold storage [24]. In agreement with our results, 1-MCP suppressed superficial scald by 30% in 'McIntosh' and by 90% in 'Delicious' apples during cold storage at 0 °C [13]. Superficial scald was also suppressed in 'Delicious' and 'Law Rome' apples along with a reduction in a-farnesene and conjugated triols [14]. In 'McIntosh' and 'Delicious' apples, it was found that 1-MCP treatment and storage at 0 °C inhibited ethylene action and delayed ethylene-dependent ripening and superficial scald development [13]. Furthermore it was reported that 1-MCP eliminated or significantly reduced superficial scald development during cold storage at 0 °C for 6 months and subsequent ripening at 20 °C for 7 d [28].

E. TPC and TAA of the fruit

After 2.5 months of cold storage fruit TPC decreased at all treatments but slightly increased with prolonged storage up to 7.5 months of cold storage reaching similar values to initial ones (Fig. 5). In all treatments, TPC did not change with 7 d shelf life except after 7.5 months plus 7 d that TPC significantly decreased for 1-MCP treated stored at 0 °C fruit, while the opposite was found for the 1-MCP treated and stored at 2 °C apples (Fig. 5). Only after 5 months cold storage the TPC was slightly higher to the apples treated with 1-MCP and stored at 0 °C compared to the other treatments showing that storage temperature or 1-MCP treatment had not significant effect on apple TPC (Fig. 5).

Fruit TAA independently of the treatment decreased after 2.5 months of cold storage mainly at 1-MCP treated and stored at 2 or 5 °C apples (Fig. 5). With prolonged storage, TAA changed only slightly for the 0 °C stored fruit, untreated and 1-MCP treated, or increased at 7.5 months for the fruit 1-MCP treated and stored at 2 or 5 °C (Fig. 5). In addition, fruit 1-MCP treated and stored at 0 °C had higher TAA compared to 1-MCP treated and stored at 2 or 5 °C or the untreated with 1-MCP fruit after 5 months cold storage, but differences were not significant thereafter. With the additional 7 d shelf life TAA for apples 1-MCP treated and stored at 2 or 5 °C was lower than TAA for 1-MCP treated and stored at 0 °C at 2.5 months, similar at 5 months and higher at 7.5 months. It was shown that TAA may vary with storage temperature, storage duration and shelf life and probably it is necessary to measure TAA with other methods or certain constituents besides TPC to better understand the above changes.

More specifically, it was shown that during long-term cold storage at 0 °C as well as during an additional 7 d shelf life of apples at 16 °C, TPC, TAA and radical scavenging activity increased considerably either in regular cold storage or in CA [19]. In addition, it was reported that long term cold storage at 0 °C after 1-MCP application of 'Cripps Pink' apples slightly decreased or did not affect TPC and individual phenolic compounds in the peel, while in the flesh tissue these ingredients increased or remained constant [17]. In terms of TAA (as measured by the DPPH method) it was found increased during cold storage, while 1-MCP application reduced TAA only to the peel. Other researchers found that 1-MCP had a positive effect on inhibiting the reduction of TPC, flavonoids and

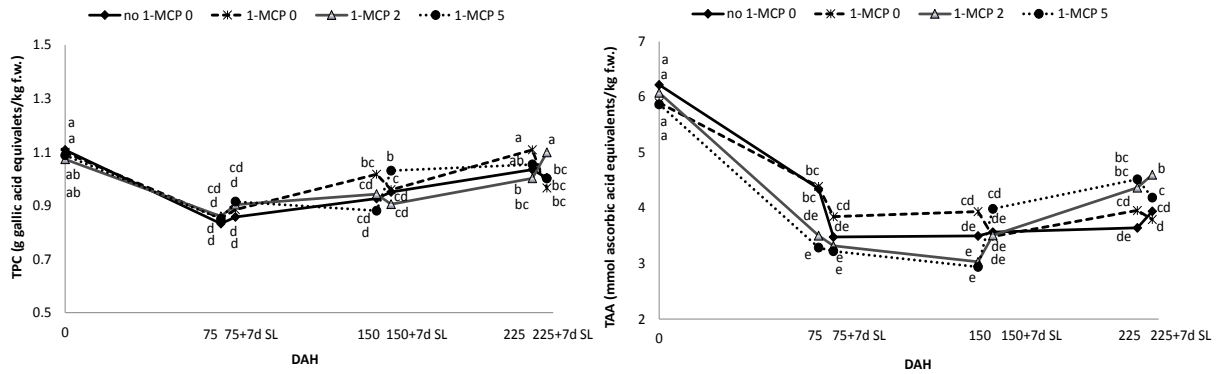

Fig. 5: Total phenolic content (TPC) and total antioxidant activity (TAA) with FRAP assay at harvest, at 75, 150 and 225 days after harvest (DAH) and after 7 d shelf-life (7 d SL) for untreated with 1-MCP and stored at 0 ^{o}C (no 1-MCP 0), 1-MCP treated and stored at 0 ^{o}C (1-MCP 0), 1-MCP treated and stored at 2 ^{o}C (1-MCP 2) and 1-MCP treated and stored at 5 ^{o}C (1-MCP 5) apples cv. 'Starking Delicious'. The values represent the mean of three 10-fruit replications (n=3) and values with the same letter are not significantly different, according to LSD at P≤0.05.

DPPH radical scavenging activity in apple peel and flesh during cold storage at 0 ^{o}C and after shelf life [20]. According to Carbone et al. [16] cold storage at 0-1 ^{o}C in air for 3 months did not significantly affect TPC and antiradical power of whole apples (using the DPPH assay). In addition other researchers found that TPC, flavonoid and anthocyanin concentrations as well as TAA in apples were relatively stable during air storage and 1-MCP treated fruit had higher TPC in the peel and slightly lower in the flesh compared to the untreated with 1-MCP fruit [29]. It is clear that literature does not give a clear trend on the effect of cold storage duration or 1-MCP treatment on TPC and TAA evolution during storage and shelf life.

IV. CONCLUSIONS

Energy costs of fruit storage facilities are relatively high in warm apple growing areas. The effect of 1-MCP application, cold storage temperature (0, 2 or 5 ^{o}C) and 7 d shelf life on fruit quality characteristics, superficial scald incidence, TPC and TAA of 'Starking Delicious' apples was evaluated. Apples treated with 1-MCP and stored at 0 ^{o}C had better FF and organoleptic quality and retained their external appearance (reduced scald) compared to the untreated with 1-MCP fruit throughout the storage period and after 7 d shelf life. After 2.5 months of cold storage, apples treated with 1-MCP and stored at 2 or 5 ^{o}C adequately retained FF compared to untreated with 1-MCP fruit even after 7 d shelf life, while all other quality parameters remained similar to apples treated with 1-MCP and stored at 0 ^{o}C. Thus, we propose, after pilot applications, commercial storage at 2 or 5 ^{o}C for 2.5 months for 'Starking Delicious' apples after 1-MCP treatment. This could result in energy costs savings especially in warm apple-growing areas. The application of 1-MCP, storage at 0, 2 or 5 ^{o}C for extended period of time plus shelf life had minor effects on TPC and TAA of 'Starking Delicious' apples.

ACKNOWLEDGMENT

The authors thank Mrs. Evangelia Panagiotaki for her technical assistance during laboratory work.

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