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COMBINATIONAL IMPACT OF *Debaryomyces hansenii* BIOAGENT AND 1-METHYLECYCLOPROPEN (1-MCP) ON SHELF LIFE AND QUALITY ATTRIBUTES OF KINNOW MANDARIN

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ABSTRACT: Due to rising consumers' concern over chemical/pesticides residue free eatables and international food safety laws, it is felt world wide to find out an alternative approach for postharvest food loss reduction and quality retention. To find out safe postharvest treatment alternatives, Kinnow mandarin fruits were treated individually with 1-MCP (250 nl L⁻¹), *Debaryomyces hansenii* (10⁹ cfu ml⁻¹ for 2 minutes) and their combination (250 nl L⁻¹ + 10⁹ cfu ml⁻¹). Treated fruits were stored at 10°C temperature and 85% RH. Their impact on pathological, physical and quality parameters was investigated after 45 days storage. Fruits treated with 1-MCP (250 nl L⁻¹) + *Debaryomyces hansenii* bioagent (10⁹ cfu ml⁻¹) resulted in minimum incidence of moulds (green 3.61% and blue 2.05%) over all natural decay (7.25%) and higher sensory score (7.50). Higher fruit firmness and lower PLW was recorded equally good with 1-MCP alone and in combination of *Debaryomyces hansenii* bioagent. Postharvest fruit quality parameters viz TSS, acidity, total sugars and vitamin C content were not affected with the 1-MCP and *Debaryomyces hansenii* either alone or in combination.

Keywords: Kinnow mandarin, 1-MCP, Debaryomyces hansenii, postharvest decay, quality.

Kinnow is hybrid mandarin between King and Willow leaf mandarins, developed by Dr. H.B. Frost at California during the year 1935. In India, it was introduced during mid sixties at Regional Fruit Research Station, Abohar (Punjab) for conducting feasibility trials. Where other citrus varieties failed to adopt, Kinnow consistently performed well in semi-arid and sub-mountainous tract of India. Besides India, Kinnow is now a well established citrus fruit in Pakistan and Afghanistan. Basically, characters like precocity and prolificacy in bearing, attractive fruit colour, higher juice content, absence of granulation disorder and heavy returns have attracted the growers to intensify its cultivation in India (Bindra, 6). Several factors have been reported to be associated with postharvest losses of Kinnow mandarin. Postharvest losses caused by fungal diseases and physiological activities (respiration, ethylene liberation and enzyme) are the major factors limiting the shelf life of the Kinnow (Singh and Mandal, 23).

Presently, the control of the postharvest losses

relies mainly on use of the synthetic chemicals. Practising non-chemical control methods to reduce postharvest losses is most important and becoming popular world over. Consumers are demanding minimal pesticide load on the fresh produce and many spoilage causing pathogens are developing resistance to several synthetic chemicals (Conway et al., 7). Most of the fungicides have been phased out and rest are used with restriction due to the health concerns. Therefore, it is imperative to find out the safer alternative to minimise environmental degradation and ensure consumer safety. Biocontrol strategy has made much progress during the last decade. At present, several biocontrol treatments have been approved for commercial applications, and now research is focused on improving the bioefficacy of the antagonist. One of the approaches has been the selection of combinations of antagonists, which may work more effectively (Barkai-Golan and Phillip, 5). It is a very challenging work, as microorganisms have differential growth habits, requirements for nutrition and cultural conditions.

Naturally occurring micro organisms, which are found to be adhered on the fruits and vegetables surface have been shown potential to protect the fresh produce against postharvest disease causing pathogens. During last decade several products viz Serenade (Bacillus subtilis based), Messenger (Erwinia amylovora based), Biosave (Pseudomonas syringae strain 10 LP), Aspire (Candida oleophila strain 1-18), AQ-10 bio-fungicide (Ampelomyces quisqualis) have been isolated and registered in the United States and Germany (El-Neshawy et al., 9; Fravel, 10, Plaza et al., 19, Zhao et al., 31,). Use of some safe bioactive compounds have been proved beneficial in bringing down the physiological activities of fruits during transportation, storage and minimizing the over all qualitative and quantitative losses (Porat et al., 20). 1-methylecyclopropen (1-MCP) is a synthetic cyclic olefin that inhibit ethylene by blocking access to the ethylene biding receptor (Sisler et al., 24). Eduardo and Kader (8) investigated that 1-MCP bound at the ethylene receptor in many fruits is still capable of inhibiting cell wall degrading enzymes such as PG secreted by pathogens and thus prevent pathogenesis. The yeast, Debaryomyces hansenii has exhibited a wide spectrum of biological activity against many pathogens (Wilson and Chaulutz, 25); it reduces the incidence of the green mould development by competing for space and nutrients at any injury site on the rind of the citrus fruit and thus inhibiting mould development. The bioefficacy of the Debaryomyces hansenii has been reported to be enhanced by several workers when it was applied in combination with calcium salts, sodium salts, salicylic acid and waxes (Yu et al., 30, Singh and Mandal, 23). However, there is no information concerning the combinational effect of bio agent and 1-MCP on postharvest diseases and quality attributes of the Kinnow mandarin fruits.

The objectives of this study were to determine postharvest loss reduction and quality retention of the Kinnow mandarin fruits by combined application of the *Debaryomyces hansenii* and 1-MCP.

MATERIALS AND METHODS

Kinnow mandarin fruits were manually harvested at commercial maturity stage from 10-year old orchard in Abohar (Punjab) during January 2008. After preliminary selection, fruit that had blemishes or otherwise appeared over mature were discarded. Selected fruit were washed with tap water, air dried prior to treatment with 1-MCP and *Debaryomyces hansenii*. 1-MCP (3.3% powder) was obtained from Rohm and Hass, Italy.

Freshly harvested fruits were treated for 2 minutes each with distilled water, 1-MCP (500 nl L^{-1}), 1-MCP (500 nl L^{-1}) + *Debaryomyces hansenii* slurry (10⁹ cfu ml⁻¹) and *Debaryomyces hansenii* slurry (10⁹ cfu ml⁻¹). Ten fruits were taken in each treatment and replicated thrice. Treated fruits were packed into CFB boxes and placed into cold storage (10°C; 85% RH) just after drying off surface water.

Weight of individual lot containing 10 fruits each was recorded at day 0 (A) and at the scheduled sampling date (B). Physiological loss in weight was calculated as (A-B)/A x100 and expressed as percentage loss in original fresh weight.

Respiration rate was measured based on 'closed system' by using auto gas analyzer (Model: Checkmate 9900 O₂/CO₂, PBI Dansensor, Denmark) and expressed as ml CO_2 kg⁻¹ h⁻¹. Known weight of whole Kinnow mandarin fruits were trapped in two-liter airtight container having twist-top lid fitted with a silicon rubber septum at the centre of the lid. The containers were kept at 25°C for 2-3 hours for accumulation of respiratory gases in the head space. After specified time, the head space gas was sucked to the sensor of analyzer through the hypodermic hollow needle and the displayed value of evolution rate of CO₂ concentration (%) was recorded. Rate of respiration was calculated on the basis of rate of evolution of CO_2 from the sample per unit weight per unit time (Asrey et al., 3).

Total sugars were determined by the method described by AOAC (2) by taking a known quantity

of homogenized pulp, using lead acetate to remove excess of lead free aliquot were examined by titrating against boiling Fehling's solution, which had previously been standardized using methylene blue indicator. Total sugars were determined after complete inversion of non-reducing sugars by acid hydrolysis and aliquot of this lead free solution were analyzed by similar method as described earlier. The data were expressed in percentage.

Total soluble solids contents were determined by measuring refractive index of the juice samples with hand refractometer and the results were expressed in percentages (Larrigaudiere *et al.*, 15).

By using 2,6-dichloroindophenol indicator titrimetric method, the ascorbic acid content of the fruit juice was determined. Results were expressed as milligrams of ascorbic acid per 100 g sample (Ozden and Bayindirili, 18).

Pressed fruit juice acidity was measured by titration with 0.1N NaOH to pH 8.1, 4g of juice diluted with 20 ml of double distilled water. Titratable acidity was calculated and expressed as per cent malic acid (Wright and Kader, 29).

The anti-oxidant capacity of the fruit pulp was determined by the FRAP (ferric reducing ability of plasma) method. Fruit hardness was determined using a texture analyzer (model: TA+Di, Stable Micro-systems, UK) using compression. The sample was compressed using a cutting and hardness was defined as maximum force (kgf) during compression.

Obtained data were subjected to analysis of the variance (ANOVA) using SAS package. Statistical significance was assessed at P=0.05 and least significant difference was used for pair-wise comparison of the means.

RESULTS AND DISCUSSION

1. Effect on disease control, physiological and physical parameters

Application of 1-MCP and *Debaryomyces* hansenii individually as well as in combination, significantly affected the disease incidence, physiological and physical parameters of stored Kinnow mandarin fruits (Table1). Minimum incidence percentage of both the moulds (3.61 green and 2.05 blue) was recorded in *Debaryomyces hansenii* (10^9 cfu ml⁻¹) + 1-MCP (750 nl L⁻¹) treated fruit after 45 days of cold storage (10° C temperature and 85% RH). Significnat variation in incidence (P=0.05) was found within the treatments. The incidence suppression efficacy of *Debaryomyces hansenii* and 1-MCP was found higher against blue mould rot over green mould rot. The decay and physiological loss in weight was also found lower with MCP + *Debaryomyces hansenii* treated fruits (4.45 and 7.25%, respectively) (Fig. 1 and 2).

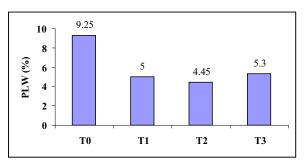


Fig. 1. Impact of 1-MCP and *Debaryomyces hansenii* on PLW of Kinnow madarin after 45 days storage at 10°C and 85% RH. Each value is the mean of three replications.

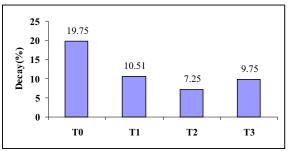


Fig. 2. Impact of 1-MCP and *Debaryomyces hansenii* on decay of Kinnow madarin after 45 days storage at 10°C and 85% RH. Each value is the mean of three replications.

where,

T₀-Control

 T_1 -1-MCP (250 nl L⁻¹)

 T_2 -1-MCP (250 nl L⁻¹ + *Debaryomyces hansenii* 10⁹ cfu ml⁻¹)

T₃-Debaryomyces hansenii (10⁹ cfu ml⁻¹)

The fruit firmness was decreased in all the treatments in due course of time during storage. Firmness of fruit was not significantly changed due to sole treatment of *Debaryomyces hansenii* while it was found affected with 1-MCP and in combination (1-MCP + *Debaryomyces hansenii*). Maximum firmness (6.67 Kgf) was recorded in 1-MCP treated fruits.

Respiration rate of stored fruits was neither significantly changed due to sole application of *Debaryomyces hansenii* nor its combined application with 1-MCP. Interestingly, fruit treatment with 1-MCP alone, reduced the respiration rate (1.17 μ L k⁻¹ h⁻¹) of stored Kinnow mandarin fruit. In respect to organoleptic rating of the Kinnow mandarin fruits 45 days after storage, all the treatment significantly affected the fruit acceptability score. 1-MCP + *Debaryomyces hansenii* application followed by 1-MCP alone (250 nl L⁻¹) retained the higher organoleptic rating of 7.50 and 6.58, respectively.

2. Effect on post harvest fruit quality parameters

Debaryomyces hansenii and 1-MCP neither individually nor in combination had affected the postharvest quality related chemical traits of the treated Kinnow mandarin fruits. The difference in the total soluble solids (TSS), titratable acidity, total sugars, vitamin C and total antioxidant capacity of the fruit juice was found non significant in all the treatments after 45 days storage at 10°C and 85% RH (Table 2).

The fruit treated with *Debaryomyces hansenii* were found least prone to blue mould and green mould rot caused by the *Penicillium digitatum* and *Penicillium italicum*. Furthermore, the blue and green mould controlling efficiency of *Debaryomyces hansenii* was increased when it was used along with 1-MCP (10^9 cfu ml⁻¹ + 250 nl L⁻¹). These findings indicate that *Debaryomyces hansenii* has potential to supress the blue and green mould causing pathogens in Kinnow mandarin. 1-MCP has given synergistic strength to bioagent (*Debaryomyces hansenii*) in sharpening its antagonism against blue and green mould caused by Penicillium italicum and Penicillium digitatum, Debaryomyces hansenii exerts respectively. antagonism through competition with pathogen for space and nutrients (Singh and Mandal, 23). As the 1-MCP has ability to reduce respiration rate, cell wall softening enzyme activities (Eduardo and Kader, 8; Win et al., 28) and fruit firmness retention; its application with Debaryomyces hansenii might have reduced the nutrient availability to the pathogen which intern resulted into less incidence percentage of moulds in stored Kinnow mandarin fruits. These findings are in agreement with the results of other work carried out on these aspects (Singh, 22; Wilson et al., 26). They found that the competition for nutrients and growing space is the main mode of action of antagonistic yeast to control postharvest storage diseases of fruits. The bio-control of antagonistic yeast encompasses several modes of action viz. mycoparasitism, induced resistance, production of lyetic enzymes (chitinase and glucanase), limiting spore germination and elongation of germ tube (Wilson et al., 27; Zheng et al., 32). 1-MCP is a synthetic cyclic olefin that inhibits ethylene by blocking access to the ethylene binding receptor, lowers action of maturation associated genes (PC-PG1 and PC-PG2) and enzymes (Sisler and Serek, 24; Khan and Singh, 14, and Martinez et al., 17). The combined application of 1-MCP and Debaryomyces hansenii have been succeeded in keeping over all fruit decay percentage at its minimum level (7.25%), while their impact on fruit firmness and respiration rate was at par with 1-MCP or bioagent treated fruits, respectively. Minimum fruit decay loss by the combined application of 1-MCP and Debaryomyces hansenii may be attributed due to the higher fruit firmness (1-MCP treatment retained higher fruit firmness), which has likely prevented the pathogen invasion in stored fruits. Aguayo et al. (1) also recorded less decay loss in 1-MCP + CaCl₂ treated strawberry fruit kept at room temperature and cold storage.

Respiration rate, PLW and fruit firmness of the stored fruits remained unaffected with the bioagent

Table 1: Impact of 1-MCP and Debaryomyces hansenii on quality attributes and mould rot control of Kinnow after 45
days storage at 10°C temperature and 85% R.H.

Treatment	Firmness (Kgf)	Respiration (ml CO ₂ k ⁻¹ h ⁻¹)	Green mould incidence (%)	Blue mould incidence (%)	Sensory score
Control	6.47b	1.23ab	8.32a	6.23a	5.58c
1-MCP (250 nl L ⁻¹)	6.67ab	1.17b	5.66b	3.78b	6.58b
1-MCP (250 nl L ⁻¹ + <i>Debaryomyces hansenii</i> 10 ⁹ cfu ml ⁻¹)	6.52a	1.28b	3.61c	2.05c	7.50a
Debaryomyces hansenii (10 ⁹ cfu ml ⁻¹)	6.46b	1.33a	4.20c	3.59b	5.83c
Initial value	7.01	1.16	0.00	0.00	7.60
CD (P=0.05)	0.17	0.12	1.30	0.96	0.70

Means with the same letter, do not differ significantly as per Duncan Multiple Range Test

Table 2: Impact of 1-MCP and *Debaryomyces hansenii* on quality attributes of Kinnow mandarin after 45 days storage at 10°C temperature and 85% R.H.

Treatment	TSS (%)	Acidity (%)	Sugars (%)	Vitamin C (mg 100 ml ⁻¹ juice)	Antioxidant (mg Fe ⁺⁺ 100 ml ⁻¹ juice)
Control	11.42a	1.07a	10.16a	19.61a	123.51b
1-MCP (250 nl L ⁻¹)	10.75a	1.09a	10.32a	20.21a	127.07a
1-MCP (250 nl L ⁻¹ + <i>Debaryomyces</i> hansenii 10 ⁹ cfu ml ⁻¹)	11.17a	1.07a	10.35a	20.23a	126.82a
Debaryomyces hansenii (10 ⁹ cfu ml ⁻¹)	11.26a	1.09a	10.39a	20.23a	124.55b
Initial value	11.17	1.05	10.25	21.26	128.37
C.D. (P=0.05)	NS	NS	NS	NS	2.14

Means with the same letter, do not differ significantly as per Duncan Multiple Range Test.

(*Debaryomyces hansenii*) application. As all above parameters are interdependent and any change in an individual affect the value of other one. These results are in expected lines, because the *Debaryomyces hansenii* treatment was given to the intact fruits (no wounding and artificial inoculation). So, practically there will be no difference in the physiology of bioagent treated healthy fruits and control fruits. When there is interaction of pathogen, fruits and bioagent cells, the physiological activities will increase according the severity of pathogenicity level. These findings got support of Hiwasa *et al.* (12); Manganaris *et al.* (16) and Singh and Mandal *et al.* (23).

variation postharvest The in quality parameters (TSS, acidity, sugar, vitamin C) of stored Kinnow mandarin fruits were insignificant in all the treatments, whereas antioxidant capacity of the stored Kinnow fruit shown significant difference under 1-MCP and 1-MCP + Debaryomyces hansenii treated fruit juice. The results are in conformity of earlier workers (Bai et al., 4; Gutierrez et al., 11; and Itai et al. 13, and Singh, 21).

In conclusion, our findings show that the 1-MCP and *Debaryomyces hansenii* are compatible with each other. For controlling postharvest deterioration and diseases, 1-MCP and *Debaryomyces hansenii* are utilized individually or 1-MCP with any fungicide. For evolving further more eco-friendly green postharvest treatment technology, it is required to find out alternatives for 1-MCP with the use of bio-based formulations. Their respective antifungal and antisenescense potential may be gainfully utilized in postharvest disease control and shelf life extension of Kinnow fruits.

REFERENCES

- Aguayo, E., Jansasithorn, R. and Kader, A.A. (2007). Combined effects of 1-methylcyclopropene, calcium chloride dip, and/or atmospheric modification on quality changes in fresh-cut strawberries. *Postharvest Biol. and Tech.*, 40: 269–278.
- AOAC (1995). Official Methods of Analysis, 16th ed. 45.1.14. AOAC, Arlington, Virginia.
- Asrey, Ram, Jain, R.K. and Kumar, A. (2008). Maturity, transportation and storage study in strawberry fruit. J. Food Sci. Tech., 45: 540-542.
- Bai, J., Baldwin, E.A., Goodner, K.L., Mattheis, J.P. and Brecht, J.K. (2005). Response of four apple cultivars to 1-methylcyclopropene treatment and controlled atmosphere storage. *Hort. Sci.*, 40: 1534-1538.
- Barkai-Golan, R. and Phillip, D.J., (1991). Postharvest fruit treatment of fresh fruits and vegetables for decay control. *Plant Diseases*, 75: 1085-1089
- Bindra, A.S., (1990). In:*Citriculture in north* western India, K.S. Gill,K.S., Kanwar,J.S., Singh,R.(eds),Prospects and problems of citrus growing in Punjab with special reference to 'Kinnow'pp- 83-99.
- Conway, W.S., Leverentz, B., Janisiewicz, W.J., Blodgett, A.B., Saftner, R.A. and Camp, M.J., (2004). Integrating heat treatment, biocontrol and sodium bicarbonate to reduce postharvest decay of apple caused by *Colletotrichum acutatum* and *Penicillium expansum*. *Postharvest Biol. and Tech.* 32: 11-20.
- 8. Eduardo, V.B. and Kader, A.A. (2007). Effect of

1- methylecyclopropen on softening of fresh cut kiwifruit, mango and persimmon slices. *Postharvest Biol. and Tech.*, **43:** 238-244.

- El-Neshawy, S.M. and El-Sheikh, M.M., (1998). Control of green mould on oranges by *Candida oleophila* and calcium treatments. *Annals Agric. Sci. Cairo* (Special Issue), 3: 881-890.
- Fravel, D.R. (2005). Commercialization and implementation of biocontrol. *Annual Rev. Plant Biol.*, 43: 337-359.
- Gutierrez, S.M., Trinchero, D.G., Cerri, M.A., Vilella, F. and Sozzi, O.G. (2008). Different responses of goldenberry fruit treated at four maturity stages with the ethylene antagonist 1-methylcyclopropene. *Postharvest Biol. and Tech.* 48: 199-205.
- Hiwasa, K., Kinugasa, Y., Amano, A., Hashimoto, A., Nakano, R., Inaba, A. and Kubo, Y. (2003). Ethylene is required for both the initiation and progression of softening in pear (*Pyrus communis* L.) fruit. *J. Experi. Bot.*, 54: 771-779.
- Itai, A. and Tanahashi, T. (2008). Inhibition of sucrose loss during cold storage in Japanese pear (*Pyrus pyrifolia* Nakai) by 1-MCP. *Postharvest Biol. and Tech.*, 48: 355-363.
- Khan, A.S. and Singh, Z. (2007). 1-MCP regulates ethylene biosynthesis and fruit softening during ripening of 'Tegan Blue' plum. *Postharvest Biol. and Tech.*, 43: 298-306.
- Larrigaudiere, C., Pons, J., Torres, R. and Usall, J. (2002). Storage performance of Clementines treated with heat water, sodium carbonate and sodium bicarbonate dips. *J. Hort. Sci. Biotech.*, 77: 314-319.
- Manganaris, G.A., Crisosto, C.H., Bremer, V. and Holcroft, D. (2008). Novel 1-methylcyclopropene immersion formulation extends shelf life of advanced maturity 'Joanna Red' plums (*Prunus salicina* Lindell). *Postharvest Biol. and Tech.* 47: 429-433.
- 17. Martinez, X.C., Alonso, J.M., Sanchez, B. and Granel, A. (2002). Ethylene perception and response in citrus fruit. In: *Kanellis*, A.K.,

Grierson, D. (Eds.), Kluwer Academic Publisher, The Netherlands, 137-142.

- Ozden, C. and Bayindirli, L. (2002). Effect of combinational use of controlled atmosphere cold storage and edible coating applications on shelf life and quality attributes of green peppers. *European Food Res. Tech.*, 21: 320-326.
- Plaza, P., Torres, R., Teixido, N., Usall, J., Abadias, M. and Vinas, I. (2001). Control of green mould by the combination of Pantoea agglomerans (CPA-2) and sodium bicarbonate on oranges. *Bulletin-OILB/SROP*, 24(3): 167-170.
- Porat, R., Cohen, B.W.L., Avinoam, D., Raphael, G. and Samir, D. (1999). Effects of ethylene and 1-methylcyclopropene on the postharvest qualities of 'Shamouti' oranges. *Postharvest Biol. and Tech.* 15: 155-163.
- Singh, D. (2002). Bioefficacy of *Debarymyces* hansenii on the incidence and growth of *Penicillium italicum* on Kinnow fruit in combination with oil and wax emulsions. *Annals Plant Protec. Sci.*, 10: 272-276.
- Singh, D. (2004). Effect of *Debaryomyces* hansenii and calcium salt on fruit rot of peach (*Rhizopus macrosporus*). *Annals Plant Protec. Sci.*, **12**: 310-313.
- Singh, D. and Mandal, G. (2006). Improved control of *Rhizopus stolonifer* induced storage rot of peach with hot water and antagonistic yeast, *Debaryomyces hansenii*. *Indian Phytopath.*, **59**: 168-173.
- 24. Sisler, E.C. and Serek, M. (1997). Inhibitors of ethylene responses implants at the receptor level: recent developments. *Physiol. Plant*, **100**: 577-582.
- 25. Wilson, C.L. and Chaultz, E. (1989). Postharvest biocontrol of *Penicillium* rots of

citrus with antagonistic yeasts and bacteria. *Scientia Hort.*, **40:** 105-112.

- Wilson, C.L., Wisniewski, M., El-Ghaouth, A., Droby, S. and Chalutz, E. (1996). Commercialization of antagonistic yeasts for the biological control of post harvest diseases of fruits and vegetables. *J. Microbiol. Biotech.*, 46: 237-242.
- Wilson, C.L., Wisniewski, M.E., Biles, C.L., McLaughlin, R., Chalutz, E. and Droby, E. (1991). Biological control of postharvest disease of fruits and vegetables: alternatives to synthetic fungicides. *Crop Protec.*, 10: 172-177.
- Win, T.O., Srilaong, V., Julian, H., Khin, L.K. and Sirichai, K. (2006). Effects of different concentrations of 1-MCP on the yellowing of West Indian lime (*Citrus aurantifolia*, Swingle) fruit. *Postharvest Biol. and Tech.* 42: 23-30.
- 29. Wright, K.P. and Kader, A.A. (1997). Effect of controlled-atmosphere storage on the quality and carotenoid content of sliced persimmons and peaches. *Postharvest Biol. and Tech.* **10**: 89-97.
- Yu, T. and Zheng, X.D. (2007). An integrated strategy to control postharvest blue and grey mould rots of apple fruit by combining biocontrol yeast with gibberellic acid. *International J. Food Sci. Tech.* 42: 977-984.
- Zhao, Y., Shao, X.F., Tu, K. and Chen, J.K. (2007). Inhibitory effect of *Bacillus subtilis* B10 on the diseases of postharvest strawberry. *J. Fruit Sci.*, 24: 339-343.
- Zheng, X.D., Zhang, H.Y. and Xi, Y.F. (2004). Effects of *Cryptococcus laurentii* (Kufferath) Skinner on biocontrol of postharvest decay of arbutus berries. *Bot. Bull. Acad. Sinicia*, 45: 55-60.