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USE OF MODIFIED ATMOSPHERES TO CONTROL INSECT PESTS ON HORTICULTURAL PRODUCTS

A literature review

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SUMMARY

This literature review discusses the use of MA to control insect pests in horticultural products and the effects on and tolerances of horticultural products. Quarantine treatments are required for im- and export of many fruits and vegetables. Until now chemical fumigants are used to disinfest horticultural commodities, but they may be prohibited in the near future, because they form risks to humans and may damage the horticultural commodities. Therefore, alternative methods have to be developed for postharvest insect control. Generally, most countries have very low levels of tolerance to the presence of insects on imported agricultural products. If the levels of tolerance would be decided in a more rational way, other treatments but fumigating with chemicals might be accepted.

Generally, low O2 and/or high CO2 concentrations cause mortality of insects. The mortality rate is dependent on the concentration of the gases, RH, temperature, insect species, insect stage and length of treatment period. Use of other gases, e.g. helium and nitrogen, may increase mortality. Insects may develop resistance to MA treatments after several generations. Several potential MA quarantine treatments are described. Differences are made for controlled atmopsheres for external and internal pests, film wraps and coatings.

Insecticidal MA may have beneficial effects on horticultural commodities. Respiration rate is decreased, ethylene production and action are inhibited, colour change and softening are retarded, composition and nutritional values are maintained, the incidence of some physiological disorders is reduced and decay is inhibited. The specifice beneficial effects observed depend on the commodity, maturity stage, storage temperature, atmospheric composition and time of exposure.

The possible detrimental effects of insecticidal MA on fresh commodities include low O2 and/or high CO2 injury and off-flavour development. The period of tolerance of fruits and vegetables to insecticidal MA conditions is, therefore, limited to the time before the onsec of these effects. Models are described, which predict tolerances of fruits and vegetables to insecticidal CA.

Several MA treatments, which may be potentially used as quarantine treatments are discussed. A reduction in quarantine mortality requirements by importing countries would advance the development of other potential MA quarantine treatments. Much of the research on MA was done for specific, practical reasons with little control over some modifying factors, making broad comparisons and conclusions precarious. Entomologists and plant physiologists should coordinate their activities to perform comparable research and increased communication is needed between scientists working in CA grain storage and fresh commodity storage. So far, vegetables and ornamentals have been mainly ignored. More attention should be paid to the insect pests of those commodities and to the development of MA treatments of those pests. Also some general suggestions for future research are mentioned.

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1 INTRODUCTION

Quarantine treatments are required for im- and export of many fruits and vegetables. Ethylene dibromide and methyl bromide used to be the most common fumigants for postharvest insect disinfestation. In the USA unaccepted shipments are fumigated. In the Netherlands, fumigation with methyl bromide is only allowed if the products are to be exported, while in Japan fumigation is standard. However, these gases are extremely toxic to humans and constitute serious hazards to workers. Besides, methyl bromide may cause damage to the product and reduce storage life. In recent years, people have become more and more concerned about the use of chemicals in food and environment. Ethylene dibromide was prohibited as a fumigant in many countries. The use of methyl bromide may be restricted in the near future. Therefore, alternative methods have to be developed for postharvest insect control. Currently, cold and hot treatments are used as quarantine procedures for a few commodities. Controlled atmospheres with O2 concentrations below 1% and/or CO2 concentrations above 50% have been reported to kill effectively certain insects in many agricultural products (Ke and Kader, 1992a). This phenomenon is not quite new. Already in 1919 Dendy reported, that insects, when sealed in containers, caused their own death by exhaustion of the oxygen and the buildup of CO2 (Cotton, 1961) and Froggatt (1921) suggested that CO2 could be used effectively to control insects infesting shelled corn. These treatments have the advantages of leaving no chemical residues in the fresh commodities and being safe to workers and the environment. If very low O2 and/or very high CO2 atmospheres can kill the insects of concern without detrimental effects on the fresh commodities, they may be used as postharvest insect control treatments. Currently no CA treatment is approved for use as a quarantine treatment, but there is a definite need for this type of treatment (Klag, 1986).

Every country has its own level of tolerance to the presence of insects on imported agricultural products. In Japan zero tolerance is the standard. Not a single insect is accepted in a shipment of agricultural products. In the USA 'Probit 9' is the standard on which insect control is based. Probit 9 was defined by Baker (1939) as 99.9968% mortality with 95% fiducial limits (Gould and Sharp, 1990). Recently, serious questions have arisen about whether Probit 9 mortality or some proportion thereof is too rigourous for many of the host-insect systems that are subjected to quarantine. Use of the concept (<32 survivors in 1 million treated insects) has never been justified (Vail *et al.*, 1993). Development of the Probit 9 concept assumed high rates of infestation. High-level infestations occur with certain species of tephritid fruit flies (e.g., the Mediterranean fruit fly, *Ceratitis capitata* Wiedemann, and the melon fly, *Dacus cucurbitae* Coquillet) in artificially infested fruits. Baker

(1939) provided no explanation as to why Probit 9 was required, other than to ensure no survival in products where fruit flies were involved. However, such infestations do not occur in marketed fruit. In the case of most lepidopterous pests, high-level infestations (e.g., larvae of diapausing codling moth, *Cydia pomonella* L., in walnuts or other fruits) cannot be obtained (Vail *et al.*, 1993).

Landolt *et al.* (1984) stated that risk should not be based on the number of insects killed but rather on the risk remaining after quarantine efforts. They refer to risk as the probability of a potential mating pair surviving the quarantine procedures, usually fumigation. Probability can be calculated from the infestation rate and shipment size. Thus, treatments can be tailored to the crop, pest, infestation level, and shipment sizes to provide security and yet avoid unnecessary overkill.

Especially in the USA and some other countries, e.g. Israel, much research has been carried out on the possible use of MA as quarantine treatment on insect pests. This research was mainly focused on stored products, such as grains, beans, nuts and dried fruits, and on tropical fruits. Other commodities, such as vegetables, cutflowers and potplants are hardly being discussed in regard of MA to control insect pests. In the Netherlands there are some problems with insects on cutflowers, potplants and fruity vegetables, especially bellpeppers. Export destinations causing problems are USA, Japan, and in a less degree UK and Sweden. The most important insect pests on ornamentals are thrips, aphids and mites. On other commodities some diptera and lepidoptera cause most of the problems.

This report reviews the available literature on the effects of MA treatments on insects. Several techniques will be mentioned, e.g. CA, MA, film wrapping and coating. The effect of CO2 and O2 concentrations, duration of the treatment, and other conditions, such as temperature, relative humidity and insect stage, will be discussed. Furthermore, the effects of insecticidal MA on horticultural products will be reviewed with focus on storage time, quality, decay, flavour and aroma. From the conclusions recommendations for research can be given.

2 EFFECTS OF MA TREATMENTS ON INSECTS

This chapter will discuss the effects of MA on insects. Many factors are involved and decide on the success of the MA treatment in controlling insect pests. There are several ways of creating MA, such as CA, film wrapping and coating. Of great influence on insect killing are the concentrations of CO2, O2 and other gases. The length of the treatment is another important factor as are the conditions under which the treatment takes place, such as temperature and relative humidity. MA treatments may not have the same effect on all stages of the insect.

2.1 Components of modified atmospheres

Initially, it was suggested that if a low O2 content of less than 2% was achieved and maintained, the remaining mixture of atmospheric gas was biologically unimportant. This assumption is still valid in the case of hermetic storage for insect control in grain. However, holding time is an important variable in this assumption. Intensive research has demonstrated that controlled atmospheres can be effective at O2 levels above 2% and that the percent of other gases can play an important role in effecting insect control (Davis and Jay, 1977).

The gas mixture produced by atmospheric burners containing both CO2 and CO produces a more rapid kill of test insects than pure N2/O2 mixtures at the same O2 concentrations. Atmospheres with high concentrations of CO2, such as 60% CO2 and 40% normal air, also have been shown to be insecticidal and may kill insects more rapidly than the lower O2 mixtures produced by atmospheric burners (Davis and Jay, 1977).

The use of N2 for insect control has the advantage that 78% of this gas is already available in the interstitial spaces of a grain mass. Yet O2 in these spaces must be reduced to less than 2% to obtain effective control. This level is often difficult to attain and maintain in semi-tight storage facilities. Therefore CO2 is generally considered to be more efficient than N2 in situations where tight sealing is physically impossible or not economically feasible (Davis and Jay, 1977).

CA research was carried out with almonds and raisins, the most abundant nut and dried fruit crops, respectively (Soderstrom, 1977). First, the effect of a low O2 atmosphere (consisting of ca. 0.5% O2, 9.5-11% CO2 and the balance principally N2) on the taste quality of almonds and raisins was determined. At 1, 3, 6, 9, and 12 months, samples were analyzed for taste and odour. Taste revealed that the atmosphere did not alter either the flavour or odour of the almonds (Guadagni *et al.*, 1978) or raisins. Second, mortality of the major species of insects infesting dried fruits and tree nuts was determined. At 27°C two days were required to kill all stages of these insects, whereas at 16°C one week was required.

A CO2 concentration of approximately 60% will give good control of most stored-grain pests in 4 days at temperatures of at least 27°C and higher (Jay, 1971). The CO2 concentration can fluctuate ±10% around 60% and still give effective control: with N2, mixtures with more than 2% O2 become ineffective for controlling most life stages of stored-grain pests. Laboratory studies have shown that eggs of the red flour beetle, *Tribolium castaneum* Herbst, do not hatch in atmospheres above 20% CO2 when the O2 level is ca. 19%. Similarly, AliNiazee and Lindgren (1970) reported that egg hatch of the red flour beetle and the confused flour beetle, *T. confusum* Jacquelin duVal, was delayed or partially or fully inhibited in CO2 atmospheres; in similar N2 atmospheres, there was as much egg hatch as there was in those eggs exposed to air. Laboratory studies also showed that a concentration of about 62% CO2 and 9% O2 gave over 90% control of 0- to 25-hr-old eggs of the cowpea weevil, *Callosobruchus maculatus* F., in 1 day, but concentrations of 99.2% N2 (balance O2) took 3 days to produce the same results. In 1-week-old adult cowpea weevils, 90% mortality was achieved in 2 days in an atmosphere containing 62% CO2, but with a concentration of 99.7% N2, it took over 3 days to achieve this mortality (Davis and Jay, 1977).

Aphids and thrips are common insect pests on flowers and leafy vegetables, and when present at destination ports, may require fumigation. The effects of a range of CO2 (0, 10, 30, and 50%) and O2 (0, 0.5, 2, and 21%) concentrations, separately and in combination, were tested on insect mortality at 0 or 5°C (Zheng *et al.*, 1993). Nonfeeding aphids and thrips were completely killed in atmospheres containing 10% CO2 in as little as 6 days at 0 or 5°C. Low O2 atmospheres were not nearly as effective: mortality was achieved after 2 to 3 weeks in 0.5 or 2% O2. Mortality in low O2 was greater at 5°C than at 0°C, presumably because of higher respiratory demand. Feeding aphids were killed under the same conditions as nonfeeding ones, whereas feeding thrips required longer treatment periods than nonfeeding insects (Appendix 3).

Argon and atmospheric N2 are considered to be biologically inert and have been ignored in controlled atmosphere research, except as diluents. However, AliNiazee (1972) found that 100% mortality of *T. confusum* and *T. castaneum* adults occurred in half the time when the insects were exposed to 100% helium rather than 100% N2 at 21.1 or 26.7°C and 38±6% RH. The differences in lethal time were less for pupae and not significant for eggs and larvae exposed to 100% N2 or helium.

Klaustermeyer *et al.* (1977) have investigated CO as a possible component of insecticidal MA on head lettuce against green peach aphids, *Myzus persicae* Sulzev, corn earworms, *Heliothis zea*

Boddie, and cabbage loopers, *Trichoplusia ni* Hübner. Neither short-term exposures (1 or 2 hours at 20°C to 0, 10, 30, 70, or 100% CO) nor continuous exposure treatments (1, 3, or 7 days at 2.5°C to 1, 5, 10, 20, 40, or 80% CO in combination with 5 or 21% O2 with or without added CO2 at 1%) effectively controlled the insects.

2.2 Factors influencing efficacy of modified atmospheres

Factors that affect the susceptibility of insects to MA will be discussed in this section. Pests of fresh commodities, even pests found on the fruit surface, are not subjected to RH as low as those to which stored product pests are subjected. Low RH increases the susceptibility of pests to MA (Hallman, 1994). The adult and pupal stages of many pests of fresh commodities, such as fruit flies, are not present in the marketed commodity and need not be considered when developing treatments; generally, all stages of stored product pests are present. Stored products can tolerate greater extremes of temperature and atmospheric composition than fresh commodities. Stored product mites were more difficult to kill with CA than other stored product pests. Mite pests of fresh commodities also appeared harder to kill using CA (Lidster *et al.*, 1981, 1984). However, CO2 levels and temperatures tested were low; higher levels might provide more rapid mortality.

Host acceptability probably affects the susceptibility of some pests of fresh commodities to CA. Many hosts of quarantine pests of fresh commodities are poor hosts. Consequently, pests developing on poor hosts may be more susceptible to a quarantine treatment than when developing on favourable hosts (Hallman, 1994). Generally, as the O2 concentration decreases and the CO2 concentration increases beyond certain thresholds, pest mortality increases. However, lower mortality (64-66%) occurred to exposed blueberry maggots, *Rhagoletis mendax* Curran, in 100% CO2 for 48 h compared with 45-90% CO2 plus 2 or 5% O2 (77-90% mortality) (Prange and Lidster, 1992). This same phenomenon has been observed with adult granary and rice weevils, *Sitophilus granarius* L. and *S. oryzae* L., respectively (Lindgren and Vincent, 1970). Mortality of navel orangeworm, *Amyelois transitella* Walker, decreased with increasing CO2 content from 1 to 10% when the atmosphere contained 0.3% O2, while at 20% CO2, mortality did not decrease (Brandl *et al.*, 1983).

Caribbean fruit fly , *Anastrepha suspensa* Loew, eggs and larvae in laboratory diet were exposed to combinations of 2, 10, or 20% O2 and 20, 50, or 80% CO2 at 15.6 or 10°C for 3, 5, 7, or 10 days. Multiple linear regression analysis indicated that, within the limits of this experiment, mortality did not vary with O2 concentration and was only slightly influenced by temperature. Insect response varied directly with CO2 concentration and time of exposure (Benschoter, 1987).

Temperature has been positively correlated to mortality in MA (Ke and Kader, 1992a). For example, as temperature increased in 2.5°C steps between 0 and 7.5°C, considerable increases occurred in mortality of overwintering European red mite, *Panonychus ulmi* Koch, eggs exposed to 5.0% CO2 and 3.0% O2 or 1.5% CO2 and 1.0% O2 (Lidster *et al.*, 1984). However, Hallman (1994) mentions a study in which 2-24-h-old codling moth eggs in an atmosphere of 20% CO2 and 0% O2 were killed with lower exposure times at 15°C than at 25°C.

Soderstrom *et al.* (1986) determined that the navel orangeworm was relatively more susceptible to CA treatment by reductions in RH or O2 than the Indianmeal moth, *Plodia interpunctella* Hübner. They hypothesized that this was because the Indianmeal moth, a storage pest, was adapted to the low RH and O2 found in storage conditions, whereas the navel orangeworm, a field pest, was adapted to higher RH and ambient O2 levels found in the field. Diapausing stages are usually the hardest to kill with MA (Soderstrom *et al.*, 1990; Whiting *et al.*, 1992).

2.3 Physiological effects of modified atmospheres on insects

Almost all of the research on MA as quarantine treatments has been on high levels of CO2 (>20%) and/or low levels of O2 (<5%). Other, unsuccessful, work involved CO (Klaustermeyer *et al.*, 1973) and low levels of ethylene, which is explosive at concentrations >3% (Hallman, 1994). AliNiazee (1972) did research on the effect of 100% N2 or helium, which is mentioned above. Nicolas and Sillans (1989) have discussed the physiological and biochemical responses of arthropods to low O2 and high CO2. Low O2 and elevated CO2 cause different physiological lesions with CO2 having multiple sites of action. However, resistance to CO2 has been induced in the granary weevil and the rice weevil, indicating non-multiple modes of action in those two insects (Bond and Buckland, 1979; Navarro *et al.*, 1985). Treatment of successive generations of these species with high CO2 concentrations produced insects with a 3-fold increase in tolerance to the gas in 7 generations. However, these two stored product pests responded differently to MA than other insects studied. Therefore, they should not be used as general examples of insect responses to MA, especially for pests of fresh commodities.

It is difficult to separate the effects of high CO2 from those of low O2. Friedlander (1983) suggested that, among other effects, high CO2 prevented the anaplerotic production of nicotinamide adenine dinucleotide and inhibited the regeneration of choline to acetylcholine. In the absence of O2, insect development virtually stops and survival depends on the accumulation of glycolytic products and reduction in metabolism; O2 deficiency inactivates the mechanism of selective permeability of the nerve sheats (Miller, 1966), while O2 deficiency and excess CO2 cause spiracles to open resulting in water loss.

Actual death of the insect in low O2 treatments may occur when metabolic products accumulating in the insect nervous system due to halted or slowed metabolic pathways reach toxic levels (Hallman, 1994). Death caused by high CO2 may be the result of dehydration due to open spiracles which then cause changes in the ionic balance in nerve ganglia. Severe dehydration could not occur to insects inside fresh commodities and maybe not to surface pests of fresh comodities stored under high humidity, either. However, chemical changes occurring in fruit with MA, such as an increase in the products of anaerobic metabolism, including ethanol, ethyl acetate, and acetaldehyde (Nisperos-Carriedo *et al.*, 1990), may be toxic and contribute to insect mortality. Acetaldehyde (2.0% for 4 h at 21°C) killed 100% of western flower thrips, *Frankliniella occidentalis* Pergande, on strawberries (Aharoni *et al.*, 1979).

2.4 Modified atmosphere quarantine treatments

2.4.1 Controlled atmospheres

2.4.1.1 External pests

Some studies that tested the effect of standard CA storage on pests of fresh commodities determined that the low temperatures and not the atmospheric conditions used were responsible for insect mortality (Glass *et al.*, 1961; Dickler, 1975). Klaustermeyer *et al.* (1973) were the first to publish a study using CA to attempt control of surface pests of fresh commodities. Various CA combinations did not cause significant mortality to one aphid and two caterpillar species on lettuce at 20°C. However, the lowest level of O2 used was 5%, and many of the treatments lasted only 1 or 2 h; lower O2 levels and longer treatment times should be tested. Complete mortality of San Jose scale, *Quadraspidiotus perniciosus* Comstock, on apples was achieved in 2 days with >90% CO2 and <1% O2 at >12°C and in one day with 96% CO2 at 22°C (Morgan and Gaunce, 1975).

Aharoni *et al.* (1981) attained 100% mortality of western flower thrips on strawberries after holding the berries in 88.7-90.6% CO2, 1.9-2.3% O2 at 2.5°C for 48 h. This procedure could be a viable method to replace methyl bromide fumigation for shipment of strawberries from California to Japan. However, a difference in flavour of treated strawberries was noted by 80% of the tasters and should be further investigated.

All apple rust mite, *Aculus schlechtendali* Nalepa, and European red mite eggs on apples were killed by storage in 1.0% O2 plus 1.0% CO2 at 2.8°C for 160 days without reducing fruit quality (Lidster *et al.*, 1981). Mortality of European red mite eggs was increased significantly if the atmosphere was 1.5% CO2 and 1.0% O2 when the temperature was increased from 0 to 7.5°C (Lidster *et al.*, 1984). Both of these studies, and another showing a minor loss of apple quality (Lidster, 1982), suggest the possibility of providing quarantine security for apples infested with mite eggs.

Mites from five different families, the greenhouse thrips, *Heliothrips haemorrhoidalis* Bouché, and the lightbrown apple moth, *Epiphyas postvittana* Walker, on Japanese persimmons were killed within 7 days' exposure to 0.5% O2, 5.3% CO2 (Hallman, 1994). However, survival of longtailed mealybug, *Pseudococcus longispinus* Targioni-Tozetti, was 0.2% after 7 days under the same conditions. Hallman (1994) mentions a CA treatment that resulted in one successful shipments of asparagus from New Zealand to Japan. The asparagus were held in 60% CO2 in air at 0-1°C for 4.5 days to kill New Zealand flower thrips, *Thrips obscuratus* Crawford, and green peach aphid, *Myzus persicae* Sulzer.

2.4.1.2 Internal pests

CA storage conditions for apples (2.5% O2, 1-1.5% CO2 at approximately 1°C) did not provide quarantine security of diapausing codling moth larvae within 133 days (Moffitt and Albano, 1972). A lower O2 (0.5%), higher CO2 (10%) atmosphere at 25°C was also unsuccessful, although 60% CO2 might provide quarantine security for walnuts stored at 60% RH (Soderstrom *et al.*, 1990). Toba and Moffitt (1991) achieved 100% mortality of an estimated 142 021 non-diapausing larvae of codling moth in apples treated with 1.5-2.0% O2 and >1% CO2 at 0±0.28°C, 95% RH for 13 weeks. These conditions could provide a feasible quarantine treatment for apples infested with codling moth where diapausing larvae were not of concern.

The apple maggot, *Rhagoletis pomonella* Walsh, is a tephritid fruit fly, which causes great damage to apple in the USA. AliNiazee *et al.* (1989) achieved 100% mortality of all stages after 8 days in 100% at 20°C. Lowering the temperature to 0°C prolonged the time needed for 100% mortality to 21 days. 'Golden Delicious' apples developed off-flavours in 21 and 2 days in 100% N2 at 0 and 21°C respectively. 'Bartlett' pears required 12 days in N2 at 0°C to develop detectable off-flavours, whereas 'Granny Smith' apples and 'Anjou' pears did not exhibit anaerobic off-flavours when held in N2 at 0°C for up to 24 days.

Delate et al. (1990) achieved 100% mortality of sweetpotato weevil, *Cylas formicarius elegantulus* Summers, adults in 7 days using several low O2 and high CO2 combinations at 25°C, 75% RH. These treatments can only be applied to cured sweet potatoes, as curing does not occur at O2 concentrations <8%. Curing (4-7 days, 30°C, 95% RH) is required to allow wound healing prior to storage.

Maximum mortality of blueberry maggots subjected to 2, 5, or 20% O2, plus 0.033-985 CO2 at 5 Or 21°C for 48 h was 91% (Prange and Lidster, 1992). Lower O2 levels would probably be needed to achieve mortality required for quarantine security.

Four species of tortricids (Lepidoptera), that attack apples, were killed by 0.4% O2 and 5% CO2 at 40°C in 14.3 h (Whiting *et al.*, 1992). It remains to be seen whether apples could tolerate these treatments.

Additional research is needed on combining CA with other treatments. For example, CA and heat have been shown to be synergistic. The highest temperatures used in most CA studies have been ambient. Whiting *et al.* (1992) reduced the LT99 for fifth instar codling moth from 350 to 14.3 h in 0.4% O2 and 5% CO2 when the temperature was raised from 20 to 40°C. In some instances, cold may be combined favourably with CA. High levels of CO2 synergized fumigants (Bond and Buckland, 1978).

Reduced atmospheric pressure was used to control the green peach aphid on harvested head lettuce. Exposing lettuce heads to a pressure of 2.66 kPa at 2°C for 52 hr resulted in 100% aphid mortality, whereas the lettuce marketing quality was not affected. When the atmospheric pressure is reduced, the partial pressure of the atmospheric gases is reduced in direct proportion. Since the incoming air to the reduced pressure chamber was water-saturated and the water pressure at 2°C is

0.705 kPa, the total gas pressure in the chamber at 2.66 kPa is only 1.95 kPa, and the O2 partial pressure is 0.399 kPa, as compared with 21.1 kPa at atmospheric pressure (Aharoni *et al.*, 1986).

2.4.2 Film wraps

Film wraps are used to extend the storage life of fresh commodities. Film wraps modify the atmosphere inside of a fruit; O2 is generally lowered while CO2 levels are increased. Shetty *et al.* (1989) were the first to use plastic film wrap to kill insects inside fruits. Film wrap was found promising as a quarantine treatment though improved wrapping techniques and wraps were needed to ensure quarantine security. Gould and Sharp (1990) obtained 99.95% mortality of Caribbean fruit fly immatures in mangoes after the fruits had been film wrapped for 15 days. Unfortunately, the fruit deteriorated after being film wrapped for only 6 days. Additionally, it was found that third instar Caribbean fruit flies were able to penetrate film wrap on grapefruits even when two layers were applied (Hallman, 1994).

Table 2.1. Studies of film wraps used to kill insects inside different fruits (Hallman, 1994).

Insect	Fruit	Film	Mortality at	Temperature	Reference
Drosophila melanogaster	Mango	Cryovac D-955	100% at 3 days[1]	24-25°C	Shetty *et al.* (1989)
Bactrocera dorsalis	Papaya	Cryovac D-955	>95% at 6 days	27°C	Shetty *et al.* (1989)
Bactrocera cucurbitae	Papaya	Cryovac D-955	≈85% at 6 days[2]	22-24°C	Jang (1990)
Ceratitis capitata	Papaya	Cryovac D-955	≈90% at 6 days[2]	22-24°C	Jang (1990)
Anastrepha suspensa	Mango	Clysar EHC-150	99.95% at 15 days	24-26°C	Gould+Sharp(1990)
Anastrepha suspensa	Grapefr.	ClysarEHC-50-F	92.2% at 35 days	24-26°C	Sharp (1990)
Anastrepha suspensa	Grapefr.	ClysarEHC-150-F[3]	97.5% at 35 days	24-26°C	Sharp (1990)

[1] Number of insects tested not given; therefore, confidence limits for 100% mortality cannot be determined.

[2] Jang (1990) suggested that improper wrapping may have reduced mortality level.

[3] Fruit double wrapped.

Individual film wraps have not provided quarantine security (Probit 9) against any of the fruit flies studied (Table 2.1). However, the fact that 100% of the pomace fly, *Drosophila melanogaster* Meigen, was killed after 3 days of shrink wrap is significant as this insect was much harder to kill in raisins stored in <0.5% O2 and 12-14% CO2 than several beetle and moth species (Soderstrom and Brandl, 1984).

It was suggested to combine film wrap with heat or cold treatments (Hallman, 1994). Combining film wrap with heat treatments may be synergistic for insect kill as was the combination of heat and CA (Ke and Kader, 1992a). Cold was usually not synergistic with CA (Gaunce *et al.*, 1982).

Film wrap should also be studied in combination with irradiation and fumigation as plastic films vary in their permeability to fumigants (Houck *et al.*, 1989). the mode of action of films in killing insects should be examined. All available films should be tested for quarantine purposes. Films should be designed specifically for killing insects inside wrapped commodities.

2.4.3 Coatings

Coatings, like film wraps, were developed to preserve the quality of fresh commodities. Hallman (1994) mentions several reports about coatings applied to cherimoya, which killed the mite *Brevipalpus chilensis* Baker, present on the fruit surface, and which provided quarantine security. The result of this study was the first use of a coating as a quarantine treatment. The mode of acting probably involes physically restricting the mites and sealing of the tracheal and alimentary systems. Therefore, the mortality is not caused entirely by the MA treatment.

Studies on the effect of methoprene-impregnated waxes on larvae of the Mediterranean fruit fly and oriental fruit fly, *Dacus dorsalis* Hendel, in papayas and peaches presented data showing that wax alone killed insects inside fruits (Saul *et al.*, 1985, 1987). Controls comprising wax-coated fruit without methoprene yielded estimated mortalities of 20-89%. However, this observation was not examined further. Coatings varied in their ability to achieve mortality (Hallman, 1994). Coatings showed a synergistic reaction with hot air treatment but not with cold, irradiation, or methyl bromide treatments (Hallman, 1994). Available coatings should be tested for their ability to kill insects inside fruits. Coatings differ significantly in permeability to atmospheric gases (Hagenmaier and Shaw, 1992). Novel materials, such as chitosan and soyabean proteins, are being investigated as coatings to maintain quality of fresh commodities. Coatings should be designed with the specific purpose of killing insects as well as preserving commodity quality; information on the mode of action of coatings would aid in their development. Besides reduction in O2 and increase in CO2, coatings also may cause increased production of volatiles inside the fruits, which could affect insect survival (Nisperos-Carriedo *et al.*, 1990). Combination treatments using coatings with other treatment procedures, such as forced hot air, need to be studied.

The fruit coatings Primafresh 31, Sta-Fresh 360HS, Sta-Fresh 600, and Nature Seal were studied for their ability to kill Caribbean fruit fly larvae infesting fruits. Mortality in grapefruit ranged from 33% (Primafresh 31) to 100% (Sta-Fresh 600). Methyl cellulose and shellac, known to restrict gaseous exchange, were identified as substances in Nature Seal that reduced survival of

Caribbean fruit fly; Nature Seal with 4% methyl cellulose and 17% shellac caused 99.2% mortality in grapefruits. Coating of cold-stored mangoes and carambolas did not increase Caribbean fruit fly mortality. Hot-air treatment of Nature Seal (2% methyl cellulose plus 10% shellac)-coated grapefruits increased Caribbean fruit fly mortality compared with uncoated fruits; when both were exposed to hot air at 48°C for 60 min, no larvae survived treatment of coated grapefruits, whereas 24% survived treatment of uncoated grapefruits. Carambolas immersed in 46°C Nature Seal heated slower than those immersed than those immersed in 46°C water. Also, more damage occurred to carambolas immersed in Nature Seal than in water at 46°C. CO2 and alcohol levels were higher in coated grapefruits, and O2 levels were lower compared with uncoated grapefruits. Coatings may have potential as possible quarantine treatments of interior pests, if not alone then in combination with other treatments. Coatings might also be considered as part of an integrated pest management system of fruit pests (Hallman *et al.*, 1994).

2.4.4 Other modified atmosphere treatments

Dentener *et al.* (1992) investigated the use of polymeric film bags containing an O2 remover to kill insect pests found on persimmons. Complete mortality of lightbrown apple moth, *Epiphyas postvittana* Walker, and longtailed mealybug, *Pseudococcus longispinus* Targioni-Tozetti, was achieved after 4 days at 20°C followed by 14 days at 0°C. Although atmospheric conditions were not precisely controlled, the treatment shows potential.

3 EFFECTS OF INSECTICIDAL MA ON HORTICULTURAL COMMODITIES

Fresh horticultural products vary greatly in their relative tolerance to low O2 concentrations and elevated CO2 concentrations during long-term storage (Kader *et al.*, 1989). These tolerance limits are the levels below which (O2) or above which (CO2) physiological damage would be expected, and can be different at temperatures above or below those recommended for each commodity. A given commodity may tolerate much higher CO2 concentrations and/or lower O2 concentrations for a short duration. The tolerance to low O2 concentrations decreases with the increase in temperature and/or duration since O2 requirements for aerobic respiration of the commodity increase with rise in temperature (Boersig *et al.*, 1988). Depending on the commodity, damage associated with elevated CO2 may either increase or decrease with an increase in temperature. CO2 production increases but its solubility decreases with temperature. Further, the physiological effects of CO2 could be temperature dependent. Tolerance limits to elevated CO2 decrease with a reduction in O2 level, and similarly the tolerance limits to reduced O2 decrease with the increase in CO2 level (Kader and Ke, 1994).

Generally, the effect of reduced O2 and/or elevated CO2 on lowering the respiration rate has been assumed to be the primary reason for the beneficial effects of MA on fresh produce. Kader (1986) indicated that postharvest deterioration of fresh produce can be caused by many factors in addition to high respiration rates, including biochemical changes associated with respiratory metabolism, ethylene biosynthesis and action, compositional changes, anatomical changes associated with growth and development, physical injuries, water loss, physiological disorders, and pathological breakdown. CA containing less than 1% O2 and/or more than 60% CO2 have been shown to be effective in controlling insects of stored products, such as nuts and dried fruits and vegetables.

3.1 Responses of horticultural commodities to insecticidal MA

Keeping fresh horticultural products in very low O2 and/or very high CO2 atmospheres may have beneficial effects, such as reducing respiration rate, inhibiting ethylene production and action, retarding colour change and softening, maintaining composition and nutritional value, reducing the incidence of some physiological disorders, and inhibiting decay. The specific beneficial effects observed depend on the commodity, maturity stage, storage temperature, atmospheric composition, and time of exposure.

As O2 level is reduced to 1-3% or CO2 level raised to 5-20%, respiration rate usually decreases. However, if O2 concentration is reduced to near 0% or CO2 level increased to 50-90%, respiration rate may be similar to or even higher than that of the commodities kept in air since anaerobic respiration could occur (Ke *et al.*, 1991b). For most commodities, decreasing O2 level to 1% or below and/or enhancing CO2 level to 50% or above greatly inhibits ethylene production and action (Ke *et al.*, 1990, 1991b,c). Investigation was done on the effects of 20% O2 + 60% CO2 on respiration and ethylene production rates of 18 kinds of fruits and vegetables. It was found that this atmosphere decreased respiration rates in climacteric fruits and broccoli, but the effect on respiration rates of non-climacteric commodities was small. They proposed that the respiratory responses of horticultural crops to high CO2 might be mediated by the effects of CO2 on the action and/or synthesis of ethylene (Kubo *et al.*, 1990).

Insecticidal low O2 and/or high CO2 atmospheres retard skin yellowing in apples and 'Bartlett' pears and skin darkening in plums and cherries (Ke *et al.*, 1990, 1991b; Ke and Kader, 1992b). Exposure to insecticidal MA also retards flesh softening in apples, 'Bartlett' pears, plums, and strawberies (Ke *et al.*, 1990, 1991a,b). For Asian pears, oranges, peaches, and nectarines, insecticidal MA do not influence their normal changes in colour and flesh firmness (Ke and Kader, 1990, 1992b; Ke *et al.*, 1991b,c).

Short-term exposures to insecticidal MA usually do not significantly influence soluble solids content. In some commodities, such as peach and apple, loss of organic acids may be retarded (Ke *et al.*, 1991b,c). Insecticidal MA treatments do not significantly influence vitamin C content in strawberries and oranges (Ke and Kader, 1990; Ke *et al.*, 1991a). For long-term storage of apples and pears, however, reducing O2 level to 1-3% with or without the addition of 3-5% CO2 maintains higher levels of soluble solids content, titratable acidity, and amino acids (Mellenthin *et al.*, 1980; Chen and Mellenthin, 1982). Through these beneficial effects, dessert quality is maintained.

Shaw *et al.* (1991) stored 'Hamlin', 'Pineapple', and 'Valencia' oranges in CA of 100% N2, 100% CO2, or 0.1-0.7% acetaldehyde in air, for 8 to 24 h. These treatments caused two- to threefold increases in acetaldehyde, ethyl acetate, ethyl butyrate, and ethanol within 24 h after treatment, but no consistent flavour changes in juice were noted for up to 8 days after treatment. Pesis *et al.* (1991) found that treating feijoa fruits with 98% N2 for 24 h enhanced their flavour quality.

Insecticidal MA treatments reduce severity of internal breakdown (chilling injury) in peaches (Ke *et al.*, 1991c) and some browning and scald disorders in apples and pears (Mellenthin *et al.*, 1980; Lau, 1983; Chen *et al.*, 1985). Lowering O2 level to below 1% or increasing CO2 level to 10-30% substantially reduces decay incidence in strawberies, blueberries, and cherries (Patterson, 1982; Smittle and Miller, 1988; Ke *et al.*, 1991a). These treatments may have a good potential for controlling postharvest decay, especially for the soft fruits that usually have very short storage life under normal cold storage in air. The effects of MA on decay may include the direct effect on inhibiting pathogen growth and the indirect effect on delaying ripening and senescence of the commodities and consequently maintaining their resistance to pathogen attack (El-Goorani and Sommer, 1981).

3.2 Tolerance of horticultural commodities to insecticidal MA

The possible detrimental effects of insecticidal MA on fresh commodities include low O2 and/or high CO2 injury and off-flavour development. The period of tolerance of fruits and vegetables to insecticidal MA conditions is, therefore, limited to the time before the onset of these effects. The tolerances of some fruits to insecticidal low O2 and/or high CO2 atmospheres are summarized in Table 2.2. Delate and Brecht (1989) found that sweet potato roots tolerated exposure to 2% O2 + 40% CO2 at 25°C for up to 10 days.

Table 2.2. Possible controlled atmosphere quarantine treatments (Hallman, 1994).

Commodity	Pest	O2 (%)	CO2 (%)	Temp. (°C)	Time (days)	Reference
Apple	San Jose scale	<1	>90	>12	2	Morgan + Gaunce (1975)
Apple	San Jose scale	0	96	22	1	Morgan + Gaunce (1975)
Apple	Codling moth larvae	1.5-2	<1	0	91	Toba + Moffitt (1991)
Apple	Apple rust and European red mites	1.0	1.0	2.8	160	Lidster *et al.* (1981)
Apple	Four torticid species	0.4	5.0	40	>0.6	Whiting *et al.* (1992a,b)
Asparagus	Green peach aphid and New Zealand flower thrips	8.4[1]	60	0-1	4.5	Carpenter + Potter (1994)
Strawberry	Western flower thrips	1.9-2.3	88.7-90.6	2.5	2	Aharoni *et al.* (1981)
Sweet potato[2]	Sweet potato weevil	4	60	25	7	Delate *et al.* (1990)
Sweet potato[2]	Sweet potato weevil	2	40	25	7	Delate *et al.* (1990)
Sweet potato[2]	Sweet potato weevil	2	60	25	7	Delate *et al.* (1990)
Walnut	Codling moth larvae	8.4[1]	60	25	>14	Soderstrom *et al.* (1990)

[1] O2 level estimated from 40% air.

[2] Cured sweet potato only.

For a few commodities such as Asian pear, nectarine, and papaya, low O2 injury could be observed after exposure to 0-0.25% O2 for 3-14 days (Yahia *et al.*, 1989; Ke and Kader, 1990, 1992b; Ke *et al.*, 1991b). Smilanick and Fouse (1989) observed MA-induced damage in nectarines after only 3 days at 0.5% O2. For most other commodities, low O2 injury is not observed during short-term low O2 treatments. Most commodities could tolerate CO2 levels in the 10-30% range for only a few days. The exceptions include cherry, strawberry, and blueberry that can be kept in these CO2 atmospheres for more than 10 days without detrimental effects (Chen *et al.*, 1981; Patterson, 1982; Smittle and Miller, 1988; Ke *et al.*, 1991a). With CO2 levels at 50-100%, however, injury could be observed in all commodities after 3-8 days of treatment (Mitchell *et al.*, 1984; Yahia *et al.*, 1989; Ke and Kader, 1990; Ke *et al.*, 1990, 1991a). Symptoms of low O2 and/or high CO2 injury may include internal and/or external browning and skin pitting (Ke and

Kader 1990, 1992b; Ke *et al.*, 1991a,b). the injured fruits may fail to ripen after removal to air and show increased susceptibility to decay.

Another very common detrimental effect on exposing fruits and vegetables to insecticidal MA is off-flavour development. This is generally thought to be caused by the accumulation of fermentation products such as ethanol, acetaldehyde, ethyl acetate, and probably some other volatiles under the very low O2 and/or very high CO2 conditions. At the early stage of off-flavour development, slight alcoholic flavour could be detected; at a later stage, unpleasant off-flavour occurs. Ethanol and acetaldehyde contents greatly increase during off-flavour development, with ethanol as the predominant volatile (Nichols and Patterson, 1987; Patterson and Nichols, 1988; Ke and Kader, 1990, 1992b; Ke *et al.*, 1991a,b).

For many commodities, such as apple, 'Bartlett' pear, plum, cherry, orange, and strawberry, off-flavour development is the only detrimental effect observed following short-term exposure to O2 level at or below 1% (Ke and Kader, 1990, 1992b; Ke *et al.*, 1990, 1991a,b). The major factors that influence off-flavour development include the O2 level used, storage temperature, respiration rate, resistance to gas diffusion, soluble solids content, and ethanol accumulation rate of the commodity under a low O2 treatment (Ke *et al.*, 1991b; Ke and Kader, 1992b).

Physiological and biochemical responses of mango, papaya and avocado fruits to atmospheres that have potential as insect quarantine treatments (<0.5% O2 and >50% CO2) were evaluated (Yahia, 1993). Avocado fruit were very sensitive, mango fruit were very tolerant, and papaya fruit were intermediate in their level of tolerance for such extreme atmospheres. Insecticidal atmospheres resulted in changes in glycolytic and tricarboxylic acid cycle activities and slightly increased the activity of alcohol dehydrogenase (ADH) and pyruvate decarboxylase (PDC) in mango, but caused no fruit injury nor detrimental organoleptic changes after exposure for up to 5 days. Papaya developed decay and off-flavours, and had increased activity of PDC and lactate dehydrogenase (LDH) after 2 days of treatment. Avocado developed injury, had increased production of ethanol and acetaldehyde, had increased activity of anaerobic enzymes, and had decreased concentration of 3 glycolytic intermediates after one day of exposure to these atmospheres. Because of the differences in their responses and level of tolerance, these 3 fruits could serve as excellent systems to study the basis of fruit tolerance to MA.

The physiological response of mango fruit to vapour-heat treatments was explored. Mangoes, cv.

Tommy Atkins, were treated at 46°C for 160, 220 or 280 min and at 50°C for 120, 180 or 240 min. 'Keitt' mangoes were treated at 46°C for 180 or 240 min and at 48°C for 300 min. In vapour-heated 'Keitt' mangoes, CO2 accumulated to 13% and O2 decreased to 6% in the internal atmosphere. Ethanol, methanol and acetaldehyde concentrations increased, electrolyte leakage increased and ethylene-forming enzyme (EFE) activity decreased immediately after heat treatment. Three days after treatment, EFE activity recovered and electrolyte leakage decreased to control levels. Vapour-heat treatment also reduced the rate of fruit softening and mesocarp colour development. Mango fruit apparently have the capacity to recover from vapour-heat quarantine treatments (Mitcham and McDonald, 1993).

The effects of a range of CO2 (0, 10, 30, and 50%) and O2 (0, 0.5, 2, and 21%) concentrations, separately and in combination, were tested on the quality of roses, carnations, broccoli and iceberg lettuce at 0 or 5°C (Zheng *et al.*, 1993). The test products survived these storage conditions with minimal phytotoxic symptoms (Appendix 3). An atmosphere of 10% CO2 at 0 or 5°C had no detrimental effect on the quality and vase-life of roses or carnations at 20°C. the overall quality (visual quality, injury, off-odours evaluated 4 h after transfer to air at 20°C) of iceberg lettuce was satisfactory after exposure to 10% CO2 at 0°C for 6 days, and that of broccoli was unaffected by this atmosphere at either 0 or 5°C. Under low O2 atmospheres (0.5 or 2%) there was no significant damage or loss in quality of roses or broccoli after 3 weeks at 5°C. In contrast, lettuce showed injury in 0.5% O2 after 2 weeks. Analysis of major anaerobic volatiles showed that large increases in ethanol and acetaldehyde concentrations correlated with off-odour detection.

3.3 Models to predict tolerances of fruits and vegetables to insecticidal CA

Kader and Ke (1994) developed a multiple regression model for quick prediction of fruit and vegetable tolerance to insecticidal low O2 atmospheres, which will be described here. Decreasing O2 level to near 0% causes faster off-flavour development. Increasing storage temperature dramatically enhances off-flavour. If the relative tolerance of fruits to 0.25% O2 at 0°C is used as a reference point (100%), then increasing O2 concentration to 1.05 at the same temperature usually increases the tolerance threefold; on the other hand, raising the temperature to 10°C at the same O2 concentration decreases the tolerance to about 35%. A commodity with higher respiration rate and greater resistance to gas diffusion is less tolerant to insecticidal low O2 atmospheres (Ke and Kader, 1992b). On the other hand, higher soluble solids content requires more ethanol to cause off-flavour, probably due to the interaction between soluble solids and ethanol in determining flavour

(Ke *et al.*, 1991b; Ke and Kader, 1992b). For example, Asian pear, 'Yellow Newtown' apple, and plum had 10.7%, 13.4%, and 15.3% average soluble solids contents and their threshold ethanol concentrations for off-flavour detection were about 300, 1000, and 3000 $\mu l\ l^{-1}$, respectively. Flavour and ethanol content appear to have a logarithmic relationship and the threshold ethanol content for off-flavour detection (E_o, $\mu l\ l^{-1}$) could be predicted by soluble solids content (*S*, %) of the commodity as follows: $E_o = 10^{0.228\,S}$ (Ke *et al.*, 1991b). It should be pointed out that tolerance to low O2 is not determined by a single factor and all the related factors should be considered. More mature fruits may have higher respiration rates and resistance to gas diffusion and they are consequently less tolerant to low O2. Over-ripe commodities should not be treated with low O2 and/or very high CO2 atmospheres due to their low tolerance to these CA conditions.

By using soluble solids content (%) and average ethanol accumulation rate per day (V_E, $\mu l\ l^{-1}$ day^{-1}), a model has been developed for predicting fruit tolerance limit (T_l, day) to insecticidal low O2 atmospheres: $T_l = (10^{0.228\,S})/V_E$ (Ke *et al.*, 1991b). Using storage temperature (*T*, °C), O2 concentration (*C*, %), respiration rate (*R*, ml CO2 $kg^{-1}\ h^{-1}$), resistance to gas diffusion (*r*, % $(ml\ kg^{-1}\ h)^{-1}$), and soluble solids content of the commodity as variables, the following multiple regression model was developed (Ke and Kader, 1992b) for quick prediciton of fruit tolerance to insecticidal low O2 atmospheres:

$$T_l = -0.484T + 64.6C - 0.722R - 4.20r + 3.00S - 5.90T \times C - 18.2 \qquad [2.1]$$

Equation [2.1] indicates that relatively high temperature, respiration rate, and resistance to gas diffusion reduce fruit tolerance to low O2 atmospheres; relatively high O2 concentration and *S* increase fruit tolerance. The interaction between temperature and O2 concentration is also considered in this model.

A correlation coefficient of 0.902 was obtained for Equation [2.1] at $P<0.0001$. This model could reasonably predict T_l in most cases and the average difference between observed tolerance from experiment (T_{le}) and predicted tolerance is about 4 days. In Equation [2.1], *T* and *C* are predetermined or specified, *R* and *r* are measured after 3 days of low O2 treatment, and *S* is commodity specific, that is measured after the fruits are allowed to ripen. It should be noted that this regression model is limited to *T* of 0, 5, and 10°C and *C* of 0.25% and 0.02% O2. For other temperatures and O2 concentrations, this model should be used with care or modified as needed.

Since T influences both R and r, variables R and r are not completely independent in Equation [2.1]. The interaction between T and C also reduces the accuracy of using this model for predicting T_l. To improve predition accuracy, the following individual models for each temperature (0, 5, and 10°C) were developed (Ke and Kader, 1992b):

For 0°C

$$T_l = 54.2C - 12.2R - 7.23r + 6.56S - 39.8 \qquad [2.2]$$

For Equation [2.2], correlation coefficient = 0.984, $P = 0.013$, average difference between T_{le} and $T_l = 1.6$ days.

For 5°C

$$T_l = 14.5C - 9.90R - 1.07r + 7.68S - 56.0 \qquad [2.3]$$

For Equation [2.3], correlation coefficient = 0.986, $P = 0.011$, average difference between T_{le} and $T_l = 1.6$ days.

For 10°C

$$T_l = 11.4C + 0.352R - 1.88r + 1.30S - 10.4 \qquad [2.4]$$

For Equation [2.4], correlation coefficient = 0.963, $P = 0.046$, average difference between T_{le} and $T_l = 0.7$ days. In Equation [2.4], the regression coefficient of 0.352 is small compared to those of C, r, and S and statistically not significant ($P = 0.224$). Since the influence of R on T_l should be negative according to Equation [2.1], the involvement of R data in the model did not appear to be appropriate and it was removed to obtain the following model:

$$T_l = 10.3C - 2.60r + 1.06S - 4.35 \qquad [2.5]$$

For Equation [2.5], correlation coefficient = 0.934, $P = 0.029$, average difference between T_{le} and $T_l = 0.9$ days.

By predicting the tolerances of fruits and vegetables to insecticidal CA conditions, optimum temperature/O2 concentration/time combinations could be selected so that insects of concern may be controlled without detrimental effects on the fresh commodities (Kader and Ke, 1994).

4 POTENTIAL USE OF MA AS QUARANTINE TREATMENTS

Only when fruits and vegetables tolerate the low O2 and/or high CO2 conditions for a period longer than the time required to kill completely the insects of concern, can MA be used as a quarantine procedure. Such information can be obtained by comparing Appendix 1 and 2. For apple, most insects (brownheaded (*Ctenopseustis obliquana*) and greenheaded (*Planotortrix excessana*) leaf rollers, apple rust mite (*Aculus schlechtendali*), European red mite (*Panonychus ulmi*), codling moth (*Cydia pomonella*), light brown apple moth (*Epiphyas postvittana*), San Jose scale (*Quadraspidiotus perniciosus*), and wheat bug (*Nysius huttoni*)) may be completely killed by MA before detrimental effects occur.

For nectarine, the treatment involving exposure to 0.5% O2 and 25°C for up to 12 days which is required to control codling moth (Soderstrom *et al.*, 1987) is beyond the limits of tolerance of the fruits. However, other stone fruits such as cherry and plum are generally more tolerant to low O2 than nectarine. Further work is required to evaluate the feasibility of using low O2 and/or high CO2 to control insects in stone fruits.

For orange, 0.25% O2 may be effective in controlling Caribbean fruit fly (*Anastrepha suspensa*) without detrimental effects on the commodity. Thrips (*Heliothis spp.*) could be completely killed by 50-90% CO2 without injury to strawberry. For persimmon, leaf roller, mealy bug (*Pseudococcus longispinus*), and thrips could be completely killed by low O2 treatment before detrimental effects occur. Sweet potato weevil (*Cylas formicarius elegantulus*) may also be controlled by 2-4% O2 + 40-60% CO2 atmospheres.

Therefore, MA appears to have a good potential for postharvest quarantine treatments for some insects in certain fruits and vegetables, but much more research is needed before specific recommendations can be made.

5 CONCLUSIONS AND RECOMMENDATIONS

MA quarantine treatments of pests of fresh commodities have been developed and may provide quarantine security without damaging the commodity (Table 2.2). Other potential quarantine treatments cited by Ke and Kader (1992a,b) lack the information needed to indicate whether they would work (Appendixes 1 and 2). For example, in some studies few insects were tested, mortality may have been affected by other factors not controlled (such as low RH) or 100% mortality was estimated and not achieved. Other studies lack data on the tolerance of the commodity to the treatment. All of the treatments listed (Table 2.2) require further research before they can be applied; the exception may be shipment of asparagus from New Zealand to Japan, because a successful pilot shipment was made (Hallman, 1994). Approval of a MA treatment of apples from the nortwestern USA for codling moth (Toba and Moffitt, 1991) depends on convincing importing countries that diapausing larvae will not be present. A reduction in quarantine mortality requirements would advance the development of other potential MA quarantine treatments. Quarantine requirements often appear too strict for commodities with extremely low pest infestation levels (Vail *et al.*, 1993).

As with many quarantine treatment techniques, the precise modes of action of MA are not known. Also, the effects of other variables, such as host, on the efficacy of the treatments are not clear. Much of the research on MA was done for specific, practical reasons with little control over some modifying factors, making broad camparisons and conclusions precarious.

It bears repetition that entomologists and plant physiologists should coordinate their activities to perform comparable research and increased communication is needed between scientists working in CA grain storage and fresh commodity storage (Ke and Kader, 1992a; Hallman, 1994).

So far, most research has been focused on stored products and fruits. Vegetables and ornamental have been mainly ignored. Especially those commodities are produced in the Netherlands for the export market. If research is undertaken in the Netherlands special attention should be paid to the insect pests of vegetables and ornamentals and to the development of MA treatment of those pests.

General suggestions for future research are (Ke and Kader, 1992a):

1. Investigating the mode of action of low O2 and high CO2 atmospheres on various insects of quarantine significance in the marketing of fresh fruits and vegetables.
2. Studying the responses of insects at various developmental stages to those combinations of time, temperature, and MA composition which have been shown to be tolerated by the host commodities.
3. Developing practical methods for MA application as quarantine treatments during temporary storage and/or transport in gas-tight vehicles.
4. Elucidating the mechanism of low O2 and/or high CO2 injury in fresh horticultural commodities.
5. Understanding the mechanism of biochemical regulation of off-flavour development in fresh fruits and vegetables kept under stress levels of O2 and/or CO2.

6. Developing methods to reduce low O2 and/or high CO2 injury and off-flavour development and consequently increasing commodity tolerance to insecticidal MA.

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Appendix 1 (Ke and Kader, 1992a)

Table 1. Time required to kill certain insects in some fresh fruits and vegetables by controlled atmospheres.

Commodity	Insect		Temperature (°C)	CA compostition		Days for 100% mortality	Reference
	Species	Stage		O_2%	CO_2%		
Apple							
	Greenheaded/ (*Planotortrix excessana*)	First/third	0.5	3	3	59	Batchelor *et al.* (1985b)
	brownheaded (*Ctenopseustis obliquana*) leafrollers	First/fifth instars	0.5	3	3	32-60	Waddell *et al.* (1990)
	Apple maggot	Egg	0	0		24	Ali Niazee *et al.* (1989)
	(*Rhagoletis pomonella*)	Egg	20	0		7	Kosittrakun (1989)
	Apple rust mite (*Aculus schlechtendali*)	Egg	2.8	1	1	160	Lidster *et al.* (1981)
	European red mite	Egg	2.8	1	1	160	Lidster *et al.* (1981, 1984)
	(*Panonychus ulmi*)	Egg	7.5	1	1.5	14	
	Codling moth	Larvae	0	1.5-2	<1	91	Toba and Moffitt (1991)
	(*Cydia pomonella*)	Larvae	27	1	95	2	Gaunce *et al.* (1982)
	Light brown	First/fifth	0.5	3	3	32-60	
	apple moth	First/third	2	3	3	90	Waddell *et al.* (1990)
	(*Epiphyas postvittana*)	First	20	0.4	5	0.9	Batchelor *et al.* (1985b)
		First	30	0.4	5	0.5	Whiting *et al.* (1991)
		Fifth instars	40	0.4	5	0.2	
	San Jose scale	Cap/adult	2	3	3	112	Dickler (1975)
	(*Quadraspidiotus perniciosus*)	Cap/nymph	12	1	90	2	Morgan and Gaunce (1975)
	Wheat bug (*Nysius huttoni*)	Adult	0.5	3	3	59	Waddell *et al.* (1988)
Stone fruits							
	Codling moth	Adult	25	0.5	10	2-3	
	(*Cydia pomonella*)	Egg	25	0.5	10	2-3	Soderstrom *et al.* (1987, 1990)
		Pupa	25	0.5	10	6-12	
		Egg	25	8.4	60	2-3	
		Pupa	25	8.4	60	6-10	
Citrus							
	Caribbean fruit fly	Egg	15.6	0.5-2	30	10	Benshoter (1987)
	(*Anastrepha suspensa*)	Pupa	15.6	0.5-2	30	10	
Strawberry							
	Western flower thrips (*Frankliniella occidentalis*)	Adult/pupa	2.5	1	50-90	1-2	Aharoni *et al.* (1979, 1981)
Persimmon							
	Leafroller (*Planotortrix excessana*)	Third instar	20	0.5	5	4	
	Mealy bug (*Pseudococcus longispinus*)	Larvae/adult	20	0.5	5	7	Dentener *et al.* (1990, 1992)
	Thrips (*Heliothrips spp.*)	Larvae/adult	20	2-3	8-11	7	
Asparagus							
	Thrips	Adult	2	7	7	14	Batchelor *et al.* (1985a)
	Aphids	Adult	2	7	7	14	
Sweet potato							
	Sweet potato weevil	Adult	25	2-4	40-60	2-7	Delate *et al.* (1990)
	(*Cylas formicarius elegantulus*)	Adult	30	8	40-60	4-8	

Appendix 2 (Ke and Kader, 1992a)

Table 2. Tolerance of some fresh fruits and vegetables to controlled atmospheres.

Commodity	Temperature (°C)	CA composition		Tolerance (days)	Reference
		O_2%	CO_2%		
Apple	0	0.02		12	
	0	0.25		23	
	0	0.5		100	
	0	1	1-3	220	
	0	2-3	1-3	300	Chen *et al.* (1985)
	5	0.02		9	Couey and Olsen (1977)
	5	0.25		11	Dewey and Bourne (1982)
	5	0.5		55	Ke *et al.* (1991b)
	5	1	1-3	180	Lau (1983)
	5	2-3	1-3	210	Little *et al.* (1982)
	10	0.02		5	
	10	0.25		7	
	18		10-20	10-15	
European pear	0	0.02		>10	
	0	0.25		40	
	0	0.5		60	
	0	1	1-3	90	Chen and Mellenthin (1982)
	5	0.25		18	Ke *et al.* (1990)
	5	1		25	Mellenthin *et al.* (1980)
	10	0.25		10	Yoshida *et al.* (1986)
	10	1		>10	
	25	0.25		3-5	
	0-10	10-16	20-50	6	
	5-10	4.2	80	4	
Asian pear	0	0.02		14	
	0	0.25		14	Ke *et al.* (1991b)
	2	1-3		60-120	Zagory *et al.* (1989)
	5	0.02		14	
	5	0.25		14	
Cherry	0	0.02		25	
	0	0.25		35-44	
	0	1		56	Chen *et al.* (1981)
	5	0.02		21	Ke and Kader (1992)
	5	0.25		38	Patterson (1982)
	0		20-40	42	Patterson and Melsted (1977)
	0		60	7	
	0		80-100	4-7	
Nectarine	0	0.02		14	
	0	0.25		28	
	5	0.5		3-6	Ke and Kader (1992)
	10	0.02		10	Mitchell *et al.* (1984)
	10	0.25		13	Smilanick and Fouse (1989)
	15	0.5		2-3	Soderstrom *et al.* (1987)
	25	0.5		2-3	
	0	4.2	80	10	
	0	1	99	6	
Peach	0	0.02		40	
	0	0.25		>40	
	5	0.02		14	Ke *et al.* (1991c)
	5	0.25		14	Mitchell *et al.* (1984)
	0	4.2	80	6	
	0	1	99	3	
Plum	5	0.02		32	
	5	0.25		41	
	10	0.02		9	Ke *et al.* (1991b)
	10	0.25		14	Mitchell *et al.* (1984)
	0	1	99	5	
	5	4.2	80	6	
Orange	5	0.02		16	
	5	0.25		23	
	10	0.02		15	Ke and Kader (1990)
	10	0.25		20	
	5	8.4	60	5	

Appendix 2 (continued)

Table 2. (*Continued*)

Commodity	Temperature (°C)	CA composition		Tolerance (days)	Reference
		O_2%	CO_2%		
Strawberry	0	0.02		6	
	0	0.25		10	
	5	0.25		8	Ke *et al.* (1991a)
	5	1		>10	
	0-5		20	10-15	
	0-5	4.2-10	50-80	8	
Blueberry	5	0		14-21	Smittle and Miller (1988)
	5	5	10-20	42	
Persimmon	20	0.5	5	7-10	Dentener *et al.* (1990)
	20	3	33	10	Gazit and Adato (1972)
	20	0-4	80-100	2-5	Pesis and Ben-Arie (1986) Pesis *et al.* (1986)
Mango	20	0.1-0.2		5	
	20	2	50	5	Yahia *et al.* (1989)
	20	0.5	70-80	4	
Papaya	20	<0.5		2-3	Yahia *et al.* (1989)
Sweet potato	25	2	40	7-10	Delate and Brecht (1989)

Appendix 3 (Zheng *et al.*, 1993)

Table 1. Comparison of insect and commodity tolerances to controlled atmospheres.

Temperature and atmosphere		Days for 100% mortality[z]		Commodity tolerance (days)[y]			
		Aphids	Thrips	Rose	Carnation	Broccoli	Lettuce
0C	Air	>21	>21	>21		>21	>21
	2% O_2	>21	>21	>21		>21	>21
	0.5% O_2	>21	21	>21		>21	21
	0.02% O_2	7-14	14-21	15-20		9	13
5C	Air	>21	21	>21	>21	21	17
	2% O_2	14-21	14-21	>21	>21	>21	>21
	0.5% O_2	14-21	14-21	>21		>21	>21
	0.02% O_2	7	7-14	7-14		4	11
0C	Air	>6	>6	>14	>14	>14	>6
	10% CO_2	>6	6	>14	>14	>14	6
	30% CO_2	4	4	4	6	2	5
	50% CO_2	4	4	2	6	2	5
5C	Air	>6	>6	>6	>6	>6	>6
	10% CO_2	6	>6	>6	>6	>6	5
	30% CO_2	6	4	2	6	2	4
	50% CO_2	6	4	1	6	2	3
0C	Air	>14	>14	>14	>14	>14	>14
	2% O_2 + 10% CO_2	>14	7	>14	>14	>14	6
	1% O_2 + 10% CO_2	>14	7	>14	>14	>14	6
	0.5% O_2 + 10% CO_2	>14	7	>14	>14	>14	6
5C	Air	>14	>14	>14	>14	>14	>14
	2% O_2 + 10% CO_2	>14	14	>14	>14	>14	5
	1% O_2 + 10% CO_2	>14	7	>14	>14	>14	5
	0.5% O_2 + 10% CO_2	>14	7	>14	>14	>14	5

[z] Data was obtained with non-feeding insects. It took more time to completely kill feeding thrips, but not feeding aphids.

[y] Commodity tolerance was the number of days until slight injury and/or slight off-odor occurred, or visual quality decreased to fair (<7).