Do High Oxygen Atmospheres Control Postharvest Decay of Fruits and Vegetables?

Annette Wszelaki[1], Shimshon Ben Yehoshua[2], and Beth Mitcham[1]

Introduction

With the continued loss of currently used postharvest decay control measures (i.e. pesticides), there is a perpetual need to search for alternatives. One potential alternative, proposed recently by Day (1996), is the use of elevated oxygen atmospheres. He proposed that these atmospheres could control decay while also preventing off-flavor and odor development in fruits, which can be encountered with the traditionally used atmospheres of elevated carbon dioxide and low oxygen. In this article we will examine the potential of elevated oxygen as a postharvest decay control measure.

Oxygen Toxicity

Except for organisms especially adapted to live under anaerobic conditions, all plants and animals need oxygen for the production of energy and maintenance of life. Yet, oxygen is toxic to life at concentrations only slightly greater than in air (hyperbaric oxygen). Bert (1878) gave us the clear notion that oxygen at increased concentrations was toxic to living cells, showing both the universal nature of oxygen toxicity as well as the variation in sensitivity among species.

Toxicity of hyperbaric oxygen may be due to the unfavorable effects on the oxidation- reduction potential of the system, the oxidation of certain enzymes, especially those having sulfhydryl groups or disulfide bridges and the accumulation of injurious reactive oxygen products. Moreover, Gerschman et al. (1964) and Haugaard (1968) suggested lipid peroxidation as another cause of oxygen toxicity. However, the major explanation for oxygen toxicity, which gains widespread acceptance, is the formation of superoxide radicals (O_2^-), which are destructive to some aspects of cell metabolism (Fridovitch, 1975).

In general, aerobic bacteria and fungi are more resistant to oxygen toxicity than are mammalian cells. Their growth usually resumes when the organisms are returned to air (Haugaard, 1968). It appears that fungi are substantially more resistant to high oxygen than are plant (Caldwell, 1956) and animal tissues (Bean, 1945), although not as resistant as some bacteria (Caldwell, 1965). Oxygen is a substance universally toxic to living cells. It is only by developing special defense mechanisms that organisms can survive the ever present oxidizing potential of the oxygen in their surroundings (Haugaard, 1968).

Searching the available literature suggests these atmospheres might be more damaging to the fruit than to the microorganisms. However, too little research on commodity response and on decay control has been done. Studies on the effects of hyperbaric oxygen on ripening parameters, such as respiration, ethylene production and color formation on plums (Claypool & Allen, 1951), avocados (Biale, 1946) and tomatoes (Frenkel & Garrison, 1976), have shown that generally these attributes increase with increasing oxygen levels. No obvious phytotoxicity was reported.

Case Studies

Studies on the effect of elevated oxygen on microorganisms themselves have been few and quite variable in their results. Caldwell (1965) found that exposure of bacteria and fungi to 10-atm oxygen suppressed their growth completely. On return to air, the fungi began to grow after a variable delay period, whereas the bacterial cultures, with one exception, resumed growth immediately. It is worthy to note that this effect was not solely due to elevated oxygen concentrations, but also to the increase in atmospheric pressure.

Robb (1966) reported that of 103 species of fungi exposed to high oxygen concentrations (10-atm pressure) for 7 days, 52 species resumed growth after treatment. Of these species, 22 recovered after 14 days of exposure. On resumption of growth, after a lag period between decompression and recovery, growth rates were the same as those of untreated colonies. This group of 22 fungi included several species of *Aspergillus*, as well as six species of *Penicillium*.

More detailed investigation of the reactions of *Fusarium solani, Rhizopus arrhizus* and *Mucor racemosus* showed that the lag period of growth following exposure to high oxygen generally increased with increasing exposure times. The extinction points, that is the exposure that killed all replicates, varied and at exposures approaching the extinction point there was selective survival either of spores or strains of these three fungi.

Recently, Day et al. (1996) suggested that high oxygen atmospheres are advantageous for use in modified atmosphere packaging to directly inhibit decay-causing organisms. Gonzalez and Day (1998) later suggested that 99% oxygen alone did not prevent the growth of any of the following microorganisms tested: *Pseudomonas fragi, Aeromonas hydrophila, Yersinia enterocolitica* and *Listeria monocytogenes*. They found that the *P. fragi* and *A. hydrophila* were inhibited only slightly (14 and 15%, respectively). However, the combination of high oxygen and 20% carbon dioxide significantly inhibited all the organisms tested. They also found that the combined effect of oxygen and carbon dioxide was enhanced at lower temperatures. This is not surprising as atmospheres of 10% carbon dioxide and greater are well-known fungistats and are commercially used for some commodities (Mitchell, 1992). However, many commodities cannot tolerate these high carbon dioxide levels, which can initiate or aggravate physiological disorders; cause irregular fruit ripening, off flavors and odors; and even increase decay susceptibility (Kader, 1995).

Amanatidou et al. (1999) reported on the effect of elevated oxygen and carbon dioxide on the surface growth of various microorganisms associated with vegetables and considered important human pathogens. They found that in general exposure to 80 to 90% oxygen alone did not strongly inhibit microbial growth, but caused a significant reduction in the growth rate of some of the microorganisms tested, such as *Salmonella enteritidis, Salmonella typhimurium* and the biological control yeast *Candida guilliermondii*. In contrast, growth of other microorganisms was actually stimulated.

Carbon dioxide at 10 to 20% was much more effective in significantly reducing the growth of *Pseudomonas fluorescens* and of *Salmonella enteritidis*. The combined application of 80 to 90% oxygen together with either 10 or 20% carbon dioxide had an inhibitory effect on the growth of all microorganisms tested. In general, a notable prolongation of the lag phase and a reduction in the final population density was observed. The most prominent effect was with the yeast antagonists that were completely inhibited in their growth. They concluded that when high oxygen or high carbon dioxide were applied alone, the inhibitory effect on microbial growth was highly variable. Stronger and much more consistent inhibition of microbial growth was obtained when the two gases were used in combination (Amanatidou et al., 1999).

Recent Experiments

In a preliminary experiment, the effects of various concentrations of oxygen with and without 15% carbon dioxide on the decay of grapefruit were investigated. Most decay was caused by *Penicillium digitatum*. The only effective reduction of decay occurred with 80% oxygen, or its combination with 15% carbon dioxide, but not with 40% oxygen, or its combination with 15% carbon dioxide. It is interesting that 100% oxygen enhanced the *Penicillium* decay of grapefruit in this experiment (Kader and Ben Yehoshua, unpublished data).

In other experiments (Wszelaki and Mitcham, unpublished), the effects of elevated oxygen atmospheres *in vitro* on mycelial growth of *Botrytis cinerea*, as well as the effects of the atmospheres *in vivo* on strawberry decay were examined. Eight atmospheres (21 (air), 40, 60, 80, 90 & 100% oxygen, 40% oxygen + 15% carbon dioxide, and 15% carbon dioxide (balance air)) were applied to plugs of mycelia growing in Petri plates at 5°C (41°F). Diameter of the colony was measured after 5, 7 and 14 days of treatment, and also after 1 and 3 days at 20°C (68°F) to determine if the atmospheres provided any residual protection. After 5 days, the 80, 90 and 100% oxygen treatments showed significantly slower mycelial growth compared to the 21, 40 and 60% oxygen treatments, which were nearly identical in their growth patterns. However, 15% carbon dioxide and the combination of 40% oxygen + 15% carbon dioxide was much more effective at retarding *Botrytis* growth than any of the elevated oxygen treatments alone (Fig.1). After 7 days, the results were similar. However, after 14 days under elevated oxygen, the 100% oxygen treatment had surpassed both carbon dioxide treatments in myce-

lial growth suppression. The 90% oxygen treatment was intermediate between the carbon dioxide treatments, with greater suppression of growth than the 15% carbon dioxide, but less than the combination treatment (Fig.1). No residual protection in the delay of mycelial growth after 1 and 3 days at 20°C (68°F) was observed in any treatment.

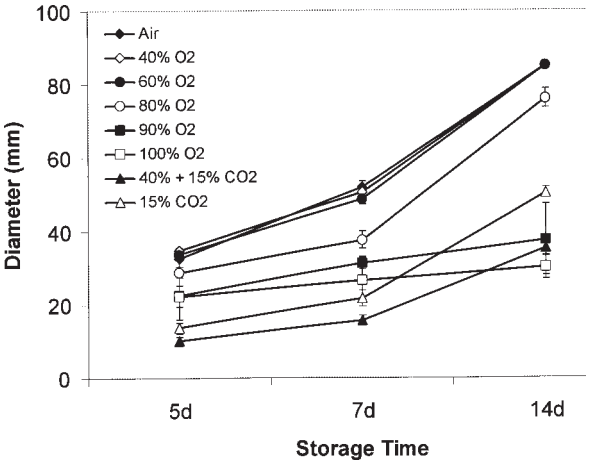

Figure 1. *Mycelial growth after 5, 7 and 14 days elevated oxygen treatment at 5 °C (41 °F). Each data point represents the average of 3 reps of 6 plates each. Error bars indicate one standard deviation.*

While the *in vitro* studies provide essential knowledge, they are not always correlated to decay growth on the fruit. Atmospheric effects on both the pathogen and the fruit can influence decay development on the commodity. Therefore, it was important to look at the effects of the atmospheres directly on the decay development of strawberries (cv. Camarosa). The same eight atmospheres were used as for the mycelial growth experiment. The fruit were evaluated for decay after 5 and 14 days of treatment, as well as after an additional 2 days at 20°C (68°F) in air to simulate market conditions. After 5 days of treatment, decay levels were the same for all treatments. While after 14 days of treatment, there was a decrease in decay with an increase in oxygen concentration above 40% (Fig.2). The 90 and 100% oxygen treatments had significantly less decay than either the 15% carbon dioxide by itself or in combination with 40% oxygen. However, after 14 days under the atmospheres plus 2 days at 20 °C (68°F), decay of the fruit from the 90 and 100% oxygen and 15% carbon dioxide treatments was similar, although it remained less than that of the other treatments (Fig.2). The strawberry

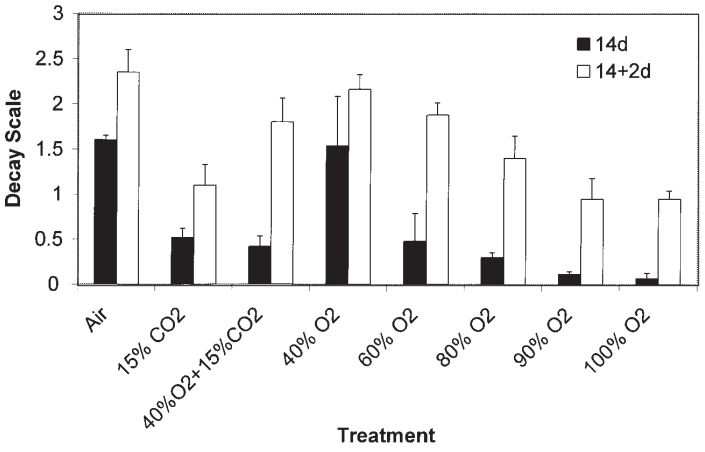

Figure 2. *Strawberry (cv. Camarosa) decay after 14 days elevated oxygen treatment at 5 °C (41 °F). In 14+2d, the additional two days indicate the time the fruit was held at 20 °C (68 °F) to simulate market conditions. Decay scale: 0=no decay, 1=slight, 2=moderate, 3=severe. Error bars indicate one* standard deviation. n=20

fruit did not appear damaged by the high oxygen treatment and quality parameters were similar for all treatments. However, fermentative volatiles accumulated in all treatments.

Conclusion

While these treatments have shown some promise in our and others data, the question of commercial feasibility must be addressed. It appears that the oxygen atmospheres that are most effective for decay control are those close to 80% or those in combination with carbon dioxide. These could be difficult to maintain either in a package or on a larger scale, as well as perilous in a commercial situation. Furthermore, the decay control does not seem to be greatly superior to currently used methods of 10-15% carbon dioxide. One aspect of the potential benefits of elevated oxygen treatment that is still under analysis in our laboratory is the effect on fruit quality and anaerobic fermentation products. If elevated oxygen can hinder decay without compromising fruit flavor or odor, unlike some high carbon dioxide treatments, which can produce off-flavors and odors, it deserves further investigation as a control measure. Alternatively, assuming that hyperbaric oxygen will ameliorate the undesirable effects of carbon dioxide, then its commercial use will most likely involve combinations with fungi-

static concentrations of carbon dioxide on commodities that do not tolerate such carbon dioxide levels alone.

References

Amanatidou, A., E.J. Smid and L.G.M. Gorris 1999. Effect of elevated oxygen and carbon dioxide on the surface growth of vegetable associated microorganisms. J. of Applied Microbiology (in press).

Bean, J.W. 1945. Effects of oxygen at increased pressure. Physiological Reviews 25:1-147.

Bert, P. 1878. Barometric Pressure: Researches in Experimental Physiology. Translated by M.A. Hitchcock and F.A. Hitchcock. College Book Co., Columbus, OH, 1943.

Biale, J.B. 1946. Effect of oxygen concentration on respiration on the Fuerte avocado fruit. Amer. J. Botany 33:363-373.

Caldwell, J. 1956. Studies in respiration of apples at various pressures of oxygen. J. Exp. Botany 7:326-334.

Caldwell, J. 1965. Effects of high partial pressures of oxygen on fungi and bacteria. Nature 206:321-323.

Claypool, L.L. and F.W. Allen 1951. The influence of temperature and oxygen level on the respiration and ripening of Wickson plums. Hilgardia 21(6):129-160.

Day, B.P.F. 1996. High oxygen MAP for fresh prepared produce. Postharvest News and Information 7(3):31N-34N.

Frenkel, C. and S.A. Garrison 1976. Initiation of lycopene synthesis in the tomato mutant rin as influenced by oxygen and ethylene interactions. HortScience 11:20-21.

Fridovitch, I. 1975. Superoxide dismutase. Annual Review of Biochemistry 44:147-159.

Gerschman, R. 1964. Biological effects of oxygen. In: Oxygen in the Animal Organism, F. Dickens and E. Neil (eds.) Macmillan, New York, NY, pp.475-492.

Gonzalez, M. and B.P.F. Day 1998. The effect of elevated oxygen and carbon dioxide modified atmospheres on psychrotrophic pathogens and spoilage microorganisms associated with fresh prepared produce. Research Summary Sheet, Campden & Chorleywood Food Research Association.

Haugaard, N. 1968. Cellular Mechanisms of Oxygen Toxicity. Physiological Reviews 48(2):311-373.

Kader, A.A. 1995. Regulation of fruit physiology by controlled/modified atmospheres. Acta Hort. 398:59-70.

Mitchell, F.G. 1992. Postharvest handling systems: Small fruits. In Postharvest Technology of Horticultural Crops, ed. A.A. Kader, 223-231, Oakland, CA: The Regents of the University of California DANR.

Robb, S.M. 1966. Reactions of fungi to exposure of 10 atmospheres pressure of oxygen. Journal of General Microbiology 45:17-29.

---

[1] *Department of Pomology, University of California, Davis, CA.*

[2] *State of Israel, Ministry of Agriculture, Agricultural Research Organization, The Volcani Center, Israel*