

16 Modified Atmosphere Packaging

16.1 INTRODUCTION

16.1.1 DEFINITIONS

There is a continuous search for improved methods of transporting food from producers to consumers. It has long been known (see Section 16.1.2) that the preservative effect of chilling can be greatly enhanced when it is combined with control or modification of the gas atmosphere during storage. Such methods have been used commercially for over 100 years for the bulk storage and transport of fresh meat and fruits and are referred to as controlled atmosphere storage (CAS). Since the 1970s and the widespread availability of polymeric packages, this approach has been applied to consumer packs and given the name *modified atmosphere packaging* (MAP) because the atmosphere surrounding the food is modified but not controlled.

MAP can be defined as the enclosure of food in a package in which the atmosphere inside the package is modified or altered to provide an optimum atmosphere for increasing shelf life and maintaining food quality. Modification of the atmosphere may be achieved either actively or passively. *Active modification* involves displacing the air with a controlled, desired mixture of gases, a procedure generally referred to as *gas flushing*. *Passive modification* occurs as a consequence of the food's respiration and/or the metabolism of microorganisms associated with the food; the package structure normally incorporates a polymeric film, and so the permeation of gases through the film (which varies depending on the nature of the film and the storage temperature) also influences the composition of the atmosphere that develops.

Vacuum packaging of respiring foods or foods containing viable microorganisms such as flesh foods is clearly a form of MAP, because after initial modification of the atmosphere by removal of most of the air, biological action continues to alter or modify the atmosphere inside the package. In vacuum packaging, elevated levels of CO₂ can be produced by microorganisms or by respiring fruits and vegetables. Even when no gas is produced inside the package after sealing, vacuum packaging still qualifies as MAP because removing the air has modified the atmosphere inside the package.

Two terms are in widespread use concerning procedures that involve changes in the gas atmosphere in bulk storage facilities. In CAS, the gas composition inside a food storage room is continually monitored and adjusted to maintain the optimum concentration within quite close tolerances. In contrast, the less common *modified atmosphere storage* (MAS) typically involves some initial modification of the atmospheric composition in an airtight storage room, which changes further with time as a result of the respiratory activity of the fresh food and the growth of microorganisms. Because CAS is capital intensive and expensive to operate, it is more appropriate for those foods that are amenable to long-term storage such as apples, kiwifruit, pears and meat.

Controlled atmosphere packaging (CAP) is, strictly speaking, the enclosure of food in a gas-impermeable package inside which the gaseous environment with respect to CO₂, O₂, N₂, water vapor and trace gases has been changed and is selectively controlled to increase shelf life. Using this definition, there are no CAP systems in commercial use. However, the combination of in-package or in-film O₂ and C₂H₄ absorbers, together with CO₂ release agents (i.e., active packaging as discussed in Chapter 15), could be classed as CAP, at least during the early stages of the storage life of the packaged product.

An associated technique is *hypobaric storage*, which consists of placing the food in an environment in which pressure, air temperature and humidity are precisely controlled, and the rate at which air in the storage environment is changed is closely regulated (Burg, 2004). Unlike CAS and MAS, no gases other than air are required. The total pressure within the hypobaric chamber is important because the O₂ concentration is directly proportional to that pressure. Although much research has been carried out into the use of hypobaric conditions for refrigerated storage of flesh foods and horticultural products, it has not been employed commercially to any great extent for the storage or transportation of foods. However, it is used commercially by growers of cut flowers.

16.1.2 HISTORY OF MAP

The first recorded scientific investigation into the effect of modified atmospheres on fruit ripening appears to have been conducted by Jacques Etienne Berard, a professor at the School of Pharmacy at Montpellier in France, who published his findings in 1821. Berard recognized that harvested fruits utilize O₂ and give off CO₂, and that fruits placed in an atmosphere deprived of O₂ did not ripen as rapidly. There is no record of commercial use of this information for nearly 100 years.

However, a remarkable application of the principles of CAS took place in 1865 in Cleveland, Ohio, when Benjamin Nyce built a reasonably airtight store that used ice for cooling and a special paste for filtering the atmosphere to remove CO₂. He operated this store for a few years but refused to permit others to use his patented procedures; there is no record of expanded use of his system.

The first American scientists to investigate CAS were Thatcher and Booth of Washington State University. Around 1903, they performed 2 years of testing that proved promising but the work was discontinued. In the period between 1907 and 1915, research personnel at the U.S. Department of Agriculture and Cornell University studied the response of several fruits to both lower O₂ and higher CO₂ levels in storage atmospheres. This work was reported in various scientific journals but did not result in commercial applications.

The first intensive and systematic research on CAS of fruits was initiated in England in 1918 by Franklin Kidd (later knighted for his efforts) and Cyril West at the Low Temperature Research Station at Cambridge. Various temperatures and atmospheres were used with apples, pears, plums, strawberries, gooseberries and raspberries. The atmospheres were generated passively by fruit respiration, and were dependent on the O₂ consumed and the CO₂ evolved by the fruit within a gas-tight building. The first commercial CA store appears to have been constructed by a grower near Canterbury in Kent in 1929 and, by 1938, there were over 200 commercial CAS facilities in England.

The knowledge that CO₂ inhibits bacterial growth is not new; in 1877, Pasteur and Joubert observed that *Bacillus anthracis* could be killed using CO₂, and 5 years later, the first paper on the preservative effect of CO₂ on extending the shelf life of beef was published in Germany by Kolbe. During the period from 1880 to late 1920, about 100 reports were published on the inhibitory effects of CO₂ on microorganisms. In 1930, Killefer in England demonstrated that lamb, pork and fish remained fresh twice as long in 100% CO₂ compared with storage in air at chill temperatures, and similar improvements were reported by other English researchers for bacon and beef. In 1933, Haines found that the doubling time of some common bacteria on meat stored in 10% CO₂ at 0°C was twice that in air at the same temperature. Practical application of these results was made in the shipment of chilled beef carcasses from Australasia to England from the early 1930s, with an atmosphere of 10% CO₂ and a temperature of -1°C providing a storage life of 40–50 days without spoilage.

Coyne from the United Kingdom reported in 1932 that fillets and whole fish at ice temperature could be kept twice as long if stored in an atmosphere containing a minimum of 25% CO₂ but that undesirable textural and visual changes occurred if the CO₂ concentration exceeded 80%. Although his results were taken to a semicommercial stage, the technique was never adopted by industry.

A comprehensive study into the use of CO₂-enriched atmospheres for extending the shelf life of poultry meat (chicken portions) was carried out in the United States by Ogilvy and Ayres in 1951. The maximum usable CO₂ concentration was 25% because, above this, the meat became discolored; even at 15%, a loss of bloom was sometimes noted.

In the 1950s, Whirlpool Corporation, the U.S. makers of household washers, driers and refrigerators, tried to develop small gas generators for household food preservation of fresh meats and produce using controlled atmospheres together with refrigeration. Its efforts did not prove successful, but resulted in the Whirlpool Corporation building larger generators (Tectrol® units) for CA warehouses and, later, truck transports for apples, lettuces and a host of fresh foods. In the 1960s, this technology was spun off to a joint venture company called Transfresh (now Fresh Express and owned by Chiquita Brands International), and it is the world's leading producer of fresh cut vegetables (Brody, 2003).

The first patent for MAP of red meat was issued in France in 1969 to Georgala and Davidson, two workers at Unilever. It described an atmosphere containing $\geq 70\%$ O₂ and $\geq 10\%$ CO₂, the balance being an inert gas. Under such an MA in a gas-impermeable container, beef was still in a fresh condition after 15 days at 4°C.

In 1931, Skovholt and Bailey showed that storage of bread in atmospheres containing at least 17% CO₂ delayed appearance of mold, with concentrations of 50% doubling the mold-free shelf life. Aulund from Norway confirmed these findings in 1961, achieving a mold-free shelf life of 16 days for rye bread packaged in an atmosphere of CO₂. During the 1960s, more extensive research was undertaken in the United Kingdom at the Chorleywood Flour, Milling and Baking Research Association into the gas packaging of bakery products using elevated levels of CO₂ to retard mold growth. Unlike flesh foods and fruits and vegetables, baked goods such as bread, pastries and cakes do not benefit from storage at chill temperatures because the rate of staling increases as the temperature is lowered. A large U.K. bakery used MAP in the late 1960s for cake and achieved shelf life extensions of 4–5 days. However, MAP of baked goods did not become significant until the late 1970s when new labeling regulations in Europe required a listing of all preservatives on the label. Adoption of MAP avoided the need to use, and thus list, preservatives and also gave a longer shelf life. Today, there is very little MAP of soft bakery goods in the United States despite the relatively widespread use of MAP for baked goods in Europe.

In the United States, vacuum packaging of poultry was introduced by Cryovac®, followed by the “boxed beef” concept in 1967, both involving vacuum packaging in low O₂ barrier materials. In these circumstances, the atmosphere around the meat becomes depleted in O₂ (often <1% v/v) and enriched in CO₂ (>20% v/v), resulting in microbial changes quite different from those observed during aerobic storage. Vacuum-packaged boxed beef is then distributed to retail outlets where it is converted into consumer units; vacuum-packaged, boxed pork and lamb followed in the 1970s.

In summary, the successful commercialization of MAP in the late 1970s was preceded by over 150 years of scientific research on the inhibitory effects of CO₂ on microbial growth, as well as the effect of gaseous atmospheres on respiring produce. It required the convergence of scientific knowledge, polymeric films, gas flushing and vacuum packaging equipment, and cold distribution chains to achieve the commercial success it enjoys today. Surprising to many is that MAP, in its many manifestations, is now well ahead of the more widely publicized canning, freezing, aseptic packaging, and retort pouch and tray packaging in terms of volume of food preserved (Brody, 2003). Although extension of shelf life is the most apparent advantage of MAP, there are also several other advantages (as well as disadvantages) as shown in Table 16.1.

16.2 PRINCIPLES

MAP is used to delay deterioration of foods that are not sterile and whose enzymic systems may still be operative. With the exception of baked goods, MAP is always used in association with chill temperatures. Chill temperatures are those close to but above the freezing point of fresh foods, and

TABLE 16.1
Advantages and Disadvantages of MAP

Advantages	Disadvantages
Shelf life increase from 50% to 400%	Added costs for gases, packaging materials and machinery
Reduced economic losses due to longer shelf life	Temperature control necessary
Decreased distribution costs, longer distribution distances and fewer deliveries required	Different gas formulations for each product type
Provides a high quality product	Special equipment and training required
Easier separation of sliced products	Potential growth of food-borne pathogens due to temperature abuse by retailers and consumers
Centralized packaging and portion control	Increased pack volume adversely affects transport costs and retail display space
Improved presentation—clear view of product and all-around visibility	Loss of benefits once the pack is opened or leaks
Little or no need for chemical preservatives	CO ₂ dissolving into the food could lead to pack collapse and increased drip
Sealed packages are barriers against product recontamination and drip from package	
Odorless and convenient packages	

Source: Adapted from Sivertsvik, M. et al., Modified atmosphere packaging, in: *Minimal Processing Technologies in the Food Industry*, Ohlsson, T. and Bengtsson, N. (Eds), CRC Press, Boca Raton, FL, pp. 61–86, 2002.

are usually taken as -1°C to $+7^{\circ}\text{C}$. Holding food at chill temperatures is widely used as an effective short-term preservation method, which has the effect of retarding the following occurrences:

1. Growth of microorganisms
2. Postharvest metabolic activities of intact plant tissues, and postslaughter metabolic activities of animal tissues
3. Deteriorative chemical reactions, including enzyme-catalyzed oxidative browning, oxidation of lipids, chemical changes associated with color degradation, autolysis of fish and loss of nutritive value of foods in general
4. Moisture loss

The effect of chilling on the microflora in a particular food depends on the temperature characteristics of the organisms as well as the temperature and time of storage. As the temperature is lowered from the optimum, growth slows and eventually stops. Microorganisms which can grow within the 0°C – 7°C range are defined as *psychrotrophs*. The most important psychotropic bacteria as far as chill temperature preservation of food is concerned are from the genus *Pseudomonas*, but the pathogens *Clostridium botulinum* type E, *Yersinia enterocolitica*, *Listeria monocytogenes*, enterotoxigenic *Escherichia coli* and *Aeromonas hydrophila* are also able to grow at or below 6°C . Thus, chill temperatures cannot be relied on absolutely to keep foods safe because of the possible survival and growth of these pathogens at chill temperatures.

If cooling is too fast or if the temperature is reduced too near to the freezing point of the food, then chilling injury can result. This can manifest itself in various ways, for example, cold shortening of muscle and physiological disorders of fruits and vegetables. In general, each food has a minimum temperature below which it cannot be held without some undesirable changes occurring in that food.

The preservative effect of chilling can be greatly enhanced when it is combined with modification of the gas atmosphere. This is because many deteriorative reactions involve aerobic respiration in which the food or microorganism consumes O₂ and produces CO₂ and water. By reducing

O₂ concentration, aerobic respiration can be slowed. By increasing CO₂ concentration, microbial growth can be slowed or inhibited (see the following text).

In addition to the benefits resulting from modification of the atmosphere inside the package, other benefits from MAP for fresh foods can include maintenance of high RH and reduction in water loss, as well as improved hygiene by reducing contamination during handling. In the case of fresh produce, surface abrasions are minimized by avoiding contact between the produce and the shipping container and there is a reduced spread of decay from one item to another. In many cases, the benefits of using MAP relate more to one or more of these positive effects than to changes in the O₂ and CO₂ concentrations inside the package. Negative effects of MAP of fresh produce include a slowing down in the cooling rate of the packaged products, and increased potential for water condensation within the package, which may encourage fungal growth.

Although there is considerable information available regarding suitable gas mixtures for different foods, there is still a need for additional scientific information regarding many aspects relating to MAP, including

1. Mechanisms of action of CO₂ on microorganisms
2. Influence of CO₂ on microbial ecology of a food
3. Safety of MAP packaged food products
4. Interactive effects of MAP and other preservation methods
5. Effect of MAP on nutritional quality of packaged foods

16.3 GASES USED IN MAP

The normal composition of air by volume is 78.08% N₂, 20.95% O₂, 0.93% argon (Ar), 0.03% CO₂ and traces of nine other gases in very low concentrations. The three main gases used in MAP are O₂, CO₂ and N₂, either singly or in combination. Noble or “inert” gases such as Ar are being used commercially for a wide range of products although the literature on their application and benefits is limited (Spencer, 2005). The solubilities in water of these gases are given in Table 16.2. Gases also have significant solubility in the lipid phase of foods. For example, at 0°C and atmospheric pressure, 1 L of vegetable oil can absorb approximately 6 mL of N₂, 12 mL of O₂ and 1 L of CO₂ (Lencki, 2005). Use of carbon monoxide (CO), sulfur dioxide (SO₂) and nitrous oxide (N₂O) in MAP has also been reported.

16.3.1 CARBON DIOXIDE

CO₂ is the most important gas in the MAP of foods, due to its bacteriostatic and fungistatic properties. It inhibits the growth of many spoilage bacteria, with the degree of inhibition increasing with increasing concentration. It is particularly effective against molds and Gram-negative, aerobic spoilage bacteria such as *Pseudomonas* sp., but much less effective in controlling yeasts or lactic acid bacteria.

TABLE 16.2
Solubilities of Gases in Water at Atmospheric Pressure and Various Temperatures

Temperature (°C)	Gas Solubility (mg kg ⁻¹)				
	Oxygen	Nitrogen	Carbon Dioxide	Argon	Carbon Monoxide
0	69	29	3350	100	44
5	62	27	2770	89	40
10	58	24	2320	78	36
20	42	18	1690	59	28

Solubilities given for carbon dioxide include all chemical species of the gas and its reaction products with water.

CO₂ is a colorless gas with a slight pungent odor at very high concentrations. It dissolves readily in the aqueous phase and forms a significant quantity of other hydrated carbonate species depending on the pH. At the slightly acidic pH found in many foods, the dissolved CO₂ first hydrates to form carbonic acid H₂CO₃ (Equation 16.1); this is known to disrupt microbial cell membranes and inhibit respiratory enzymes. It then ionizes to release bicarbonate and an H⁺ ion (Equation 16.2) that can lower pH depending on the buffering capacity of the food (Lencki, 2005). When the pH of a CO₂ solution rises to 8.0 (most unlikely in foods), the HCO₃⁻ may even dissociate further to form CO₃²⁻ (Equation 16.3):



As with all gases, the solubility of CO₂ increases with decreasing temperature and therefore the antimicrobial activity of CO₂ is markedly greater at lower temperatures. This has significant implications for MAP of foods. The high solubility of CO₂ in high moisture/high fat foods such as meat, poultry and seafood can result in package collapse due to the reduction of headspace volume. High levels of CO₂ can also result in increased drip or exudate from flesh foods, and the addition of absorbent pads in the base of the package is used to compensate for this.

16.3.2 OXYGEN

O₂ is a colorless, odorless gas that is highly reactive and supports combustion. It has a low solubility in water. O₂ promotes several types of deteriorative reactions in foods including fat oxidation, browning reactions and pigment oxidation. Most of the common spoilage bacteria and fungi require O₂ for growth. For these reasons, O₂ is either excluded or reduced to as low a concentration as possible. Exceptions occur where O₂ is needed for the respiration of fruits and vegetables or the retention of color in red meat.

16.3.3 NITROGEN

N₂ is an inert gas with no odor or taste. It has a lower density than air and a low solubility in water and other food constituents, making it a useful filler gas in MAP to counteract package collapse caused by CO₂ dissolving in the food. N₂ indirectly influences the microorganisms in perishable foods by retarding the growth of aerobic spoilage microbes but it does not prevent the growth of anaerobic bacteria. MAP mixtures based on N₂ have a similar density to air, and a great deal of turbulence is introduced when such mixtures are used to displace air inside packages as a result of the interaction of two columns of similar density. Sufficient force must be applied to introduce a mass of at least eight times the volume of airspace into the package, although applying a vacuum first can reduce the volume of air that must be displaced (Spencer, 2005).

16.3.4 CARBON MONOXIDE

CO is a colorless, tasteless and odorless gas, which is highly reactive and very flammable. It has a low solubility in water but is relatively soluble in some organic solvents. CO has been studied in the MAP of meat where it has the potential to inhibit metmyoglobin formation and promote metmyoglobin reduction, even when O₂ is present. Lipid oxidation and browning are reduced and the shelf life of the food is prolonged. CO combines with myoglobin to form the bright cherry-red pigment carboxymyoglobin,

which is much more stable than oxymyoglobin; a CO concentration of 0.4% in a MAP of meat is sufficient to give a bright red color (see Chapter 17). CO at 5%–10% (combined with less than 5% O₂) is an effective fungistat, which can be used on commodities that do not tolerate high CO₂ levels.

CO has not been approved by regulatory authorities for commercial use in the EU, and is not included in the list of allowed food additives (Directive 95/2/EC). It was legal in Norway for retail packaging of red meat, but Norway has since banned the use of CO in compliance with EU rules. It has been sanctioned for use in the United States to prevent browning in packaged lettuce, and for pretreating meat in a master pack system where it is considered a processing aid. Commercial application has been limited because of its toxicity, its explosive nature at 12.5%–74.2% in air and the fact that it has a limited effect on microorganisms.

16.3.5 NOBLE GASES

The *noble gases* are a family of elements characterized by their lack of reactivity and include helium (He), argon (Ar), xenon (Xe) and neon (Ne). Although the noble gases are chemically inert, it has been suggested that they are biologically active and several patents have been issued for their use in MAP. There has been some research into the biochemical and physiological effects of noble gases on specific enzymes, and the majority of these studies have been related to browning in fresh fruits and vegetables and respiratory metabolism.

For example, although Ar has been considered to be completely inert, some research suggests that it is a competitive inhibitor of respiratory enzymes, including oxidases. It has also been claimed that Ar slows down the rate of production of volatile amino bases in seafood, inhibits enzymic discoloration, delays the onset of textural softening, extends the microbial lag phase, inhibits microbial oxidases and enhances the effectiveness of CO₂ by weakening microbes, thereby enabling the use of less CO₂ in MAP. A possible reason for these effects could be the greater solubility of Ar compared to O₂ and N₂ and its similar atomic size to O₂. It is also claimed that Ar is more effective at displacing O₂ from cellular sites and enzymic O₂ receptors. O'Beirne et al. (2011) suggested that the greater inhibitory effect exhibited by Ar on oxidases was proportional to the greater molecular space occupied by Ar compared to N₂ and was directly related to the van der Waals radius values of Ar against N₂ (1.91 against 1.54 Å).

Ar is a much denser gas than N₂ (1.650 compared to 1.153 kg m⁻³) and therefore can be made (unlike N₂) to flow in a laminar fashion like a liquid through air space. Thus, if it is introduced into the bottom of a tray, it displaces the air column upward as it fills the tray (Spencer, 2005). For the same reason, Ar is commonly used instead of N₂ to flush the neck of wine bottles immediately prior to corking. Because Ar is denser than N₂ and four times more efficient at displacing air, the difference in cost (Ar costs approximately five times that of N₂) is negligible.

Despite few scientific publications, noble gases are being used in a number of food applications including potato chips, processed meats, nuts, beverages, fresh pasta, chilled prepared meals and lettuce with claims of an average 25% improvement in shelf life. Some products, such as fresh pizza, have shelf life improvements of 40%–50%. At present, nearly 200 different argon-packaged foods can be found on supermarket shelves in the United Kingdom and elsewhere in Europe (Spencer, 2005).

16.3.6 GAS MIXTURES

The gas mixtures used for MAP of different foods depend on the nature of the food and the likely spoilage mechanisms. Where spoilage is mainly microbial, the CO₂ levels in the gas mix should be as high as possible, limited only by the negative effects of CO₂ (e.g., package collapse) on the specific food. Typical gas compositions for this situation are 30%–60% CO₂ and 40%–70% N₂. For O₂-sensitive products where spoilage is mainly by oxidative rancidity, 100% N₂ or N₂/CO₂ mixtures (if microbial spoilage is also important) are used. For respiring products, it is important to avoid too high a CO₂ level or too low an O₂ level, so that anaerobic respiration is prevented.

16.4 METHODS OF CREATING MA CONDITIONS

16.4.1 PASSIVE MA

In this approach (also known as *commodity-generated MA*), an atmosphere high in CO₂ and low in O₂ passively evolves within a sealed package over time as a result of the respiration of the product. Ideally, the gas permeabilities of the packaging film are such that sufficient O₂ can enter the package to avoid anoxic conditions and the occurrence of anaerobic respiration, while at the same time excess CO₂ can diffuse from the package to avoid injuriously high levels. Passive modification is commonly used for MAP of fresh respiring fruits and vegetables. Given the simplicity of this approach and the many interrelated variables which affect respiration rate, considerable research is required to develop appropriate passive MA systems for horticultural products.

16.4.2 ACTIVE MA

Several methods can be used to actively modify the gas atmosphere inside a packaged product. These include vacuum packaging where the air is removed under vacuum and the package sealed. This method finds widest application for the packaging of flesh foods, particularly red meat. A two-stage method involves first removing the air inside the package using a vacuum followed by flushing with the desired gas mixture. This creates the desired MA immediately after packaging compared to the passive approach, which may require a week or longer before achieving the same gas composition. In a third active MA method, no vacuum is used but a gas mixture is injected into the package and the air swept or flushed out immediately prior to sealing, resulting in residual O₂ levels of 2–5%. For O₂-sensitive products, the two-stage method is preferred.

Regardless of whether vacuum or gas flush packaging is used to create an MA, the package itself must provide a barrier to permeation over the expected shelf life, otherwise the beneficial effects of reducing O₂ will be lost. In the case of vacuum packaging, the barrier issue relates only to O₂ ingress, but, in gas flushing, both O₂ ingress and N₂ egress must be considered. An elegant analysis of the situation has been presented (Brown, 1992) and is repeated here.

In vacuum packaging, both O₂ and N₂ are potential entrants into the package. O₂ is a faster permeant than N₂ by a factor of about 4–6 (see Table 4.3); however, air contains only a quarter as much O₂ as it does N₂. Therefore, O₂ and N₂ are about equally as likely to enter the package.

On the other hand, gas flush packaging (assuming that only N₂ is used) imposes a 100% N₂ atmosphere inside the package, working against a 79% N₂ atmosphere in the surrounding air. Thus, the driving force for N₂ exiting the package is $1.00 - 0.79 = 0.21$ atmospheres, while the driving force for O₂ to enter is the partial pressure of O₂ in the air ≈ 0.21 atmospheres. Because the driving forces (i.e., the partial pressure differences inside and outside the package) are equal, and the permeation rate for N₂ egress is about a quarter to a sixth that of O₂ ingress, the package will slowly increase in pressure. If the package is made from a flexible film or has a film lid sealed to a plastic tray, a “pillowing” effect is likely to occur. It is important that the reasons for this pillowing effect are clearly understood as it is not uncommon for it to be ascribed by the uninitiated to microbial growth, resulting in perfectly safe product being removed from sale.

In addition to the mechanical methods described earlier, absorbers can be used inside the package, for example, to delay the climacteric rise in respiration for some fruits by adsorbing C₂H₄, to prevent the build-up of CO₂ to injurious levels by adsorbing CO₂ as well as to lower the concentration of O₂ through the use of O₂ absorbers. Obviously, the use of gas absorbers adds considerably to the cost, and therefore their use is limited to those commodities for which it is cost-effective. An alternative method for quickly reducing the O₂ content and increasing the CO₂ content within a package involves the use of ferrous carbonate inside a gas-permeable sachet; in the presence of moist air, the amorphous material oxidizes with the release of CO₂. The quantity of ferrous carbonate used must be carefully calculated since if too much is present inside the package, then anoxic conditions will be established.

The practical upper level of dissolved CO₂ in a food is limited by the degree of filling (DoF) and the flexibility of the chosen packaging material. A high CO₂ level and high DoF is hampered by package collapse. One approach to address this problem is *soluble gas stabilization* (SGS) and involves dissolving CO₂ in the food at low temperature (ca. 0°C) and elevated pressures (>2 atm) prior to packaging (Sivertsvik et al., 2002). This results in packages with smaller gas:product ratios and thus decreased package sizes for a given weight of product. SGS has shown promising results on extending the shelf life of a range of seafood, and has also been used successfully on dairy products. In addition, a mathematical model has been developed to estimate the equilibrium gas composition in MAP and SGS systems (Rotabakk et al., 2008).

16.5 EQUIPMENT FOR MAP

Equipment for MAP must generally be capable of removing air from the package and replacing it with a mixture of gases. There are basically three types of packaging equipment used for MAP: horizontal or vertical form-fill-seal (FFS) machines using pouches or trays, snorkel machines using preformed bags or pouches and chamber machines using preformed pouches or trays.

16.5.1 FORM-FILL-SEAL MACHINES

FFS machines can either form pouches (vertically or horizontally), or thermoformed trays with a heat sealed lid, from rollstock. In the pouch version, the desired gas mixture is introduced into the package in a continuous countercurrent flow to force out the air, after which the ends of the web are heat sealed and the packages cut from one another. In the tray version, product is placed in the tray and a vacuum drawn, after which the desired gas mixture is introduced and the top web of film heat sealed to the base tray.

16.5.2 CHAMBER MACHINES

Here, the filled package (either a preformed pouch or tray inside a bag) is loaded into a chamber, a vacuum is pulled and the package is then flushed with the gas mixture and heat sealed. This is a batch process, is relatively slow and most suitable for bulk or master packs.

16.5.3 SNORKEL MACHINES

Snorkel machines operate without a chamber. The product is placed inside a large flexible pouch (or bag) and positioned in the machine. Snorkels or probes are inserted into the pouch and remove air, after which the vacuum is broken by the addition of the desired gas mix. The probes are then removed and the package is heat sealed. These machines are used mainly for bulk packaging and for so-called *master packs* in which individual retail packs are packaged in a large MA pouch or bag.

16.6 PACKAGING FOR MAP APPLICATIONS

The main characteristics to be considered when selecting packaging materials for MAP are permeability of the package to gases and water vapor, mechanical properties, heat sealability and transparency. For nonrespiring products, all the common high gas barrier structures have been used in MAP, including laminates and coextruded films containing PVdC, EVOH and PAs as a barrier layer. The inside layer is usually LDPE to provide a good heat seal and moisture vapor barrier.

The OTR of packaging materials used for MAP of chilled products varies extensively with temperature, RH and material thickness after the thermoforming of packages. Gnanaraj et al. (2005) reported OTRs for a range of films at 10°C, 15°C, 23°C, 30°C and 35°C and 0% and 50% RH. The OTRs at 10°C were typically half those at 23°C. Jakobsen et al. (2005) studied two different

polymer combinations: an APET-LDPE tray and a PA-LDPE lid. A temperature reduction of 8°C (in the interval 7°C–23°C) caused an OTR reduction of 26%–48%, depending on material type, degree of thermoforming and RH. An increased OTR was observed as a result of material thinning; however, the increase was not always directly proportional to the degree of material thinning. The changes observed in OTR emphasize the necessity of evaluating the performance of packaging materials under realistic storage conditions to estimate the real O₂ content of a chosen package solution. The same comments apply to the TR of other gases used in MAP.

For the MAP of respiring produce such as fruits and vegetables, the choice of suitable packaging materials is much more complex, and no easy solutions are available due to the dynamic nature of the food. Ideally, the packaging material should maintain a low O₂ concentration (3%–5%) in the headspace and prevent CO₂ levels exceeding 10%–20%. Polyolefin films are normally used, but to achieve the desired MAP, it has been necessary to perforate the film or (more recently) use a special patch; O₂ and CO₂ absorbers have also been used on a limited scale. MAP materials for fruits and vegetables are discussed in more detail in Chapter 18.

To prevent condensation of water vapor on the inside of the package as a result of temperature differentials between the package contents and the packaging material, antifogging agents are used. These amphiphilic additives function by decreasing the interfacial tension between the polymer and the condensed moisture vapor, enabling the water droplets to coalesce and spread as a thin transparent layer across the surface of the film. Typical antifogging agents include nonionic ethoxylates or hydrophilic fatty acid esters.

For the trays used in MAP, there are no special requirements other than good thermoformability. Typical tray materials are PS and PVC, although the latter is much less common in Europe because of environmental concerns among consumers. Preformed plastic-coated paperboard trays have also been used. Regardless of the type of tray, it is essential that the lidding film can be adequately sealed onto the tray.

16.7 MICROBIOLOGY OF MAP

The species of microorganisms that cause spoilage of particular foods are influenced by two factors: the nature of the foods and their surroundings. These are known as *intrinsic* and *extrinsic* parameters and were discussed in Chapter 11 (see especially Table 11.4). The two extrinsic factors most relevant in MAP are the gaseous composition of the in-pack environment and the temperature. The specific microbiology of various foods is dealt with in subsequent chapters of this book. However, general comments on the effects of MAs on food spoilage and pathogenic microorganisms will be outlined here.

Microbial food spoilage is characterized by undesirable sensory changes to the odor, color, flavor and sometimes the texture of the food, making it inedible or unsealable. Spoilage is an important safeguard in preventing food poisoning, because the deterioration in food quality normally (but not always) warns the consumer that the food may be unsafe.

Microorganisms have different respiratory and metabolic needs and can be grouped according to their O₂ needs as shown in Table 16.3. The effect of CO₂ on microbial growth is shown in Table 16.4.

Concentrations of CO₂ in excess of 5% v/v inhibit the growth of most food spoilage bacteria, especially psychrotrophic species such as *Pseudomonas*, which grow on a wide range of refrigerated foods. The effect of CO₂ on bacterial growth is complex and the growth inhibition of microorganisms in MA is determined by the concentration of dissolved CO₂ in the product. However, CO₂ does not retard the growth of all types of microorganisms; for example, the growth of lactic acid bacteria is enhanced in the presence of CO₂ and low O₂ concentrations. Another example is *L. monocytogenes* which, although inhibited by 100% CO₂, is unaffected by CO₂ if at least 5% O₂ is present.

Much research has been carried out regarding the safety and the health hazards of MAP of foods, especially on those pathogens able to multiply at chill temperatures and those able to multiply in anaerobic conditions. There are seven food-borne pathogenic bacteria known to be capable of growth

TABLE 16.3
Oxygen Requirements of Some Microorganisms of Relevance in MAP

Group	Spoilage Organisms	Pathogens
Aerobes (require atmospheric O ₂ for growth)	<i>Micrococcus</i> sp. Molds, e.g., <i>Botrytis cinerea</i> <i>Pseudomonas</i> sp.	<i>Bacillus cereus</i> <i>Yersinia enterocolitica</i> <i>Vibrio parahaemolyticus</i> <i>Campylobacter jejuni</i>
Microaerophiles (require low levels of O ₂ for growth)	<i>Lactobacillus</i> sp. <i>Bacillus</i> spp. <i>Enterobacteriaceae</i>	<i>Listeria monocytogenes</i> <i>Aeromonas hydrophila</i> <i>Escherichia coli</i>
Facultative anaerobes (grow in presence or absence of O ₂)	<i>Brochothrix thermosphacta</i> <i>Shewanella putrefaciens</i> Yeasts	<i>Salmonella</i> spp. <i>Staphylococcus</i> spp. <i>Vibrio</i> sp.
Anaerobes (inhibited or killed by O ₂)	<i>Clostridium sporogenes</i> <i>Clostridium tyrobutyricum</i>	<i>Clostridium perfringens</i> <i>Clostridium botulinum</i>

TABLE 16.4
Effect of CO₂ Atmosphere on Growth

Microorganism	Type of Growth	Effect on Growth in CO ₂ Atmosphere
<i>Aeromonas</i> spp.	Facultative	Inhibited (weakly)
<i>Bacillus cereus</i>	Facultative	Inhibited
<i>Campylobacter jejuni</i>	Microaerophilic	Inhibited, survival ^a
<i>Clostridium botulinum</i> proteolytic (A, B, F)	Anaerobic	Unaffected ^b
<i>C. botulinum</i> nonproteolytic (B, E, F)	Anaerobic	Unaffected ^b
<i>Clostridium perfringens</i>	Anaerobic	Inhibited
<i>Escherichia coli</i>	Facultative	Inhibited (weakly)
<i>Listeria monocytogenes</i>	Facultative	Unaffected/inhibited ^c
<i>Plesiomonas</i> spp.	Facultative	Inhibited
<i>Salmonella</i>	Facultative	Inhibited ^b
<i>Staphylococcus aureus</i>	Facultative	Inhibited (weakly)
<i>Vibrio cholerae</i>	Facultative	Inhibited
<i>Vibrio parahaemolyticus</i>	Facultative	Inhibited
<i>Yersinia enterocolitica</i>	Facultative	Inhibited

Source: Adapted from Sivertsvik, M. et al., Modified atmosphere packaging, in: *Minimal Processing Technologies in the Food Industry*, Ohlsson, T. and Bengtsson, N. (Eds), CRC Press, Boca Raton, FL, pp. 61–86, 2002.

^a The bacteria survive better in CO₂ as compared to air, but growth is (weakly) inhibited.

^b One report of growth stimulation under CO₂.

^c Unaffected by CO₂ atmosphere if at least 5% O₂ present; inhibited under 100% CO₂.

at or below 5°C: *C. botulinum* Type E, *L. monocytogenes*, *Y. enterocolitica*, *Vibrio parahaemolyticus*, enterotoxigenic *E. coli*, *Bacillus cereus* and *Aeromonas hydrophila*. Two others are capable of growth at temperatures just above 5°C: *Staphylococcus aureus* at 6°C (10°C for toxin production) and *Salmonella* sp. at 7°C. Thus, it is vitally important that the MAs inhibit the growth of these organisms in foods under refrigerated storage. Fortunately, most of these organisms do not compete well with harmless bacteria, such as the *Lactobacillus* sp., which grow rapidly if temperature abuse occurs.

The spores of *C. botulinum* types B, E and F can grow and produce toxin after 5 weeks at 3°C, 3–4 weeks at 4°C and 2–3 weeks at 5°C. Growth occurs more frequently from spores of type F strains than for types B and E. It has been demonstrated that 100% CO₂ can have an inhibitory effect on the growth of *C. botulinum* at chill temperatures, and an increased inhibitory effect has been observed when combining 100% CO₂ with increased NaCl levels and decreased pH. Of most concern in MAP is the possible growth of *C. botulinum* Type E, which is associated with fishery products. It is tolerant of low temperatures, is anaerobic and may grow and produce a potent neurotoxin on the food before spoilage is detectable by the consumer. Storage at temperatures below 3°C should provide an adequate safeguard for foods where *C. botulinum* may be present.

The concept of barrier or hurdle technology was introduced in the 1980s and refers to combinations of different preservation factors (“hurdles”) that are used to achieve multitarget, mild preservation effects. Hurdles that are used in addition to chill temperatures to limit or prevent the growth of psychrophilic pathogens during low temperature storage or limited temperature abuse of the food include CO₂, thermal processing, pH reduction (<5), preservatives, reduced a_w (<0.97) and competitive microflora.

16.8 SAFETY OF MAP

The shelf life and safety of any MAP food is influenced by a number of factors including the nature of the food, the gaseous environment inside the package, the nature of the package, the storage temperature and the packaging process and machinery. In discussing the safety of MAP foods, it is useful to divide them into two categories: those products such as smoked salmon, cured meats, fruits and salad vegetables that are eaten without any prior heat treatment, and those such as fresh fish, raw meats and poultry products that are usually subjected to a sufficient heat treatment to kill all vegetative pathogens (Walker and Betts, 2008). Clearly, the first category presents more risks from a microbiological point of view.

Chilled foods have been subjected to detailed regulatory controls in many countries, particularly with respect to temperature requirements, and at the international level, HACCP-based approaches to hygiene have been established (Codex, 1997, 1999). One of the major concerns of MAP foods is temperature abuse, because the biostatic effects of CO₂ are temperature dependent, and a rise in temperature during storage could permit the growth of microorganisms which had been inhibited by CO₂ at lower temperatures. If O₂ were present in the package, then growth of aerobic spoilage organisms during periods when the food was at nonrefrigerated temperatures would alert consumers to temperature abuse due to the appearance of undesirable odors, colors or slime. However, the absence of O₂ will favor the growth of anaerobic microorganisms (including *C. botulinum*) over aerobic spoilage organisms. It has been demonstrated that anaerobic pathogens can grow at temperatures as low as 3°C and produce toxin without any sensory manifestation of food deterioration.

An area of active research is edible films for use in MAP systems. However, these films can create a very low O₂ environment where anaerobic pathogens such as *C. botulinum* may thrive. Antimicrobial compounds that can be incorporated into the coating are also being investigated.

Successful control of both product respiration and ethylene production by MAP can result in fruit or vegetable products of high sensory quality. However, control of these processes is dependent on maintaining optimum temperature control along the entire food chain continuum, from processing, storage, transportation and retailing to in the home. Maintaining proper storage temperatures is often most difficult at retail and domestic level.

Currently, there is concern with psychrotrophic food-borne pathogens such as *L. monocytogenes*, *Y. enterocolitica* and *A. hydrophila*, as well as nonproteolytic *C. botulinum*, although clearly a number of other microorganisms, especially *Salmonella*, *E. coli* O157:H7 and *Shigella* spp., can be potential health risks when present on MAP produce. Although only two MAP produce products (*C. botulinum* in coleslaw mix in the United States in 1990 and *Salmonella* in ready-to-eat salad vegetables in the United Kingdom in 2001) have been implicated in food-borne illness outbreaks, the potential for growth of pathogens exists. The success and microbiological safety of MAP is dependent on controlled low temperature storage and the product’s characteristics.

It is difficult to evaluate the safety of MAP foods solely on the basis of the growth of certain pathogens at abusive temperatures because (1) most food pathogens do not grow at chill temperatures and (2) CO₂ is not highly effective at nonrefrigeration temperatures. To minimize problems with pathogens inside MAP foods, the foods should be of the highest microbiological quality at the time of packing, they should be processed and packaged under high standards of hygiene and sanitation, their temperature should be reduced as rapidly as possible and the temperatures during distribution should be rigidly maintained as low as required to avoid anaerobic pathogen growth. If the aforementioned requirements are compromised in any way, serious public health hazards could result from ingestion of the MAP food.

Predictive microbiology was discussed in Chapter 12 and is being increasingly used as a tool to provide rapid and reliable answers concerning the likely growth of specific organisms under defined conditions. There are many advantages to the use of predictive models with chilled foods, especially in the decision-making processes of HACCP and risk analysis, and reference should be made to a recent review (McClure and Amézquita, 2008).

16.9 REFRIGERATED, PASTEURIZED FOODS WITH EXTENDED DURABILITY AND *SOUS VIDE*

There is an increased interest in “fresh,” preservative-free food with extended durability. In addition to foods packaged in MAs, there are refrigerated, pasteurized foods with extended durability (REPFEDs) such as *sous vide* and cook-chill foods that are produced by the following general process. Meals or components of meals (which may include both raw and cooked components) are sealed in a heat-stable pouch and the packaged product cooked at temperatures ranging from 65°C to 95°C. The product is then cooled and stored at refrigeration temperatures (1°C–7°C) and has a shelf life of up to 42 days dependent on the heat treatment and the storage temperature (Gorris and Peck, 1998).

If the pasteurization process is adequately carried out, then all vegetative bacteria present are killed, but bacterial spores can survive this heating process. Since REPFEDs are mostly packed under vacuum or in an anaerobic atmosphere, growth of aerobic microorganisms is restricted while growth of anaerobic bacteria is favored. The ecological niche found in REPFEDs favors colonization by microorganisms that produce heat-resistant spores and grow in the absence of O₂ at refrigeration temperatures. Nonproteolytic *C. botulinum* is the principal microbiological safety concern in REPFEDs (Gorris and Peck, 1998). Proteolytic *C. botulinum* and *Clostridium perfringens* are of concern when the storage temperature exceeds 10°C for a prolonged time.

Hurdle technology has been applied to REPFEDs to control growth of spoilage and pathogenic microorganisms, although each hurdle is insufficient on its own to achieve the same effect. Typical hurdles include pH ≤ 5, salt concentration ≥ 3.5% and $a_w \leq 0.97$ throughout the food.

A food processing technology known as *sous vide* (literally “under vacuum”) has been developed to enhance the shelf life of refrigerated foods and is a particular type of cook-chill process. Though lauded as a revolution by some, it is really the result of an evolution of the conventional cook-chill process. It uses vacuum packaging, heating and rapid cooling, followed (normally) by chilled storage, although two of the largest producers in the United States specialize in frozen *sous vide* products.

The precursor of *sous vide* was first discussed in 1960 (Kohman, 1960) and the name “Frigi-Canning” given to this new process, which involved thermal processing in hermetically sealed cans or jars, followed by rapid chilling since only vegetative forms of microorganisms were destroyed. The first formal application of *sous vide* technology was developed and tested at two hospitals in Sweden from 1960 to 1965, the process being termed the “Nacka System” after one of the hospitals. *Sous vide* technology appeared in France in 1972 for the processing of ham, and the first large-scale application of modern *sous vide* technology was undertaken in 1985 by the French national railway company SNCF (Bailey, 1995). The *sous vide* method was developed for restaurant use in the early 1970s by a French chef George Pralus, who was commissioned to find a way to reduce the shrinkage of *foie gras*; he developed a vacuum packaging and cooking process that not only

dramatically reduced shrinkage (from 46% to only 5%) but also enhanced flavor. Today, *sous vide* products range from institutional foodservice to in-home convenience foods to restaurant meals.

Low-acid, refrigerated foods, which are precooked, free of microbial inhibitors and vacuum packaged, present some very serious challenges, because the partial cooking results in the destruction of the vegetative microflora, leaving heat-resistant spores as survivors. However, *sous vide* may be produced safely if proper controls (including HACCP) are in place.

16.10 APPLICATIONS OF MAP

MAP is being successfully used by many food processing companies around the world to extend the shelf life and retain the quality of a wide variety of foods. Table 16.5 gives examples of foods

TABLE 16.5
Examples of Gas Mixtures for Selected Food Products

Product	Temperature (°C)	O ₂ (%)	CO ₂ (%)	N ₂ (%)
Meat products				
Fresh red meat	0–2	40–80	20	Balance
Cured meat	1–3	0	30	70
Pork	0–2	40–80	20	Balance
Offal	0–1	40	50	10
Poultry	0–2	0	20–100	Balance
Fish				
White fish	0–2	30	40	30
Oily fish	0–2	0	60	40
Salmon	0–2	20	60	20
Scampi	0–2	30	40	30
Shrimp	0–2	30	40	30
Plant products				
Apples	0–4	1–3	0–3	Balance
Broccoli	0–1	3–5	10–15	Balance
Celery	2–5	4–6	3–5	Balance
Lettuce	<5	2–3	5–6	Balance
Tomatoes	7–12	4	4	Balance
Baked products				
Bread	RT ^a		60	40
Cakes	RT		60	40
Crumpets	RT		60	40
Crepes	RT		60	40
Fruit pies	RT		60	40
Pita bread	RT		60	40
Pasta and ready meals				
Pasta	4		80	20
Lasagna	2–4		70	30
Pizza	5		50	50
Quiche	5		50	50
Sausage rolls	4		80	20

Source: From Brody, A.L., Modified atmosphere packaging, in: *Encyclopedia of Agricultural, Food, and Biological Engineering*, Heldman, D.R. (Ed.), Marcel Dekker, New York, pp. 666–670, 2003.

^a Room temperature; staling is accelerated at refrigerated temperatures.

currently packaged in MAs, together with the gas mixtures typically used. A detailed discussion of the MAP of these various foods can be found in subsequent chapters.

Several novel technologies offer the potential of further improvements in the shelf life and safety of MAP foods. These include the use of active and intelligent packaging as discussed in Chapter 15. In particular, O₂ and C₂H₄ absorbers and CO₂ emitters, used either alone or in combination with MAs, are likely to find wider application as more cost-effective and innovative designs are commercialized.

The use of TTIs (also discussed in Chapter 15) on individual MA packs could enable the temperature to be monitored throughout the supply chain and a warning given if the food had suffered temperature abuse. Although not currently practicable due largely to cost, innovative developments enabling each individual package to have its own TTI could make this a commercial reality.

The use of hurdle technology involving MAP in combination with deliberate manipulation of a_w , pH and/or redox potential, as well as the use of preservatives, bacteriocins, ultra high pressure and edible coatings, could lead to better control of potential pathogens and thus a safer product.

In conclusion, MAP is a simple concept and is increasingly applied to many different foods, providing substantial extensions in shelf life and significant economies in production and distribution. However, MAP is not a substitute for good manufacturing practices, HACCP programs and proper temperature control during storage and distribution. Used intelligently and responsibly, MAP is likely to become the dominant form of food preservation in the twenty-first century.

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