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Module – 24: Modified Atmosphere Packaging & Controlled Atmosphere Packaging

1.0 Introduction

The normal gaseous composition of air is nitrogen (N₂) 78.08% (v/v), oxygen (O₂) 20.96% and carbon dioxide (CO₂) 0.04%, variable concentrations of water vapour and traces of noble gases. Food spoils rapidly in air due to moisture loss or gain, reaction with oxygen and the growth of aerobic microorganisms, i.e. bacteria and moulds. Microbial growth causes changes in texture, colour, flavour and nutritional value of the food which can make food unpalatable and potentially unsafe for human consumption. Storage of foods in a modified gaseous atmosphere can maintain quality and extend product shelf life, by slowing chemical and biochemical deteriorative reactions and by slowing or preventing the growth of spoilage organisms.

Modified atmosphere packaging (MAP) is defined as *the packaging of a perishable product in an atmosphere which has been modified so that its composition is other than that of air* (Hintlian & Hotchkiss, 1986) while controlled atmosphere storage (CAS) involves maintaining a fixed concentration of gases surrounding the product by careful monitoring and addition of gases, the gaseous composition of fresh MAP foods is constantly changing due to chemical reactions and microbial activity. Gas exchange between the pack headspace and the external environment may also occur as a result of permeation across the package material.

1.1 Historical development

The first fresh carcass meat was exported from New Zealand and Australia under CAS in the early 1930s. Early developments were generally for storage and transportation of bulk foods. Scientific investigations on the effect of gases on extending the shelf life of foods were conducted in 1930 on fresh meat and poultry. Fresh poultry was stored in an atmosphere of 100% CO₂ which was found to considerably extend shelf life (Killefer, 1930).

Commercial retailing of fresh meat in MAP tray systems was introduced in the early 1970s. European meat processing and packaging developed during the 1980s with centralised production of MAP meat in consumer packs for distribution to retail outlets. In the past few years, there has been a considerable increase in the range of foods packed in modified atmospheres for retail sale including meat, poultry, fish, bacon, bread, cakes, crisps, cheese and salad vegetables. UK retail sales of products packed under MAP grew from approximately 2 billion packs in the mid 1990s to 2.8 billion packs in 1998. Carcass meat and cooked meat and meat products accounted for 29% and 15% of the total volume of MAP retail foods (Anon, 1999).

2.0 Gaseous environment

2.1 Gases used in MAP

The three main gases used in MAP are O_2 , CO_2 and N_2 . The choice of gas depends upon the food product being packed. Used singly or in combination, these gases are commonly used to increase shelf life with optimal organoleptic properties of the food. Inert or noble gases, such as argon, are used commercially for products like coffee and snacks, however, the literature on

their application and benefits is limited. Experimental use of carbon monoxide (CO) and sulphur dioxide (SO₂) has also been reported.

2.1.1 Carbon dioxide (CO_2)

It is a colourless gas with a slight pungent odour at very high concentrations. It is a suffocating and slightly corrosive in the presence of moisture. It dissolves readily in water (1.57 g/kg at 100 kPa, 20°C) to produce carbonic acid (H_2CO_3) that increases the acidity of the solution and reduces the pH. It is also soluble in lipids and some other organic compounds. The solubility of CO_2 increases with decreasing temperature. Therefore, the antimicrobial activity of CO_2 is markedly greater at refrigeration temperature. It has significant implications for MAP of foods. The high solubility of CO_2 can result in pack collapse due to the reduction of headspace volume.

2.1.2 Oxygen (O₂)

It is a colourless, odourless and highly reactive gas which supports combustion. It has a low solubility in water (0.040 g/kg at 100 kPa, 20°C). Oxygen 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 O_2 for growth. Therefore, to increase the shelf life of foods, the pack atmosphere should contain a low concentration of residual O_2 .

2.1.3 Nitrogen (N₂)

It is an inert gas with no odour, taste or colour. It has a lower density than air, non-flammable and has a low solubility in water (0.018 g/kg at 100 kPa, 20°C) and other food constituents. Nitrogen does not support the growth of aerobic microbes and therefore inhibits the growth of aerobic spoilage but does not prevent the growth of anaerobic bacteria. The low solubility of N_2 in foods can be used to prevent pack collapse by including sufficient N_2 in the gas mix to balance the volume decrease due to CO_2 going into solution.

2.2 Effect of the gaseous environment on the activity of bacteria, yeasts and moulds

Food may contain a wide range of microorganisms including bacteria and their spores, yeasts, moulds, protozoa and viruses. The major concern is to prevent the growth of bacteria, yeasts and moulds in foods. Some microorganisms may survive during the shelf life period and cause food poisoning or disease in consumers.

2.2.1 Effect of oxygen

Bacteria, yeasts and moulds have different respiratory and metabolic needs and can be grouped according to their O_2 needs (Table 1).

Table 1: Oxygen requirements of some microorganisms of relevance in modified atmosphere

Group	Spoilage organisms	Pathogens
Aerobes	Micrococcussp	Bacillus cereus
	Moulds e.g. Botrytis cinerea	Yersinia enterocolitica
	Pseudomonas spp.	Vibrio parahaemolyticus
		Camplobacter jejuni
Microaerophiles	Lactobacillus spp.	Listeria monocytogenes

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	Bacillus spp.	Aeromonas hydrophilia
	Enterobacteriaceae	Escherichia coli
Facultative anaerobes	Brocothrix thermosphacta Salmonella spp.	
	Shewanella putrefaciens	Staphylococcus spp.
	Yeasts	Vibrio spp.
Anaerobes	Clostridium sporogenes	Clostridium perfringens
	Clostridium tyrobutyricum	Clostridium botulinum

2.2.2 Effect of carbon dioxide

The antibacterial properties of CO_2 have been known for some time (Valley & Rettger, 1927). More recent work has shown that CO_2 is effective against psychrotrophs (King & Nagel, 1975) and has potential for extending the shelf life of food stored at low temperatures.

There are several theories regarding the actual mechanism of CO_2 action. In general, CO_2 increases the lag phase and generation time of microorganisms, and these effects are enhanced at lower temperatures. There appears to be an array of antimicrobial mechanisms including CO_2 lowering pH, inhibition of succinic oxidase at CO_2 concentrations greater than 10%, inhibition of certain decarboxylation enzymes and disruption of the cell membrane (Valley & Rettger, 1927; King & Nagel, 1975; Gill & Tan, 1979; Enfors & Molin, 1981, Daniels et al., 1985).

The sensitivity of selected spoilage and pathogenic bacteria to CO_2 is shown in Table 2. The growth of Gram-negative bacteria is inhibited more than that of Gram-positive bacteria. The effects of CO_2 are markedly temperature dependent, and it is therefore imperative that the integrity of temperature control across the supply chain be maintained. It has been observed that germination of spores of *Clostridium botulinum* may be stimulated by CO_2 (Eklund, 1982).

Table 10.2 Sensitivity of microorganisms relevant to modified atmosphere packaging to carbon dioxide

Inhibited by CO ₂	CO ₂ has little or no effect on	Growth is stimulated by CO ₂
	growth	
Pseudomonas spp.	Enterococcus spp.	Lactobacillus spp.
Aeromonas spp.	Brochothrix spp.	Clostridium botulinum
Bacillus spp.	Lactobacillus spp.	
Moulds including Botrytis	Clostridium spp.	
cinerea		
Enterobacteriaceae including	Listeria monocytogenes	
E. coli		
Staphylococcus aureus	Aeromonas hydrophilia	
Yersinia enterocolitica		

 CO_2 is soluble in water and lipids at low temperature, and adjustment for adsorption is required. A high concentration of CO_2 can lead to defects, e.g. increased drip in fresh meats, and to container collapse. The latter can occur where CO_2 is the major gas present, and where the gas goes into solution in the water and lipid phases of the product. To counteract this effect, an insoluble gas such as nitrogen may be added to the gas mix. When CO_2 is required to control the bacterial and mould growth, a minimum of 20% is generally used. Optimal levels appear to be in the region of 20–30%. Concentrations of 100% CO_2 may be used in bulk packs of meat and poultry.

2.2.3 Effect of nitrogen

Nitrogen is a relatively inert gas. It is used to displace air and, in particular, O_2 from MAP. Since air and consequently O_2 have been removed, growth of aerobic spoilage organisms is inhibited. It is also used to balance gas pressure inside packs, so as to prevent the collapse of packs containing high moisture and fat-containing foods, e.g. meat. Because of the solubility of CO_2 in water and fat, these foods tend to absorb CO_2 from the pack atmosphere.

2.3 Effect of the gaseous environment on the chemical and biochemical properties of foods

Food spoilage can also be caused by chemical and biochemical, including enzyme-catalysed, reactions in food.

2.3.1 Effect of oxygen

Apart from its effect on microorganisms, O_2 can promote oxidation of lipids, influence the colour of some food pigments, contribute to enzymic browning and promote off-flavours in some foods.

2.3.1.1 Lipid oxidation

Lipid oxidation is often called oxidative rancidity and is promoted by O_2 . Oxidative rancidity is a major cause of food spoilage. The reaction of O_2 with unsaturated fatty acids in fat-containing foods is a major cause of deterioration. Oxidation of unsaturated fat is referred to as autoxidation, since the rate of oxidation increases as the reaction proceeds. Hydroperoxides are the predominant initial reaction products of fatty acids with oxygen. Subsequent reactions control both the rate of reaction and the nature of the products formed. Some of these products, viz. acids and aldehydes, are largely responsible for the off-flavour characteristics of rancid foods. Removal of O_2 and its replacement with N_2 or CO_2 or mixtures thereof can inhibit the development of rancidity.

2.3.1.2 Pigment colour in meat

There are three major pigments in meat, oxymyoglobin (red), myoglobin (purple) and metmyoglobin (brown). The colour cycle in fresh meat is reversible and dynamic with constant formation and reformation of the three pigments. Brown metmyoglobin, the oxidised or ferric form of the pigment, cannot bind O_2 . The purple myoglobin, in the presence of O_2 , may be oxygenated to the bright red pigment oxymyoglobin, producing the familiar bloom of fresh meat, or it may be oxidised to metmyoglobin, producing the undesirable brown. The conversion of myoglobin to oxymyoglobin or metmyoglobin is depended on O_2 concentration. Under low O_2 environments, the reduced myoglobin is oxidized to the undesirable brown metmyoglobin pigment. Conversely, high O_2 environments favour the formation of oxymyoglobin.

2.3.1.3 Photo-oxidation of chlorophyll

The green colour of chlorophyll changes to brown/grey when oxidised to pheophytin which is not desirable. The photo-oxidation of chlorophyll and loss of desirable green colour can be significantly reduced by MAP under low O_2 levels and in opaque packages.

2.3.1.4 Oxidative off-flavours

Oxidative off-flavours can be caused by numerous oxidative reactions in food and drink products. Oxidative warmed-over flavour is a characteristic off-flavour primarily associated with cooked meats and poultry. Commercially, this affects mainly the chilled ready meals and other cook-chill products. In cooked meats and poultry held at chilled storage temperatures, this stale, oxidised flavour may become apparent within a short time.

Meat, fish, poultry, beverage and dairy products are highly susceptible to oxidative processes which can initiate a chain of reactions resulting in flavour impairment. This can occur relatively quickly. MAP under low O_2 levels can delay the onset of oxidative off-flavours.

2.3.2 Effects of other MAP gases

Nitrogen is inert and has no direct effect on the chemical and biochemical properties of foods. Because of the high solubility of CO_2 and its reaction with water to form carbonic acid, there is potential for some adverse effects on particular foods. These are probably due to the production of localised areas of low pH on or near the food surface. These effects if they do occur, and there is debate whether they occur in practice, may result in the loss of blooming some meats for example. The mechanism is likely to be associated with pH-induced protein changes including denaturation and other changes in conformation, resulting in atypical values for light absorption and reflection from the product surface.

Carbon monoxide can combine with myogloblin to form the bright red compound carboxymyoglobin that is similar in colour to oxymyogloblin. This compound is much more stable than oxymyogloblin and is one of the reasons why CO is toxic. CO also retards fat oxidation and the formation of metmyogloblin. Currently, CO is not approved for use in MAP.

3.0 Packaging materials

Plastic packaging materials may consist of a monolayer formed from a single plastic, but most MAP films are multilayer structures formed from several layers of different plastics. Plastics packaging for MAP applications is most commonly found in the form of flexible films for bags, pouches, pillow packs and top webs or as rigid and semi-rigid structures for base trays, dishes, cups and tubs. Commonly used plastic flexible laminates are produced from polyethylene (PE), polypropylene (PP), polyamide (nylons), polyethylene terephthalate (PET), polyvinyl chloride (PVC), polyvinylidene chloride (PVdC) and ethylene vinyl alcohol (EVOH). Rigid and semi-rigid structures are commonly produced from PP, PET, unplasticised PVC and expanded polystyrene.

Material	Application
UPVC/PE	Thermoformed base tray, Pre-formed base tray
PET/PE	Thermoformed base tray
XPP/EVOH/PE	Thermoformed base tray
PS/EVOH/PE	Thermoformed base tray
PET/EVOH/PE	Thermoformed base tray
PVdC coated PP/PE	Lidding film
PVdC coated PET/PE	Lidding film
PA/PE	Lidding film, Flow wrap film
PA/ionomer	Flow wrap film
PA/EVOH/PE	Flow wrap film
PET	Pre-formed base tray

 Table 10.3
 Typical plastic-based packaging structures for MAP applications

PP Pre-for	ned base tray
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3.1 Selection of plastic packaging materials

Several factors must be considered when selecting package materials for MAP applications.

3.1.1 Food contact approval

Packaging materials in contact with food must not transfer components from the packaging to the food product in amounts that could harm the consumer.

3.1.2 Gas and vapour barrier properties

Packaging materials for MAP must have the required degree of gas and vapour barrier for the particular food application.

3.1.3 Optical properties

Good optical properties, such as high gloss and transparency, are essential for bag, pouch and top web materials to satisfy consumer demand for a clear view of the product. To provide attractive appearance and shelf impact, some base tray materials are available in various colours which enhances the visual appeal of the product. PET, PP and EPS trays are supplied in a range of colours. PVC trays are generally used in their natural form to provide a transparent pack.

3.1.4 Antifogging properties

Condensation (fogging) of water vapour on the inner surface of food packs can occur when the temperature of the pack environment is reduced, resulting in a temperature differential between the pack contents and the packaging material. Fogging of the inner surface of lidding film is a result of light scattering by the small droplets of condensed moisture that leads to poor product visibility and an aesthetically unpleasing appearance of the pack. This can be overcome by applying antifogging agents to the plastic heat sealing layer, either as an internal additive or as an external coating. These chemicals decrease the surface energy of the packaging film which enables moisture to spread as a thin film across the under surface of the pack rather than collecting as visible droplets. Antifogging agents include fatty acid esters. Most lidding materials are available with antifog properties, and commonly treated plastics include LDPE, LLDPE, EVA and PET.

3.1.5 Mechanical properties

Resistance to tearing and puncture and good machine handling characteristics are important in optimising the packaging operation and maintaining pack integrity during forming and subsequent handling and distribution.

3.1.6 Heat sealing properties

Effective heat seals are essential for maintaining the desired gas composition within the pack. Heat seal quality is dependent on many factors including seal material, seal width and machine settings such as temperature, pressure and dwell time.

3.2 Modified atmosphere packaging machines

The function of MAP machines is to retain the product on a thermoformed or pre-formed base tray, or within a flexible pouch or bag, modify the atmosphere, apply a top web if required, seal the pack and cut and remove waste trim to produce the final pack. Different types of MAP machines are discussed below.

3.2.1 Chamber machines

They are generally used for low production throughput, with pre-formed pouches, though tray machines. The filled pack is loaded into the machine, the chamber closes, a vacuum is created on the pack and back flushed with the modified atmosphere. Heated sealing bars seal the pack, the chamber opens, packs are removed and the cycle continues. These machines are cheap and easy to operate but relatively slow and labour intensive. Some chamber machines can handle large packages and are suitable for bulk packs.

3.2.2 Snorkel machines

They operate without a chamber and use pre-formed bags or pouches. The bags are filled and positioned in the machine. The snorkel is introduced into the bag, draws a vacuum and introduces the modified atmosphere. The snorkels withdraw and the bag is heat sealed. Bag inbox bulk products and retail packs in large MAP master packs can be produced on these machines.

3.2.3 Form-fill-seal tray machines

Form-fill-seal (FFS) machines form pouches from a continuous sheet of roll stock (flow wrap), or form flexible or semirigid tray systems comprising a thermoformed tray with a heat sealed lid. FFS machines may be orientated in a vertical plane or a horizontal plane. FFS machines using pre-formed trays or producing thermoformed trays are almost exclusively horizontal machines. Horizontal form-fill-seal MAP machines are used extensively in the food industry.

4.0 Modification of the pack atmosphere

4.1 MAP machines use mainly one of the two techniques to modify the pack atmosphere.

4.1.1 Gas flushing

This method employs a continuous gas stream that flushes air out from the package prior to sealing. This method is less effective at flushing air out of the pack results in residual oxygen levels of 2–5%. Hence, it is not suited for oxygen-sensitive food products. Generally, gas flushing machines have a simple and rapid operation and therefore a high packing rate.

4.1.2 Compensated vacuum gas flushing

This method uses a two-stage process:

The evacuation stage – a vacuum is pulled on the pack to remove air. Generally, it is not possible to achieve a full vacuum, since reduced pressures will result in water to boil. The vacuum achieved is generally between 5 and 10 mm of Hg. As a general rule, the cooler and drier the food, the lower the achievable vacuum.

Gas flushing stage – the pack is flushed with the modified gas mix. The evacuation of air from the pack results in lower residual oxygen levels than that achieved by gas flushing, and therefore this method is better suited for packing oxygen-sensitive products.

The two-stage process employed by the compensated vacuum method results in a lower packaging rate than that possible with gas flushing.

4.2 Sealing

An effective heat seal is critical for maintaining the quality and safety of the packaged product. Film factors (thickness and surface treatments) and plastic composition (resin type, molecular weight distribution and presence of additives) will determine the machine settings for the sealing operation. The correct combination of time, temperature and pressure of the seal bars is necessary to produce a good seal. Insufficient dwell time or temperature can result in ineffective seals that separate at the bond interface. Excessive dwell time or temperature can result in weakness adjacent to the seal area.

4.3 Cutting

Packs are discharged as a continuous arrangement of filled and sealed packs from a thermoform-fill-seal machine, and therefore, the final operation is to separate into individual packs. This can be carried out by two methods - die cutting and longitudinal and transverse cutting.

Die cutting is achieved in one operation. A shaped blade is forced through the film which is clamped in place by a frame assembly. Transverse cutting separates packs into rows and is carried out by guillotines or punches which are driven through the film that is supported by anvils. This may be carried out in conjunction with longitudinal cutting where circular knives cut Post Grad through the tray flanges parallel to the length of the film.

5. MAIN FOOD TYPES

5.1 Raw red meat

The major causes of spoilage of meat are microbial growth and oxidation of the red oxymyoglobin pigment. The desirable red colour of the oxymyoglobin pigment has to be maintained, by having an appropriate O₂ concentration in the pack atmosphere, and at the same time minimise the growth of aerobic microorganisms. Highly pigmented red meats, such as venison and wild boar, require higher concentrations of O₂.

Aerobic spoilage bacteria, such as Pseudomonas species, normally constitute the major flora on red meats. Since these bacteria are inhibited by CO₂, it is possible to achieve both red colour stability and microbial inhibition by using gas mixtures containing 20–30% CO₂ and 70–80% O₂. These mixtures can extend the chilled shelf life of red meats from 2-4 days to 5-8 days. A gas/product ratio of 2:1 is recommended.

5.2 Raw poultry

Microbial growth, particularly growth of *Pseudomonas* and *Achromobacter* species, is the major factor limiting the shelf life of raw poultry which are effectively inhibited by CO₂. The inclusion of CO₂ in MAP at a concentration in excess of 20% can significantly extend the shelf life of raw poultry products. CO₂ concentrations higher than 35% in the gas mixture of retail packs are not recommended because of the risks of pack collapse and excessive drip. Nitrogen is used as an inert filler gas, and a gas/product ratio of 2:1 is recommended. Since pack collapse is not a problem for bulk MAP master packs, gas atmospheres of 100% CO₂ are frequently used. It is advisable to maintain refrigeration temperature as some pathogens are not inhibited by CO₂.

5.3 Cooked meat products

The principal spoilage mechanisms that limit the shelf life of cooked meat products are microbial growth, colour change and oxidative rancidity. For cooked meat products, the heating process should kill vegetative bacterial cells, inactivate degradative enzymes and fix the colour. Hence, spoilage of cooked meat products is primarily due to post-process contamination by microorganisms. The colour of cooked meats is susceptible to oxidation, so low levels of O₂ is maintained in packs. MAP using CO_2/N_2 mixes (25–50% CO_2 and 50–75% N_2) along with a gas/product ratio of 2:1 is widely used to maximise the shelf life and inhibit the development of oxidative off-flavours and rancidity.

5.4 Fish and fish products

Spoilage of fish and shellfish results from changes caused by three major mechanisms: (i) the breakdown of tissue by the fish's own enzymes (autolysis of cells), (ii) growth of microorganisms, and (iii) oxidative reactions. MAP can be used to control mechanisms (ii) and (iii) but has no direct effect on autolysis which is major cause of spoilage and reduces the effect of MAP.

The major spoilage bacteria in processed fish are *Pseudomonas, Moraxella, Acinetobacter, Flavobacterium, Cytophagaspecies* and *Clostridium botulinum*.

Generally, O_2 is included in MAP of white non-processed (i.e. non-fatty) fish to control the growth of *clostridia*. Gas mixtures of 30% O_2 , 40% CO_2 and 30% N_2 are used but it will not significantly enhance the shelf life of oily or fatty fish. High, 40%, CO_2 mixes along with 60% N_2 are generally used for smoked and fatty fish.

5.5 Fruits and vegetables

Fresh produce continues to respire after harvesting and produce CO_2 , water vapour and ethane. Ethane promotes ripening and softening of tissues and reduces the shelf life if not controlled. Hence, the goal of MAP for fruits and vegetables is to reduce respiration to extend shelf life while maintaining quality. Respiration can be reduced by lowering the temperature, lowering the O_2 concentration, increasing the CO_2 concentration and by the combined use of O_2 depletion and CO_2 enhancement of pack atmospheres. If the O_2 concentration is reduced beyond a critical concentration, anaerobic respiration will be started and produce ethanol, acetaldehyde and organic acids, which leads to undesirable odours and flavours and a marked deterioration in product quality. While increasing the CO_2 concentration will also inhibit respiration, high concentrations may cause damage in some species and cultivars. O_2 concentrations below about 3% can induce anaerobic respiration in many species of fresh produce.

The use of low concentrations of O_2 and elevated levels of CO_2 can have a synergistic effect on slowing down respiration and ripening. While the mechanisms whereby MAP can extend the shelf life of fresh produce are not fully understood, it is known that the low O_2 /high CO_2 conditions reduce the conversion of chlorophyll to pheophytin, decrease the sensitivity of plant tissue to C_2H_4 , inhibit the synthesis of carotenoids, reduce oxidative browning and discolouration and inhibit the growth of microorganisms.

Major pathogens as far as fresh produce is concerned are *L. monocytogenes* and *C. botulinum*. *L. monocytogenescan* grow under reduced O_2 levels and is not markedly inhibited by CO_2 and can also grow at lower temperature.

Equilibrium MAP has been used for fresh produce. Essentially, this involves using knowledge of the permeability characteristics of particular packaging films, along with the respiration characteristics of the product to balance the gas transfer rates of O_2 and CO_2 through the package with the respiration rate of the particular product.

Increasingly, gas packing fresh produce along with $CO_2/O_2/N_2$ gas mixtures is being used. This approach may have benefit in reducing enzymic browning reactions before a passively generated equilibrium modified atmosphere has been established.

5.6 Dairy products

MAP can increase the shelf life of a number of dairy products viz. whole milk powders, cheeses and fat spreads.

Whole milk powder is particularly susceptible to the development of off-flavours due to fat oxidation. Commercially, the air is removed under vacuum and replaced with 100% N_2 or N_2/CO_2 mixes and the powder is hermetically sealed in metal cans.

Cheddar cheese is traditionally vacuum packed. Increasingly MAP is being used with high CO_2 concentration gas mixes. This has the advantage of obtaining a low residual O_2 content and a tight pack due to the CO_2 going into solution. It is important to balance this process using the correct N_2 level in the gas mix so as to avoid excessive pressure being put on the pack seal.

Use of N_2/CO_2 atmospheres has significant potential for extending the shelf life of cottage cheese. The cottage cheese is a high-moisture, low-fat product that is susceptible to a number of spoilage organisms including *Pseudomonas* spp. Use of gas mixtures containing 40% CO₂ balanced with 60% N₂ can increase the shelf life significantly.

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