

SESSION 5 - MICROBIOLOGY AND STORAGE LIFE OF CHILLED FRESH MEATS

Microbiology and storage life of chilled fresh meats

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Introduction

The storage life of fresh meats held under refrigeration may be extended by several methods, including vacuum packaging, storage in modified gas atmospheres and irradiation. Such procedures may also be combined with pre-treatment of the meat to reduce the initial bacterial contamination.

In this paper the effects of the procedures listed above on the microbiology and storage life of red meats stored at 0-5°C are discussed. The significance of environmental factors in controlling the growth of the various types of spoilage bacteria are compared. Methods of assessing the acceptability of meats and the possibility of developing chemical definitions of spoilage are considered. Areas requiring further research are identified.

It has been possible to list only some of the relevant literature here. Background information may be found in the articles of Ingram and Simonsen (1980) and Dainty, Shaw and Roberts (1983).

Aerobic storage

Although the total aerobic count on beef carcasses following the completion of dressing may be as high as $10^5/cm^2$, commonly less than 1% of these bacteria are psychrotrophs and hence able to grow below 8-10°C. The flora on refrigerated fresh meats is predominantly composed of Gram-negative bacteria and *Pseudomonas* spp. become dominant as storage progresses, particularly at high relative humidity. Spoilage usually becomes significant when numbers reach ca. $10^7/cm^2$ and is characterized by putrefactive "off" odours followed by slime production (Ayres, 1960). The time taken to reach spoilage is directly related to the initial number of (psychrotrophic) bacteria on the meat. Even with a low initial count, the maximum storage life at 0°C is only about two weeks.

The available data indicate that the pseudomonads have faster growth rates than the other types of bacteria present and according to Gill and Newton (1977) this advantage increases as the temperature is reduced. These workers examined the utilization of substrates by pure cultures of spoilage organisms growing on meat and found that substrate exhaustion at the meat surface did not limit bacterial growth under aerobic conditions. They suggested that the maximum cell density of aerobic spoilage cultures is determined by oxygen limitation of growth. Thus it appears that growth rate determines the composition of the aerobic spoilage flora. This situation seems to be a simple one and significant interactions between the various components of the flora appear to be unlikely.

Because of their importance in spoilage, there has been considerable interest in the taxonomy of pseudomonads isolated from meats, with most workers reporting that many strains could not be readily identified with the described

species and biotypes (Dainty, Shaw and Roberts, 1983). Recently there have been two numerical taxonomic studies of isolates from meat. Most strains were non-fluorescent and have been designated as *Pseudomonas fragi*. Fluorescent strains were also present and were identified as *Pseudomonas fluorescens* and *Pseudomonas putida* (Molin and Ternström, 1982; Shaw and Latty, 1982).

Vacuum-packaged fresh meat

If conditions in a culture are made anaerobic, then aerobes such as *Pseudomonas* cease to grow and facultative organisms grow more slowly. Storage of meats in gas atmospheres other than air has been used for about fifty years and this concept led to vacuum packaging which is a form of modified atmosphere storage. Vacuum packaging is now widely used especially for cuts of beef and forms the basis of a considerable world trade in chilled meat.

When fresh meat is vacuum packaged in bags made of plastic film of low gas permeability, the oxygen is consumed and carbon dioxide is produced (recently discussed by Enfors and Molin, 1984). Providing the packaging has been correctly done, there is very little headspace within a vacuum-packaged cut of beef. This makes accurate gas analysis difficult but the atmosphere contains less than 1% oxygen, some 20-40% carbon dioxide with the remainder being nitrogen (Dainty et al., 1979).

The storage life of vacuum-packaged beef at 0°C is 10-12 weeks (Newton and Rigg, 1979; Egan, 1983), providing the following criteria are met:-

- (i) the meat must have been produced using good manufacturing practice,
- (ii) only meat of normal ultimate pH is used (pH must be lower than 6.0 and preferably 5.8 or lower),
- (iii) the packaging film used must have a low permeability to gases ($<100 ml$ of $O_2/m^2/24h/atm$ measured at 25°C and 98% rh. Films with permeability $<30 ml$ of $O_2/m^2/24h/atm$ give a greater margin of safety).
- (iv) there must be good temperature control throughout the storage period.

There is considerable literature on the microbiology of vacuum-packaged meat, with most studies having been done using beef. When reading this it is important to note that technological factors may make direct comparisons of results difficult, particularly as newer less-permeable packaging films have replaced older types. Nylon-based films have caused a particular problem since their permeability changes markedly with relative humidity (Rigg, 1979). This was not appreciated in some of the early studies. Further in some studies the pH of the meat was not quoted and presumably was not taken into account. These two factors, meat pH and packaging film permeability actually control the composition of the microbial flora of chilled vacuum-packaged beef.

The spoilage of vacuum-packaged meat

Determining when vacuum-packaged meat is spoiled presents problems. In the absence of visual spoilage, packs are opened and checked for the presence of an "off" odour. Any "off" odour must be distinguished from the "confinement"

or "vacuum-pack" odour characteristic of this type of meat prior to spoilage. Provided the meat is acceptable following these tests, it is usually cooked under controlled conditions and subjected to evaluation by a taste panel (Carpenter et al., 1976; Newton and Rigg, 1979; Egan and Shay, 1982).

Our studies have used a trained analytical taste panel of 15 members and have shown that spoilage is first manifested as an "off" or changed flavour. When first noted it is described by panellists as cheesy, sour and acid but later in storage also as bitter and liver-like. "Off" odours are also noted, but these occur later in storage and the panel results clearly indicate that the flavour change is the major defect (Egan, 1983).

These organoleptic changes are attributed to the accumulation of end products resulting from the growth of lactic acid bacteria, which reach a population on the lean surface in the range of $1-5 \times 10^7/cm^2$ and dominate the flora (Seideman et al., 1976; Dainty et al., 1979; Newton and Rigg, 1979; Egan, 1983). Taste panel evaluation of meat carrying pure cultures of lactic acid bacteria has confirmed that these organisms do produce the types of flavour changes listed above (Egan and Shay, 1982; Hanna et al., 1983). Significant spoilage is not usually noted by the taste panel until several weeks after the count of lactic acid bacteria reaches $10^7/cm^2$ (Egan, 1983). Thus the total count is not a useful indicator of the spoilage of vacuum-packaged normal pH beef stored in plastic film of low gas permeability. When beef is stored vacuum-packaged at 5°C in the absence of a significant population of bacteria (so called "sterile" meat) spoilage still occurs and is again first manifested by the development of an undesirable flavour (Egan and Shay, 1982). This also occurs at 0°C and the panel results indicate a storage life of ca. 16 weeks at this temperature.

It is difficult to directly compare taste panel assessments of spoilage with a consumer appreciation of the meat. Almost certainly a trained taste panel will detect organoleptic changes in the cooked meat in a controlled experiment sooner than most consumers in the home. Thus it is likely that acceptance of the panel result is erring on the side of safety.

Over a period of years a number of complaints concerning the acceptability of meat exported from Australia have been referred to our laboratory. Invariably rejection or downgrading was based on a visual assessment (usually an undesirable colour was noted). In all cases, at least one of the criteria mentioned previously was not met. It would be very useful if an objective method for evaluating the condition of vacuum-packaged meat was available.

Vacuum-packaged cuts of beef are commonly broken down at central processing plants and repacked for retail sale. Relatively little information about the microbiological characteristics and the storage life of the repacked meat is available (Vanderzant et al., 1982).

The microbiology of vacuum-packaged beef

The microbiology of packaged meats has now been studied for some 20 years. In 1962, Jaye, Kittaka and Ordal reported major differences in the bacterial flora of meat stored in gas-permeable and impermeable films. In the same year, Ingram outlined the microbiological principles involved in repacking meats. Subsequently extensive studies have established the relative

significances of the various factors that control the composition of the bacterial flora of vacuum-packaged meat.

The composition of the flora of vacuum-packaged beef is largely determined by a combination of two factors, the gas permeability of the packaging film and the muscle pH. There has been considerable discussion as to whether the inhibition of the growth of certain spoilage organisms is due to a lack of oxygen or to inhibition by carbon dioxide. This cannot be discussed in detail here, but it appears that the effect of carbon dioxide is equally as important as oxygen limitation (Erichsen and Molin, 1981; Shaw and Roncaroli, 1982).

In the case of *Pseudomonas* spp. there is very little growth provided the criteria concerning meat pH and film permeability mentioned earlier are met (Newton and Rigg, 1979; Erichsen and Molin, 1981). In certain cases where some growth has been observed, the reasons for this are not known but may possibly be related to initial inoculum effects (Sutherland, Patterson and Murray, 1975; Dainty et al., 1979). Inhibition by carbon dioxide is the main reason for the failure of pseudomonads to grow on vacuum-packaged beef but the very low concentrations of oxygen also probably contribute.

Brochothrix thermosphacta does not grow on beef under anaerobic conditions if the meat pH is 5.8 or lower (Campbell et al., 1979). On vacuum-packaged beef of normal pH the amount of growth of this organism is controlled by the film permeability. When low permeability films are used its population does not normally exceed ca. $10^4/cm^2$ and it is unlikely to be significant in spoilage. The inhibition of the growth of *B.thermosphacta* on vacuum-packaged beef when the pH is 5.8 or lower is caused by the lactate present in the muscle. Not only does the concentration of lactate in the meat increase as the pH falls but, a greater proportion of it is in the undissociated form, which is the active inhibitor. Further, this bacterium is more sensitive to inhibition under anaerobic conditions. These factors combine to produce a situation where growth cannot occur if the muscle pH is below 5.8 and oxygen is unavailable (Grau, 1980).

Thus the effect of oxygen permeability on the growth of *B.thermosphacta* is largely an indirect one. This is confirmed by the fact that this organism can grow to a high population ($>10^7/cm^2$) on vacuum-packaged beef of high pH even when the meat is in bags made of film of extremely low gas permeability (Campbell et al., 1979). *B.thermosphacta* is sensitive to inhibition by carbon dioxide under anaerobic conditions (Molin, 1983). However it seems unlikely that the concentrations reached in vacuum-packaged meat are high enough to be significant.

There have been relatively fewer studies of the psychrotrophic Gram-negative bacteria of vacuum-packaged beef. As with *B.thermosphacta* they are unlikely to exceed ca. $10^4/cm^2$ on meat of normal pH packaged in bags made of low gas permeability. Shaw and Roncaroli (1982) showed that oxygen limitation contributes to their inhibition in vacuum packs. Again, whilst carbon dioxide may have some influence on their growth, it may not be present in high enough concentration to cause significant inhibition.

The amount of growth of fermentative Gram-negative bacteria on vacuum-packaged beef is affected by muscle pH with greater populations being reached on high pH meat (Gill and Newton, 1979; Erichsen, Molin and Möller, 1981). Most of

these studies were done using temperatures of 4-5°C rather than 0°C and this particularly favours the growth of this group of organisms (Egan and Shay, 1984). The concentration of lactate in the muscle may be a factor restricting the growth of these organisms on vacuum-packaged normal pH beef as is the case with *B.thermosphaeta* (Grau, 1981).

Vacuum-packaged high pH meat

When meat pH is 6.0 or higher, a wider range of bacteria are able to grow. The reasons for this have already been discussed. In particular Gram-negative bacteria reach much higher populations. This results in more rapid spoilage which is due to putrefaction rather than to souring. Storage life may also be terminated by colour defects particularly greening. This discolouration is due to the formation of sulphmyoglobin, a green pigment produced when hydrogen sulphide reacts with myoglobin (Nicol, Shaw and Ledward, 1970). Hydrogen sulphide is produced as a result of the degradation of cysteine. *Alteromonas putrefaciens* is a strong producer of this gas, but because it is unable to grow below pH 6.0, it is only a problem on high pH meat. Other Gram-negative organisms including *Serratia liquefaciens* and *Hafnia alvei* are able to produce hydrogen sulphide during growth on meat, but their significance as a cause of greening of vacuum-packaged meat remains to be determined (Gill and Newton, 1979; Hanna et al., 1979). The spoilage of high pH vacuum-packaged beef has been studied by several groups of workers (Bem, Hechelmann and Leistner, 1976; Taylor and Shaw, 1977; Erichsen, Molin and Möller, 1981).

Vacuum-packaged lamb and pork

Although there have been fewer studies of the microbiology of vacuum-packaged lamb and pork than there have of beef, there has recently been an increase of interest in these products. Shaw, Harding and Taylor (1980) reported a storage life of six weeks for vacuum-packaged cuts of lamb stored at 0-1°C. *B.thermosphaeta* and lactic acid bacteria were the possible cause of the cheesy/sour odours which terminated storage life.

Lamb carcasses may be reduced in size by a process known as telescoping (Eustace, 1984). In this process the hind legs are forced up into the thoracic cavity and by this means there is a considerable volume reduction with significant savings in transport costs. Such carcasses are vacuum-packaged for export from Australia. However there are still voids in the packs and these, together with a considerable volume of exudate which commonly collects, create problems of a microbiological nature. Further studies aimed at increasing the storage life of this product are underway in our laboratory.

The microbiology of vacuum-packaged pork has recently been studied in some detail in our laboratory and some of the results are presented elsewhere in these Proceedings (Egan and Shay, 1984). A major problem with both pork and lamb is that there is a much higher incidence of high pH meat than occurs with beef. Further there may be a greater proportion fat. These factors combine to give a much shorter storage life than that of vacuum-packaged normal pH beef.

The lactic acid bacteria of vacuum-packaged meat

The occurrence, properties and significance of the lactic acid bacteria of

meats have been discussed recently (Egan, 1983). Since then the first numerical taxonomic study of the lactic acid bacteria isolated from vacuum-packaged meats has appeared (Shaw and Harding, 1984). This study has confirmed the significance of streptobacteria on packaged meats, since two large clusters (together 88 of the 100 isolates studied) consisted of these organisms. One cluster contained isolates not identifiable with any described species, and the other contained strains provisionally identified with *Lactobacillus sake* or *Lactobacillus bavaricus* according to the isomer of lactic acid produced.

In many of the studies of bacteria from meats, multiple isolates have been chosen from each piece of meat. In the case of the lactic acid bacteria, the population of some $10^7/cm^2$ present after 4-5 weeks storage, arises from an initial population which is probably less than $10/cm^2$. Some packs examined in our laboratory have carried what appears to be a pure culture and the taking of multiple isolates from a pack may result in the study of many siblings. Thus further studies of these organisms appear warranted.

In 1981 we reported the isolation of a *Lactobacillus* that produces hydrogen sulphide during growth on vacuum-packaged beef of normal pH (Shay and Egan, 1981). Subsequently we isolated several other strains with this property from vacuum-packaged beef and pork and in three cases these organisms appear to have been associated with mild greening. Further, we have now isolated similar organisms from vacuum-packaged sliced processed meats.

Lactic acid bacteria have been reported to inhibit the growth of *B.thermosphaeta* on vacuum-packaged beef and on beef stored under anaerobic conditions (Roth and Clark, 1975; Newton and Gill, 1978). It has been claimed that the inhibition is due to an antibiotic produced by the lactobacilli (Gill and Newton, 1978; Newton and Gill, 1978). However no direct evidence for such a compound has been produced and it is also possible that the inhibition is due to competition for substrate (Shay, Egan and Rogers, 1984).

Pre-treatment of meat with solutions of organic acids as a means of extending the storage life of vacuum-packaged meat

The problem of obtaining an adequate storage life for vacuum-packaged high pH meats has already been discussed. One approach to this problem has been to treat carcasses or cuts of meat so as to reduce the microbial population prior to processing.

The effect washing or spraying with hot or cold water and with chlorinated water has been shown to reduce microbial counts on fresh meat (Bailey, 1971; Kotula et al., 1974) and Beimüller, Carpenter and Reynolds (1973) found that spraying pork carcasses with acetic acid solutions caused a 1-2 log₁₀ reduction in contamination, depending upon the pH of solution used. Eustace (1984) has shown that immersing lamb carcasses in a 1.5% solution of acetic acid for 10 seconds at 55°C caused a 1.3 log₁₀ reduction in contamination. Further, when these carcasses were vacuum-packaged and stored at 0°C, there was a significant extension in storage life.

Recently Cacciarelli et al., (1983) reported that sanitizing boneless pork loins with a 2% acetic acid solution prior to vacuum packaging, resulted in a significantly lower total count (and count of lactobacilli) throughout 28 days

storage at 4°C. They suggested, that providing the slight discolouration caused by the acid treatment could be prevented, the technique should be useful in extending the storage life.

As a part of studies of the microbiology of vacuum-packaged pork we have examined the effect of pretreatment of the meat with acetic acid on the microbiology of vacuum-packaged pork stored at 0°C. Pork loins (pH 6.2-6.6) were cut into 4 equal portions. Two portions from each loin were immediately vacuum packaged and the remaining portions were immersed in a 1.5% (v/v) solution of acetic acid for 10 sec at 55°C. After draining for 5-10 min the meat was vacuum-packaged. The meat was sampled at various times for microbiological analysis, with all four packs from the one loin being examined at the same time.

Dipping in acetic acid caused a reduction of at least 90% in the starting count (Fig. 1a). The organisms present were predominantly mesophiles and it is not known whether the acid treatment was effective against the psychrotrophs. Whilst the total count on the treated samples was lower than that on the controls throughout the experiment the amount of inhibition was not large (Fig. 1a). There was also only slight inhibition of the growth of the lactic acid bacteria (Fig. 1b). In contrast, the treatment strongly inhibited the growth of the Gram negatives (Fig. 1a) and caused significant inhibition of the growth of *B.thermosphaeta* (Fig. 1b). Similar results were obtained for the fat surface and for meat stored at 5°C. The treatment caused a slight initial discolouration but this was hardly noticeable after 1-2 weeks storage.

These results suggest that acetic acid treatment may be of particular use under conditions where Gram-negative bacteria are likely to cause spoilage i.e. at 4-5°C. It may also be effective in reducing the amount of growth of potential Gram-negative pathogens such as *Aeromonas hydrophila*. It is largely the residual effect of the acetic acid rather than its' effect on the starting count which has the potential to make this a useful commercial procedure.

Spraying with lactic acid solutions has been shown to be effective in reducing the initial contamination of beef carcasses (Snijders et al., 1979) and the ecological consequences of this technique have been discussed by van Netten and Mossel (1980). Further studies on the usefulness of pretreating fresh meats with these compounds are required since they have the potential to increase the storage life and improve the microbiological quality of vacuum-packaged meats.

Modified atmosphere storage of fresh meats

Recently there has been renewed interest in the storage of meats in modified or controlled gas atmospheres (Christopher et al., 1979; Christopher et al., 1980; Finne, 1982). In the case of fresh meats, this technique has the potential to solve the problem of the storage of meats which have an inadequate storage life when vacuum-packaged (lamb and pork). The use of carbon dioxide as a preservative for foods has been reviewed recently (Clark and Takács, 1980). A concentration of 20%, which is commonly used for retail meats, does not prevent the growth of *B.thermosphaeta* and some Gram-negative organisms, and so high concentrations are needed to obtain a long storage life.

Molin and co-workers have made extensive studies on the storage of fresh meats in atmospheres of carbon dioxide. Storage of pork in 100% carbon dioxide at either 0°C or 4°C resulted in a flora on the lean surface consisting entirely of *Lactobacillus* spp. These organisms were also dominant on the fat but at 4°C *Aeromonas* spp. were present. These workers suggested that the storage life of pork stored in 100% carbon dioxide was 5 weeks at 4°C and 3 months at 0°C (Enfors, Molin and Ternström, 1979; Blickstad and Molin, 1983). Our own microbiological data largely agrees with that of the Swedish workers however we have detected high populations ($10^6/cm^2$) of Gram-negative bacteria on high pH pork stored for 2-3 weeks at 5°C. These organisms were provisionally identified as *Aeromonas* spp. and had grown in an atmosphere which contained more than 90% carbon dioxide.

The application of modified atmosphere storage using carbon dioxide presents a number of technical problems. To be fully effective the meat must be held in an excess of the gas. This may be accomplished using rigid containers but these present problems in commercial use. The meat may be stored sealed in plastic bags made of film of low gas permeability, but to be effective the volume of gas added must be at least twice the volume of the meat. After packaging gas is absorbed, the amount depending upon the temperature. We have varied the gas to meat ratio in packs and found that if conditions are carefully controlled we can add just sufficient gas such that it is absorbed during the initial stages of storage and the pack tightens. Unfortunately, this amount of gas is not optimal microbiologically.

Modified atmosphere storage is also widely used for retail and consumer packs. Atmospheres consisting of mixtures of carbon dioxide with air or oxygen are most commonly used. A mixture of 20% carbon dioxide with 80% oxygen has the advantage of the inhibitory effect of the carbon dioxide on bacterial growth together with the colour enhancing effect of the oxygen. Meat stored in such atmospheres remains acceptable for up to two weeks at temperatures up to 4°C (Taylor, 1983).

Potential pathogens on packaged fresh meats

Several potential pathogens have been reported to occur on vacuum-packaged meat (especially high pH meat) and on meat stored in modified gas atmospheres. For pathogens, these organisms are unusual in their ability to grow at temperatures below 5°C. *Yersinia enterocolitica* has been detected on vacuum-packaged beef, pork and lamb (Hanna et al., 1976; Seelye and Yearbury, 1979; Gill and Newton, 1979; Erichsen, Molin and Möller, 1981; Myers et al., 1982). Grau (1981) has shown that this organism will grow in pure culture to a population in excess of $10^8/g$ on beef of pH 6.0 to 6.2 stored under anaerobic conditions at 5°C. To date there appear to be no reports of food poisoning caused by its' presence on meats, apparently because the strains usually isolated from foods are non-virulent (Zink, Lachica and Dubel, 1982).

Erichsen, Molin and Möller (1981) reported the presence of *Erysipelothrix*-like organisms on beef stored at 4°C. They were isolated not only from vacuum-packaged meat but also from meat stored in an atmosphere of 100% carbon dioxide. We have isolated similar organisms from vacuum-packaged pork. They are Gram-positive catalase-negative rods which grow at 37°C and are strong

producers of hydrogen sulphide. Preliminary studies have indicated that these bacteria are not antigenically related to *Erysipelothrix rhusiopathiae* and they may yet prove to be unusual strains of lactobacilli.

Aeromonas hydrophila has been isolated from vacuum-packaged pork by Myers et al. (1982) and from pork stored in nitrogen by Enfors, Molin and Ternström (1979). It has also been recovered from the fat surface of pork loins stored in 100% carbon dioxide (Blickstad and Molin, 1983). These isolations were all from meats stored at 4-5°C. The significance of food-borne strains of this organism as human pathogens is not yet clear, however the bacteria isolated by Myers et al. (1982) were cytotoxic to tissue cell cultures. This organism has been reported as being responsible for an outbreak of food-borne illness in the Soviet Union. The vehicle in this outbreak was mackerel fillets (Kalina, 1977).

The available information suggests that these organisms may not be a problem on meats stored at 0°C. However since they may occur in very high numbers on meat at 4-5°C further studies are needed.

Irradiation

Several years ago the Joint Expert Committee on Wholesomeness of Irradiated Foods (convened by the World Health Organization) concluded that any food irradiated to 10 kGy is safe for human consumption. Since then there has been a great increase in interest in the preservation of meat by irradiation (Ingram and Roberts, 1980; Teufel, 1981; Niemand, van der Linde and Holzappel, 1981; Gibbs, 1982; Kampelmacher, 1983). Whilst there is no doubt about the effectiveness of irradiation in improving the microbiological quality of meats, consumer acceptance and organoleptic changes induced in sensitive foods remain as problems. Packaged meats are obviously most suitable for irradiation since the problem of recontamination is avoided. In Australia, vacuum-packaged sliced cooked meats present particular problems because of short storage lives.

We have recently been using the laboratory's trained analytical taste panel to evaluate the effect of radiation pasteurization (radurization) on the storage life and organoleptic quality of packaged meats. Vacuum-packaged sliced corned beef ($a_w = 0.950$, pH 6.5; 90 x 150g packs) was obtained from a local processor immediately after slicing and packaging. The packs were allocated at random to the three treatments (frozen-stored control, normal unirradiated stored and irradiated to 4 kGy). All packs except the frozen controls were stored at 5°C in the dark. Details of taste-panel procedures have been published (Egan, Ford and Shay, 1980). Fig. 2a shows the effect of irradiation on the microbial flora. The initial quality of the meat was not good with a total count of psychrotrophs in excess of $10^7/g$ (determined by incubation of plates at 5°C). The bacteria grew readily and reached a count in excess of $10^8/g$ after about 14 days. Lactic acid bacteria were the major component of the flora and *B.thermosphacta* reached a population in excess of $10^6/g$. Following irradiation the starting count was reduced by more than five log₁₀ units (Fig. 2a). On this meat viable organisms were first detected after 13 days storage. They grew slowly and had reached a population of only ca. $10^5/g$ by the end of the experiment. In contrast to the normal flora on the unirradiated packs which was dominated by Gram-positive organisms, the bacteria which eventually grew on the irradiated meat were Gram negative.

Taste panel evaluation of the meat the day following irradiation showed that the irradiated meat had undergone significant changes in flavour ($P < 0.05$) and aroma ($P < 0.01$) when compared to the normal and frozen control samples. The unirradiated meat samples gradually spoiled due to bacterial growth. "Off" aroma was significantly different from that of the frozen control samples after 22 days storage ($P < 0.01$) (Fig. 2b). This indicates a storage life of about three weeks for the normal product. It was at about that time that the "off" aroma of the (unirradiated) normal samples became stronger than the results (but different) odour of the irradiated meat (Fig. 2b). Similar results were obtained for flavour. Thus the panel considered the irradiated meat less acceptable than the non-irradiated samples until three weeks of storage had elapsed (Macfarlane, Shay, Wills and Egan, unpublished results).

Corned beef is a relatively bland product and these results will be criticized on that basis and also because of the inclusion of the control treatments. Consumers presented with irradiated meat in an uncontrolled test would almost certainly be less discriminating.

There is no doubt about the effectiveness of irradiation which has the potential to produce pathogen free meats, but further studies of its use are needed. In addition, there is no point in irradiating meats which already have an adequate storage life such as vacuum-packaged normal pH beef.

The chemistry of spoilage

Spoilage of meat is usually assessed by determining the number of bacteria present and/or by evaluating its organoleptic condition. The growth of the spoilage flora leads to chemical changes in the meat and it is these changes which alter its acceptability. Thus appropriate chemical tests might prove useful in determining the quality of meats and predicting spoilage. Any rapid objective measurement of spoilage would be of considerable use to the meat industry and regulatory authorities.

A number of chemical tests have been proposed as indicators of spoilage. These include ammonia production, high pH, extract release volume and dye reduction tests. However none of these has proved useful in predicting spoilage (Ingram and Simonsen, 1980). In recent years there has been increased interest in attempting to correlate the accumulation of end products of bacterial metabolism with the spoilage of meat. Much of this research has been done at the British Meat Research Institute (reviewed by Dainty, 1982 and Dainty, Shaw and Roberts, 1983). Under aerobic conditions, spoilage is due to the growth of pseudomonads. Whilst these organisms are proteolytic, it is now clear that proteolysis only becomes significant after spoilage (Margitic and Jay, 1970; Dainty et al., 1979).

Recently, it was suggested that the estimation of the combined cadaverine and putrescine content of meat might be a useful indicator of spoilage (Slemm, 1981). However, it has now been shown that significant changes in diamine concentration do not occur until the total aerobic count exceeds $4 \times 10^7/cm^2$ by which time spoilage is obvious (Edwards, Dainty and Hibbard, 1983). These workers have also examined the production of volatiles by pure cultures of five strains of meat spoilage pseudomonads during growth on beef. They found a good correlation between the odour descriptions and the chemical data for

three of the strains. Esters and sulphur containing compounds were most commonly detected (Dainty, Edwards and Hibbard, 1984).

The growth of bacteria on meat is primarily at the expense of the low molecular weight soluble constituents - carbohydrates and amino acids. To date there have been few studies of the utilization of substrates by bacteria growing on meats. Two recent reports suggest that very few compounds are actually utilized (Gill and Newton, 1977; Newton and Gill, 1978) but the situation is almost certainly more complex than indicated by these workers. More detailed experiments may yield results which aid in the interpretation of studies in which the end products of bacterial metabolism are estimated. Further they will assist in obtaining a greater understanding of the factors controlling bacterial growth.

Since lactic acid bacteria dominate the flora of vacuum-packaged normal pH beef, the sour/cheesy/acid flavours detected by taste panels are likely to be caused by the end products of the metabolism of these organisms. Two groups have shown that acetic acid is the major short chain fatty acid present in vacuum-packaged beef after 9 weeks storage at 0-2°C (Sutherland et al., 1976; Dainty et al., 1979). Smaller concentrations of other low molecular weight fatty acids were also detected together with amines including methylamine, dimethylamine and trimethylamine. Unfortunately the interpretation of the results was complicated by the fact that the production of the amines and some of the fatty acids was shown to be non-microbial (Dainty, Shaw and Roberts, 1983). On vacuum-packaged high pH beef, hydrogen sulphide and high concentrations of trimethylamine were detected and these were attributed to the high numbers of Gram-negative bacteria present.

Recently, Edwards, Dainty and Hibbard (1984) measured the concentrations of putrescine and cadaverine in vacuum-packaged normal pH beef during storage at 10°C. Measurable increases in the concentrations of these compounds were evident before maximum bacterial numbers were attained. The source of the diamines is not clear since the concentrations correlated better with the total viable count than with the count of Gram-negative bacteria (which are known to produce these compounds). The authors concluded that estimation of diamine concentration might be of value in the objective assessment of meat quality.

To-date it appears that no chemical method is suitable for use in predicting spoilage. Rather chemical analyses may serve as confirmation of spoilage. However further studies may change this situation.

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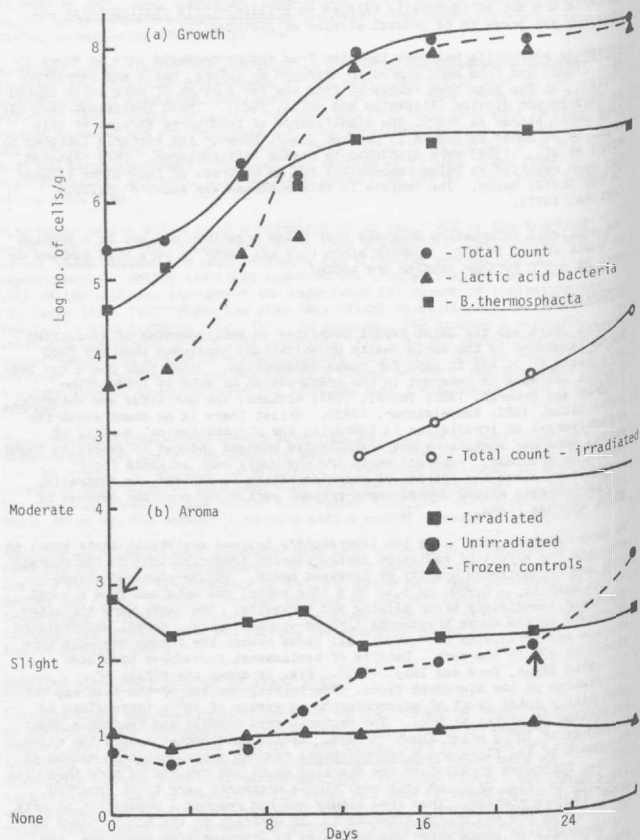


Fig. 2: Growth of the bacteria (a) and concomitant aroma changes (b) in irradiated and control samples of vacuum-packaged sliced corned beef stored at 5°C. Arrows indicate first time at which the aroma was significantly different from that of the frozen control samples.

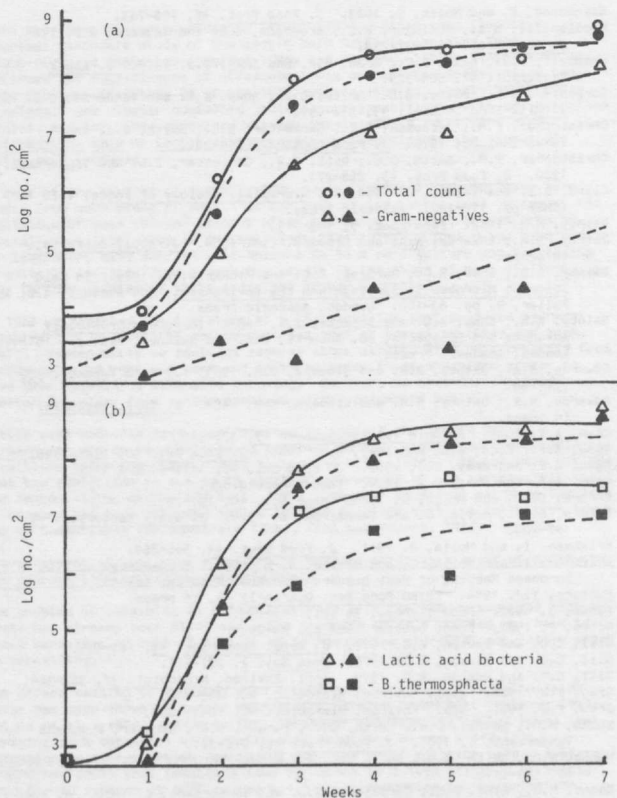


Fig. 1: Growth of bacteria on the lean surface of vacuum-packaged pork (pH 6.2-6.6) at 0°C. Open symbols control samples, closed symbols acetic acid treated samples.