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### Microbial risks associated with salt reduction in certain foods and alternative options for preservation.

**Technical Report** 

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#### CONTENTS

	Section	Page
	SCOPE OF THE REPORT	4
	SUMMARY	5
1	<ul> <li>INTRODUCTION</li> <li>1.1 The importance of salt in nutrition</li> <li>1.2 Salt in food preservation and safety</li> <li>1.3 Analysing the safety of products reformulated with reduced salt</li> </ul>	6
2	<ul> <li>MAINTAINING SAFETY MARGINS IN REDUCED SALT FOODS</li> <li>2.1 Reformulation, safety assessment and predicting problems</li> <li>2.2. Predicted growth in reduced salt products</li> <li>2.3 Potential problem foods</li> </ul>	8
3	SALT REDUCTION IN CURED MEAT PRODUCTS 3.1 Introduction 3.2 Nitrite	11
	3.3 Pathogens of concern	
	3.3.1 Listeria monocytogenes	
	3.3.2 Non-proteolytic <i>Clostridium botulinum</i>	
	3.3.3 Bacillus cereus	
	3.3.4 Yersinia enterocolitica	
	3.3.5 Aeromonas hydrophila	
	3.3.6 Proteolytic Clostridium botulinum	
	3.3.7 Clostridium perfringens	
	3.4 Types of cured meat	
	3.4.1 Bacon and gammon	
	3.4.2 High a <sub>w</sub> ham	
	3.4.3 Partial F <sub>0</sub> products	
	3.5 Non-safety aspects of salt reduction in cured meats	
	3.5.1 Texture	
	3.5.2 Flavour	
	3.5.3 Spoilage	
	3.6 Alternatives to salt in cured meats	
	3.6.1 Salt reduction with shelf-life adjustment	
	3.6.2 Process modification to reduce the need for Salt	
	3.6.3 Replacing salt with alternative CI salts	
	3.6.4 Reducing salt and substituting functionality with corresponding	
	non- Na or lower Na containing additives	
	3.7 Cured meats additives with antimicrobial activity	
	3.7.1 Phosphates	
	3.7.2 Sodium ascorbate, isoascorbate or erythorbate	
	3.7.3 Sugars	
	3.7.4 Smoke	

	<b>3.8</b> 3.8.1 3.8.2 3.8.3 <b>3.9</b>	Alternative antimicrobial additives Organic Acids Alternative solutes Alternative texture and flavour enhancers Summary of salt reduction in cured meats	
4	GENE 4.1 4.2 4.3 4.4	RATION OF AN EQUIVALENT TREATMENT MODEL Predictive modelling ComBase Creation of new models to calculate equivalent growth conditions Limitations of current models and suggested improvements	24
5	DISC	USSION	28
6	CONC	CLUSIONS	29
	RENCES	31	
	<ol> <li>Illus mo</li> <li>The and</li> <li>App</li> <li>App</li> <li>Mod</li> <li>Pre</li> <li>The</li> <li>Typ</li> <li>Effet</li> </ol>	<b>ENDICES</b> strative reductions in salt for non-cereal based foods from the FSA salt del e relationship between sodium and salt contents, aqueous salt (s/w) d brine % proximate growth limiting conditions for food pathogens dels available in Growth Predictor edicted effect of salt reduction on pathogen growth curves e growth/no growth boundary for non-proteolytic <i>C. botulinum</i> bical processes for cured meat products ect of salt, pH and heat treatment on time to growth from 10 <sup>6</sup> spores of n-proteolytic <i>C. botulinum</i>	35

#### **SCOPE OF THE REPORT**

The aim of this report is to consider the microbiological food safety implications surrounding salt reduction in non-cereal foods with particular attention paid to the safety of cured meat products. This includes:

- To evaluate current scientific and other relevant literature and data to inform the FSA's position with regard to microbiological safety:
  - By assessing the implications of salt and nitrate/nitrite reduction in those foods where these preservatives contribute to microbiological safety.
  - Identifying those foods and microorganisms where reductions in salt and nitrate/nitrite will present the greatest risk in terms of consumer safety.
  - Identifying alternative processing/preservation approaches which could be used to compensate for reductions in salt and/or nitrate/nitrite whilst maintaining an equivalent level of safety.
  - To produce models to help evaluate the extent to which reducing shelf-life could compensate for reductions in salt and/or nitrate/nitrite in those foods considered to be at highest risk.
  - > To consider possible minimum achievable salt levels in different products which would still maintain the products main physical characteristics, such as texture.
  - Highlight where there are gaps in current knowledge and where there is a need for further research.
  - ➤ To provide the findings in a paper to the Food Standards Agency and make a presentation to the Advisory Committee on the Microbiological Safety of Food.

#### SUMMARY

The aim of this report was to assess the implications of salt reduction on the microbiological safety of foods and to calculate alternative preservative conditions that could compensate for NaCl reduction whilst maintaining an equivalent safety margin.

The effect of changing salt concentrations on the potential for growth by food pathogens was modelled for hypothetical foods based on the pH, moisture contents and salt concentrations of bacon, ham, chicken roll, smoked salmon, cottage cheese and beef burger. In all cases the rate of growth of foodborne pathogens was much greater in the reduced salt product. The greatest changes were noticed in salt sensitive organisms. For example, non-proteolytic *C. botulinum* would not grow in a product containing 5.5% aqueous salt, but has the potential to grow within 4 weeks at 8°C in a product of pH 5.5 if the aqueous salt concentration is 2.85%. However, it is important to remember that even small increases in growth rates may have implications for public health so all foodborne pathogens must be considered.

When salt levels are reduced it may be necessary to include other preservative factors or decrease the shelf-life of a food to ensure that an adequate safety margin is maintained. It is essential that all producers understand the consequences of salt reduction and are able to reformulate their products safely. As part of this study models were produced that could calculate different combinations of treatments that resulted in the same growth rates. These models illustrate the type of tools that could be developed to aid manufacturers in product reformulation.

There is scope to reduce salt in foods, and reduced salt products, or products where salt has been partially replaced with potassium chloride or sodium lactate are already available. However, as the inhibitory effect of salt varies widely with the organism of concern, environmental conditions, the presence of other preservative factors and processing conditions it is difficult to make blanket recommendations on appropriate salt levels. The safety of each reformulated food should be evaluated on case to case basis and consistent with a HACCP based approach.

#### 1. INTRODUCTION

Throughout this report the word salt has been used to represent sodium chloride unless otherwise stated.

#### **1.1** The importance of salt in nutrition

Although sodium is vital to our health, an excess of sodium in the diet has been linked to hypertension. The major source of dietary sodium is salt. In its report on salt and health, the Scientific Advisory Committee on Nutrition (SACN) concluded that reducing population salt intake would confer significant public health benefits by contributing to a reduction in the burden of cardiovascular disease (SACN, 2003). They supported previous recommendations by the Committee on Medical Aspects of Food and Nutrition Policy (COMA) that the average salt intake for adults should be reduced from 9g to 6g per day (DoH, 1994). This was considered to be an achievable goal.

In 2004, the Food Standard Agency (FSA) adopted a plan to reduce average salt intake in adults from 9.5g to 6g per day by 2010. It is not possible to meet this target by simply reducing the amount of discretionary salt added to food by consumers as a large proportion of salt intake is non-discretionary. The National Food survey 2000 showed that manufactured foods contributed 86% of the total household dietary salt intake (DEFRA, 2001). A previous study by the British Nutrition Foundation found manufactured foods accounted for 60-70% of dietary sodium (BNF, 1994), of which cereal products provided nearly 40% and meat and meat products approximately 21%. It is vital that food manufacturers and retailers assist by reducing the salt in their products if the proposed salt reduction targets are to be met.

In order to explore the effect of salt reductions in different food groups on total dietary sodium intake, the FSA produced a salt model (FSA, 2003). Foods were divided into nutritionally similar groups and data on food consumption and sodium content were combined to produce weighted average sodium contents for each group. Illustrative averages were assigned to each of the food groups based on levels towards the lower end of existing products. These figures permit reductions in each food group to be considered and combined to achieve the desired target dietary sodium intake. The illustrative reductions for non-cereal based foods are shown in Appendix 1 and the conversion of the weight of sodium to salt is shown in Appendix 2.

Although desirable from a nutritional point of view, reducing salt in foods is not a simple issue. Salt affects the flavour, texture, water holding capacity, and in some products microbial stability and safety. It is particularly important that the implications of salt reduction on microbiological safety are understood. Lowering salt has the potential to reduce safety in certain products unless alternative preservative factors are included or shelf-life reduced.

#### **1.2** Salt in food preservation and safety

The use of salt to preserve foods is an empirically developed practice dating back many thousands of years. Although primarily evolved to prevent spoilage, salting also prevented the growth of food poisoning organisms. Salting was very important as it allowed the nutritional benefits of meat, fish and vegetables to be extended from times of plenty to times of scarcity. The development of other preservation techniques, such as refrigeration, means salting is no

longer necessary for survival, and foods using salt as the primary preserving mechanism are uncommon as they are judged to be too salty. However, products using salt for partial preservation are common, both because consumers appreciate their organoleptic properties and because salt has functional properties in foods.

The antimicrobial activity of salt relates largely to its affect on lowering water activity (a<sub>w</sub>). Adding ions to the media surrounding bacterial cells causes water efflux through their semipermeable membranes. As cells must maintain a suitable level of cytoplasmic water for effective functioning of cell components, they try to maintain homeostasis by active accumulation of ions or uptake or synthesis of compatible solutes. The energy expended in these activities reduces growth rates and eventually growth is prevented. The ability of microorganisms to tolerate salt stress in otherwise optimal conditions varies widely between species (see Appendix 3) and will be reduced by sub optimal pH, temperature, redox potential, nutrient availability and the presence of other antimicrobial agents.

If salt is part of a combined preservative system of a food, reducing the salt level will require adjustment of one of the intrinsic or extrinsic properties of the food to ensure the same degree of preservation is maintained. Any reformulation requires analysis of hazards arising from those changes and appropriate action must be taken if new hazards are identified.

#### **1.3** Analysing the safety of products reformulated with reduced salt

Analysing the potential risk associated with reformulated products can be achieved from knowledge of conditions that prevent growth, challenge testing, modelling the new environments using predictive models, or frequently a mixture of all three. Challenge testing is specific to the product but can be very expensive particularly if a range of products, formulations and different bacteria have to be tested. It can also take several weeks for data to be produced. If predictive models are available they offer a much cheaper and quicker way of predicting the consequences of change. However, as models tend to be based on a limited number of key variables and are not specific to one food type, challenge testing may still be required, but at a reduced level. The models currently available in Growth Predictor are listed in Appendix 4 and see Section 4 more details of predictive modelling.

#### 2 MAINTAINING SAFETY MARGINS IN REDUCED SALT FOODS

#### 2.1 Reformulation and safety assessment

Even small changes in recipe or production method can affect the microbiological stability of a food product. It is therefore vital to review product safety whenever changes are proposed. If additional components are included in a product to compensate for any loss of functionality associated with salt reduction, it will be necessary to assess any associated changes in microbiological flora. Normally, reducing salt concentrations will not change the number or species of bacteria present initially but may affect their survival and growth. The magnitude of any increased pathogen growth associated with salt reduction in a food will depend on the contribution made by salt to the safety of that product. There is no microbiological safety implication from reducing salt for many food groups such as:

- Frozen foods
- Foods heated to the equivalent of 121°C for 2.5 min or more
- Acid foods (pH<3.8)
- Low  $a_w$  foods ( $a_w < 0.86$ ) where  $a_w$  is not related to salt
- Foods with added salt treated the same as an unsalted equivalent

Milder preservation systems may prevent growth of specific pathogens. Effective refrigerated storage will usually prevent growth of *Campylobacter*, proteolytic *Clostridium botulinum*, *Clostridium perfringens*, *Escherichia coli*, *Salmonella*, *Shigella*, *Staphylococcus aureus*, *Vibrio* and mesophilic *Bacillus cereus* in foods. A pasteurisation heat treatment of 70°C for 2 min or equivalent will deliver a 6 decimal (6D) reduction in numbers of non-sporeforming food pathogens (Anon, 1992) and therefore eliminate them in products where post processing contamination can be prevented.

The use of such preservation strategies to destroy pathogens or prevent their growth results in foods which are safe regardless of the salt content. However, using such preservation strategies may be unacceptable if the essential quality of the product, whether taste, texture, nutrients or convenience (for example ambient stability or shelf-life), is lost. Many foods do not rely on a single controlling factor but use a combination of preservation methods to ensure safety. These include heat treatment, storage temperature, storage time,  $a_w$ , pH, redox potential, atmosphere, preservatives such as nitrite or organic acids and the presence of competitive organisms.

#### 2.2. Predicted growth in reduced salt products

The groups used to classify foods in the salt reduction model (FSA, 2003) do not necessarily correspond to their mechanisms of preservation. For example, fresh and frozen shepherd's pie are similar nutritionally but will have very different safety issues. It is also important to note that while the FSA salt model is concerned with the quantity of sodium per 100g of food, the effect on micro-organisms will depend on the amount of salt in the aqueous phase of the food. Salt reductions per weight of food therefore have greater impact on  $a_w$  in foods with low moisture contents. For example, if the salt content of a butter which contained 16% water was reduced from 1.85 to 1.02 g per 100g butter, the aqueous salt content would fall from 11.6% to 6.4% (s/w). Variability in food composition and processing regimes means safety will have to be assessed for individual products and not according to group in the salt reduction model.

To model the effect of the proposed changes in salt concentration on pathogen growth it is necessary to know other food parameters such as pH, storage temperature and time.

The effect of reducing salt concentrations on the predicted growth of pathogens in conditions based on the pH values and moisture contents of selected foods is shown in Appendix 5. These growth curves illustrate that reducing salt concentrations in foods will decrease lag time and increase bacterial growth rates unless alternative inhibitory strategies are used. The magnitude of the increase will depend upon the organism, food and proposed salt reduction.

#### 2.3 **Potential problem foods**

The growth curves in Appendix 5 illustrate that lowering salt levels would have a considerable impact on growth, particularly for salt sensitive organisms such as non-proteolytic *C. botulinum*. In some foods the reductions listed in the illustrative salt model would decrease the concentrations of aqueous salt to below the level recommended by the ACMSF for the prevention of growth of non-proteolytic *Clostridium botulinum* in refrigerated processed foods of extended durability (ACMSF 1992, 1995). The current recommendations are maintenance of chill temperatures throughout the chill chain and at least one of the following controlling factors:

- storage at  $<3^{\circ}C$
- storage at  $\leq$ 5°C and a shelf-life of  $\leq$ 10 days
- storage at 5°-10°C and a shelf-life of  $\leq$ 5 days
- a heat treatment of 90°C for 10 min or equivalent lethality (e.g. 70°C for 1675 min, 75°C for 464 min, 80°C for 129 min, 85°C for 36 min ).
- a pH of 5 or less throughout the food.
- a minimum salt concentration of 3.5% in the aqueous phase throughout the food.
- an  $a_w$  of 0.97 or less throughout the food.
- a combination of heat and preservative factors which can be shown consistently to prevent growth and toxin production by *C. botulinum* (recommends a 6D reduction in growth).

Reducing salt levels below 3.5% s/w could be dangerous in MAP/vacuum packed chilled foods where salt was the only factor controlling growth of non-proteolytic *C. botulinum*.

Any increased risk of food poisoning associated with salt reduction will depend not only on altered growth rates but also on the prevalence of the organism of concern. *Listeria monocytogenes* is relatively salt resistant so growth rate may increase only slightly for the proposed salt reduction, but small increases in growth rate could greatly increase the risk of food poisoning in the population. A recent risk assessment on the microbiological food safety risk due to *L. monocytogenes* in processed red meat in Australia suggested decreasing microbial growth rates by relatively modest amounts can have a large effect on the risk of listeriosis from processed meats. Reducing the growth rate of *L. monocytogenes* by 50% decreased the risk of illness in the population by 80 to 90% (T. Ross, personal communication). This suggests even a small increase in growth rate resulting from a decrease in salt without adequate adjustment of other preservative factors could increase considerably the risk to the susceptible population.

As salt reduction can impact on risk of food poisoning through increasing the potential for growth of both salt sensitive pathogens and less sensitive but commonly encountered pathogens, it is important to consider all pathogens in any food where salt is one of the inhibitory factors. Foods that will require analysis include, processed meats, bacon, ham, sausages, shelf-stable canned cured meats, pâtés, meat spreads, dips, chilled pies, chilled soups and sauces, ambient stable cooking and pasta sauces, salted fishery products, cheese and cheese spreads, yellow fats, and composite products, for example pizzas.

When reducing salt levels it is important to take into account variability in a product and leave a safety margin for samples containing less salt than the average value. Consider bacon as an example. The calculated aqueous salt concentration of different types of bacon subjected to identical reductions in salt per 100g are shown in Table 1. The protective effect of salt per g food will increase with decreasing water content. The variability in moisture content between different types of bacon largely reflects the amount of fat in the product. A drive toward fat reduction would indirectly increase the amount of salt per g food required to maintain product safety.

Product	oduct Water		Current	Reduced	Reduced	
	content	salt	salt	salt	salt	
	(g/100g)	(g/100g)	(%s/w)	(g/100g)	(%s/w)	
Back	63.9	3.79	5.93	1.91	3.00	
Middle	59.9	3.79	6.33	1.91	3.19	
Streaky	57.7	3.79	6.57	1.91	3.31	
Gammon	68.6	3.79	5.52	1.91	2.78	

Table 1. The effect of moisture content on the relationship between illustrative salt reduction and reduction in aqueous salt for bacons with different moisture contents (Chan et al. 1995).

Inconsistencies in muscle structure between animals and even along the length of an individual back means aqueous salt contents will also show variability within a single bacon type. Unsmoked back bacon from one UK producer has an average aqueous salt of 5.13% s/w and a standard deviation of 0.87. Assuming salt concentrations amongst reduced salt bacon were normally distributed with the same variability, 50% of samples prepared to a target level of 1.91g/100g of food would contain less than the target and 2.5% of samples would contain less than 0.8 g/100g food. Different food types and production conditions will result in different distributions of salt levels. The variability of a product must be taken into account when deciding suitable safety margins.

It must also be noted that salt is required in some foods to aid development of the correct microbial flora. Salt is important in controlling fermentation processes such as the production of Sauerkraut and is important in cheese where it influences the flavour, body and texture, encourages growth of lactic acid bacteria and discourages organisms that cause proteolysis.

#### **3** SALT REDUCTION IN CURED MEAT PRODUCTS

#### 3.1 Introduction

Designing preservative strategies to maintain microbial safety in reduced salt products is complicated. It is difficult to generalise about products as each relies on different combinations of preservative factors to ensure safety and so must be assessed individually on a case by case basis. It is also difficult to consider microbiological safety in isolation; the degree of safety that is built into a product is often constrained by sensory properties. Alternatives to salt need not only be effective at maintaining product safety but must also be practical, safe, economical and maintain product quality. Salt reduction will affect the microbiological safety of a wide range of foods making it difficult to consider them all in detail. Cured meats are a good example of foods that rely on multiple preservative systems and this group has been assigned the largest illustrative reductions in the FSA salt model and so is a good example for more detailed consideration.

Cured meats have been consumed for hundreds of years and are thought to have developed empirically from meats preserved using salt contaminated with saltpetre (potassium nitrate). The presence of salt and nitrite helped preservation and generated a desirable red colour and a distinctive cured meats flavour that differs from fresh meat. Modern cured meats are manufactured with lower levels of salt and nitrite than would be required for ambient stability. Instead foods are produced for their organoleptic properties *per se* and are mildly cured combined with the need for refrigeration or thermal treatment.

In addition to current moves to reduce salt levels in foods there are also moves to re-examine nitrite levels (Anon, 2003). The antibotulinal properties of nitrite interdepend on the presence of chloride ions as well as factors including pH, heat treatment and the presence of phosphates and ascorbates and it is more inhibitory under anaerobic conditions. It is therefore important to consider any reduction in nitrite levels if the implications of reducing salt levels on product safety are to be understood fully.

#### 3.2 Nitrite

Sodium or potassium nitrites are multifunctional food additives. As well as affecting product colour and producing a cured meat flavour they have an antioxidant effect, protect against warmed over flavour and are antimicrobial (Pearson and Tauber, 1984). In particular they are used to provide protection from growth of *C. botulinum*: the level of nitrite used does not necessarily prevent growth by *C. botulinum* but products containing nitrite are more inhibitory than the equivalent product without.

The levels of nitrite required to impart the typical cured meat flavour, colour and aroma on a product are below those used for safety. Several studies have shown that low levels of nitrite are required to give meat a distinctive cured character, although taste panel scores increase with increasing nitrite concentration (Cassens, 1995). Froehlich *et al.* (1983) suggested that, based on the evidence of an untrained taste panel, consumers would be as likely to buy ham made with 50ppm nitrite as 100 or 150ppm although experienced tasters could appreciate the difference. Touraille and Goutefongea (1985) found as little as 10ppm was enough to develop a cured like aroma. Levels of 20 to 30ppm have been shown as sufficient for colour formation (Pierson and Smoot, 1982). Higher levels of nitrite are included mainly for their antibotulinal qualities.

Under mildly acidic conditions, nitrite is converted to nitrosating agents that react with carbohydrates, lipids and proteins including enzymes such as peroxidase, catalase and cytochromes (Singhal and Kulkarni, 2000). Although there are many possible sites of antimicrobial action, part of nitrites antimicrobial effect is thought to result from the sequestration of iron which prevents activity of iron containing co-factors or enzymes (Tompkin, 1983). Nitrosating agents can also act directly on cells and it is thought that at least part of the inhibitory affect against *C. botulinum* arises from the destruction of iron-sulphur enzymes such as ferredoxin (Reddy *et al.*, 1983)

Kinetic growth models including nitrite and salt as inhibitory factors are available for many organisms. Growth Predictor contains growth models for *L*. monocytogenes and *Salmonella* and PMP has growth models for *A. hydrophila*, *B. cereus*, *E coli* O157:H7, *L. monocytogenes*, *Shigella flexneri* and *Staph. aureus*. Neither have models for proteolytic *C. botulinum*, non-proteolytic *C. botulinum* or *C. perfringens* where nitrite is a controlling factor. Models to predict the probability of toxin production in cured meat by proteolytic *C. botulinum* within a six month period are available for temperatures above 15°C and salt concentrations greater than 3.5%. (Roberts and Gibson, 1986; Roberts and Jarvis, 1983).

#### 3.3 Pathogens of concern

Changing salt concentrations will not affect the occurrence of organisms in foods but may affect their growth, survival or death. In the absence of gross temperature abuse, psychrotrophic organisms are the greatest concern in chilled products, and spore forming bacteria a concern in heat-treated shelf-stable products.

#### 3.3.1 Listeria monocytogenes

*Listeria monocytogenes* is a Gram positive non-sporulating organism that occurs commonly in the environment and has been isolated from a wide range of foods including pork products. It is generally considered to be the most heat resistant of the non-spore forming foodborne pathogens but can be effectively eliminated from food by a heat treatment of 70°C for 2 min or equivalent (Anon, 1992). Its ubiquitous nature means that preventing post process contamination (often in the production environment) is important.

Outbreaks of listeriosis have been associated with several meat products including pâté, rillettes, pork tongue, poultry based deli-meats and turkey frankfurters. A recent risk assessment of listeriosis by the FDA/USDA (Anon, 2003) estimated that processed meats including deli meats, frankfurters, pâtés and fermented products, caused >50% of the cases of listeriosis in USA.

#### 3.3.2 Non-proteolytic Clostridium botulinum

Non-proteolytic and proteolytic *C. botulinum* are physiologically and phylogenetically distinct (Lund and Peck, 2000) and would not be described as the same species other than that fact that they both produce botulinum neurotoxin.

Non- proteolytic *C. botulinum* is a Gram positive spore forming rod that is widespread in soils and can be isolated from many food products. Its ubiquitous nature means it must be assumed that this organism could be present in a food, although any contamination is likely to be low level. Non-proteolytic *C. botulinum* has a long association with cured meat: it was first

isolated in 1897 from ham involved in a food poisoning outbreak in Belgium. Many other outbreaks of botulism in cured meats have been recorded but they are most frequently associated with home prepared rather than commercial products.

The conditions recommended by the ACMSF to control this product are listed in section 2.3. Inhibitory concentrations of salt at suboptimal pH values and temperatures have been identified (Appendix 6).

#### 3.3.3 Bacillus cereus

*Bacillus cereus* is a Gram positive spore forming organism. It is widespread in the environment and is commonly isolated from a wide range of foods. It is responsible for two types of food poisoning, emetic and diarrhoeal. The emetic form results from the ingestion of a toxin preformed in food and the diarrhoeal form from the release of an enterotoxin in the small intestine. Food poisoning is usually associated with foods that have been heated then subsequently held in unsatisfactory conditions allowing outgrowth from surviving spores. Many strains of *B. cereus* are mesophilic having a minimum growth temperature of  $10^{\circ}$ C, but some strains are psychrotrophic and can grow at  $4^{\circ}$ C.

#### 3.3.4 Yersinia enterocolitica

*Yersinia enterocolitica* is a Gram negative non-sporulating organism. It is widespread in the environment although many strains are non-pathogenic. Pathogenic strains are most frequently associated with pigs, and are thought to colonise the tonsil area (Nesbakken, 2000). It can be isolated from pork and a wide range of other foods and refrigeration is relatively ineffective at preventing growth. The organism is relatively sensitive to salt and heat and is not thought to compete well with other organisms.

#### 3.3.5 Aeromonas hydrophila

The main source of *A. hydrophila* is generally accepted to be water but it is a common contaminant of foods and has been isolated from ham (Gobat and Jemmi, 1993). It is known to cause gastrointestinal illness in humans, although its link with foodborne illness is not clear. *A. hydrophila* is less likely to be a problem than other organisms, although it must be remembered that reformulated products may allow the growth of organisms that had not previously caused problems.

#### 3.3.6 Proteolytic *Clostridium botulinum*

Proteolytic *C. botulinum* is a Gram positive spore forming organism that is widespread in soils and can be isolated from many food products. It is a potential problem in raw, cooked or canned cured meats. The organism cannot grow at 10°C or below so is not a major problem in refrigerated foods unless they are subjected to temperature abuse. Greater concern surrounds foods subjected to sub-sterilisation heat-treatments and subsequently stored above 10°C as it has the ability to produce very heat resistant spores. *C. botulinum* spores are the most heat resistant of the food poisoning bacteria and their destruction forms the basis for the heat-treatment of low acid, anaerobic, shelf stable foods (see section 3.4).

Kinetic growth models are available for proteolytic *C. botulinum* but, as they do not include nitrite or heat treatment, their application to growth in canned cured meats is limited. A large amount of data on the effect of curing parameters on the probability of toxin production as a function of salt concentration, nitrite, phosphate, ascorbate and temperature (15, 17.5, 20, 35°C) was collected in a series of experiments conducted in the early 1980s at The Meat Research Institute (Roberts and Gibson, 1986). These data indicate that the probability of

toxin production rises rapidly as salt concentration drops below 4% if no other factors change (Robinson *et al.*, 1982). Probability of toxin production models for parts of cured meat systems, including comparing 2.5 and 3.5% s/w were also created by Leatherhead Food RA. (Jarvis and Robinson, 1983). Unfortunately most of the salt concentrations used in both models were above the illustrative targets of the salt model so can not be used to predict changes in the probability of toxin production

#### 3.3.7 *Clostridium perfringens*

*C. perfringens* is a Gram positive spore forming anaerobe. It is widespread in nature and has been isolated from many foods especially meat and poultry. It does not grow below 15°C and so doesn't grow during correct storage of chilled products. It's spores, although heat-resistant, are destroyed by processes designed to prevent growth by proteolytic *C. botulinum*. Food poisoning is normally associated with growth during inadequate cooling of bulk products. Perfringens Predictor (www.ifr.ac.uk/Safety/GrowthPredictor) is a model that allows growth of this organism to be predicted from measured cooling curves using salt concentration and pH as the additional controlling variables.

#### 3.4 Types of cured meat

To understand the implication of salt reduction on the microbiological safety of a meat system it is important to understand the preservation systems already in place. These will depend on the type of cured meat.

Cured meats can be divided into five product groups. These are:

- i. Bacon and Gammon. A raw product, stored refrigerated, usually cooked before consumption,
- ii. High moisture hams. Products subjected to a pasteurisation heat treatment and stored refrigerated.
- iii. Low a<sub>w</sub> hams. Cured with high salt content and dried to reduce water activity.
- iv. Fractional F<sub>0</sub> products. Ambient stable products subjected to a sub-botulinum cook heat treatment.
- v. Full F<sub>0</sub> products. Products subjected to a botulinum cook to kill all food poisoning organisms.

The F<sub>0</sub>-value is the number of minutes at a 121.1°C (250°F) required to destroy a specific number of specific organism. It can be used to compare the lethality of heat treatments performed at different temperatures. Proteolytic *C. botulinum* is the most heat resistant food pathogen and low acid canning is based reducing the population of viable proteolytic *C. botulinum* spores by a factor of  $10^{12}$ , the "botulinum cook". Reducing the number of viable proteolytic *C. botulinum* spores by a factor of  $10^{12}$  has an F<sub>0</sub> value of 2.5 min. Full F<sub>0</sub> products have been subjected to a heat treatment equivalent to more than 2.5 min at 121°C.

Typical production processes are outlined in Appendix 7. Products subjected to a full  $F_0$  cook will not be considered further in this article as salt is not required for the safety of these products. Low  $a_w$  hams will not be considered as they are not commonly produced in the UK.

#### 3.4.1 Bacon and gammon

Predicting microbial risk associated with a product such as bacon is difficult as it depends on many interacting factors. Modelling the growth of pathogens based only on salt concentration, pH and storage temperature/time of bacon would greatly overestimate actual times or probabilities of growth as they do not take into account the presence of nitrite, modified atmosphere packaging or the natural flora.

Bacon is an unheated product and the addition of salt and nitrite lead to a distinctive spoilage flora which differs from that of fresh meat. The curing salts tend to encourage the growth of Gram positive spoilage organisms such as Streptococci, Micrococci, Lactobacilli and Leuconostoc (Brown, 2000), which are responsible for the less unacceptable spoilage by souring, and discourage the growth of Gram negative spoilage organisms normally associated with meat, such as *Pseudomonas*, which cause the more unpleasant putrification. Reducing salt concentrations may alter spoilage patterns. Applegate *et al.* (1987) showed bacon produced with 1% salt w/w produced vinegary off odours after 2 week storage at 6°C compared to 3 weeks for bacon containing 2 or 3% salt w/w. They stated the rate of acetic acid, tyramine and tryptamine formation was inversely related to the salt content of the bacon.

The presence of a fermentable carbohydrate along with *Lactobacillus planterum* or *Pediococcus acidilactici* (the Wisconsin process) has been shown to be effective in preventing growth of proteolytic *C. botulinum* in bacon at ambient temperature (Tanaka *et al.*, 1980; Tanaka *et al.*, 1985). Such systems are also likely to influence growth by other pathogens.

The complexity of the preservative systems in foods such as bacon makes it difficult to estimate the magnitude of any safety factor associated with them. Hauschild (1982) estimated the probability of toxin production by proteolytic *C. botulinum* in bacon at 27°C as between  $10^{-4}$  and  $4\times10^{-7}$  using data from inoculated pack studies. The same types of analyses are not available for psychrotrophic organisms such as *L. monocytogenes* and non-proteolytic *C. botulinum*. More data on growth in reduced salt products and better exposure assessments are required before the risk of food poisoning associated with reduced salt products can be calculated.

#### 3.4.2 High a<sub>w</sub> ham

Typical high moisture hams (Appendix 7) are heated throughout for a minimum of 70°C for 2 min or equivalent as recommended to give a  $10^6$  fold reduction in *L. monocytogenes* (Anon, 1992). Products cooked and stored in hermetically sealed packages are stable for prolonged periods if stored at <3°C. The potential for growth by non-proteolytic *C. botulinum* exists if storage temperatures are not maintained below 3°C and post-process contamination can occur if the product is sliced repacked or portioned. Post process contaminants are most likely to be organisms from the processing area or equipment. These will mainly be spoilage organisms but *L. monocytogenes* is a potential hazard in sliced meats as it can often be found in the processing environment.

Spores of non-proteolytic *C. botulinum* are of moderate heat resistance with D-values in the range of  $D_{82.2^{\circ}C} = 0.5$ -2.4 min (Lund and Peck, 2000). Cooking may damage spores such that heat treatment may combine with other preservative factors to provide an appropriate degree of protection against growth and toxin production by non-proteolytic *C. botulinum*. Combinations of heat treatment, salt concentration, pH and storage temperature on time to

growth from  $10^6$  spores of non-proteolytic *C. botulinum* in meat slurry have recently been reported (Peck and Stringer, 2005) (see Appendix 8).

Cooked ham is likely to contain antimicrobial factors in addition to those included in available models. These may reduce growth rate and narrow conditions that allow growth but will not prevent growth completely. Pivnick and Bird (1965) showed non-proteolytic *C. botulinum* type E could grow in commercial cooked sliced ham (2.3-3.2 salt g/100g meat, 3.2-4.5% s/w) within 4 weeks at 15°C, but did not find toxin within 8 weeks at 10°C or 5°C. Further reductions in salt levels may allow growth at refrigeration temperatures. It has been shown that 4.6% s/w prevented toxin production by non-proteolytic *C. botulinum* type E within 32 days at 20°C in cured jellied pork tongue; whereas product made with only 3.6 or 2.4% s/w could become toxic after 8 and 4 days respectively (Pivnick and Barnett, 1965)

#### 3.4.3 Partial F<sub>0</sub> products

Shelf-stable canned cured meat products (SSCCMP) are not subjected to a full botulinum cook but are heated to  $F_0$  values between 0.5 and 1.5 to avoid deterioration of product appearance and flavour. Heat treatments of this magnitude will destroy vegetative cells and spores of non-proteolytic *C. botulinum* and will reduce the number of viable spores of proteolytic *C. botulinum* by a factor of between  $10^2$  and  $10^7$ . The safety of such products depends on the heat-treated spores being inhibited by salt, nitrite and pH, combined with good control of the ingoing ingredients.

Hauschild and Simonson (1985) combined theoretical calculations of safety from industrial data and health reports (i.e. data on number of units produced without illness) with data from challenge test studies to produce broad guidelines for levels of brine and heat-treatments for the manufacture of SSCCMP assuming 150ppm nitrite: Their guidelines were:

Luncheon meat

3.0 - 4.0% brine F<sub>0</sub> 1.0 -1.5 4.0 - 4.5% brine F<sub>0</sub> 1.0 4.5 - 5.0% brine F<sub>0</sub> 0.5 -1.0 5.0 - 5.5% brine F<sub>0</sub> 0.5

Ham and shoulder 3.3% brine F<sub>0</sub> 0.3-0.5 3.7% brine F<sub>0</sub> 0.2 -0.3 4.0% brine F<sub>0</sub> 0.1 -0.2

Frankfurter in brine 2.5% brine F<sub>0</sub> 1.5

(For a definition of  $F_0$  see section 3.4)

These recommendations are based on a 7-8D process (reducing population by 7 to 8 decimal reductions or  $10^{-7}$ - $10^{-8}$  of the initial count), less than the traditional 12D process used for low acid foods and therefore also require the levels of spores on raw materials to be strictly controlled. There seems no scientific evidence for why ham and shoulder should require a less stringent heat treatment than luncheon meat; these values are more likely to reflect industrial practices evolved based on the sensory characteristics of the products. Shelf-stable hams are

formulated with less salt and given a lower heat treatment than luncheon meat for organoleptic reasons.

Despite reduced protection factors, the safety record of commercial SSCCMP products is good. There is also little evidence of growth by spoilage Clostridia which are more numerous than *C. botulinum* suggesting the probability of growth of *C. botulinum* is small. However, when product safety is based on a good historical record it is difficult to say how far conditions can be changed before the product becomes a risk to public health. Hauschild and Simonson (1985) suggested use of conditions less stringent than their recommendations should be based on extensive product testing. This would include products reformulated to meet the illustrative salt levels listed in the FSA salt model. It must also be noted that nitrite, salt and heat-treatment have synergistic effects so that if nitrite and/or salt are reduced the  $F_0$  value required to deliver the same level of protection will increase.

Salt not only influences pathogen growth but can also affect survival during heat treatment. The effect is species dependant but thermal inactivation is often greater at higher  $a_w$  so reducing salt is unlikely to increase heat resistance. However, heat-treated cells or spores can be sub-lethally damaged and less able to grow subsequently in the adverse conditions such as the presence of salt. The apparent thermal reduction may increase with increasing salt concentration. For example, non-proteolytic *C. botulinum* is less able to recover from heat-treatment in media containing high concentration of salt than media with low concentrations (Stringer and Peck, 1997).

#### 3.5 Non-safety aspects of salt reduction in cured meats

Product safety cannot be considered in isolation as products have to be organoleptically acceptable as well as safe. Salt is a multifunctional agent: As well being as a preservative, inhibiting growth by both pathogenic and spoilage organisms, it also aids meat protein solubilation, thus water-holding capacity, fat retention and binding of meat, and contributes to flavour. The degree of safety that is built into a product is often constrained by sensory properties.

#### 3.5.1 Texture

Salt is critical to the texture of meat products as it solubilises the structural proteins permitting them to act as binding agents. The chloride ion is thought to be the major contributor to protein solubilisation (Whiting and Richards, 1978). Extracted myofibular proteins help bind meat pieces together, entrap free water and help to trap fat globules. Fat retention and water binding capacity are critical to the texture of the meat product.

Water binding capacity of meat increases with increasing pH. It is possible that water retention and texture could be improved by increasing the pH of low salt meat products but the combination of reduced salt and increased pH is undesirable from a safety point of view. Conversely, reducing pH to retard bacterial growth could increase water exudation from the raw product and water loss during cooking.

#### 3.5.2 Flavour

Salt is a flavour enhancer and in particular the sodium ion is thought to contribute to the salty taste. It has been known for many years that perceived salty flavour of bacon and ham is not closely related to concentration of salt in a product but depends on salt release in the mouth (Ingram, 1949). This in turn depends on the structure of the meat. Determining what is an acceptable salty flavour in a product is difficult as preferences vary between individuals and products and can vary with the perception of the product or its intended use.

Studies of Finnish ham made with 1.1, 1.4, 1.7, 2.0, 2.3 and 2.6% added salt (1.5, 1.8, 2.2, 2.6, 2.9, 3.3% brine) concluded that the lowest salt content which did not taste significantly less salty than normal cooked ham (2.3% salt) was 1.7% salt (the level of the control sample) (Ruusunen *et al.*, 2001). Cooking losses were similar for all the hams except that made with 1.1% salt where it was increased. Microbial safety and stability was not considered as part of these experiments.

Manufacturers have suggested they feel the current lower limits for an acceptable taste is 2.5% salt in bacon, 2.25 to 2.5% salt in traditional ham, and perhaps as low as 1.5% in sandwich ham (BMPA, personal communication).

It may be necessary to co-ordinate gradual reductions in salt. Although consumers may find the reduced salt product acceptable, it does not mean they could not tell the difference between the original and reduced salt product judged side by side.

Individuals taste tolerance to salt is related to exposure. It is said that when salt intake is reduced, the salt taste receptors become more sensitive so foods would not become unacceptable if salt levels were reduced gradually (Antonios and MacGregor, 1997). The FSA aims to reduce sodium intake over a period of five years which should allow for taste preferences to change.

#### 3.5.3 Spoilage

Although spoilage may not make food injurious to health, it makes food unpleasant. Spoilage problems can be detrimental to the reputation of a food type or a manufacturer so it is particularly important that increased spoilage problems are not associated with a new or reformulated product. Lower salt products must be reformulated carefully to ensure they are not rejected by the consumer.

The salt, nitrite and anaerobic atmosphere used to preserve cured meats aids growth of lactic acid bacteria and these become numerically dominant during refrigerated storage. Even products containing high concentrations of lactic acid bacteria usually remain organoleptically acceptable as lactic acid bacteria result in acceptable souring in products such as ham (Brown, 2000). Reducing salt and nitrite could allow more rapid growth by lactic acid bacteria and decrease shelf-life or alter the pattern of spoilage towards purification by allowing growth of more proteolytic organisms that deaminate protein and peptides.

#### 3.6 Alternatives to salt in cured meats

Reducing sodium content in meats can be achieved by

- Reducing salt with shelf-life adjustment if necessary
- Modifying processing conditions to reduce the need for salt.
- Replacing salt with another Cl salt
- Reducing salt and substituting functionality with corresponding non- Na or lower Na containing additives

#### 3.6.1 Salt reduction with shelf-life adjustment

The most obvious way of reducing Na concentrations in food is by adding less salt. Ruusunen *et al.* (2001) suggest that the simplest way to reduce the Na content in the diet is to reduce the highest salt contents in foods down to the levels found in equivalent lower salt products in the market. They suggested this is possible without any taste or technological problems. Reduction of salt alone has the advantage of avoiding the inclusion of extra additives in products but, if salt is part of the preservative system, the shelf-life of the product may need to be reduced. One manufacturer producing reduced salt bacon has already used this technique; when the salt content of bacon was reduced from 3.5% w/w to 2.3% w/w the shelf-life was reduced from 56 days to 28 days.

#### 3.6.2 Process modification to reduce the need for Salt

Tumbling helps protein extraction and is critical to texture development in products such as processed hams. In studies on the effect of tumbling parameters on reduced salt restructured ham, it was found that tumbling for 15.6h at 12rpm with 18.3% replacement of NaCl with KCl was optimal for cooking yield and 12.h at 17rpm optimal for sensory quality (Lin, Mittal and Barbut 1991 reported in Collins, 1997). Extraction also depends on the time and temperature of pre blending and resting and it has been suggested that high pressure and ultrasound treatments could increasing protein extraction. Using these techniques might help compensate for the effect of small reductions in salt on texture. As the use of tumbling and other physical aids to protein extraction are already routine in the manufacture of many processed meats, margins for improved protein extraction may be small.

It has been suggested that air drying individual slices of bacon to reduce their  $a_w$  can be used as an anti-Clostridial alternative to nitrite (Konstance and Panzer, 1985). Reducing water content would increase the antimicrobial effectiveness of any salt present but it may also affect the organoleptic qualities of the product.

The growth rates of many bacteria can be reduced by packing meats under vacuum or in modified atmospheres containing carbon dioxide rather than in air. Manipulation of atmosphere to counterbalance reductions in salt is probably limited as they are already widely used. Cured meats are packed in anaerobic atmospheres as the presence of curing salts makes them more liable to spoilage through oxidative rancidity of the fat than fresh meat.

Improving raw material quality and factory hygiene, and inhibiting growth using techniques such as spraying products with protective cultures may reduce undesirable microbiological changes associate with reduced salt products.

#### 3.6.3 Replacing salt with alternative CI salts

The protein and water binding effects of salt are thought to be largely due to the ionic strength of the Cl ion so substitution using alternative chloride salts such as potassium, calcium, magnesium or lithium can be made without loss of functionality. LiCl has the most salt like flavour but is not used as it is generally considered to be toxic to humans (Price, 1997). The most frequently used substitute is KCl. Several researchers have reported that 35-50% salt can be substituted with KCl in meat products without loss of functionality but levels over 50% can lead to bitter or metallic off-flavours (Collins, 1997). As flavour intensity increases, for example with spicier, more acid or saltier foods, a higher proportion of KCl substitution can be tolerated (Price, 1997).

It should be noted that osmotic effects of the different salts will depend upon their molar concentration and not their weight. Weight for weight KCL is less effective than NaCl.

It may also be worth considering that the potassium ion is an osmoprotectant accumulated by organisms such as *L. monocytogenes* (Pachett *et al.*, 1992). Terrell *et al.* (1984) found replacement of salt (2.5%) in ground pork with KCl or CaCl did not influence growth of the spoilage organisms *Micrococcus* or *Moraxella* and only slightly increased *Lactobacillus* counts in refrigerated conditions.

## 3.6.4 Reducing salt and substituting functionality with corresponding non- Na or lower Na containing additives

Sodium can be added to foods in forms other than NaCl. Substituting sodium salts such as sodium phosphate, lactate or glutamate, with a non-sodium containing alternative would reduce sodium content while maintaining product quality.

Salt is a multifunctional additive, affecting microbial stability, flavour, texture and water holding capacity. This multifunctional nature means it may be difficult to find a direct replacement for salt and multiple additives may be required. This must be balanced against consumer demand for foods containing fewer additives.

#### 3.7 Cured meats additives with antimicrobial activity

#### 3.7.1 Phosphates

The addition of phosphates to cured meat products has become standard practice as they increase the water holding capacity of the meat which helps to minimise shrinkage, reduce cook out losses, improves slicing and increases yield. Polyphosphates solubilise and dissociate actomyosin into actin and myosin. Although the mechanism of protein solubilisatation differs from that of salt, it enhances the effectiveness of salt and so can be used as a partial alternative. There are a range of phosphates available and blends are often used to achieve the correct balance between solubility and functionality. Tests on low salt hams showed no significant difference between flavour, appearance or texture of the hams made using either 1.4 or 1.8% potassium phosphate or sodium phosphate for sodium phosphate

could be used to extend sodium reduction in low salt meat products without loss of functionality.

Phosphates can influence the growth of bacteria in meat products with some being more inhibitory than others (Tompkin, 1984). There is a limit to the use of phosphate as too much can impart a soapy flavour or produce a rubbery texture (Pearson and Tauber, 1984)

#### 3.7.2 Sodium ascorbate, isoascorbate or erythorbate

Ascorbates are primarily used to accelerate cured meat pigment development and stability but they also have an antioxidant effect. Although primarily included for other purposes, ascorbates can also contribute to bacteriological stability. The addition of 200 mg/kg Isoascorbate enhanced the antibotulinal effect of nitrite in shelf stable canned cured meats, possibly through ion sequestration, but at higher levels the effects are reversed, possibly through accelerated nitrite depletion (Tompkin, 1978).

#### 3.7.3 Sugars

Sugars are added to cured meat for colour stabilisation, flavouring and as a substrate for lactic fermentation thus indirect inhibition of pathogen growth. Tanaka *et al.* (1985) described how using a fermentable carbohydrate and lactic acid bacteria (Wisconsin process) was effective as a replacement for nitrite in bacon.

Sugars also moderate the harsh flavour of salt. Reducing sugar content may increase the salty taste of reduced salt products.

#### 3.7.4 Smoke

Smoke is mainly used to develop colour and flavour. Both smoke direct from wood or as liquid smoke contains phenols, alcohols, organic acids, carbonyls, hydrocarbons and formaldehyde and has antimicrobial properties (Lawrie, 1979).

#### 3.8 Alternative antimicrobial additives

Previous pressure to reduce nitrite levels in cured meats facilitated the search for a number of alternative preservatives. Pierson and Smoot (1982) estimated that over 700 compounds have been examined for their suitability for use in curing systems. Although a large number of potential preservatives have been described, relatively few are permitted in food. Also, effects seen *in vitro* are not necessarily transferable to foods. For example, fat soluble compounds such as parabenes are effective in broth systems but ineffective in foods where they partition into the fat. Other antimicrobial additives have been used in cured meat systems.

#### 3.8.1 Organic Acids

Organic acids and their salts are widely used as additives for food preservation. Their antimicrobial effects are due to both pH depression and metabolic inhibition caused by the undissociated molecules. The undissociated form is able to diffuse through the cell membrane. Once inside the cell it dissociates, releasing protons and acidifying the cytoplasm.

Cells have to expend energy to eliminate the excess protons and maintain a neutral internal pH. The effectiveness of organic acids therefore depends on pH and they tend to be more effective in acid than neutral foods.

As well as being antimicrobial, adding salts of organic acids will reduce water activity and the presence of certain concentrations of organic acid, particularly lactic, acetic or citric, enhance or intensify the salty flavour of meats (Price, 1997) and so can be used as a salt substitute to a degree. The level of organic acid included in a product formulation can be limited by the generation of tangy flavours. Acetic acid in particular has a strong flavour whereas lactate has a milder taste.

Several authors have shown that lactate can delay growth by *L. monocytogenes* and nonproteolytic *C. botulinum* and it is effective in cured meats. Lactates have been promoted as a means of extending shelf-life in cured meats and pre-blended curing salt mixtures containing sodium lactate or potassium lactate are available commercially. Mixtures of lactate and diacetate are also commercially available and these two salts are thought to act synergistically to prevent growth from *L. monocytogenes*.

Sorbates have also been extensively studied for use in meat products in combination with nitrites. In 1978 the USDA proposed 40ppm nitrite and 0.26% sorbate as an effective treatment to protect from *C. botulinum* growth in cured bacon. The proposed regulation was later withdrawn as it was suggested sorbate gives bacon a chemical like flavour and prickly mouth sensation (Berry and Blumer, 1981).

Similarly glucono-delta-lactone has been used to control microbial growth in semidry sausage. Its use up to a maximum of 1% decrease pH by 0.5 units but has a biting acid taste.

#### 3.8.2 Alternative solutes

The antimicrobial effect of reduced  $a_w$  caused by salt can in theory be achieved using other solutes. Alternative salts have already been mentioned. Adding sufficient sugar to get the desired osmotic effect would probably make products taste too sweet. Gou *et al.* (1996) suggested that up to 30% of the salt in dry cured pork loin containing 3.5% w/w salt could be substituted with glycine but microbiological testing was not performed on these products. Above 40% substitution, glycine adversely affected flavour, decreasing saltiness and producing an undesirable sweet taste. It also resulted in an inconsistent texture which may have related to glycine having less ability to dissolve proteins.

#### 3.8.3 Alternative texture and flavour enhancers

Hydrolysed vegetable proteins, yeast extract, monosodium glutamate or spices can be used to increase flavour and help compensate for salt reduction. The use of protein or carbohydrate binding agents or gelling agents can be used to improve water binding or improve texture (Collins, 1997). It is not clear what effect the inclusion of these compounds would have on the microbiological safety or spoilage of products.

#### 3.9 Summary of salt reduction in cured meats

Although originally developed as a mechanism of preserving meats, cured meats are now valued for their organoleptic qualities and contribute to variability in the diet. In 2004, 478.2K tonnes of bacon and 127.7K tonnes of sliced and canned ham were consumed (MLC, personal communication).

The safety of cured meats is a complex, multi-component system, depending on the interaction of salt, nitrite, pH, phosphates, ascorbate, and other added antimicrobial agents with heat treatment, storage temperature, atmosphere and shelf-life. Reducing salt in these products will make them more susceptible to growth by micro-organisms, but the magnitude of the effect is difficult to predict as the complicated preservative systems are not fully understood. Adjustments to salt levels can be made but, as there is no rationale for altering the combinations of protective factors, challenge testing will be required to show the intended formulation will remain safe during shelf-life.

#### 4 GENERATION OF AN EQUIVALENT TREATMENT MODEL

The microbiological safety of food products frequently relies not on single factor but combinations of inhibitory conditions. Where many factors contribute to the safety of a food, the desired safety margin may be obtained using different combinations of these factors. Thus increasing other inhibitory factors can be used to compensate for a reduction in salt. As part of this report, predictive models have been generated to determine how factors must be changed to maintain constant growth rates in compensation for salt reduction. The models were created from data in ComBase.

#### 4.1 **Predictive modelling**

Predictive microbiology is the science of using mathematics to describe and predict the response of micro-organisms to their environment. It is based on the hypothesis that the growth, survival or death of an organism in a particular set of conditions is an intrinsic characteristic of that organism and will occur reproducibly in identical conditions. Thus past observations can be used to predict future responses. Trends in primary variables such as growth rate, death rate or probability of growth, can be described using a mathematical function, the primary model. Secondary models can then be created to describe how the parameters of primary models depend on environmental factors such as temperature, pH and water activity. The advantage of using models is that, if observations are made under a sufficient number of combinations of the environmental factors, it is possible to predict how the organism would behave in any combination of conditions within the observation region, not only those used in generating the model.

Models can either be kinetic or probalistic. Kinetic models describe rates of growth or death. Probability models calculate the chances of a given event, such as growth or toxin production, occurring within a given time. The probability of growth falls as conditions reach the limits of growth. Models that define the growth/no growth boundary can be helpful in identifying combinations of conditions that prevent growth

Kinetic growth models have been produced for all food poisoning organisms against what is considered the key independent variables of temperature, pH, and water activity. Many models that include additional variables such as organic acids or nitrite are also available and hundreds of models for specific growth conditions are available in the scientific literature. The largest sets of models are available in Growth Predictor (based on the data formerly used to create Food Micromodel, available at www.ifr.ac.uk/Safety/GrowthPredictor) and Pathogen Modeling Program (PMP available at www.arserrc.gov/mfs/pathogen.htm), developed by national modelling programs in the UK and USA respectively. The original raw data from these two programs have now been combined into a single database, ComBase-PMP (www.combase.cc), and a new set of models are to be produced using the combined data. The models currently available in Growth Predictor are listed in Appendix 4. There are models based on three controlling factors (temperature, pH and a<sub>w</sub>) for all food poisoning organisms and models incorporating a fourth factor for some. None of the current models in Growth Predictors.

#### 4.2 ComBase

Good predictive models require large amounts of quality quantitative data. In recent years there has been a desire to have a standard format for electronic storage of raw microbiological

data in order to facilitate data sharing. Compiling all data, whether collected in laboratory media or food matrices, into a single database, increases the efficiency of data analysis, enhancing model performance and validation and allows flexibility in the application of new modelling techniques. This ideal has led to the creation of ComBase (www.combase.cc), a database on microbial responses to food environments (Baranyi and Tamplin, 2004) which in August 2004 contained over 32,000 growth or survival curves. A universal database allows rapid searching for data from previous studies carried out using the conditions of interest. Holding composite data in a unified format provides flexibility to allow the application of new modelling techniques. For example, the data held in ComBase was used in this report to create a model to allow comparison of the effect of different inhibitory factors on growth of food poisoning organisms as part of this study (see section 4.3).

#### 4.3 Creation of new models to calculate equivalent growth conditions

As part of this study, new models were created to allow the calculation of equivalent growth conditions. One way to compare environmental factors is to quantify how much each must change to induce a unit change in a growth parameter. This can be done using a Z-value concept (Pin *et al.*, 2001). The Z-value is similar to the z-value concept used for comparisons of thermal inactivation treatments except that growth Z-values are not constant, varying with the environmental conditions. The Z-value concept can be applied to any growth parameter such as growth rate or lag time or time to a set increase in cell number. It has been shown that, for cells with similar histories, growth rate, doubling time and time to reach a fixed concentration will be the same (apart from the sign) making it necessary only to calculate the Z-value of one of these parameters to compare the effectiveness of different variables (e.g. salt, temperature and shelf-life).

Data in ComBase were used to calculate generalised Z-values for major pathogens. These were incorporated into a model that allow the preservative effect of salt to be compared to other factors allowing the identification of changes (either in formulation or storage conditions) required to maintain the same level of microbiological safety in a reduced salt food. The models included are the same as in Growth Predictor listed in Appendix 4. An example of the output for *L. monocytogenes* is shown in Figure 1.

The example in Figure 1 shows data for growth of *L. monocytogenes* in a food pH 6.0, 3% s/w stored at 5°C. To use the model first select the worksheet for the pathogen of interest. The original parameters of the food and the desired reduction in salt concentration are entered in the blue boxes. The model calculates the percentage increase in growth rate between the original and reduced salt condition which is shown in cell C7. Each row shows different sets of conditions in which *L. monocytogenes* has the same growth rate. The data in rows 14 to 19 shows conditions where only one additional factor has been changed. For example, if the salt concentration was reduced by 1.6% s/w, the same growth rate could be maintained by reducing storage temperature to 4.6°C, decreasing pH to 5.76 or adding 8598ppm lactic acid, 227ppm acetic acid, 17ppm nitrite or including 8.4% CO<sub>2</sub> in the headspace. Rows 23 to 27 shows equivalent conditions if the temperature was to change in addition to the salt concentrations. This indicates the levels of additional factors required to give equivalent protection in the case of temperature abuse. The model limits for each factor are shown in rows 29 and 30.

In practice, such adjustments to product design are constrained by quality and operational limits. It may also not be possible to adjust all factors, for example lowering storage temperature would compensate for reductions in salt but it may not be possible to guarantee that the reduced temperature is maintained throughout the chill chain. The additional factors may also have already been optimised in some foods, for example foods packed in modified atmospheres containing carbon dioxide.



Figure 1. Screen dump showing the equivalent growth rate model for *Listeria monocytogenes* based on temperature, pH and aw with levels of acetic or lactic acids, nitrite or carbon dioxide as a forth factor. For explanation see text.

#### 4.4 Limitations of current models and suggested improvements

Many of the available predictive models are based on the three key variables of temperature, pH and  $a_w$  (salt). As pH is frequently a function of the food and temperature a function of the distribution system, these cannot be adjusted to compensate for salt reduction so the models can only be used to predict increased growth rates or, conversely, decreased storage times. Models incorporating a fourth factor have greater flexibility, but none of the models include interactions between more than four factors so you cannot consider multiple combinations of factors, such as increased lactate and carbon dioxide, that could compensate for reduced salt. Models using more variables would allow a greater range of alternative conditions that maintained product safety to be predicted.

Although theoretically possible to model as many factors as desired, models tend to be limited by the quantity of quality raw data required. Increasing the number of variables increases the number of combinations of factors that must be measured before a model can be produced. The advent of large databases of pooled data, such as ComBase should make the creation of multiple factor models possible. There are currently data for more than four variables for some organisms but models combining more than four variables cannot be created as there are no data on the interactions between the selected variables. Data from a small number of strategic experiments would allow multifactor models to be created to cover a wide range of growth conditions. Increasing the amount of data on growth of organisms in real foods available in databases would make it much quicker and simpler to validate new models.

One obvious problem when using Growth Predictor or PMP models to predict growth in products such as cured meats is that the models for non-proteolytic *C. botulinum*, proteolytic *C. botulinum* and *C. perfringens* do not include nitrite as a controlling factor. This problem cannot be quickly rectified as there are no data on the effect of nitrite on growth of *C. botulinum* in the ComBase database. Considerable data exists in the literature on the effect of nitrite on the probability of growth of proteolytic *C. botulinum* and it may be possible to include some of this data in modern models. There are fewer reports of the effect of nitrite on growth kinetics. Data on the effect of nitrite on growth of non-proteolytic *C. botulinum* is scarce.

In addition to extending the range of conditions for which predictive models are available it is also important to simplify their use. This would be helped by the development of more user friendly software. One basic principle of empirical modelling is that results should not be extrapolated outside the region from which observations were made. However, it is not easy to determine the interpolation region for multi-factor models. Incorporating areas of interpolation into models would prevent users accidentally extrapolating data. Further developments to combine growth/no growth and kinetic data into a single model would simplify model choice.

#### 5 **DISCUSSION**

Any change in food formulation or processing conditions must be analysed to identify any new hazards and appropriate action must be taken to manage these hazards as required. It is clear that reducing salt levels in foods has the potential to lead to more rapid growth of microorganisms so reducing levels across the board without careful consideration of compensatory measures could increase the risk of food poisoning. Previous cases have highlighted that failure to understand product reformulation can have deadly consequences. In 1989, canned hazelnut conserve used in the manufacture of hazelnut yoghurt caused the UK's largest botulism outbreak with 27 people ill and one dead (O'Mahony et al., 1990). The manufacturers of the conserve had made a low calorie product by replacing sugar with aspartame. Although aspartame replaces the sweetness it does not reduce the a<sub>w</sub> to a similar extent and C. botulinum was able to grow. It is important that strategies are developed and implemented to ensure all food manufacturers are aware of the full implications of salt reduction. While large manufacturers and retailers have the technical resources to analyse products and develop safe approaches, small manufacturers may not. It may therefore be necessary to draw up guidelines or develop better product analysis tools to aid processors in reformulation.

No activity is risk free. Agreement on what is an acceptable level of protection from pathogen growth would help when designing preservative systems. Risk modelling may also be required as it is not necessarily obvious how changes in processes relate to the prevalence of food poisoning in a population. For example, if spoilage also increased, it is possible that more product would be discarded causing a reduction in food poisoning. Analysing risk using hazard domain models would allow sensitivity analysis to be performed. This would determine the importance of small changes in salt concentration on the risk of a particular adverse effect in the population. Risk assessment models would also allow any increased hazard associated with salt reduction to be compared with reduced risk of cardiac disease. New tools for quantitative risk analyses may be required to cope with the complexity and uncertainty of the preservative systems in some foods. It is particularly important to understand the effect of inherent variability on product safety and set safety margins that take this variability into account.

The implication of salt reduction will vary from food to food such that each food will have to be considered individually. For foods where salt is not a controlling factor, this will be relatively rapid. Where foods are partially preserved by salt, predictive models may be helpful. If available, models can greatly aid product design and reduce the number of challenge tests required. However, the number of conditions covered by models is currently limited. Increasing the number of preservative factors included and modelling interaction between factors would increase the number of situations in which modelling would be beneficial. The increased availability of raw data in databases also means it is possible to generate models to target specific questions such as what combinations of preservative factors give the same growth rates. The availability, ease of use, and number of model based tools should be increased. Creating expert systems that could convert measured parameters in foods into advice on product safety may also help decision making by food scientists.

It is important that reformulated, reduced salt foods are not only safe but are stable and have desirable organoleptic properties. Foods that are unsafe, unpleasant or lack convenience will not be accepted by consumers. It is therefore important that nutritionists, microbiologists, food technologists, retailers and consumers work together to come up with an acceptable solution.

#### CONCLUSIONS

6

- There is scope to reduce salt in foods. However, as salt influences bacterial growth, survival and recovery after adverse treatments, reducing salt in certain foods will have consequences for food safety that must be considered. It may be necessary to reformulate these foods or reduce shelf-life to maintain product safety. Any change in formulation, processing or storage conditions means product safety and shelf-life must be re-evaluated and action must be taken if new hazards are identified.
- The inhibitory effect of salt varies with the microorganism of concern, the environmental conditions, the presence of other preservative factors, processing and storage conditions. It is therefore not possible to make overall recommendations on safe levels of salt. Each food must be considered separately.
- Products with reduced salt or where potassium chloride or lactate have been used as partial substitutes for salt are already commercially available.
- The inherent variability of a product should be considered when salt levels are reduced. Safety margins must take this variability into account.
- Salt is not the only source of added sodium in foods. Substitution of additives other than salt with their non-sodium equivalents would help reduce sodium levels. Such reductions would not be apparent where salt concentration is deduced from chloride analysis.
- Additional pressures to alter food formulations, such as the desires to reduce nitrites, lower fat content and reduce additives must be considered along with salt reduction as they interact to effect product safety.
- Validated predictive models are useful tools when growth is affected by multiple factors and they can greatly reduce the need for product testing. Further development would extend their usefulness.
- Improvements to predictive models would include:
  - Inclusion of additional factors such as nitrite in models for non-proteolytic *C. botulinum*, proteolytic *C. botulinum* and *C. perfringens*
  - Creation of models based on more than four factors so interactions between factors can be determined.
  - Combined thermal death and growth models to predict the effects of heat treatment on subsequent growth
  - Identification of critical limits for combinations of factors
  - Improved definition of areas of interpolation in multifactor models
  - More data on growth and survival in foods
  - Development of tools for specific problems such as models to determine equivalent inhibitory factors
- Improved risk models are required to quantify the risks associated with changes to products. While it is possible to predict growth of a pathogen in a specific food it is

much harder to determine the often subtle impact changes will have on the risk of illness in the population.

- Product safety cannot be considered in isolation. The organoleptic and technological properties of reduced salt products must also be acceptable to consumers.
- It is essential that all producers, both large and small, understand the consequences of reducing salt on product safety. Plans for effective dissemination of information to all producers are required. This may require co-operation from ingredient manufacturers, trade organisations, regulatory bodies and research organisations.

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# **Appendix 1.** Illustrative reductions in salt for non-cereal based foods from the FSA salt model (FSA, 2003)

Food group	Current average sodium value (mg/100 g food)	Illustrative Average Sodium value (mg/100 g food)	Current average salt (g/100g food)	Illustrative average salt (g/100g food)	Illustrative average reduction in salt (g/100g food) and %reduction	% of total reduction contributed by food group
Bacon and ham	1491	750	3.79	1.91	1.88 (50%)	13.4
Sausages	962	550	2.45	1.40	1.05 (43%)	4.8
Meat roll/sliced meat products	848	450	2.16	1.14	1.01 (47%)	2.7
Cook-in and pasta sauces	627	250	1.59	0.64	0.96 (60%)	4.0
Other processed meat products	823	450	2.09	1.14	0.95 (45%)	0.6
Crisps and savoury snacks	909	550	2.31	1.40	0.91(40%)	3.2
Fat spreads	726	400	1.85	1.02	0.83 (45%)	4.8
Table sauces retail	914	600	2.32	1.53	0.80 (34%)	2.4
Quiche	550	250	1.40	0.64	0.76 (55%)	0.6
Other processed fish products	896	650	2.28	1.65	0.62 (27%)	0.7
Soup retail	440	200	1.12	0.51	0.61 (55%)	4.8
Canned vegetables	256	50	0.65	0.13	0.52 (81%)	2.1
Burgers and kebabs	503	300	1.28	0.76	0.52 (40%)	2.2
Cheese	700	500	1.78	1.27	0.51 (29%)	3.7
Baked beans	549	350	1.40	0.89	0.51 (36%)	3.9
Meal centre, fish based	430	250	1.09	0.64	0.46 (42%)	0.8
Meat pies	465	300	1.18	0.76	0.42 (35%)	2.9
Ready meals meat based	400	250	1.02	0.64	0.83 (38%)	2.8
Other processed potato products	249	100	0.63	0.25	0.38 (60%)	0.8
Take away, meat based	386	250	0.98	0.64	0.35 (35%)	1.6
Processed vegetable products	393	260	1.00	0.66	0.34 (34%)	1.0
Meal centre, meat-based	461	350	1.17	0.89	0.28 (27%)	0.8
Other processed egg products	410	300	1.04	0.76	0.28 (27%)	0.1
Beverages dry weight	156	50	0.40	0.13	0.27 (68%)	0.4
Ready meals - vegetable based	300	200	0.76	0.51	0.25 (33%)	0.3
Ready meals fish based	300	200	0.76	0.51	0.25 (33%)	0.3
Ready meals, pasta based	326	250	0.83	0.64	0.19 (23%)	0.3
Meal centre, vegetable based	260	200	0.66	0.51	0.15 (23%)	0.0
Take away, fish based	248	200	0.63	0.51	0.12 (19%)	0.2
Canned fish	343	300	0.87	0.76	0.11 (12%)	0.3
Miscellaneous	279	250	0.71	0.64	0.07 (10%)	0.7
Meal centre, pasta based	128	100	0.33	0.25	0.07 (22%)	0.1
Processed pudding products	92	80	0.23	0.20	0.03 (13%)	0.1

#### UK ADULTS SALT INTAKES: MODELLING SALT REDUCTIONS

**Appendix 2.** The relationship between sodium and salt contents, aqueous salt (s/w) and brine %.

Calculation of relationship between sodium and sodium chloride content

Molecular weight Na =22.99 Molecular weight Cl =35.45 Molecular weight NaCl = 58.44

 $NaCl \equiv 2.54 \times Na$  or  $Na \equiv 0.39 \times NaCl$ 

%brine =  $\frac{NaCl}{NaCl + water}$ 

%Salt on water (s/w) =  $\frac{NaCl}{water}$
# Appendix 3. Approximate growth limiting conditions for food pathogens

Data adapted from The Microbiological Safety and Quality of Food. Vol I and II. Ed B.L. Lund. T.C. Baird-Parker and G.W. Gould. Aspen House Inc., Maryland 2000.

Organism	Minimum growth temperature	Inhibitory Water activity (salt adjusted)	Equivalent salt conc (% w/w)	Нq	Oxygen relations
Aeromonas hydrophila	0	0.97	5	4.5	F
Bacillus cereus	4,10 <sup>*</sup>	0.93	11	5.0	F
Campylobacter jejuni	32	0.98	3	4.9	М
<i>Clostridium botulinum</i> , proteolytic	10	0.94	10	4.6	An
<i>Clostridium botulinum</i> , non-proteolytic	3	0.97	5	5.0	An
Clostridium perfringens	12	0.95	7	5.0	An
Escherichia coli (VTEC)	7	0.95	8	4.0	F
Listeria monocytogenes	0	0.92	12	4.3	F
Salmonella spp.	5	0.93	11	3.8	F
Staphylococcus aureus	7	0.86	19	4.0	F
Vibrio parahaemolyticus	10	0.95	9	4.8	F
Yersinia enterocolitica	-2	0.95	7	4.2	F

Key:

F = Facultatively aerobic An = Obligatory anaerobic

M = Microaerophilic

\* = psychrotrophic and mesophilic strains

# Appendix 4. Models available in Growth Predictor

Organism	Temperature	Hq	a.	CO2	Nitrite	Lactic acid	Acetic acid
Aeromonas hydrophila	•	•	•				
Bacillus cereus	•	•	•				
Bacillus cereus + CO_2	•	•	•	•			
Bacillus licheniformis	•	•	•				
Bacillus subtilis	•	•	•				
Bacillus subtilis (a <sub>w</sub> : with glycerol)	•	•	•				
Clostridium botulinum (non.prot)	•	•	•				
Clostridium botulinum (prot)	•	•	•				
Clostridium perfringens	•	•	•				
Escherichia coli O157	•	•	•				
Escherichia coli O157 + CO_2	•	•	•	•			
Listeria monocytogenes	•	•	•				
Listeria monocytogenes + CO_2	•	•	•	•			
Listeria monocytogenes + Nitrite	•	•	•		•		
Listeria monocytogenes + Lactic	•	•	•			•	
Listeria monocytogenes + Acetic	•	•	•				•
Staphylococcus aureus	•	•	•				
Salmonellae	•	•	•				
Salmonellae + CO_2	•	•	•	•			
Salmonellae + Nitrite	•	•	•		•		
Yersinia enterocolitica	•	•	•				
Yersinia enterocolitica + CO_2	•	•	•	•			
Yersinia enterocolitica + Lactic	•	•	0.5%salt			•	
Yersinia enterocolitica + Acetic	•	•	0.5%salt				•
Brochothrix thermosphacta	•	•	•				
Brochothrix thermosphacta death	•	•					
Pseudomonads	•	•	0.5%salt				
Saccharomyces cerevisiae		•	•				

# Appendix 5. Predicted effect of salt reduction on pathogen growth curves.

Basis for values	Hq	Water content (%)	Initial salt content (g/100g food)	Reduced salt content (g/100g food)	Initial Aqueous salt (%)	Reduced aqueous salt (%)	Temperature (°C)
Bacon	5.5	67	3.79	1.91	5.66	2.85	8
Ham	6.0	68	3.79	1.91	5.57	2.81	8
Chicken roll	6.3	71	2.16	1.14	3.04	1.61	8
Smoked salmon	6.2	65	2.28	1.65	3.51	2.54	8
Cottage cheese	4.8	79	1.78	1.27	2.25	1.61	8
Beef burger	5.6	56	1.28	0.76	2.28	1.36	20

Predictions were made using Growth Predictor (<u>www.ifr.ac.uk/Safety/GrowthPredictor</u>) based on pH and moisture contents representative of a variety of foods. The values used were:

It should be noted that the predicted growth rates are likely to be faster than those observed in the food on which the pH, salt and water content were based and the data should not be taken as an indication that a given product would become dangerous within a given time. The models tend to be designed to be fail safe and assume that conditions other than the variables under test are optimal for growth. In foods many variables may be sub-optimal for growth. For example, ham many contain nitrite and lactate, be packaged under a modified atmosphere and contain competitive micro-organisms.



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents of a typical bacon sample



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents as typical ham



The effect of reducing salt and nitrite on *Listeria monocytogenes* growth in a hypothetical food with the same pH and moisture contents as typical ham.



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents as a typical chicken roll.



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents as a typical smoked salmon.



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents as a typical cottage cheese.



The effect of reducing salt on pathogen growth in a hypothetical food with the same pH and moisture contents as a typical beef burger



## Appendix 6. The growth/no growth boundary for non-proteolytic *C. botulinum.*

Combinations of pH (adjusted with HCl) and salt that enabled growth and toxin production from spores of non-proteolytic *C. botulinum* types B, E and F in PYGS medium at 4°, 5°, 8° and 10°C within 3 weeks (a) and 6 weeks (b). Data adapted from A.F. Graham, D.R. Mason, F.J. Maxwell and M.W. Peck. Effect of pH and salt on growth from spores of non-proteolytic *Clostridium botulinum* at chill temperatures. Letters in Applied Microbiology, (1997) 24:95-100.

#### Appendix 7. Typical processes for cured meat products

#### **Carcass preparation**

Stunning Exsanguinaton Scolding Dehairing Scraping Evisceration Slitting Meat grading Chilling to <5°C.

### Typical process for brine cured bacon and gammon

Selection and trimming of raw materials

Injection with curing brine 10-20% brine by weight added 16-22% salt 250-300ppm Na nitrite

Immersion in cover brine 20% salt 0.15% Na nitrate

#### Holding

2 to 5 days at ca 5°C

Drainage and cooling

Smoking (if required) 52°C for 2 to 4 hours

Cooling to <5°C

Slicing and vacuum packaging

Shelf-life

Up to 8 weeks at <5°C (shelf-life tests have 20% of time at 6-8%)

## Typical process for dry cured Bacon

Selection and cutting of raw materials Remove bone

Rub surface with salt/nitrite/nitrate, typically 3.5% Store 2-5°C, can be under vacuum Smoke 52°C for 2 to 4 hours Slice Shelf-life Up to 8 weeks at <5°C

#### Typical process for high-moisture hams

Selection and cutting of raw materials Injection with 15% curing brine Tumbling with brine Salt (2.5-3.5%) Nitrite (300ppm ingoing, 0-50ppm residual after processing) Ascorbate Disodium phosphate Sugars Chilling at 0-5°C Stuffing into casing/moulds Thermal processing Cooked at 60 to 75°C (>2min at 70°C equivalent) Chilling Storage Up to 2 years at <5°C if pasteurised pack is unopened Slicing Packing Shelf-life of sliced product 21-28 days at <5°C in modified atmosphere pack with O<sub>2</sub> scavenger.

### Typical process for ambient stable, fractional F<sub>0</sub> cured meat product

Selection and cutting of raw materials Whole muscle, pieces or comminuted products Adding ingredients Salt (2.5-3.5%) Nitrite (300ppm ingoing, 0-50ppm residual after processing) Ascorbate Disodium phosphate Mixing/chopping/mincing as required Chilling Filling Thermal processing Temp 105-112°C  $F_0 0.5$  to 2 (heat treatments <  $F_0$  of 2.5 required for botulinum cook) Cooling to 20-30°C in 1-2 hours Shelf-life

3-5 years at ambient temperature.

**Appendix 8.** Combined effect of pH, salt concentration (% w/w), heat treatment (20 min) and subsequent storage temperature on time to visible growth from 10<sup>6</sup> spores of non-proteolytic *C. botulinum* types B, E and F in meat slurry (no added lysozyme). Data from Peck and Stringer 2005.

		ĺ	Recovery conditions																											
	-					16°C				12°C							8°C						5°C							
	pН	Salt	0.6	1.8	2.4	3.0	3.6	4.2	4.9	0.6	1.8	2.4	3.0	3.6	4.2	4.9	0.6	1.8	2.4	3.0	3.6	4.2	4.9	0.6	1.8	2.4	3.0	3.6	4.2	4.9
		6.5																												
None	-	6.0																						1						
Z		5.6																												
	-	6.5								1																				
80°C	-	6.0								1																				
00		5.6																												
		6.5																												
85°C	-	6.0																												
00		5.6																												
	-	6.5																												
0°06	-	6.0																												
0		5.6																												
		6.5																												
95°C	-	6.0																												
<u>ග</u>		5.6																												
				0 –	14 da	ys		15 -	- 28 d	ays		29 -	- 42 d	ays		43 -	- 90 d	lays		> 90	) days	6								