1. SUMMARY

The knowledge requirements of the food microbiologist working with thermally processed products are very challenging; particularly in relation to minimally processed foods. In the past, the minimum criteria for sterilized canned foods was very well-defined, however, the diversity of food products, packages and related heat processes are now very difficult to summarize in terms of minimum requirements. The overall heating characteristics of these products can be complex, particularly for mixed particulate products or multicompartment packages. Similarly, the death kinetics of the contaminating micro-organisms can be markedly different dependent on the composition of the food product. These factors combined with the consumer demands for fresher foods has resulted in product, process and package development and innovation progressing at a much faster rate than in the past. While food safety is the most important factor, there are also other issues such as shelf-life and visual quality that have strong market influences and must therefore also be considered by the microbiologist.

2. INTRODUCTION

New food preservation systems are a fascinating area, particularly for the microbiologist, many of the developments resulting from growing consumer demand for fresher foods. There are major differences in the effects on micro-organisms of newer processes compared with the traditional canned product. Canned foods would receive a substantial heat sterilization process compared with a short shelf-life chilled ready-meal which receives a lower heat process (Anon 2004). One of the key parameters to be considered in any thermally processed foods is the target organism, which could be a pathogen or spoilage organism, which should be adequately reduced by the process. While a full canning sterilization process will have been designed to achieve at least 12 log reductions of key spore-forming pathogens (mesophilic Clostridium botulinum), a short shelf-life chilled product would have been designed to achieve 6 log reductions of key vegetative pathogens (Salmonella, Listeria, E. coli) (Gaze et al. 1989). The main reasons for this difference relate to temperature and length of storage before consumption. For example, a sterilized canned product would be expected to remain microbiologically stable for up to 2 years at ambient temperatures whereas a short shelf-life chilled meal would be consumed within 10 days of manufacture.

The well-established canning technology was based on microbial inactivation kinetics related to the ‘line of best fit’ concept used to calculate D and z values. Indeed such calculations are still used today for many food manufacturing, pasteurization and sterilization processes. However, it is frequently noticed by workers in this area that micro-organisms often die in nonlinear formats. Some of the reasons for this will be considered in this document. The presence of shoulders and tails in death kinetic curves are not new and were observed in a number of substrates in published data over 20 years ago (Hersom and Hulland 1980) and has been reviewed by Juneja and Sofos (2001). An additional factor to consider when setting thermal processes is the initial microbiological loading of the raw materials, that is the range of different types and the concentration of micro-organisms present. Any process that is set, must be able to reduce these initial levels to those
acceptable for the product. Shapton and Shapton (1991) considered many aspects of the safe processing of foods, criteria for ingredients and finished products in the context of microbiological safety. Since this publication there have been a number of documents with data relevant to virtually all sectors of the food industry, e.g. Institute of Food Science and Technology, UK IFST (1999), all with the same objective of assisting food manufacturing teams with the design and implementation of their Hazard Analysis Critical Control Point system.

3. RANGE OF FOOD PRODUCTS

The range of food products that are thermally processed is very diverse; it can include low, medium and high viscosity liquids, some with particulates, Ranken (1997). The effect of these different substrates on the heat resistance of a micro-organism can be quite marked, with some proteins, fats and high total solids increasing the heat resistance by a factor of 2 or 3 when compared with the standard heat resistance of a similar micro-organism in a broth (Gaze 1992a). The influence of these components on the heat resistance of micro-organisms must therefore be carefully considered during product development.

Changes in design of food products in recent years have been driven by consumer demand, in particular for convenience and speed of food availability. The time spent preparing a meal has on average been reduced by over 50% over the past 10 years, indeed in the late teenage and early twenties age groups this has been reduced further. This has been attributed to the busy life-styles of modern society. The time spent preparing a meal has on average been reduced by over 50% over the past 10 years, indeed in the late teenage and early twenties age groups this has been reduced further. This has been attributed to the busy life-styles of modern society. The requirement for fast convenient food is coupled with the desire to improve the ‘fresh-like’ qualities of such foods, often by reducing the heat process applied. The major challenge to the thermally processed food manufacturer has therefore been to prepare food, which is safe in terms of the pathogenic organisms, microbiologically stable over its shelf life, but minimally processed to preserve good quality (Dewanto et al. 2002).

One of the very noticeable changes to product formulation and design over this time period has been the combination of different types of solid particle within foods and the trend towards one-pack meals.

The microbiologist and food process engineer have to consider the viscosity of the carrier fluid, e.g. low – thin soup, or medium – gravy or high such as concentrated starch. This fluid has a great effect on heat penetration into the product. The presence and the numbers of micro-organisms in a product before processing will depend on the quality of the raw materials used and how they have been stored. This will be true for both the ingredients of the carrier fluid and any particulates, therefore the effective heat process must be designed to adequately reduce these organisms.

The type of food particle in a product can pose a serious challenge for the design of a safe process. The most simple particle would be of uniform composition, shape and size; however, in reality food ingredients such as vegetables, rarely grow uniformly and indeed many manufactured food particulates have a wide range of shapes and sizes. Heat penetration into particles will vary with size, and so will microbial death kinetics.

Certain constituents of foods directly affect the ability of micro-organisms to survive heat treatments. If the food product contains a high proportion of protein, fats or high total solids it is very likely that the basic heat resistance characteristics of contaminating organisms will be greatly increased (Gaze 1992a) and so the published or accepted rates of thermal inactivation, which are often based on studies performed in broth cultures may be underestimates of that required in such foods. In some cases the increase in process required may be very difficult to achieve as it may begin to denature the remaining constituents of the product making it unacceptable to the consumer. The current trend to reduce salt or acids in foods would also affect the inhibitory properties of the food and therefore the required heat process to maintain stability. This point must be borne in mind by any manufacturer making seemingly minor changes to product recipes and such apparent minor change to a salt or acid level could have a major effect on heat resistance of contaminating organisms rendering foods unstable or unsafe.

There can be no doubt that the heat resistance characteristics of the contaminating micro-organisms will have a major effect on the overall design of the manufactured food. Recent advances in the use of curve fitting equations have given technologists an extra tool to assess inactivation curves that are nonlinear. This is particularly useful if you require a process time to reduce a target population by a certain factor but the death kinetics follow a sigmoid pattern or have an extended tail (Fig. 1). These types of curves have been found in our laboratories with both sporeformers and vegetative organisms heated in a very wide range of substrates and temperatures. Juneja et al. (2001) reported on the modelling of nonlinear survival curves to calculate thermal inactivation of Salmonella in poultry of different fat levels. Their findings are very useful to poultry processors allowing them to vary their thermal treatments in a safe manner in order to achieve adequate reductions in numbers.

When designing and setting any thermal process it will be designed to inactivate a particular type or group of organisms (e.g. sporeformers and vegetative pathogens); this tends to be known as the target organism for the process. For example, if the product was pasteurized and chilled a typical target organism may be Listeria monocytogenes where it has been established that a typical heat process...
at 70°C for 2 min would be required to achieve 6 log reductions (Gaze et al. 1989). However, if the product was acidified and pasteurized the choice of target organism could be *Clostridium butyricum* where it is recognized that if the product is below pH 4.2 the process required may be 95°C for 5 min in order to achieve this reduction (Gaze 1992a).

Considering a commercially sterile, hermetically sealed container or indeed an aseptically processed product at ultra high temperature, the target organism would be proteolytic *Clostridium botulinum* and the traditional minimum of F0 3 to achieve 12 log reductions still applies (Gaze and Brown 1988).

The target organism selected for a given process will vary according to the raw material used in the product and similarly the heat resistance characteristics will vary according to the behaviour of the organism when heated in the food. The diversity and variety of food product composition and its resultant effects on microbial composition and heat resistance has meant a far greater understanding of thermal microbiology is needed to ensure multicomponent food products are manufactured safely.

### 4. THERMAL PROCESSING EQUIPMENT

In recent years, there have been considerable advances in the range of processing equipment available for the manufacture of food products (Richardson 2001).

Foods may be processed in static vessels, or as a continuous flow. They may be processed in container or may be processed first then aseptically filled or hot filled. The effective temperature monitoring of such processes can be very difficult and it is in these situations that biological process verification is essential. The accuracy and reliability of the technique has improved greatly and forms a key part of many innovative process technologies.

The static heating methods tend to be used for in-pack processing, and these will follow the principles of canning retorts where the process would be designed to achieve the required temperature in the slowest heating container. The traditional approach to these investigations includes the use of thermocouple probes, which are inserted into packs and placed at specific places in the processing equipment in order to determine the slowest heating points. These investigations will be designed to establish the slowest heating point of the product (which may be within specific particles) within individual containers and the processing equipment (many vessels have cold spots). It is often necessary to include numerous probes in any one trial and these can effectively be simultaneously registered using a data logger, which could record and store at least twenty different sets of data at the same time. It is then possible to manipulate the data after processing in order to determine overall process lethality and the slowest heating points.

When multicomponent packs are considered, the slowest heating point in the pack will vary considerably depending on the number of components in the pack, the recipe and pack size. The thermal diffusivity or speed at which the heat will penetrate throughout the pack will strongly influence the ability of contaminating organisms to survive. The heating curves represented in Fig. 2 illustrate the heating characteristics of different sized particulates. It is evident that considerably more time is required to achieve process temperature in the larger particulates. This is a very important factor to be considered if microbial contamination is likely to be present in the body of the particle itself.

It is also important to note that historically only the process ‘hold time’ was used for lethality calculations not the come up and cool down time. More recently, some manufacturers do consider the heating up and cooling down times in process calculations, particularly for long come up
times. Any reductions in these margins of safety in order to produce ‘fresher’ products and higher throughput could cause serious microbiological problems.

Continuous flow processing methods are much more difficult to monitor with regard to the minimum process, which will be based on the fastest moving particulate through the heating system. The establishment of heating profiles and the heat processing of certain foods in heat exchangers can be very difficult. Some products can be sticky and may change phases as they are heated to different temperatures (Marshall and Lundblad 2003). The microbiological complexity is often compounded in a product when continuous flow processing is then aseptically filled. Aseptic filling requires not only adequate thermal processing, but also a chemical decontamination step for both the filler head and each individual container.

Many products are now also manufactured with a cold or hot clean fill where the potential for cross-contamination must be very carefully assessed to avoid serious spoilage and food poisoning issues. There are many problems that face the food manufacturer of such products when the question of ‘proof of process’ is asked. In many cases, it is impossible to obtain accurate thermocouple readings in order to assess the temperatures achieved during the filling process.

When undertaking a validation of a continuous flow thermal process the most important data with regard to microbial reduction would be the overall effect of the process on micro-organisms present in particulates as they flowed through the system. Campden and Chorleywood Food Research Association have developed a method for the validation of such processes using alginate to immobilize marker organisms in particles that simulate those in the food product (Gaze and Brown 1990). In this way survival data can be calculated and the overall process lethality assessed biologically.

The basic principles of the alginate technique are to immobilize the marker organism in an homogenate of the food to be tested and an alginate mixture; this is made into the appropriate shape and size to mimic a food particle in that product and allowed to set. A minimum number of these alginate particles (based on a statistical population) are then added to the bulk product and given the scheduled process in production equipment. These particles are often coloured with charcoal to identify them among the bulk food. Once retrieved, the microbiological assessment will determine the number of surviving organisms and consequently the lethality of the process. When using this technique the choice of marker organism is of critical importance: pathogens are never used, instead a nonpathogenic marker having a similar heat resistance to a pathogen is employed.

Another process validation technique for use in the evaluation of specific-pasteurization processes that has been recently developed is the use of time–temperature indicators. Tucker et al. (2002) discussed the potential applications of such a technique utilizing enzyme activity to indicate process lethality. The enzyme used must have a known denaturation rate when heated and measurement of this can give an indication of applied process.

In principle, the time–temperature indicators comprise silicone tubes of length 10 mm and bore 3 mm which are prepared by injecting 15–20 μl of amylase inside the tube and sealing either end with a silicone plug. These capsules can then be included in the container to be processed, heated and cooled as normal, then removed and the solution extracted in order to determine the remaining enzyme activity. A control experiment is always included in order to confirm the enzyme denaturation rate at the test temperature; this can be expressed as a \( D \) value, which allows a comparison to be made with microbial levels. These data can then be used to demonstrate the process lethality achieved in the container.

There are a number of new innovative heating technologies that may apply to the pasteurization and sterilization of food products (Anon 2002; Clark 2002). Some of these use techniques other than direct heat to achieve inactivation of contaminating organisms. It is very important to understand the microbial inactivation kinetics achieved by these techniques, for example high pressure, pulsed electric field, Ohmic heating and microwaves. The assurance of adequate microbial inactivation in these processes will be a constant challenge. The ever-growing trends for minimally processed (pasteurized) foods will require validation techniques that assure the safety of these treatments. The consequences of combining too many changes to a safe design could be severe. It is essential that the implications of changes to the manufacturers process and product recipe are fully understood and that safety margins are not reduced to levels that compromise stability. It is essential that all due care and diligence be given during the manufacture of these products to ensure safety.

5. ADVANCES IN FOOD PACKAGING

In recent years, considerable advances have been made in the type and design of packaging materials used for heat processed foods. Packaging materials are not necessarily rigid; flexible laminates can be formed into pouches and heat-sealed. This in turn brings a number of potential issues in relation to microbial leakage into packs, particularly if seal and pack integrity is not sound. In addition, there is a growing trend to develop multicomponent and multicompartment foods; these also present very demanding microbiological issues.

Multicomponent products may be in one pack and mixed together or in special packaging with several different
compartments within one overall pack. It is important to be aware that many of these developments may be market-driven as the packaging materials lend themselves to far more adventurous labelling and direct on-pack advertising. There are many microbiological criteria to consider for these types of products. For example, for the multicomponent product, each compartment may contain different types of micro-organism and while each may be stable individually, when these are all placed together, the presence or the by-products of one micro-organism may influence the ability of other organisms to grow or survive. In addition, the heat resistance characteristics of different organisms heated in the different food components could be markedly increased.

The thermal process design must therefore be structured to ensure that the most heat resistant pathogen is adequately reduced. In addition it should be noted that the presence of contaminating organisms in one component, for example, an acidic sauce, may not pose a threat. If, however, these organisms leak from one compartment of the package to another and into a neutral pH food component, it is possible that growth could occur, Gaze (1992b).

One of the most popular type of development of pasteurized foods has been ready-meals. One particular process known as ‘Sous vide’ employs the vacuum packing of the product followed by cooking. Provided the minimum safest heat process is achieved, this technique can produce very high quality ‘à la carte’ type dish that can be preserved for several weeks at chilled temperatures. Again one of the key parameters for safety would be to establish where the slowest heating point would be in the pack in order to ensure microbial stability. The question of pack integrity must also feature highly at the design stages, particularly as the product is in-pack pasteurized and any leakage of micro-organisms into the packs will compromise safety. Considerable research has been published on the use of antimicrobial packaging, the main principle of which is to deposit coatings onto packaging materials and assess the inhibitory effects on specific-organisms. del Nobile et al. (2004) published one recent example using Alicyclobacillus acidoterrestris, where active packaging was shown to inhibit the growth of this organism, such findings could have very important impacts on fruit and fruit product packaging.

A useful review of active polymer-based packaging technologies was published by Lopez-Rubio et al. (2004); it includes discussions on oxygen scavengers, carbon dioxide emitters/absorbers, enzymically active packages and antimicrobial packaging. There are a number of antimicrobial agents used in food packaging such as weak organic acids, enzymes, bacteriocins, triclosan, grape fruit seed extract, EDTA, essential oils, fungicides and chitosan some are included as slow release components of packaging (Chu and Chinnan 2004). There are a number of other antimicrobials that are used in edible films and coatings such as benzoic acid, sodium benzoate, potassium sorbate and nitrites (Cagri et al. 2004). There are a number of microbiological questions that must be considered when designing and developing a new packaging format:

i. How is the packaging material itself decontaminated?
ii. Are there opportunities for the packaging to become contaminated with pathogenic micro-organisms?
iii. Can microbial growth be sustained on the packaging material?
iv. Can contamination occur during filling?
v. What is the maximum temperature that can be achieved during filling and processing without causing distortion to the packaging material?
vi. Is the suggested packaging design and subsequent required heat process maintaining product quality?

vi. Are there any risks of breach of container integrity throughout the product life, from manufacture, storage and distribution?

6. OVERALL CONCLUSIONS

The food industry has used heat processing for many decades as a method for producing safe stable foods, and it can be tempting to assume that we know everything we need to, about the use of heat in this way. It is apparent, however, that heat processing is a complex issue: the simple application of a known temperature for a given time does not in itself assure a safe, stable product. The food microbiologist must understand many points including:

i. Knowledge of raw material microbiology.
ii. An understanding of product chemistry and its effects on microbial heat resistance.
iii. An understanding of the product recipe, e.g. preserve and size of particles.
iv. Packaging issues – type of pack, single or multicomponent, potential for leakage, etc.
v. Correct microbial type and system to use for process validation.

Only by a full understanding of these factors can a correct process be given and the safety and stability of the food assured.

7. REFERENCES


