Your product, the fridge and L. monocytogenes – how do bacteriophages help?

by Steven Hagens, chief scientific officer, EBI Food Safety.

t is of great concern that whilst statistical data from around the world on listeriosis is fragmented, there is enough evidence to show a definite increase in the number of cases.

The Community Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Foodborne Outbreaks in the European Union in 2001 and 2006, shows a steady rise in reported listeriosis cases for Denmark, the UK, Germany, Spain, France and Finland, all of which are developed countries with rigorous food safety controls.

In the USA, the Center for Disease Control and Prevention estimates that 2,500 people become seriously ill with listeriosis each year. Of these, around 500 die. We only have to look at the recent devastating impact of a single outbreak in Canada, which has seen 16 deaths at the time of writing, to see the consequences to human life and to business that a listeria outbreak can cause.

Scientific advice

In 2007 the Scientific Panel on Biological Hazards was asked by the European Commission to deliver scientific opinion and provide scientific advice on different levels of Listeria monocytogenes in ready-to-eat foods and the related risk for human illness.

The Report is extensive, looking at all aspects of listeria development and pertaining risk. An area of particular interest concerns the temperature of refrigerators in the home and the potential impact on listeria growth. Omnipresent around us, the pathogenic species, Listeria monocytogenes, is an extremely hardy bacterium, able to survive under low oxygen conditions and at low refrigeration temperatures, even as low as 0°C.

For the food manufacturer, control over the temperature of the product is removed when it enters the retail chain. Once in the chiller cabinet the temperature is the responsibility of the retailer and once in the home, that of the homeowner. Depending on the shelf life of the product, foodstuff

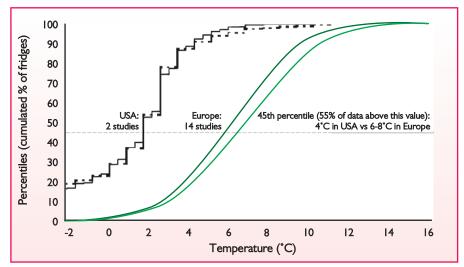


Fig. 1. Temperature distribution in domestic refrigerators in Europe and the USA (2000).

could, in fact, spend the majority of its life in the domestic refrigerator which, depending on the temperature setting, brings about its own food safety risks.

Whilst the Panel on Biological Hazards found that data on the temperature of domestic refrigerators is fragmented they were able to establish a significant variation. One analysis involved 17 surveys of seven EU Member States. Of the 11 surveys (1,924 samples) for which a mean temperature was given, this ranged from 5.0-7.2°C. The weighted mean of the means was 6.5°C.

The Report also found that European temperatures seemed to be higher than those in the USA. Furthermore, the range of temperature within the fridge itself can vary from upper shelf to middle shelf, lower shelf and door panel.

Recommendations

The European Environment Agency recommends that fridge temperature should be set between $1-4^{\circ}$ C. The website states that each degree below these temperatures makes no difference as to how well the food is preserved, but it does increase energy consumption by approximately 5%.

Whilst we do not have the data to confirm why some fridges are running at tempera-

tures as high as 6 and 7°C, some possible reasons can be suggested. Locating the fridge too close to a heat source like an oven, washing machine, heating boiler or sunny window can affect the fridge. Some people may be trying to save energy by allowing it to run at a slightly higher temperature. An older refrigerator may not work to its best ability either through wear and tear or because the design is outdated.

It could also be caused by the consumer's inefficient use of the fridge which may contain too little or too much foodstuff.

A UK study in 2008 by the Health Protection Agency into listeria contamination on sliced meats, found that whilst only five of the 1,127 samples (0.4%) were unacceptable due to levels of Listeria monocytogenes on the day of purchase, after 48 hours refrigeration at 6°C the Listeria monocytogenes levels in 31 of the 82 contaminated samples (38%) had increased to more than 100 Listeria monocytogenes per gram of meat. These are numbers that are considered to pose a potential risk to health in vulnerable people, such as the elderly, pregnant women and those with weak immune systems.

So what does this mean for manufacturers? How can they establish a realistic idea of how their products will be handled and stored by the consumer?

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As stated by the Panel of Biological Hazards, the variability in temperature control means that it is very difficult to define the 'reasonable foreseeable conditions' for the chill chain as stated in the Regulation (EC) No 2073/2005.

However, the risk to manufacturers in terms of product recall, both financial and reputation and, of course, to human health, means there is an absolute need to try and wipe out the bacteria before ready to eat food hits the chill chain and the consumer's table.

To safeguard your production line control methods, such as organic acids, nisin (a bacteriocin) and similar compounds can be used to impair or inhibit the growth of listeria. However, these substances must be labelled as additives and can affect the organoleptic qualities of the end product; and crucially, none can be relied upon to completely exclude the presence of listeria.

Other methods to eradicate the bacterium from the production line include chemical sanitisers, such as chlorine or hydrogen peroxide, which are relatively cheap but do not target the food itself.

Nature has the answer

However, it is not all doom and gloom. Nature actually provided the answer billions of years ago but it is only in the past few years that we have learnt to harness this biological gift for directed and mass-volume use.

Bacteriophages (phages) may sound unfamiliar but they are omnipresent in the world around us. Indeed they are the most abundant micro-organisms in our environment, present in anything that contains bacteria, from water to soil to the human gut. On fresh and processed meat and meat products, more than 108 viable phages per gram can often be identified and high numbers of phages are routinely consumed with our food without any impact on human health or our taste and enjoyment of the product.

In nature, phages act as a balance to keep bacteria under control. Each bacteriophage has a specific 'counter' bacterium in its sights – you might say nemesis – to which it can attach itself in order to reproduce. It cannot attach itself to any other bacteria hence it is suitable also for use with foods containing desired cultures, such as cheese.

In order to survive, therefore, each type of phage is constantly seeking its distinct host and, once identified, attaches itself to the cell wall of the bacterium using specific receptors on its surface.

Having done so, the phage punctures the cell wall and its DNA is drawn into the bacterium, effectively taking over the cell and destroying the bacterium's ability to function or replicate.

This is caused by the phage's own process of reproduction via its DNA, which produces numerous phage proteins. Some of the early proteins sequester the host cellular machinery and force it exclusively to produce new phages.

Once the reproduction is complete, specific proteins weaken the cell wall structure and osmotic pressure causes the cell to disintegrate (lysis) and new phages are released into the environment, destroying the bacterium in the process.

In the absence of target bacteria, the phages break down into common biological particles that are naturally absorbed back into the environment.

Phage solution launched

With backing from research institutes such as the Laboratory of Food Microbiology, Institute of Food Science and Nutrition at the Swiss Federal Institute of Technology Zurich, EBI Food Safety launched Listex P100, a solution containing a high concentration of phages.

By spraying or submerging the food in the solution at the most vulnerable stage of processing, the vast abundance of phages means that susceptible bacteria hosts are found and killed within hours without production having to cease.

Kosher and non-GMO accredited, this product is an innovative processing aid rather than ingredient and does not affect the organoleptic properties of the food in any way or provide any other function – indeed the product's integrity is fully protected, and best of all it is completely natural.

Initial contaminations of listeria during food production tend to be very low, but it is the subsequent outgrowth to high levels which pose a risk to consumers – for example if eaten past the use by date or stored in fridges that are too warm.

Predictive models

As correct domestic fridge temperatures cannot be guaranteed, the Panel on Biological Hazard states that 'temperature variability in the chill chain should be taken into account in both challenge tests and in the use of predictive models to establish shelf life of foods'.

Additionally, eradication or massive reductions of these initial low level contaminations using bacteriophages during processing would ensure that far fewer products leave the production facilities contaminated, if only slightly. The risk of subsequent outgrowth in a particular item would then be much reduced.

By using bacteriophages manufacturers can secure peace of mind as the phages kill the potentially deadly bacteria, rather than just inhibit their growth. In applying a concentrate solution at the critical point in processing, you demonstrate due diligence in prevention of listeria in your product.