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DETERMINATION OF THE RELATIVE EFFECTS OF TEMPERATURE, pH AND WATER ACTIVITY IN FOOD SYSTEMS: A META-ANALYSIS STUDY

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ABSTRACT

The aim of this study is to use ComBase to determine the relative effects of temperature, pH, and water activity in the inactivation rates of *Salmonella enterica* in a range of foods. This is performed to determine whether any of the above factors have a dominant effect on survival. The inactivation rates of *Salmonella* were obtained from original raw data in the ComBase browser and from complete ComBase data for *Salmonella*. A total of 972 data of different types of food systems and data of individual types of food from ComBase were analysed. Over the range of 0–90°C, the z values calculated for the food data is 14°C. At 0–46°C relevant to intermediate moisture foods (IMF), the z values for the food data was 22°C, indicating a moderate effect of temperature. The z value for inactivation at 47–90°C was 11°C, indicating that temperature has an important effect on survival. This study shows that the effect of temperature is clearer at high temperatures than in the low temperature region. It suggests that the inactivation of *Salmonella* in food systems is slightly dominated by temperature and that the pH and a_w levels appear to be less influential.

Key words: Meta-analysis, food system, temperature, pH, a_w

INTRODUCTION

The long-term survival of a bacterial species depends on its members being able: (a) to multiply and compete successfully with other species when conditions are suitable for growth; and (b) to remain viable under adverse conditions until growth becomes possible again. Rapid multiplication of *Salmonella* can take place within the intestines of animal hosts and in some cases in food, but the epidemiological cycle of infection requires survival for long periods in the external environment under conditions not permitting growth. Understanding the behaviour of *Salmonella* under these conditions is therefore important in assessing risks and in devising control strategies. As with all bacteria, *Salmonella* has defined environmental limits within which growth is possible (International Commission on Microbiological Specifications for Foods 1996).

Predictive microbiology is the use of mathematical models to predict the growth, death, and survival of pathogenic and spoilage bacteria in food products from knowledge of the environmental conditions (McClure *et al*., 1993; Ross *et al.*, 2000). Predictive models for the food industry, concerning the most common food-poisoning bacteria in a variety of foods, have been available in the UK since October 1992 under the designation of Food Micro Model. More recently, the predictive models have been made available for free to all users at the ComBase website (http://www.combase.cc/). Nevertheless, the models available for predictive microbiology can manage only relatively few parameters (hurdles); for example, temperature, pH, a_w, aerobic or anaerobic conditions, and some preservatives (nitrite, lactic acid or carbon dioxide). These are important hurdles, and thus, the available models can give a good estimate of the behaviour of food poisoning bacteria in foods. Furthermore, there are numerous additional hurdles, which are

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important for the stability and safety of foods such as presence and concentration of gases in the environment, osmoregulation, and oxidation-reduction potential. Survival may depend on numerous factors including temperature, pH, water activity, preservatives, and gas atmosphere. McQuestin *et al.* (2009) conducted a meta-analysis of the effect of temperature, pH, and water activity on the survival of *Escherichia coli* in fermented foods and showed that temperature had a dominant effect. Similar studies have not been conducted with *Salmonella enterica.* The effect of environmental conditions on the survival of microorganisms is often studied in broth systems that allow greater control of relevant variables. Nevertheless, the rates of inactivation of *E. coli* are greater in broth systems than in fermented meats, suggesting that the reason of these differences should be investigated.

Thus, the objectives of this study are to use ComBase data to determine the relative effects of temperature, pH and water activity on the survival of *Salmonella enterica* in a range of foods and to determine whether any single factor plays a dominant role on survival.

MATERIALS AND METHODS

Analysis of ComBase data for *Salmonella* inactivation under different conditions

The literature values of inactivation rates of *Salmonella* were obtained in two ways. In the first method, the original raw data of viable counts versus time were extracted from records in the ComBase browser (www.combase.cc) and curves were fitted to the raw data using the DMFit curve fitting programme. DMFit allows a range of curve-fitting options including linear, sigmoid, or sigmoid with or without the initial "shoulder" or the "final tail". Inactivation rates were expressed as log decreases per time interval with standard errors. In the second method, inactivation rates expressed as specific rates were obtained from an Excel spreadsheet of the complete ComBase data for *Salmonella* provided by Dr József Baranyi of IFR Norwich, UK.

972 data of different types of food systems and data records for several individual types of food from ComBase were analysed such as bread, produce (almond slice, dried apricot, alfa-alfa seed), sauces, and cheese. The temperature ranges of 0–90°C, 0–46°C and 47–90°C were selected from the browser (www.combase.cc). The temperature range for the growth of *Salmonella* is between 5.2°C and 46.2°C and therefore, the analyses covered the entire range of the available data, as well as the temperatures above and below the maximum temperature for growth. The pH range of 3.5–7.5 and a_w range of 0.60–0.94 were selected, while intermediate moisture food products have an a_w range of 0.60–0.90.

The inactivation rates were converted to "D" values (time needed for a tenfold decrease in viable numbers) and graphs were then drawn of the log-transformed D values to give "z" values, where "z" is the temperature change needed to adjust the D value by tenfold. Analogous plots were used to determine "z_{pH}" and "z_{aw}" as being the pH or water activity changes needed to change the respective inactivation rates by tenfold.

A simple linear regression analysis, R^2 that gives an estimate of the "goodness of fit" of the line was used to test for a correlation between the $\log_{10}$ transformed data and the rate of *Salmonella* inactivation. R^2 represents the percentage variation of the data explained by the fitted line. An R^2 of 1.0 indicates a perfect fit with the line explaining all of the variations.

RESULTS AND DISCUSSION

Analysis of food system data

Meta-analysis studies are able to point out the dominance of explanatory variables; thus allowing improved understanding of the main effects on microbiological inactivation kinetics in food. In the temperature range of 0–46°C, which is relevant to intermediate moisture foods (IMF), the z values for the combined food data was 22°C (Figure 1 and Table 1), which suggests that temperature has a moderate effect on inactivation such that an increase of 22°C would accelerate inactivation by tenfold. McQuestin *et al.* (2009) reported a somewhat similar z value of 19.2°C for fermented sausages in this temperature region.

McQuestin *et al.* (2009) conducted a meta-analysis of the effect of temperature, pH, and water activity on the survival of *Escherichia coli* in fermented foods and showed that temperature (0 to 47°C) had a dominant effect, explaining 61% of the variation in inactivation rates under different conditions. The study of McQuestin *et al.* (2009) was based on calculating inactivation rates from raw data presented in 44 papers.

The ComBase data was less clear-cut than the data of McQuestin *et al.* (2009). In particular, there was a lot of scatter in the Combase data and the R^2 value for the temperature of 0–46° was 0.36, indicating that factors other than temperature accounted a substantial part of the variation. The reason for the greater variation is not known, but could be due to the inclusion of a range of different foods rather than just fermented foods as in McQuestin *et al.* (2009) study.

Table 1 shows that the z value for food data in the high temperature region (47–90°C) was 68°C,

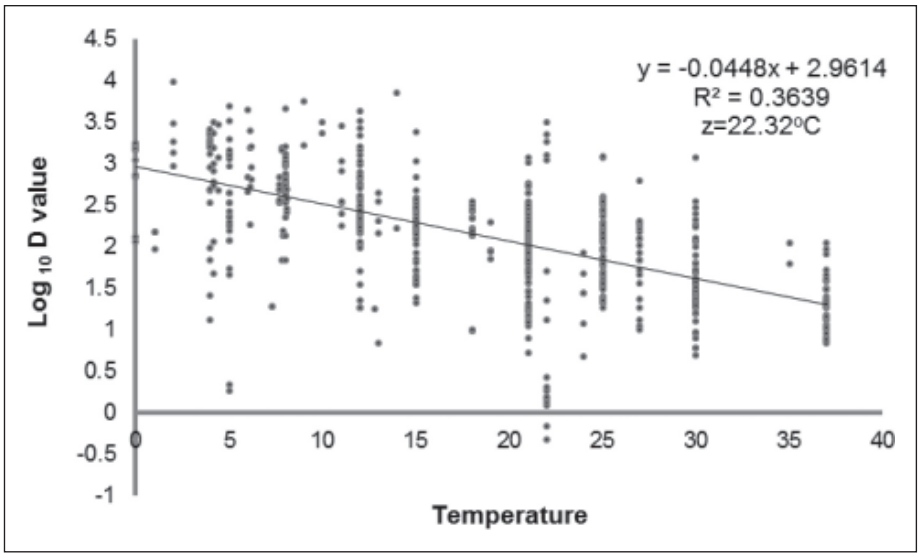

Fig. 1. The effect of temperature (0–46°C) on the survival of *Salmonella* in food system data.

Table 1. Food system data

Parameter	No. of data	Linear regression equation	R^2	z value
Temperature				
0–90°C	972	y = -0.0682x + 3.294	0.78	15[1]
0–46°C	621	y = -0.0448x + 2.961	0.36	22[1]
47–90°C	351	y = -0.0147x -0.309	0.02	68[1]
47–70°C	272	y = -0.0896x+4.0495	0.14	11[1]
pH				
0–46°C	729	y = -0.3689x + 3.2549	0.04	2.7[2]
aw				
0–46°C	375	y = -4.1188x + 4.3592	0.07	0.24[3]

[1]denotes z^t, [2]denotes z^{pH}, [3]denotes z^{aw}

indicating that temperature had no effect on inactivation rates, which was unexpected. However, some of the outlier points appeared to have very long D values even at high temperatures, which skewed the slope of the line (data not shown). The food system data was reanalysed using a temperature in the range of 47–70°C and leaving out records for very low water activity. From unpublished data, it is clear that between the temperatures of 70°C and 90°C, the types of food with low water activity are dried foods (e.g. chocolate), which are known to be protective against thermal inactivation. When these points were omitted, the z value decreased to 11°C, indicating that temperature did have an important effect on survival in this region, although there was much scatter in the data as indicated by an R^2 of only 0.14 (Table 1). McQuestin *et al.* (2009) reported a somewhat similar z value of 9.7°C for fermented sausages at temperatures above 46°C.

Looking at the overall analysis, the effect of temperature was more prominent in high temperatures than in the low temperature region relevant to IMF foods; whereas a_w and pH, surprisingly, had negligible effect in agreement with McQuestin *et al.* (2009). The authors showed that pH (2.8 to 6.14) and water activity (0.75 to 0.986) had only small effects on inactivation rates, accounting for less than 8% of variability. If this proved generally true, it would greatly simplify predictions of inactivation and survival in Hazard Analysis and Critical Control Points (HACCP) and risk assessment exercises.

Analysis of data for individual foods

In the present study, with some individual foods, appreciable effects of pH and a_w were seen, e.g. z_{pH}- = 0.6 for sauces and z_{aw}= 0.1 for cheese.

The food system data includes numerous categories of food such as beef, cheese, mixed products, dairy, egg, poultry, sausage, produce (almond slice, dried apricot, alfa-alfa seed), and sausage or dressing categories. The selected food

Table 2. Selected food data

Parameter	No. of data	Linear regression equation	R^2	z value
Temperature				
0–46°C				
Bread	10	y = -0.0529x + 3.1173	0.89	19[1]
Produce	41	y = -0.0558x + 3.3352	0.73	18[1]
pH				
Sauce	30	y = 1.5764x -4.7338	0.67	0.6[2]
a_w				
Cheese	10	y = 11.501x -6.8049	0.36	0.1[3]

[1]denotes z^t, [2]denotes z^{pH}, [3]denotes z^{aw}

data (Table 2) was analysed from the ComBase data based on "goodness of fit" of the line and z value. It is found that temperature gives a significant effect in the persistence of *Salmonella* in bread and produce (almond slice, dried apricot, alfa-alfa seed), with R^2 values of 0.89 and 0.73, respectively. The z value in the temperature range of 0–46°C was 19°C for bread and 18°C for produce (Table 2).

However, with sauce, the inactivation of *Salmonella* was dominated by pH (R^2 = 0.67, z value = 0.6 pH) and in cheese by a_w (R^2= 0.36, z value = 0.1 a_w).

Although the inactivation rates in individual foods were examined and some such as bread and produce showed a strong effect of temperature, the number of inactivation rate data points available for many other foods within the relevant range was often not very high. As more raw data for individual foods is added to ComBase, it will become possible to obtain more robust conclusions about the dominant effects on survival.

CONCLUSION

The availability of inactivation rate data already calculated in ComBase suggested a very convenient way of examining the effect of environmental conditions on the inactivation rates of *Salmonella*.

In this study, it is suggested that the inactivation of *Salmonella* in all systems was mainly dominated by temperature and that the pH and a_w levels appeared to be less influential. It is therefore important to identify representative robust strains for use in modelling studies. These strains may not necessarily be the same for all stresses. A better understanding is needed of the quantitative relationship between the severity of different non-thermal stresses and rates of inactivation.

Any effect of temperature, pH, and a_w in a combined database that has been analysed is considered masked by other factors specific to particular foods. In addition, it depends on the diversity of *Salmonella* serotypes and strain, food types, and processing conditions employed within the different studies.

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