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# The relation between temperature and growth of bacteria in dairy products

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*The effect of temperature on the growth of psychrotrophs in dairy products has been studied using the 'square root' and Arrhenius models. The square root model has been shown to accurately predict temperature effects on the growth of several psychrotrophs in a number of dairy products. The formula can be applied for individual bacteria regardless of growth medium. This was not true for the Arrhenius equation. In this case, the apparent activation energy for growth (μ) was dependent on source of isolate and growth medium. For isolates from pasteurized cream, μ values were lower during growth in double cream than those obtained during growth in full-fat milk. This was due to shorter generation times at lower temperatures when these isolates were grown in double cream.*

*Temperature-growth parameters were similar for bacteria grown in pasteurized as well as UHT products.*

## Introduction

There has been much recent interest in predictive microbiology whereby mathematical models can be established to forecast microbial growth patterns at various temperatures (Roberts and Jarvis 1983, McMeekin and Olley 1986). These models may then be used to predict shelf-lives of food products under different storage conditions (Olley and Ratkowsky 1973, Daud, McMeekin and Olley 1978, Langeveld and Cuperus 1980, Pooni and Mead 1984, Chandler and McMeekin 1985a,b).

Three equations have been proposed to relate microbial spoilage and storage temperature. Spencer and Baines (1964) proposed the equation

$$K_T = K_o (1 + cT)$$

where $K_T$ = rate of spoilage at temperature T (°C); $K_o$ = rate of spoilage at 0°C; and c = constant. This relation was found to be unreliable for storage temperatures above 6°C (Olley and Ratkowsky 1973).

Olley and Ratkowsky (1973) showed that spoilage rates of foods stored at different temperatures followed the Arrhenius equation (Arrhenius 1889)

$$K = A\, e^{(-\mu/RT)}$$

where K = spoilage rate (specific growth rate); A = constant, μ = apparent activation energy, R = universal gas constant and T = temperature (°K). The relation, however, is non-linear due to μ changing with temperature. A treatment of the Arrhenius equation has been proposed which takes into account this non-linearity (Schoolfield, Sharpe and Magnuson 1981).

The model which has received most attention is the 'square root' plot (Ratkowsky, Olley, McMeekin and Ball 1982, Ratkowsky, Lowry, McMeekin, Stokes and Chandler 1983)

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$$\sqrt{r} = b(T - T_0)$$

where r = growth rate constant; b = slope of the regression line; T = temperature (°K); $T_0$ = notional temperature below which the organism cannot grow. This equation has been applied to milk spoilage data (Chandler and McMeekin 1985a,b) and forms the basis of the technique known as temperature–function integration. This concept assumes that the main factor governing growth of bacteria in food is storage temperature and electronic devices are available which indicate growth during storage relative to growth rates expected at a fixed temperature (Owen and Nesbitt 1984).

This paper describes growth of several organisms commonly associated with dairy products in skimmed milk, whole milk, single cream and double cream at a variety of temperatures. The 'square root' and Arrhenius equations were applied to the growth data and equations derived which related various growth parameters to temperature of growth.

**Table 1. Organisms used in this study.**

| Organism | Source | Isolation temperature (°C) |
|---|---|---|
| Gram-negative | | |
| Acinetobacter spp. 55 | Pasteurized cream | 6 |
| Aeromonas hydrophila 1383 | Pasteurized cream | 6 |
| Alcaligenes faecalis G2/7 | Creamery silo milk | 6 |
| Chromobacterium spp. 12 | Pasteurized cream | 10 |
| Citrobacter freundii 1197 | Pasteurized cream | 6 |
| Enterobacter agglomerans 1498 | Farm bulk tank milk | 4 |
| Escherichia coli NCDO 1989 | Culture collection | n.k. |
| Pseudomonas fluorescens 1181 | Pasteurized cream | 10 |
| Pseudomonas fluorescens 1588 | Farm bulk tank milk | 2 |
| Pseudomonas fragi K1/22 | Creamery silo milk | 6 |
| Pseudomonas putida 1587 | Farm bulk tank milk | 2 |
| Pseudomonas stutzeri B4/4 | Creamery silo milk | 6 |
| Serratia liquefaciens 258 | Pasteurized cream | 10 |
| Serratia marcescens 755 | Pasteurized cream | 10 |
| Serratia marcescens PP | Farm bulk tank milk | 6 |
| Gram-positive | | |
| Bacillus cereus HRM 001 | Farm bulk tank milk | 30 |
| Bacillus cereus HRM 044 | Farm bulk tank milk | 6 |
| Bacillus cereus HRM 045 | Farm bulk tank milk | 6 |
| Bacillus cereus MRM 199 | Creamery silo milk | 6 |
| Bacillus circulans MRM 054 | Creamery silo milk | 6 |
| Bacillus circulans MRM 064 | Creamery silo milk | 6 |
| Bacillus lentus MRM 305 | Creamery silo milk | 6 |
| Bacillus mycoides HRM 068 | Farm bulk tank milk | 6 |
| Bacillus polymyxa MRM 304 | Creamery silo milk | 6 |
| Bacillus pumilus KRM 029 | Creamery silo milk | 30 |
| Bacillus thuringiensis MRM 218 | Creamery silo milk | 6 |
| Staphylococcus aureus C | Farm bulk tank milk | n.k. |
| Staphylococcus warneri H6/E | Pasteurized cream | 6 |
| Streptococcus faecalis NCIB 775 | Culture collection | n.k. |

n.k. = not known.

## Materials and Methods

### *Organisms*

The organisms used in the present study are listed in Table 1. Bacteria isolated at the Institute were identified using API diagnostic kits (API Laboratory Products, Basingstoke).

### *Growth of organisms*

Commercial packs of UHT skimmed milk, full-fat milk, single cream and double cream were obtained from retail outlets in the vicinity of the Institute and the contents were transferred aseptically to sterile 150 ml plastic containers (Sterilin). Cultures of bacteria grown in nutrient broth (Oxoid) at 21°C or 30°C overnight were used to inoculate the containers to a level of approximately 100 cfu $ml^{-1}$. The milk and cream samples were then incubated at 2°, 6°, 10° and 15°C and growth monitored by plate count.

For one experiment samples of pasteurized (80°C for 15 s) and UHT (140°C for 5 s) skimmed milk, full-fat milk and double cream were produced using the pilot plant at the Institute.

### *Bacterial counts*

Samples of product were removed at intervals and plated onto milk agar (Oxoid) plates using a Spiral Plate Maker (Don Whitley Scientific, Shipley, Yorks.). Plates were incubated at 21°C for 25 h or more prior to counting.

### *Analysis of growth data*

The growth curve was fitted to the data by computer using the equation described by Stannard, Williams and Gibbs (1985)

$$y = \frac{A}{\left[1 + e^{\frac{-(\lambda + \kappa x)}{\theta}}\right]^{\theta}}$$

where A, λ, κ and θ are parameters of the curve, the best values of which are obtained by the Nelder–Mead Simplex Minimization

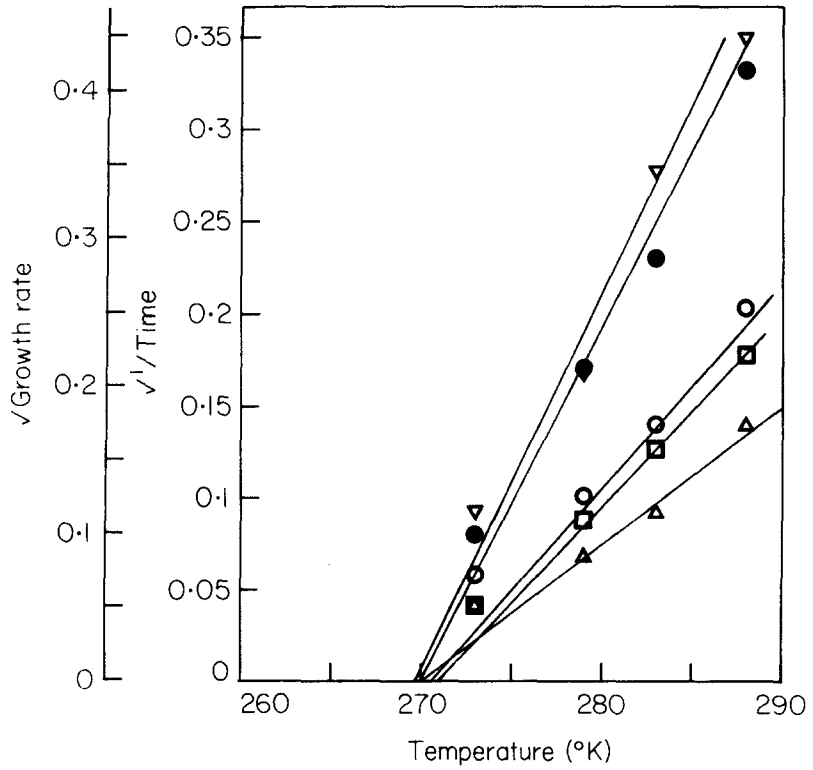


**Fig. 1.** Determination of $T_0$ values for growth of *Serratia marcescens 755* on full-fat milk using plots of length of lag phase (▽), time to achieve maximum growth rate (○), time to stationary phase (△), time for a 4 log cycle increase in growth (□) and growth rate (●) against T (°K).

**Table 2. Reproducibility of temperature/growth parameters.**

| Product | Organism | No. of experiments | $T_o$[b] | Square root plot[a] | | $\mu$[bc] | Arrhenius plot | |
|---|---|---|---|---|---|---|---|---|
| | | | | Standard deviation | % coefficient of variance | | Standard deviation | % coefficient of variance |
| Skimmed milk | Ps. fluorescens 1181 | 3 | 271·7 | 0·73 | 0·27 | 27 770 | 2360 | 8·5 |
| | Ps. fluorescens 1588 | 3 | 262·5 | 0·95 | 0·36 | 16 530 | 1760 | 10·7 |
| | Serr. liquefaciens 258 | 2 | 266·5 | 0 | 0 | 24 250 | 1960 | 8·1 |
| | Serr. marcescens 755 | 2 | 272·8 | 2·15 | 0·79 | 36 340 | 4150 | 11·4 |
| Full-fat milk | Ps. fluorescens 1181 | 3 | 271·2 | 0·45 | 0·17 | 23 920 | 2220 | 9·3 |
| | Ps. fluorescens 1588 | 3 | 263·5 | 1·08 | 0·41 | 16 350 | 970 | 5·9 |
| | Serr. liquefaciens 258 | 2 | 266·4 | 1·65 | 0·62 | 24 930 | 3390 | 13·6 |
| | Serr. marcescens 755 | 2 | 272·5 | 1·55 | 0·57 | 34 390 | 1160 | 4·8 |
| Single cream | Ps. fluorescens 1181 | 3 | 271·5 | 0·50 | 0·18 | 29 313 | 2670 | 9·1 |
| | Ps. fluorescens 1588 | 3 | 263·1 | 1·65 | 0·63 | 16 640 | 2650 | 15·9 |
| | Serr. liquefaciens 258 | 2 | 267·1 | 0·30 | 0·11 | 23 980 | 185 | 0·7 |
| Double cream | Ps. fluorescens 1181 | 3 | 269·6 | 0·90 | 0·33 | 18 680 | 260 | 1·4 |
| | Ps. fluorescens 1588 | 3 | 263·3 | 1·17 | 0·44 | 16 770 | 1920 | 11·5 |
| | Serr. liquefaciens 258 | 2 | 267·7 | 0·90 | 0·34 | 23 350 | 1180 | 5·1 |
| | Serr. liquefaciens 755 | 2 | 271·3 | 1·10 | 0·41 | 27 790 | 3070 | 11·0 |

[a] Using the reciprocal of the time taken for a 4 log cycle increase in growth.
[b] Average of data sets.
[c] $\mu$ in cals $mol^{-1}$.

Procedure (Nelder and Mead 1965, Nicol, Smith and Raggett 1985). This technique allowed excellent agreement between observed and calculated growth data at all temperatures ($r \geq 0{\cdot}991$). The Arrhenius and 'Square root' equations were applied to the results by linear regression techniques.

For the Arrhenius equation, $\log_e K$ (where K = specific growth rate) was plotted against 1/T (°K). The temperature characteristic, $\mu$, was then obtained using the equation:

$$\text{slope} = -\mu/R \text{ (where R = gas constant)}$$

For the 'square root' equation, the square root of the reciprocal of the time taken to reach a specified point on the growth curve was plotted against temperature (°K). $T_0$ was equal to the intercept of the extrapolated line on the temperature axis (i.e. when r = 0).

## Results and Discussion

*Reproducibility of temperature/growth parameters*

Growth data were obtained on a number of occasions for four organisms and the means are shown in Table 2. The reproducibility of the $T_0$ value was very good with the coefficient of variance never exceeding 1% and generally lower than 0·5%. The reproducibility of the $\mu$ value was not as good with the coefficient of variance about 10 times higher than that for $T_0$ determinations. Similar results were achieved by Stannard et al. (1985) for growth of psychrotrophs in defined media.

*Determination of $T_0$ using various growth parameters*

Values of $T_0$ were similar when obtained from plots of various growth parameters (Fig. 1). In these experiments, $T_0$ values were determined from plots of the square root of the reciprocal of length of lag phase, time to reach maximum growth rate, time to stationary phase, and time for a 4 log cycle increase in growth, as well as the square root of the growth rate.

There was remarkably good agreement between $T_0$ values assessed by the different plots. The coefficients of variation (standard deviation/mean) ranged from 0·073% to 2·43%, the average being 0·66%. Chandler and McKeekin (1985b) also found that the effect of temperature on lag phase of bacteria could be predicted by the square root model which described the actively growing culture.

*Temperature–growth relations for bacteria growing in dairy products*

$T_0$ and $\mu$ values for a number of bacteria growing in skimmed milk, full-fat milk, single cream and double cream are shown in Table 3. There was a strong correlation between $T_0$ and $\mu$ values for the organisms grown on skimmed milk ($r = 0{\cdot}93$; $n = 25$), full-fat milk ($r = 0{\cdot}90$; $n = 29$), single cream ($r = 0{\cdot}93$; $n = 25$) and double cream ($r = 0{\cdot}90$; $n = 25$). A similar relation between $T_0$ and $\mu$ ($r = 0{\cdot}88$) existed for the bacteria studied by Stannard et al. (1985) which were grown on semi-defined medium.

For the Gram-negative organisms tested, the $T_0$ values were generally in the range 258–270°K, which were the values of $T_0$ expected for psychrotrophic bacteria (Ratkowsky et al. 1982). The *Escherichia coli* strain studied together with one strain of *Serratia marcescens* had $T_0$ values within the range proposed for mesophilic bacteria (i.e. 270–280°K). The Gram-positive bacteria tested all had $T_0$ values in the mesophilic range with the exception of the *Bacillus circulans* strains and *Bacillus lentus*. This supports the findings of Langeveld and Cuperus (1980) who showed that *B. circulans* was capable of growth at lower temperatures than *B. cereus*.

As $\mu$ was directly correlated with $T_0$, then the value of $\mu$ may also be used to determine whether the organism under study was psychrotrophic (Ingraham 1958, 1961). However, Hanus and Morita (1968) found no such relation between $\mu$

**Table 3. Temperature–growth parameters for a number of bacteria grown in dairy products.**

| Organism | Growth medium | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | Skimmed milk | | Full-fat milk | | Single cream | | Double cream | |
| | μ[a] | $T_o$[b] | μ | $T_o$ | μ | $T_o$ | μ | $T_o$ |
| *Acinetobacter* spp. 55 | 17·28 | 266·6 | 18·03 | 267·0 | 18·53 | 266·9 | 17·04 | 264·3 |
| *Aeromonas hydrophila* 1383 | 28·97 | 269·4 | 27·17 | 269·2 | 27·38 | 270·4 | 27·71 | 268·9 |
| *Alcaligenes faecalis* G2/7 | 15·79 | 259·0 | 14·93 | 258·7 | 14·07 | 257·8 | 17·20 | 265·5 |
| *Chromobacterium* spp. 12 | 14·20 | 260·2 | 15·75 | 263·7 | 12·90 | 261·4 | 12·24 | 261·5 |
| *Citrobacter freundii* 1197 | 24·07 | 268·4 | 30·14 | 269·8 | 24·60 | 269·2 | 18·78 | 265·1 |
| *Enterobacter agglomerans* 1498 | 22·42 | 266·6 | 24·35 | 267·7 | 20·40 | 264·0 | 22·67 | 266·7 |
| *Escherichia coli* NCDO 1989 | 48·02 | 279·5 | 47·69 | 279·5 | 48·35 | 279·7 | 35·77 | 278·8 |
| *Pseudomonas fluorescens* 1181 | 24·93 | 269·8 | 23·97 | 269·6 | 26·93 | 270·1 | 18·85 | 269·3 |
| *Pseudomonas fluorescens* 1588 | 16·87 | 261·8 | 16·64 | 262·7 | 16·86 | 262·2 | 17·92 | 261·3 |
| *Pseudomonas fragi* K1/22 | 16·71 | 260·9 | 14·15 | 261·7 | 16·81 | 262·0 | 11·98 | 262·1 |
| *Pseudomonas putida* 1587 | 14·87 | 264·5 | 14·24 | 264·3 | 14·78 | 263·3 | 13·17 | 261·4 |
| *Pseudomonas stutzeri* B4/4 | 12·06 | 260·5 | 10·89 | 260·2 | 16·83 | 264·0 | 14·20 | 264·6 |
| *Serratia liquefaciens* 258 | 24·25 | 266·4 | 24·93 | 266·3 | 23·98 | 266·9 | 23·35 | 267·6 |
| *Serratia marcescens* 755 | 32·19 | 270·5 | 32·73 | 270·3 | 23·08 | 270·0 | 24·72 | 269·8 |
| *Serratia marcescens* PP | 39·09 | 274·2 | 39·97 | 274·0 | 39·05 | 273·2 | 41·53 | 273·8 |
| *Bacillus cereus* HRM 001 | 28·15 | 273·9 | 29·81 | 272·9 | 32·12 | 274·7 | 24·51 | 272·8 |
| *Bacillus cereus* HRM 044 | n.d. | n.d. | 43·74 | 273·9 | n.d.[c] | n.d. | n.d. | n.d. |
| *Bacillus cereus* HRM 045 | n.d. | n.d. | 41·64 | 272·1 | n.d. | n.d. | n.d. | n.d. |
| *Bacillus cereus* MRM 199 | 39·68 | 277·4 | 45·67 | 278·7 | 37·30 | 277·5 | 49·39 | 279·2 |
| *Bacillus circulans* MRM 054 | 20·61 | 266·1 | 29·32 | 265·8 | 33·70 | 269·6 | 25·65 | 268·5 |
| *Bacillus circulans* MRM 064 | 18·59 | 263·6 | 14·14 | 262·7 | 14·29 | 259·1 | 18·52 | 260·2 |
| *Bacillus lentus* MRM 305 | 27·82 | 265·8 | 34·49 | 267·0 | 29·12 | 269·2 | 27·97 | 271·4 |
| *Bacillus mycoides* HRM 068 | n.d. | n.d. | 43·03 | 274·8 | n.d. | n.d. | n.d. | n.d. |
| *Bacillus polymyxa* MRM 304 | 34·98 | 272·6 | 28·01 | 270·0 | 32·55 | 270·8 | 32·00 | 270·6 |
| *Bacillus pumilus* KRM 029 | 37·37 | 274·5 | 49·32 | 275·0 | 26·85 | 272·3 | 37·91 | 276·0 |
| *Bacillus thuringiensis* MRM 218 | n.d. | n.d. | 34·98 | 275·2 | n.d. | n.d. | n.d. | n.d. |
| *Staphylococcus aureus* C | 40·19 | 274·7 | 38·97 | 274·4 | 39·92 | 274·1 | 39·31 | 274·2 |
| *Staphylococcus warneri* H6/E | 29·67 | 272·7 | 29·17 | 272·2 | 27·82 | 270·7 | 25·34 | 271·6 |
| *Streptococcus faecalis* NCIB 775 | 28·79 | 273·8 | 28·83 | 273·6 | 37·87 | 273·2 | 36·59 | 273·6 |

[a] μ = apparent activation energy (Kcals/mol).
[b] $T_o$ = conceptual minimum temperature (°K) for growth.
[c] n.d. = not determined.

values and temperature characteristics of growth.

There was no significant change in $T_0$ or $\mu$ values for the bacteria on different products (Table 4). Ratkowsky et al. (1982) stated that the $T_0$ value was an intrinsic property of the organism and was not affected by growth medium.

$T_0$ and $\mu$ values were similar to those obtained by other workers for organisms of the same genera (Chandler and McKeekin 1985a,b; Stannard et al. 1985). $T_0$ values for 3 strains of *Ps. fluorescens* and 1 strain of *Ps fragi* grown on UHT milk were calculated from the data of Shelley, Deeth and MacRae (1986). $T_0$ values of 270·9, 268·4 and 265·2 were obtained for *Ps. fluorescens* strains AS7c1, AS11a1 and AS31a1, respectively, whilst the value for *Ps. fragi* AS24b1 was 256·2. These values were similar to those obtained in this

**Table 4. Differences between $T_0$ and $\mu$ values for bacteria grown on different dairy products.**

| Parameter | Average value for bacteria[a] grown on | | | |
|---|---|---|---|---|
| | Skimmed milk | Full-fat milk | Single cream | Double cream |
| $T_0$ | 268·5 A[b] | 268·7 A | 268·5 A | 268·8 A |
| $\mu$ | 26·30 B | 27·33 A, B | 26·24 A | 25·37 A, B |

[a] Average for 25 bacteria grown on product.
[b] Significance determined by paired t-test. A = not significant; B = $P < 0·1$.

**Table 5. Effect of source of isolate on growth parameters for bacteria grown in full-fat milk and double cream.**

| Source of isolate | No. of isolates | Average value of $\mu$ for growth in | | Average value of $T_0$ for growth in | |
|---|---|---|---|---|---|
| | | Milk | Cream | Milk | Cream |
| Raw milk | 15 | 26·99 | 26·26 | 267·7 | 268·6 |
| | | (N.S.)[a] | | (N.S.) | |
| Pasteurized double cream | 8 | 25·24 | 21·00 | 268·5 | 267·3 |
| | | (*)[b] | | (N.S.) | |

[a] N.S. = not significant ($P > 0·05$).
[b] * = $P < 0·05$.

**Table 6. Effect of source of isolate on rate of growth in milk and cream at various temperatures.**

| Source of isolate | No. of isolates | Growth temperature (°C) | Average generation time (h) in | |
|---|---|---|---|---|
| | | | Full-fat milk | Double cream |
| Raw milk | 15 | 15 | 2·9 | 2·4 |
| | | 10 | 4·9 | 5·0 |
| | | 6 | 9·9 | 9·8 |
| | | 2 | 14·8 | 16·0 |
| Pasteurized cream | 8 | 15 | 1·7 | 1·6 |
| | | 10 | 3·5 | 3·1 |
| | | 6 | 6·3 | 5·2 |
| | | 2 | 16·5 | 9·3 |

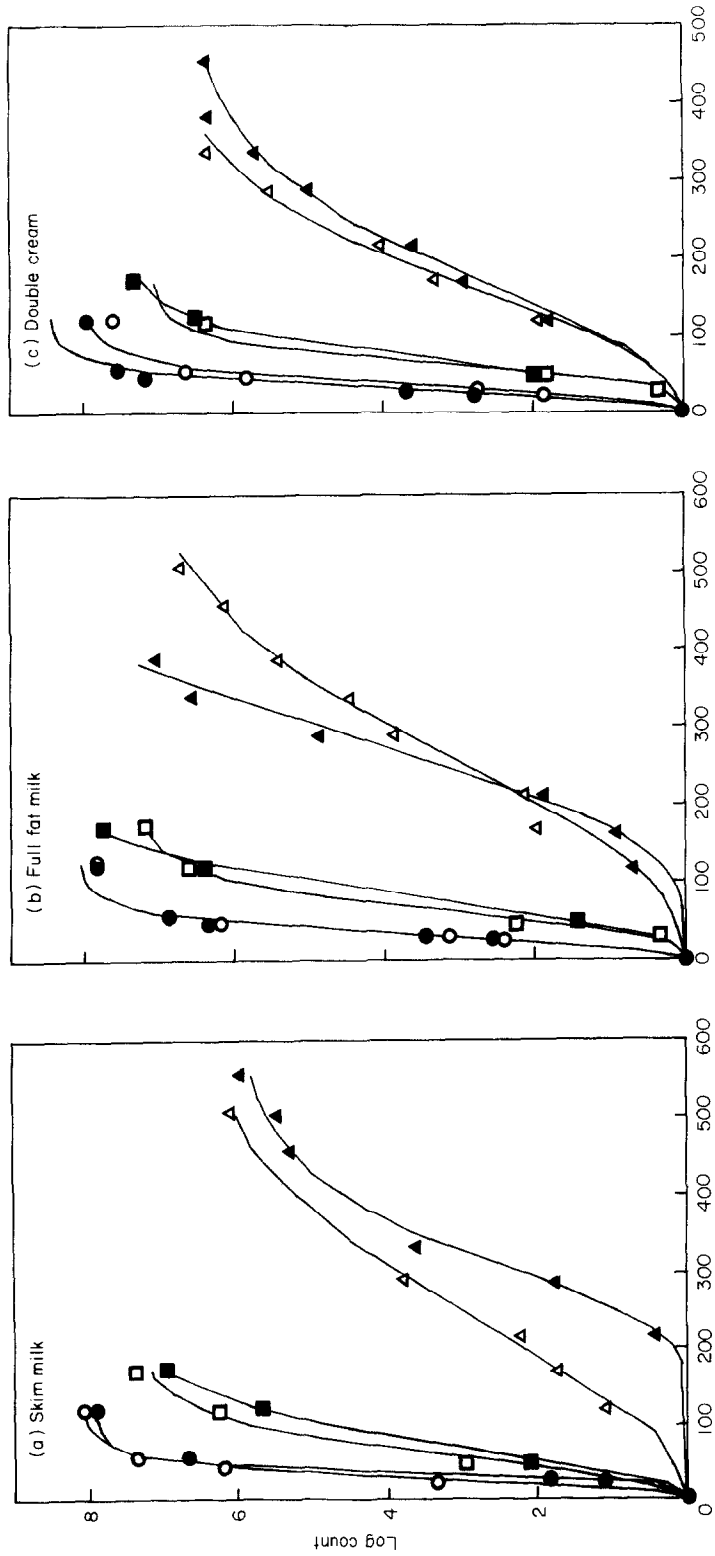


**Fig. 2.** Growth of *Serratia marcescens 755* on (a) pasteurized (open symbols) and UHT (closed symbols) skim milk, (b) pasteurized (open symbols) and UHT (closed symbols) full-fat milk and (c) pasteurized (open symbols) and UHT (closed symbols) double cream at 15°C (●, ○), 10°C (■, □) and 6°C (▲, △).

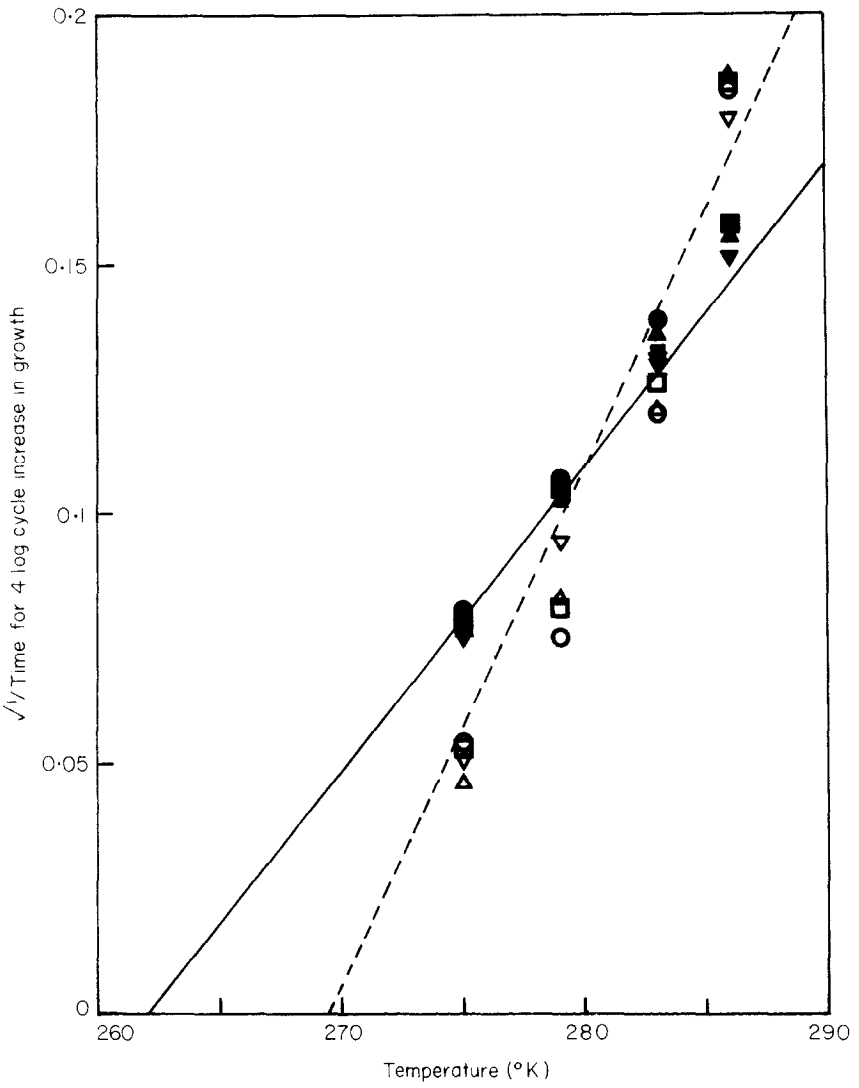


**Fig. 3.** Determination of $T_0$ values for two strains of *Pseudomonas fluorescens*. $T_0$ values were obtained from plots of $\sqrt{}$ (time for 4 log cycle increase in growth)$^{-1}$ for *Ps. fluorescens 1588* (raw milk isolate) (closed symbols) and *Ps. fluorescens 1181* (double cream isolate) (open symbols) grown on skim milk (●, ○), full-fat milk (■, □), single cream (▲, △) and double cream (▼, ▽).

study for strains of these bacteria. μ values were also similar for the strains studied by Shelley et al. (1986) and those in this study.

Maxcy (1982) proposed the use of the Arrhenius equation for prediction of the relative rate of deterioration of milk. He stated that the generation time was inversely proportional to the log of growth temperature when pseudomonads were growing at their maximum rate in milk with a correlation coefficient of 0·99. A similar relation was found in the present work but at a lower degree of correlation. Average correlation coefficients were 0·88, 0·90, 0·90 and 0·89 for skim milk, full-fat milk, single and double cream respectively.

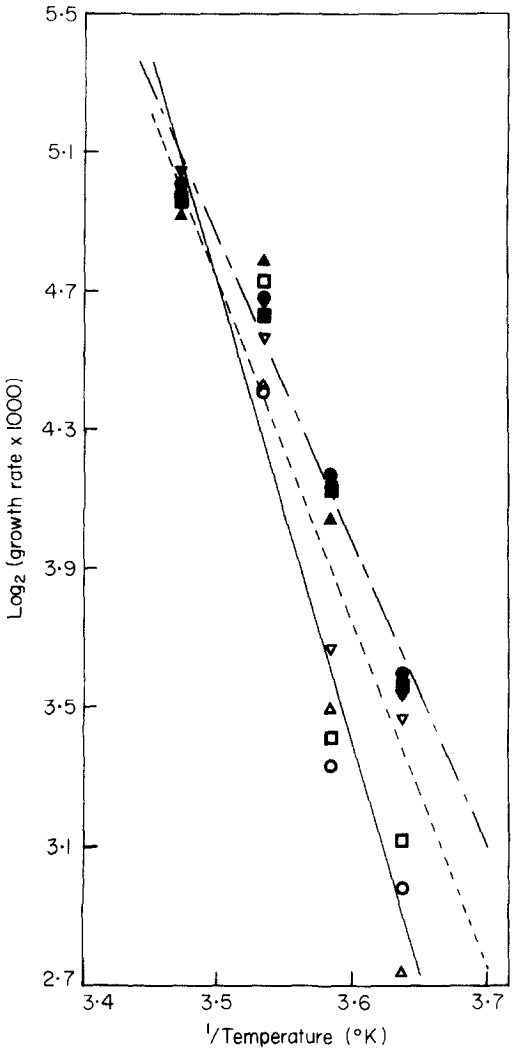


**Fig. 4.** Determination of μ values for two strains of *Pseudomonas fluorescens*. μ values were obtained from plots of $l_n$ (growth rate × 1000) for *Ps. fluorescens 1588* (raw milk isolate) grown on skim milk (●---●), full-fat milk (■---■), single cream (▲---▲) and double cream (▼---▼) and for *Ps. fluorescens 1181* (double cream isolate) grown on skim milk (○---○), full-fat milk (□---□), single cream (△---△) and double cream (▽---▽).

*Growth of bacteria in UHT and pasteurized products*

Pasteurized and UHT skimmed milks, full-fat milks and double creams were prepared from the same raw milk source in the Institute dairy. Growth of *Serratia marcescens 755* at different temperatures in these products was compared (Fig. 2). There was little difference in growth patterns of the bacterium in UHT products compared with pasteurized products. Thus, growth data generated using UHT products were applicable to pasteurized milks and creams.

**Table 7. Application of square root plot to bacteria grown in mixed culture in dairy products.**

| | | $T_o$ values | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Skimmed milk | | Full-fat milk | | Single cream | | Double cream | |
| | Organisms present | Pure culture | Mixed culture | Pure culture | Mixed culture | Pure culture | Mixed culture | Pure culture | Mixed culture |
| A | Enterobacter agglomerans 1498 | 266·6 | 270·1 | 267·7 | 270·6 | 264·0 | 269·5 | 266·7 | 268·5 |
| | Citrobacter freundii 1197 | 268·4 | 270·4 | 269·8 | 270·6 | 269·6 | 269·5 | 265·1 | 269·4 |
| B | Pseudomonas fluorescens 1181 | 269·8 | 272·2 | 269·6 | 271·1 | 270·1 | 271·1 | 269·3 | 268·4 |
| | Serratia marcescens PP | 274·2 | 272·3 | 274·0 | 272·7 | 273·2 | 272·1 | 273·8 | 272·2 |
| C | Pseudomonas fluorescens 1181 | 269·8 | 270·9 | 269·6 | 270·7 | 270·1 | 271·6 | 269·3 | 270·5 |
| | Bacillus circulans MRM 064 | 263·6 | 267·5 | 262·7 | 262·8 | 259·1 | 265·8 | 260·2 | 255·0 |
| D | Pseudomonas fluorescens 1181 | 261·8 | 263·7 | 262·7 | 264·4 | 262·2 | 261·7 | 261·3 | 263·9 |
| | Serratia marcescens PP | 274·2 | 275·2 | 274·0 | 275·7 | 273·2 | 275·2 | 273·8 | 275·1 |

*Effect of source of bacteria on growth in dairy products*

Although there seemed to be no significant variation in $\mu$ and $T_0$ values for bacteria grown in different dairy products (Table 4), when bacteria were grouped on the basis of the product from which they were isolated, differences did emerge (Table 5). There was no significant difference in either $\mu$ or $T_0$ values obtained for raw milk isolates grown on full-fat milk or double cream. However, bacteria obtained from double cream had a significantly lower $\mu$ value for growth on this substrate than that obtained during growth on milk. $T_0$ values for these organisms grown on the two substrates were not significantly different. The differences in $\mu$ for pasteurized cream isolates growing in milk and cream were mainly due to faster growth in the cream at low temperatures (Table 6). This variation of $\mu$ with growth substrate confirms the suggestion that conditions and substrates for growth have a strong influence on this parameter (Reichardt and Morita 1982).

The effect of isolate source on rate of growth in double cream presumably reflects variations in metabolic activity of the strains involved. The organisms isolated from double cream would appear better adapted to metabolize this substrate, whereas bacteria isolated from milk were able to grow equally well in the range of dairy products studied. The adaptation of double cream isolates probably involves lipid metabolism. Growth parameters for two strains of *Pseudomonas fluorescens,* one isolated from raw milk and the other from double cream are shown in Figures 3 and 4.

*Behaviour of organisms in mixed culture during growth in dairy products*

As the $T_0$ value appears to be an intrinsic characteristic of a bacterium, organisms grown in mixed culture should produce the same $T_0$ values as when they grow alone. This was shown to be the case for bacteria grown singly and in various combinations on All Purpose Tween broth (Stannard et al. 1985).

In the majority of cases in this study, $T_0$ values for organisms grown in pure culture were also comparable with those obtained from mixed culture experiments (Table 7). However, $T_0$ values for *B. circulans* differed when the organism was grown on its own and when grown in combination with a *Ps. fluorescens* strain. There was evidence that growth of the *Bacillus* spp. was inhibited in the presence of the pseudomonad and this may have affected calculations, although subsequent agar diffusion tests failed to show an interaction between the two organisms.

Shelley, Deeth and MacRae (1986) showed that generation times of strains of *Ps. fluorescens* and *Ps. fragi* grown in mixed cultures in UHT milk were not markedly different from those in pure culture.

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