

# CSIRO Food Research Quarterly



# Flame sterilization of vacuum - packed products

By D. J. Casimir, P. J. Rutledge and J. N. Huntington

CSIRO Division of Food Research, North Ryde, N.S.W.

The use of flame exhausting and flame sterilization overcomes the several processing problems which have restricted the acceptance of vacuum packing by the canning industry.

Vacuum packing was initially termed 'Geraldizing' after F. F. Fitzgerald who developed the process for vegetable products while working in the Research Department of the American Can Company. The process was first used commercially in 1928 for the production of whole kernel sweet corn. Despite the generally excellent quality and other advantages of vacuum-packed products, the process has found little acceptance in the world canning industry and is probably not used at all in the Australian industry.

The only foods that can be vacuum-packed are those that have a particulate form. A small quantity of liquid is filled into the can with the food and the can is then sealed under a high vacuum. The vacuum in the can is usually in the range of 27–22 inHg\* and the ratio of the volume of the liquid, solid, and gas space is often about 10:70:20. The cans are then processed in steam at about 120°C in stationary retorts or reel cookers.

When conventional equipment is used for vacuum packing, three production problems may arise. Firstly, the efficacy of the heat

sterilizing process depends to an important extent on the can having high vacuum and it requires special care in using vacuum closers to obtain the required level of vacuum in all cans in a production run. Secondly, because the cans have a high vacuum when cold, there is a danger they may panel or collapse when suddenly subjected to steam under pressure at the beginning of the sterilizing process. Finally, the cans float in water, so special care is needed when they are being cooled by flooding the retorts at the end of the sterilizing process.

## Flame exhausting

The flame exhausting process was described by Casimir and Lewis in 1971 and is an efficient and readily controllable method of obtaining uniform and high vacua in cans of particulate products. The required quantity of brine or syrup, say 50 ml for a 66 by 101.5-mm can, is filled into the can containing the product and the open can is passed in an upright position on a conveyor over a ribbon gas burner. The cans may be moved intermittently or in a continuous stream over the burner so that the added liquid boils and the steam displaces air from the can. Emission of steam shows that exhausting is complete and the can is immediately closed on a conventional machine at atmospheric pressure.

This procedure is adaptable to high speed lines and it gives vacua as high as 22 inHg after the cans are processed and cooled to ambient temperature.

## Heat transfer in vacuum-packed products

Jackson (1940) described how heat is

\*In this paper can vacuum is expressed in the traditional units of 'inHg'. However, the Standards Association of Australia has ruled that the approved units will be '—kPa' (minus kilopascals). The relationship of the two units is: 1 inHg = —3.386 kPa.

It is also possible to refer to the absolute pressure in a can instead of the vacuum. The absolute pressure is zero kPa when there is a total vacuum and 101.3 kPa when the can is at one atmosphere pressure. Hence a vacuum of 27 inHg is equivalent to  $27 \times (-3.386) = -91.4$  kPa or to an absolute pressure of  $101.3 - 91.4 = 9.9$  kPa.

transferred in vacuum-packed cans during processing in stationary retorts:

'The liquid in the can is easily vaporized at low temperatures in the low pressure atmosphere. This warm vapour then circulates through the can and gives up some of its heat to the cooler food particles by condensation. The condensate flows down to the warmer part of the can and is again vaporized. As the partial pressure of the gases in the can approaches the vapour pressure of the hot liquid outside the can, the rate of the heat transfer must decrease.'

Thus the rate of heat transfer in a vacuum-packed product is rapid while distillation is occurring, but later in the process, heat transfer is much slower. Consequently, the processing times for vacuum-packed products in stationary retorts are longer than comparable brine or syrup packs in which convective heat transfer continues throughout the process.

Roberts and Sognefest (1947) found that the rate of heat transfer could be increased in vacuum-packed products by rotating the cans to circulate the liquid phase continually from the can wall to all parts of the product.

### Flame sterilization

Flame sterilizers are very suitable for processing vacuum-packed products because the heating medium is at atmospheric pressure. Hence the danger of panelling the cans is minimized. Moreover, they are continually rotated during the heating and cooling operation to give high rates of heat transfer. In addition, no problem arises as a result of cans floating during cooling as they are only partly immersed in water while being showered by overhead sprays.

### Flame-processed vacuum packs

The CSIRO Pilot Flame Sterilizer described by Casimir (1975) was used to process a variety of vacuum-packed products. The processing of two of these products, diced potato and whole kernel sweet corn, is described below to illustrate the effectiveness of combining vacuum packing and flame sterilization.

#### *Diced potato*

Initial attempts to produce vacuum packs of diced potato by flame sterilization resulted in marked breakdown in texture and the product resembled lumpy potato soup. Breakdown could be prevented by treating the dice with calcium as described by

Mitchell (1972). The potatoes (variety S3012 grown by the Department of Agriculture at Orange, N.S.W.) were abrasion peeled, diced into 1-cm cubes, and the dice was divided into lots for calcium treatment. The calcium treatments were 0, 5, 10, 15, and 20 min soaking in 0.5% calcium chloride at 73°C. The treated potato dice was then drained, and 230 g of dice was filled into plain 66 by 101.5-mm cans. The cans were brined with 50 ml of a 5% salt solution and sealed in a vacuum closer to give a can vacuum of 22 inHg. The cans were then flame sterilized on the CSIRO pilot unit operating under the following conditions:

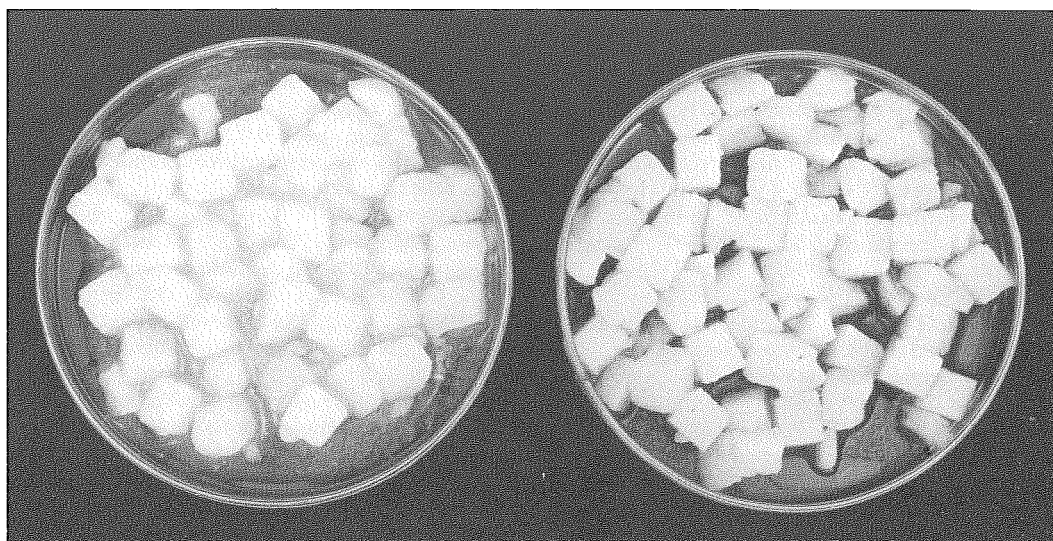
Throughput	14 cans per min
Stroke	30 cm
Rotation of can	56 cycles per min
Number of burner stations	11
Stab temperature <i>ex</i> flame	128°C
F <sub>0</sub> value of process	11.5 min

The amount of sloughing and breakdown of the tissue from the various calcium treatments was determined by firstly agitating the contents of the can with 500 ml water for 30 s. The mixture was poured onto a tared No. 9 Tyler mesh screen and the liquid portion was collected in a 1-litre measuring cylinder. The washings were used to make the volume up to 1 litre and the sloughed material was allowed to settle for 2 h before the volume of settled solids was measured. The dice on the screen was weighed after 2-min draining. Sloughed tissue volumes and drained weights for the different calcium treatments are shown in Table 1.

Table 1. Extent of sloughing in vacuum-packed diced potatoes processed in the flame sterilizer

Calcium soak (min)	Drained weight (g)	Volume of settled solids (ml)
0	157	140
5	239	50
10	241	25
15	238	25
20	242	20

The drained weights of the calcium-treated samples did not vary significantly, but they differed from that of the untreated control. The degree of sloughing as measured by the volume of settled solids was small for treatments of 10 min or longer soaking time



Calcium-treated potato dice (right) and untreated potato dice (left) after flame sterilization.

in 0.5% calcium chloride. The appearance of the control sample and the sample soaked for 10 min in the calcium solution is shown in the figure. The samples with 10-min calcium treatment had a good colour, firm texture and would be suitable for inclusion in potato salad.

#### *Whole kernel corn*

The corn was obtained from a crop of Code 22 grown at Richmond, N.S.W. After cutting and washing, 221 g of unblanched corn was filled into 66 by 101.5-mm plain cans. The cans were brined with 36 ml of a solution of 15% sugar and 5% salt, and closed under a vacuum of 22 inHg. The cans were divided into two lots for processing. One lot of cans was processed upright in a retort for 42 min at 118°C; the calculated  $F_0$  value of the process was 6.0 min. The other cans were flame sterilized in the CSIRO pilot unit operated under the following conditions:

Throughput	15.0 cans/min
Stroke	30 cm
Rotation of can	56 cycles/min
Number of burner stations	11
Stab temperature <i>ex</i> flame	128°C
$F_0$ value of process	11.0 min

The sweet corn was taste tested after storage for 18 months at ambient temperature, by 31 tasters using 7-point

Table 2. Mean taste test scores\* for vacuum-packed sweet corn

Sensation	Retorted	Flame sterilized
Colour	3.95	6.37
Flavour	5.27	6.15
Texture	5.47	6.10
General acceptability	4.58	6.05

\*Hedonic scale 1 to 7: dislike very much to like very much.

hedonic scales for colour, texture, flavour and general acceptability. The mean scores for the four factors are shown in Table 2. All pairs of results are significantly different ( $P < 0.05$ ) with colour and general acceptability being highly significantly different ( $P < 0.001$ ).

#### **Conclusions**

Flame sterilizers may be used to overcome the technical problems which have inhibited the production of vacuum-packed canned foods. It is now possible to produce a range of vacuum-packed products which are at least equal in quality to packs processed by conventional means.

Flame exhausting is a new procedure for obtaining the required level of vacuum in the can without having to use relatively slow and expensive vacuum closers. The addition of only a small amount of liquid to the can

reduces the heat capacity of the pack and gives more rapid changes in temperature during flame sterilization and subsequent cooling. Under these conditions heat damage to the product is minimized. The use of a limited amount of liquid also reduces the loss by leaching of water-soluble nutrients such as vitamin C (Brush *et al.* 1944) and sugars (Andreotti *et al.* 1974).

## References

- Andreotti, R., Tomasicchio, M., and Castelvetro, F. (1974). Ulteriori studi sulla preparazione di conserve vegetali al naturale confezionate sotto vuoto. *Ind. Conserve* **49**, 225-7.
- Brush, M. K., Hinman, W. F., and Halliday, E. G. (1944). The nutritive value of canned foods. V. Distribution of water soluble vitamins between solid and liquid portions of canned vegetables and fruits. *J. Nutr.* **28**, 131-40.
- Casimir, D. J. (1975). Flame sterilization. *CSIRO Food Res. Q.* **35**, 34-9.
- Casimir, D. J., and Lewis, P. S. (1971). Product formulation specifically for flame sterilization. Specialist Courses for the Food Industry (AIFST-CSIRO) No. 2, 59-61.
- Jackson, J. M. (1940). Mechanisms of heat transfer in canned foods during thermal processing. *Proc. Food Conf. Inst. Food Technol.* pp. 39-50.
- Mitchell, R. S. (1972). An improved method for controlling breaking of potatoes during canning. *J. Food Technol.* **7**, 401-8.
- Roberts, H. L., and Sognefest P. (1947). Agitating processes for quality improvement in vacuum-packed vegetables. *Proc. N.C.A. Tech. Sessions 40th Annu. Conv.*

# Do amines in foods trigger headache?

By E. F. L. J. Anet and D. L. Ingles

CSIRO Division of Food Research, North Ryde, N.S.W.

*'Well, that is a most astonishing thing' said Shaw.\* 'You have spent your life in trying to discover the North Pole which nobody on earth cares tuppence about and you have never attempted to find a cure for the headache which every living person is crying aloud for.'*

Cries for a cure for headache have resulted in extensive studies of this problem. In this paper we consider the relationship between one kind of headache—migraine—and amines in foods.

Migraine involves headache and usually other symptoms, e.g. disturbed vision, pins and needles, nausea and vomiting. A detailed description of migraine has been given by Lance (1973).

Numerous triggering agents of migraine attacks have been suggested, e.g. emotional or physical stress, glare, noise, hormones and alcohol. It has been known for 200 years that certain foods also trigger migraine attacks

(Fothergill 1784) but only a minority of migraine sufferers believe that their attacks are precipitated by specific foods (Hanington 1967). The basic cause of migraine is unknown, though there appears to be a genetic component.

## Chemical induction of headache

A number of substances are known to produce headaches; among these are some vasodilator drugs, used to treat angina pectoris, which have side effects including headache. For example, inhalation of amyl nitrite produces a marked flushing in the skin area of the neck and head with dilation of the meningeal vessels. This gives a transient pulsating headache. Similarly, exposure to nitroglycerine and organic nitrites results in headache, and this effect is

\*The statement above by the playwright, G. B. Shaw, a migraine sufferer, to Nansen the Arctic explorer is quoted by Pearson (1944).

an occupational hazard in the munition and pharmaceutical industries.

'Histamine headache' commonly results when histamine is given parenterally, i.e. not through the alimentary tract. This type of headache has been described (Goodman and Gilman 1970) as follows.

'The headache is preceded by flushing of the face and a gradually developing sense of pulsating fullness in the head. Following intravenous injections of the drug it appears in about a minute, reaches a peak shortly thereafter and slowly diminishes in intensity to terminate in about 5 to 10 minutes. At first it has a throbbing character and is coincident with the pulsations observed in the temporal artery; later it becomes more or less continuous and is felt as a dull ache in the head, often worst in the frontal and temporal regions.'

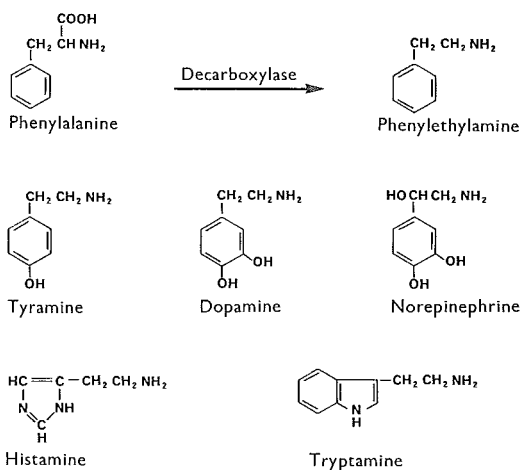
### Theory of amines as causal agents of migraine

Controversy surrounds the possible implication of histamine in the migraine syndrome (cf. Lecomte 1957). Since histamine occurs directly in many foods and may be produced in them by enzymic decarboxylation of histidine, a plausible theory of amines as a causative agent for migraine has developed.

The induction of headache by intravenous injection of histamine (tyramine produces the same effect) suggests that ingestion of food containing such amines might also produce headache in susceptible people. In a number of migraine sufferers this appears to be the case.

The enzyme, monoamine oxidase, usually protects the body against the effects of excessive amounts of circulating amine. This enzyme oxidizes the amine to ammonia and the appropriate aldehyde and is found in the intestine, blood, liver and kidneys. Consequently, the suggestion has been made that migraine results from a genetic deficiency of monoamine oxidase (Sandler *et al.* 1974).

Further evidence supporting the theory comes from the use of drugs which act as monoamine oxidase inhibitors. Such drugs, e.g. nialamide and phenelzine, are used in the treatment of psychiatric depression. It was observed that patients treated with these drugs were especially sensitive to certain foods, notably Cheddar, Camembert and Stilton cheeses. These caused hypertensive crises and headache. In some instances



intercranial bleeding occurred leading to death.

Tyramine was implicated as the causal agent in cheese (Asatoor *et al.* 1963). Presumably, the tyramine was not deaminated in the liver by the usual action of monoamine oxidase, and subsequently displaced norepinephrine in nerve endings (Anthony and Lance 1972). Overdoses of norepinephrine or conventional doses in hypersensitive persons are known to cause severe hypertension and violent headache.

Finally, studies of platelet monoamine oxidase activity in migraine patients and a control group showed significantly lower levels of activity in the former group (Sandler *et al.* 1974).

### Amines present in foods

Decarboxylase activity in foodstuffs and in man produces amines from amino acids. Thus amines may be present in the food we eat or they may be produced subsequently in the body.

Some of the amines implicated in triggering migraine and causing headache in susceptible individuals are shown in the figure while their levels in a number of foods are shown in the table.

### Correlations of migraine induction with foods

Foods which contain amines and have been implicated in migraine include: wine, beer and spirits, cheese, fish, beans, milk products, chocolate, eggs, wheat, nuts and tomatoes. Numerous references to different foods reported to trigger migraine attacks and also to precipitate a hypertensive crisis

Amine content of foods\* ( $\mu\text{g/g}$ )

Foodstuff	5-Hydroxy-tryptamine	Tryptamine	Tyramine	Dopamine	Histamine
Banana (pulp)	28		7	8	
Pineapple	50-60				
Camembert cheese			2000		
English Cheddar			0-953		
Australian Cheddar			226		
Yeast extracts			101-1639		208-2832
Chianti wine			24.5 $\mu\text{g/ml}$		22 $\mu\text{g/ml}$ †
Canned fish					1.8-156.9
Dried fish					1.8-300.9

\*Data extracted from Marley and Blackwell (1970).

†Compare also Marquardt, P., and Werringloer, H. W. J. (1965).

and headache in patients receiving monoamine oxidase inhibitors have been listed by Hanington (1967).

Ingestion of 3 mg of phenylethylamine (which occurs in chocolate) produced migraine in 18 of 36 patients with a history of dietary migraine (Sandler *et al.* 1974). Similarly, ingestion of 100 mg of tyramine (which occurs in large amounts in some cheeses) produced migraine in four persons who were completely unaffected by a placebo. The effect of tyramine was reproducible in the four patients with a clear history of dietary-induced migraine (Hanington 1967).

### Some comments on the theory of dietary migraine

Headache may be induced in anyone by the presence in blood of chemicals which are vasodilators. Headache occurs whether the chemicals enter the blood by inhalation as with amyl nitrite, by absorption through the skin or mucous membrane as with nitroglycerine or by intravenous or subcutaneous injection as with histamine or tyramine.

However, while there is good evidence that cheese containing tyramine and red wines containing histamine induce migraine attacks in some migraine sufferers, there is no evidence for an increase in the level of tyramine or histamine in the blood following ingestion of these foods. These critical measurements remain to be done.

These foods contain more than sufficient vasodilator to produce headache assuming no inactivation and complete absorption of the amine into the blood. For example, intravenous injection (Reis *et al.* 1957) of 0.05-0.1 mg histamine produces headache.

Such an injection corresponds to an average level of histamine in the blood of approximately 10-20  $\mu\text{g/l}$ . Ingestion of 1 l of red wine containing 22 mg of histamine could produce a level of *c.* 4400  $\mu\text{g}$  histamine/l in the blood of the average man. Similarly, ingestion of 100 g of tyramine-rich cheese containing 100 mg tyramine could lead to an average level of 20,000  $\mu\text{g}$  tyramine/l in the blood.

Such high levels of these amines in the blood could be lethal, but fortunately inactivation of these amines (by monoamine oxidase for example) occurs readily in the intestine and liver. It is important to know what level, if any, of these amines is attained in the blood and, in particular, what level occurs in the blood of a migraine sufferer whose attack is triggered by these foods and substances. These amines can be readily measured and satisfactory procedures for the estimation of picogram quantities of biogenic amines by mass fragmentography have been reported (see Karoum *et al.* 1975). Levels of vasoactive amines in the blood have been studied in migraine attacks induced by nitroglycerine and other agents (see Dalsgaard-Nielsen *et al.* 1975 and Anthony and Lance 1972).

If it can be shown that increased levels of vasoactive amine do not occur in the blood of migraine sufferers after they eat suspect foods, then a new theory for the mechanism of action for migraine induction by foods will be necessary.

### References

- Anthony, M., and Lance, J. W. (1972). Current concepts in the pathogenesis and interval treatment of migraine. *Drugs* 3, 153-8.

- Asatoor, A. M., Levi, A. J., and Milne, M. D. (1963). Tranylcypromine and cheese. *Lancet* **2**, 733-4.
- Dalsgaard-Nielsen, T., Bryndum, B., Fog-Moeller, F., and Kemp Genefke, I. (1975). Effect of percutaneous nitroglycerine upon the concentration of serotonin, epinephrine and norepinephrine in venous blood from migrainous subjects during attack-free intervals. *Headache* **13**, 231-4.
- Goodman, L. S., and Gilman, A. (1970). 'The Pharmacological Basis of Therapeutics.' 4th ed. p. 263. (Macmillan: London.)
- Fothergill, J. (1784). *Medical Observations and Enquiries* **6**, 103-37 (Cited by Sandler *et al.* 1974.)
- Hanington, E. (1967). Preliminary report on tyramine headache. *Br. Med. J.* **2**, 550-1.
- Karoum, F., Gillin, C., and Wyatt, R. J. (1975). Human cerebrospinal fluid (CSF) of biogenic amine metabolites. *Fed. Proc. Fed. Am. Soc. Exp. Biol.* **34**, 244.
- Lance, J. W. (1973). 'The Mechanism and Management of Headache.' 2nd ed. (Butterworth: London.)
- Lecomte, J. (1957). Liberation of endogenous histamine in man. *J. Allergy* **28**, 102-12.
- Marley, E., and Blackwell, B. (1970). Interactions of monoamine oxidase inhibitors, amines and foodstuffs. *Adv. Pharmacol. Chemother.* **8**, 185-239.
- Marquardt, P., and Werringloer, H. W. J. (1965). Toxicity of wine. *Food Cosmet. Toxicol.* **3**, 803-10.
- Pearson, H. (1944). 'Bernard Shaw.' p. 242. (Collins: London.)
- Reis, G. V., Lund, F., and Sahlgren, E. (1957). Experimental histamine headache. *Acta Med. Scand.* **157**, 451-60.
- Sandler, M., Youdim, M. B. H., and Hanington, E. (1974). A phenylethylamine oxidising defect in migraine. *Nature (Lond.)* **250**, 335-7.

## Sorbate treatment of moist prunes

By V. Stekly and D. McG. McBean

CSIRO Division of Food Research, North Ryde, N.S.W.

More than half of the prunes consumed in Australia are marketed as high-moisture fruit (35-40% water) packaged in flexible film pouches. This product, which has a water activity ( $a_w$ ) in the range of 0.82 to 0.85, supports the growth of a number of yeasts and moulds. Such moist-pack prunes are shelf stable only because contaminating organisms are inactivated when the pouches are filled at a minimum temperature of 80°C. In commercial practice, dried prunes that contain 15-20% water are washed, rehydrated in near-boiling water, inspected and then automatically weighed and sealed in flexible film pouches. The rehydrated fruit is often conveyed to the automatic weighing machine under live steam to ensure that it does not fall below the recommended sealing temperature of 80°C. The packaged prunes cool slowly as they are placed while still hot into fibreboard cartons for distribution.

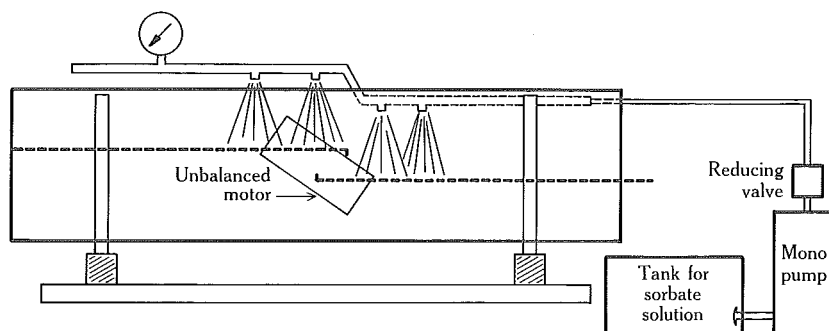
At present a small, undefined percentage of production spoils owing to:

- ▶ Filling at lower than the recommended temperature
- ▶ Recontamination by operators when they adjust the weight in a pouch by hand
- ▶ Sealing equipment being out of adjustment
- ▶ Perforation of pouches either from mishandling or by insects.

Any of these deficiencies may result in contamination, followed by noticeable growth of the invading organisms in 5-10 days at usual room temperatures. Similarly, once a pouch has been opened purposely it should be stored in a refrigerator to inhibit spoilage.

Recent amendments to food regulations in all Australian States permit the addition of the broad-spectrum fungistat, potassium sorbate, to high-moisture prunes. The use of this additive will prevent the growth of





Vibrator feeder and spraying mechanism showing unbalanced motors (one on each side); tank for sorbate solution with strainer at bottom; mono-pump; reducing valve; and rubber mountings.

microorganisms that may gain access to the product after heat processing and will increase the shelf life of the prunes after the package is opened.

The level of sorbate permitted in high-moisture prunes is 0.1% or 1000 p.p.m. (expressed as sorbic acid). The most appropriate methods for applying the fungistat are either to spray the fruit with a solution of the readily soluble potassium sorbate or to dip the fruit in the solution. In commercial packaging lines, the flow of fruit is either continuous or batchwise with about 30 kg rehydrated at a time. The points where sorbate can be applied are few. If it is dissolved in the rehydrating water, breakdown of the fungistat could be rapid in the temperature range of 90–100°C that is employed. If it is metered into the pouch, complete surface coverage is unsure. Whether the prunes are sprayed or dipped, sorbate is best applied between rehydration and packaging. The experiments conducted at North Ryde sought methods of application that would allow processors to meet the prescribed level for sorbate in packaged prunes and to test the efficiency of those methods.

### Experimental procedure

Coverage of the moist prunes by potassium sorbate solution must be complete if the fungistat treatment is to be effective. This is easily accomplished if the prunes are dipped. In practice, they are more likely to be sprayed and a small stainless-steel vibratory conveyor was built for this purpose (see figure). It consisted of a trough 1 m long and 30 cm wide and was fitted with perforated screens at two levels, so that fruit dropped from one level to the other about

half-way along its length. Spraying took place on each level to ensure that sorbate was applied to all surfaces of the prunes.

The vibrator unit consisted of two small unbalanced electric motors (30 W) adjusted to give a maximum synchronized centrifugal force of 20 kg. Their mounting on the trough at an angle of 35° caused the fruit to spread and move forward in a single layer. The capacity of the vibrator unit was 40 kg of cooked prunes per h and the average retention time was 3 min.

Potassium sorbate solution was sprayed on to the fruit through two pairs of Teejet nozzles mounted above the vibratory screens. These nozzles were selected for their uniform flat V-shaped pattern of spraying, which ensured that the fungistat was evenly applied. Sorbate solution was collected below the screens in a trough and discharged into a holding tank from which it was recirculated. A mono-pump equipped with a pressure-reducing valve provided recirculation. To avoid blockage of the nozzles the holding tank had a fine mesh screen over the outlet pipe to prevent particles of fruit from being pumped into the spraying system.

The discharges from the four jets at various pressures are shown in Table 1.

Table 1. Rate of discharge of sorbate solution through four Teejet nozzles at different pressures

Pressure (kPa)	Rate of discharge from four jets (litres/min)
140	1.1
210	1.4
280	1.5

Temperature of the sorbate solution was controlled by a heater and thermostat in the holding tank.

For processors who still handle prunes in batches, tests were done in which rehydrated prunes were allowed to drain, and then dipped for 1 min in sorbate solution before being filled into pouches and sealed.

Good-quality large prunes weighing 7–11 g each (size grades 66–44/500 g) and initially containing 19.5% to 20.5% moisture were used. They required 14–15 min immersion in boiling water to reach a moisture level of 40%.

Sorbic acid was determined by a modification of the method of Schmidt (1960.) This involves a steam distillation to separate the fungistat from the treated fruit, followed by spectrophotometric measurement of the alkaline distillate at 252 nm.

### Attainment of sorbate level

The maximum permissible level of sorbic acid in high-moisture prunes is 1000 p.p.m. but a target of 700–800 p.p.m. was set in these experiments to ensure that the prescribed level was not exceeded. Many factors influence the uptake of sorbate by prunes, but those studied were:

- ▶ Method of application—spraying or dipping
- ▶ Pressure at which spray is applied
- ▶ Concentration of sorbate solution
- ▶ Temperature of sorbate solution and of prunes
- ▶ Size of fruit.

Absorption depends on the nature of the skin of the prunes, but no attempt was made to vary this characteristic. Exposure time for spraying was about 3 min throughout and dipping time was 1 min. Each was considered to be a practical time suitable for commercial use.

### *Spraying pressure*

Three spray pressures, 140, 210 and 280 kPa, were tested using 0.5% potassium sorbate solution on size grade 44 prunes. Uptakes of the fungistat were 290, 320 and 290 p.p.m. respectively, showing that spray pressure has only a minor influence on the absorption of sorbate. Consequently all succeeding tests were done at a constant spray pressure of 210 kPa.

### *Concentration of sorbate solutions*

The average 300 p.p.m. of sorbate absorbed from a 0.5% spray solution, as shown in the preceding section, is well below the allowable limit of 1000 p.p.m. Absorption increased to 400 p.p.m. with a 1.0% solution, but it was necessary to use a 2.0% solution to reach the target level of 700–800 p.p.m.

When prunes were dipped for 1 min in potassium sorbate solutions, uptake ranged from 640 p.p.m. with a 2.0% solution to 1020 p.p.m. with a 3.0% solution. Dipping in a 2.5% solution produced an average absorption of 800 p.p.m.

### *Temperature of sorbate solutions*

The temperature of sorbate solution at the start of a test was 20°C while that of rehydrated fruit was near 70°. During the treatment the temperature of the fruit fell to about 50°, but as batches were small, that of the solution never exceeded 25°. During commercial production, the much larger throughput would cause the sorbate solution to reach higher temperatures, especially if dipping were used. The uptake of sorbate at a number of temperatures was therefore studied. Spraying tests with 1.0% sorbate solution showed that uptake increased from 380 to 480 p.p.m. when the temperature of the solution was increased from 25 to 50° and then to 500 p.p.m. when the temperature was raised to 70°. With a 2.0% sorbate solution, uptake increased from 700 to 870 p.p.m. when the temperature was increased from 25 to 50°. Similar results were observed in dipping trials.

These increases in absorption with increasing temperature are probably due to more rapid penetration of sorbate into the fruit.

### *Size of prunes*

Prunes of three sizes grades, 66, 62 and 44, were sprayed with potassium sorbate solution at about 25°C. Uptakes of the preservative were 510, 555 and 760 p.p.m. respectively. This result is opposite to that expected since the absorption area per unit weight is greatest with the smallest fruit. This unexpected result is probably due to the thinner skins of the larger fruits which allow easier penetration of the preservative.

### *Distribution of added sorbate in prunes*

Fungal growth usually commences on the surface of a food and this is the most

appropriate place for a fungistat to lodge. Results of the experiment involving size suggested that the distribution of sorbate taken up by the fruit should be investigated. Treated and packaged prunes from that experiment were left for 48 h for moisture to equilibrate. They were then carefully dissected to give a sample of skins with a trace of adhering flesh and another sample which consisted of the remaining flesh. The sorbic contents shown in Table 2 indicate that much of the sorbate penetrates deeply into the flesh even though the concentration is generally higher on the skins.

Table 2. Sorbic acid contents in the skin and flesh of prunes sprayed with 2·8% potassium sorbate solution

Size grade	Sorbic acid content (p.p.m.)	
	Skin	Flesh
66	600	420
62	640	420
44	780	600

#### Cost and re-use of sorbate

Food-grade potassium sorbate costs *c.* \$5 per kg. One hundred litres of 2% sorbate solution therefore cost \$10. In commercial practice this is enough to treat 2000 kg of prunes so the cost per kg of fruit is 0·5c or just over 0·1c per 250-g package. With the spray system described, 0·5 kg of 2% sorbate solution was used per 10 kg of prunes: a rate of 1 g of preservative per kg of prunes, again at a cost of 0·5c. Potassium sorbate solution is stable under the conditions used in these trials, and with our spray system no significant reduction in concentration was observed when 40 kg of prunes were treated with a 2% solution at 50°C during 1 h of continuous recirculation. In commercial production, however, some reduction in concentration of the recirculating solution would occur because of the carry-over of water on the surface of the fruit following

rehydration. It is good practice to discard used potassium sorbate solutions once or twice per day because they become contaminated with soluble constituents from the prunes. Thus the volumes used for spraying or dipping should be as small as possible. The cost of potassium sorbate should be offset by the prevention of losses from microbial spoilage.

#### Storage of sorbate-treated prunes

The effectiveness of potassium sorbate in inhibiting fungal growth in prunes was tested with fruit rehydrated to 40% moisture, sprayed with 2% sorbate solution and packaged in pouches of Saran-coated Cellophane laminated to polyethylene. The average sorbic acid content of these test samples was 720 p.p.m. A uniform filling temperature of 50°C was adopted for all packaged test samples. They were stored at 25° and some treatments were inoculated with *Aspergillus chevalieri*, a mould known to occur commonly on prunes and to cause serious deterioration of moist-pack fruit. Thirty pouches of each of five treatments were stored. These treatments and the results of the storage are shown in Table 3.

All sorbate-treated samples, whether they had been inoculated or not, showed no spoilage after storage for 1 year. More than half of the untreated inoculated prunes showed mould growth within 1 month while the remainder gradually disintegrated, possibly as a result of yeast growth, to become unacceptable commercially.

#### Conclusions

Spraying high-moisture prunes with 2% potassium sorbate solution or dipping them in a 2·5% solution resulted in levels of fungistat in the fruit which were close to, but below, the permitted limit of 1000 p.p.m. (expressed as sorbic acid). The above concentrations should not need to be altered

Table 3. Spoilage in moist-pack prunes in the presence of the fungistat potassium sorbate

Sorbate treatment (solution cold)	Sorbate in fruit (av. p.p.m.)	Inoculation	Incidence of spoilage in 30 pouches		
			1 month	3 months	12 months
2% spray	685	no	none	none	none
2% spray	685	yes	none	none	none
2·5% dip	665	no	none	none	none
2·5% dip	665	yes	none	none	none
Untreated		yes	17	17	30

in commercial practice despite observed variations in uptake of sorbate resulting from changes in temperature of the solution and variation in the size of the fruits and possibly in their skin characteristics. Treated fruit remained free of microbial spoilage for one year at 25°C even when inoculated with a xerophilic mould, *Aspergillus chevalieri*, which often occurs on prunes and is known to spoil high-moisture packs of the fruit. The temperature of potassium sorbate solution in commercial practice will be defined by the temperature of the fruit being treated and the rate of throughput. Although the use of

the fungistat obviates the need for packaging at a temperature of at least 80°, treated prunes should still be packed at a minimum temperature of 50°.

The cost of treating prunes with potassium sorbate is about 0.5c per kg of fruit, but this cost is offset by the reduction of spoilage in packages during distribution.

#### Reference

Schmidt, H. (1960). A specific method for the colorimetric determination of sorbic acid (in non-alcoholic foods). *Z. Anal. Chem.* **178**, 173-84.

## Microorganisms in dairy products — friends and foes

By Barbara P. Keogh

CSIRO Division of Food Research, Highett, Vic.

Microorganisms are endowed with many enzymes which are essential for their own metabolic processes and which give them the ability to act either beneficially as necessary agents in the manufacture of certain foods or adversely as food spoilage organisms. Their ability to act in these roles varies according to the species and the food, the components of which are substrates for the enzymes. The role of microorganisms in the production of dairy foods will be considered in both of these contexts.

There are three main classes of enzymes which play a part both in dairy processing and in the spoilage of dairy products: (i) the lactases which are involved in the fermentation of lactose, (ii) the proteases which are involved in the breakdown of milk proteins to peptides and amino acids, and (iii) the lipases which are involved in the breakdown of milk fat to fatty acids. These enzymes may operate advantageously or disadvantageously. The ability of microorganisms to bring about these chemical reactions is utilized to advantage in the production of fermented milks and cheeses,

while the breakdown of milk constituents by any of the three types of enzymes mentioned above is undesirable in the manufacture of products such as butter or milk powder.

#### History

For many centuries soured milk has formed a vital part of the human diet. Its use as a food was mentioned in the Bible (Deuteronomy XXXII, 14: Genesis XVIII, 8) and it was, and still is, known to the nomads of Asia Minor as part of their natural diet. It is known to have been enjoyed by the early Egyptians, Greeks and Romans and it is said that the early Romans taught cheese-making techniques in the countries they conquered. The famous Emmentaler cheese industry of Switzerland is believed to have started in this way. While the transformation of milk in flavour and texture to another very palatable food was utilized, the fact that it was the result of bacterial activity was not recognized until the time of Louis Pasteur.

In 1907 Metchnikoff, a Russian academic who worked at the Pasteur Institute,

published in his book 'Prolongation of Life', theories on the life-giving properties of the Bulgarian fermented milk, yoghurt. He believed that its consumption provided a defence against infection and the production of toxins by bacteria in the intestine. To it he attributed the good health and longevity of the Turks and Bulgars. Although his theories have since been discounted, yoghurt is a very nutritious food which is easily digestible. Despite the primitive state of bacteriology at that time, his discoveries concerning the bacterial flora of yoghurt have not, however, been discounted.

Metchnikoff attributed the special characteristics of 'Yahourth' to the activity of the 'Bulgarian bacillus' which was always present in it. His description suggests that this organism is our modern *Lactobacillus bulgaricus*. He noted also that 'a para lactic bacillus' was always present. This organism appears synonymous with *Streptococcus thermophilus* of today. It has now been established (Pette and Lolkema 1950a) that in fact the best flavoured yoghurt resulted from the associated growth of *Lactobacillus bulgaricus* and *Streptococcus thermophilus*.

There are many other types of fermented milks. Practically all nations have one or more such milk traditionally made by allowing the milk of donkeys, goats, sheep or cows to sour by the action of the lactic acid bacteria which occur naturally in the milk. Dahi of India, leber of the Bedouins, matzoon of Armenia, pinner of Lapland are but a few examples. Today, many of these products are made commercially in countries far beyond their places of origin. Probably the best known to the Western World are yoghurt, Kefir, Koumiss, acidophilus milk and some modern variations of these such as the fermented milk of Japan known as Yakult. There are too many to describe in detail, but the role played by microorganisms in the better known ones may be of interest.

### Fermented milks

#### Yoghurt

In the Middle East and southern European countries yoghurt is still prepared for local use from the milk of buffaloes, cows, donkeys, sheep and goats, but the yoghurt of the Western World is made from cows' milk. The name of the product has many spellings: yoghurt, yoghaurt, yoghourt, yahourth, yogurt, yaourt, jugurt, yaert, yaoert.

The manufacture of yoghurt in the

Western World is scientifically controlled and is a more sophisticated process than that which produces the yoghurt of the Balkans. There, the bacteria naturally occurring in the milk were used as the souring agents. In yoghurt made commercially, milk is homogenized and heated to 85–90°C, held for 15–30 min, cooled to 45° and inoculated with a mixture of *L. bulgaricus* and *S. thermophilus*. These organisms are propagated either together or separately until inoculated into milk for yoghurt manufacture. Incubation is at 43°. Heating the milk before inoculation not only destroys undesirable bacteria, but also drives off the oxygen thereby creating the right conditions for the growth of *L. bulgaricus*. Moreover, heating produces growth factors by denaturing the milk proteins. It has now been established that the *L. bulgaricus* and *S. thermophilus* stimulate each other (symbiosis). The *S. thermophilus* produces formic acid which is required as a growth factor by *L. bulgaricus* (Galeslout *et al.* 1968; Veringa *et al.* 1968). The *L. bulgaricus* with a higher proteolytic capacity, liberates essential amino acids, valine in particular, for the *S. thermophilus* (Pette and Lolkema 1950b).

During the growth of yoghurt bacteria in milk lactic acid, diacetyl and acetaldehyde are produced by the fermentation of lactose and the breakdown of proteins. All these substances contribute in some degree to the flavour and odour of the product, but the most important is acetaldehyde which is produced by *L. bulgaricus*. The selection of strains for the production of these flavour compounds does not appear to be of much importance, but the viscosity of the product depends on the ability of the strains of *L. bulgaricus* and *S. thermophilus* to produce a polysaccharide from galactose (lactose → glucose + galactose) which acts as a thickener. Viscosity plays an important part in the organoleptic quality of the yoghurt. Selection of strains on the basis of their ability to produce the thickener can eliminate the undesirable influence of seasonal variation in the milk (Veringa 1973) and obviate the need to use stabilizers such as alginates and gelatin which detract somewhat from the quality of the product.

Provided the conditions of manufacture of yoghurt are strictly controlled, the product is not very susceptible to bacterial spoilage. The high degree of acidity precludes the

growth of pathogens, but spoilage can arise from the growth of yeasts and moulds. The lactose-fermenting yeasts, e.g. *Torula cremoris*, are the most common contaminants of natural yoghurts made without added sugar, but yoghurt production today has expanded into new styles with added fruits and flavourings, and in these the risk of yeast and mould contamination is increased. Trouble also arises from types of yeasts other than those which ferment lactose. Yeast contamination can lead to abundant gas production in the body of the product, while moulds, being aerobic, form colonies on the surface. With both types of contamination there is a consequent deterioration in flavour. Obtaining infection-free cultured products is a difficult task. Yeast and mould contamination from air and equipment can be avoided by strict attention to hygiene at all stages of manufacture, and contamination from fruit and syrup is reduced by the use of good quality raw materials which are adequately heat-treated before being added to the product. Fresh and frozen fruit give the best product organoleptically, but they are more liable to yeast and mould contamination.

### *Kefir*

After yoghurt, Kefir, is possibly the best known of the fermented milks. It originated in the Caucasian mountains and until recently did not move far beyond that region. Today, Kefir is popular in many countries of Europe and other parts of the world but by far the greatest commercial production of Kefir is in Russia. In 1964 one factory in Moscow was reported to produce 100,000 kg per day (Lang and Lang 1973).

The distinctive characteristic of Kefir is the development of granules up to the size of a walnut during its production. These are composed of denatured protein containing many different strains of yeasts and species of bacteria. The characteristics of Kefir result from the combined fermentation of the milk by lactic acid bacteria and by yeasts which produce about 0.8% lactic acid, about 1% alcohol and carbon dioxide. A good Kefir foams and effervesces like beer.

After fermentation the granules are filtered off and may be used immediately to inoculate new batches of milk or may be washed and dried and stored until required. When the dried granules are inoculated into milk, they rehydrate and once more start to

ferment. The dried granules are commercially available in Europe but only to a limited extent in Australia because of quarantine regulations.

The fermented milk is a very refreshing drink. In some European countries fruit-flavoured Kefir is popular.

### *Koumiss*

Koumiss is very similar to Kefir, being mildly alcoholic and effervescent. It is traditionally made from mares' milk and originated in the Asiatic Steppes. Today in Russia, it is manufactured in large quantities from cows' milk and therefore differs little from Kefir. Currently it enjoys a reputation comparable with that of yoghurt at the time of Metchnikoff. Thus it is said to cure many ailments and is believed to be especially rich in nutrients.

The starter organisms for its commercial manufacture are *L. bulgaricus*, *L. acidophilus*, and the lactose-fermenting yeast *Saccharomyces lactis*.

### **The special qualities of *Lactobacillus acidophilus***

As it has been shown that *Lactobacillus acidophilus* may become established in the human intestine, milk fermented by this organism has achieved some recognition in the medical field. Not all strains of *L. acidophilus* can establish themselves in the intestine, and among the variants which have this capacity others arise which do not (Anon. 1974). The therapeutic properties of *L. acidophilus* have been studied in Russia for many years, and as early as 1930 strains were isolated which produced acid freely and which were claimed to possess therapeutic properties. It was subsequently found that *L. acidophilus* produced substances which suppressed *Escherichia coli* and pathogenic intestinal bacteria. Strains of *L. acidophilus* were later selected on this basis (Lang and Lang 1973).

Fermented milks containing *L. acidophilus* have some place in the treatment of intestinal disorders resulting from antibiotic treatment. It is common experience that as a result of antibiotic treatment an imbalance of the natural intestinal flora occurs. Selected strains of *L. acidophilus* are apparently able to restore this balance. In England, an antibiotic-resistant strain of *L. acidophilus* has been isolated and is marketed for consumption during the course of antibiotic

treatment, either as yoghurt which has been inoculated with the organism or as a dried powder for addition to milk.

Acidophilus milk or 'reform yoghurt', which contains only *L. acidophilus*, and Bioghurt, which contains *L. acidophilus* and *Streptococcus taette* (a variant of *S. lactis*), have been consumed in Europe and the United States for many years. Acidophilus milk is a very acid product and many attempts have been made to develop a product with a better flavour. In some instances this is done by inoculating a quantity of milk with a mixture of *S. cremoris* (or *S. lactis*) and an aroma organism such as *S. diacetylactis* or a leuconostoc or alternatively with *L. bulgaricus* and *S. thermophilus*, and mixing this fermented milk with another quantity of milk which has been inoculated with *L. acidophilus*.

In the product known as Aco yoghurt a freeze-dried acidophilus powder is added to the milk at the same time as the normal yoghurt bacteria. Another variation is a product called 'Acidophilin' which is made by fermenting milk with a mixture of *L. acidophilus*, *S. lactis*, and a Kefir culture. Yet another modification is 'acidophilus-yeast milk' which is made by fermenting milk with *L. acidophilus* and a lactose-fermenting yeast.

In Japan, cultured milk drinks are very popular and are regarded as having health-giving properties. They frequently contain autolyzed Chlorellae or Scenedesmus (algae) which stimulate the lactobacilli and extend their viability.

### Use of aroma-producing bacteria

In cultured buttermilk and cultured cream, which are popular in Europe, the United States and Australia, the fermentative organism is *S. diacetylactis* which produces diacetyl. This imparts a butter flavour to the product. Butter manufacture in Europe also has a fermentation step in the process of manufacture: before the cream is transformed to butter it is fermented with a mixed starter containing *S. diacetylactis* and leuconostocs. Both these bacteria produce the diacetyl required for the enhancement of the butter flavour. Similarly, aroma bacteria may be used in cheese manufacture to obtain a buttery flavour.

### Microorganisms in cheese manufacture

Lactic acid bacteria play an essential role in cheese manufacture. The starter

cultures may be mixtures of *S. lactis*, *S. cremoris*, *S. diacetylactis* and leuconostocs or may be single strains of *S. lactis* or *S. cremoris*, the latter being generally employed in Australia and New Zealand for the manufacture of Cheddar cheese.

A common problem encountered in Cheddar cheese manufacture is the failure of the starter organisms to produce the desired acidity in the milk because starter organisms are susceptible to bacteriophage (bacterial virus) infection. While all bacteria are susceptible to bacteriophage infections, the problem is highlighted in Cheddar cheese manufacture because of the need to produce a higher degree of acidity in a relatively shorter time than in the manufacture of other cheese varieties. If cheese starters are selected on the basis of bacteriophage sensitivity patterns, the risk of infection is reduced and this is the reason behind the choice of single strains for Cheddar cheese manufacture in Australia and New Zealand.

In all types of cheese, from the soft cheeses such as cottage cheese to the hardest cheeses such as Romano, the lactic acid produced by the starter is an important factor involved in the removal of water from the milk. In manufacturing most cheese varieties, rennet is also added to help achieve this end. The desired amount of water to retain in the curd depends on the type of cheese being manufactured; it can be controlled by changing the temperature of the curd during manufacture as well as by adjusting the amount of salt and the method of its addition to the curd. The initial step in the manufacture of all cheese varieties involves a lactic acid fermentation by either the lactic streptococci or the lactobacilli, or both. In addition, other microorganisms may be used to achieve special flavours and characteristics.

Under some conditions of manufacture certain strains of the lactic streptococci can bring about an undesirable bitter flavour, particularly in Cheddar cheese. This flavour arises from the bitter peptides produced by the action on the milk protein of the proteolytic enzymes of these strains of bacteria.

The aroma-producing bacteria, i.e. *S. diacetylactis* and leuconostocs are essential to the manufacture of cheeses such as Gouda and Edam, as well as many others where the buttery flavour of diacetyl is a characteristic of the variety. The aroma-producing starters also produce CO<sub>2</sub> which is responsible for small gas holes that are another

characteristic of these varieties but are regarded as defects in Cheddar cheese.

*Propionibacterium shermanii* plays an important role in the manufacture of Swiss-type cheeses such as Emmentaler because it produces propionic acid and CO<sub>2</sub>. The former is responsible for the slightly sweet flavour of the cheese and the CO<sub>2</sub> for the large gas holes. The size of the gas holes is controlled by carefully controlling the time the formed curd is in the brine used for salting and the maturation temperatures.

Other microorganisms are involved in the production of surface-ripened cheeses where a slime consisting of salt-resistant yeasts and *Brevibacterium linens* forms on the surface and produces enzymes which diffuse into the cheese. These bring about a very strong characteristic flavour from the breakdown of proteins and milk fats. Limburger and Tilsit are examples of this type of cheese.

Mould-ripened cheeses which are so pleasing to many palates develop their characteristic flavours from the activity of moulds. In cheeses such as Roquefort of France, Gorgonzola of Italy, Blue Vein of Denmark and Stilton of England, as well as other lesser known varieties, the mould is *Penicillium roqueforti*.

In traditional production of these cheeses the mould arose from fortuitous contamination, but in commercial manufacture today it is intentionally introduced into the curd. During maturation the mould grows in the air space around the curd particles; Roquefort, Gorgonzola and Blue Vein are additionally spiked with stainless steel wires at an early stage in maturation to allow air into the curd. The mould then grows in the channels made by the wires. The open texture of the curd in Stilton provides conditions for abundant mould growth so that it is not usually spiked.

The enzymes from the growing mould diffuse into the curd and hydrolyse the milk fats to free fatty acids giving the flavours characteristic of these varieties. The fatty acids, caprylic and caproic acids, are important flavour constituents of this type of cheese. In addition, the oxidases of the moulds oxidize some of the free fatty acids to ketones which are also important flavour constituents.

In the manufacture of cheeses such as Camembert and Brie, the mould is inoculated into the milk or applied to the surface of the formed curd after salting. During the

maturation process the mould grows and its enzymes diffuse into the centre of the curd, breaking down the milk protein as they do so. Such cheeses are matured for only about 6 weeks because if matured for longer periods some of the amino acids are reduced to ammonia which is clearly recognized when the cheese is consumed.

The white mould of Camembert cheese is *Penicillium camembertii*, *Penicillium candidum* or *Penicillium caseicolum*. Any green moulds on the surface of a Camembert are contaminants and make the cheese unpalatable. Thus this cheese must be matured in carefully controlled rooms.

### Undesirable activities of microorganisms

While mould strains such as those described above can be used to advantage, there are many that cause food spoilage and some that are a health hazard. Certain *Aspergillus* strains, in particular, produce mycotoxins and some of these have been found to be carcinogenic in animals.

The lactic acid bacteria which are used to advantage in the production of fermented milks and cheese may cause defects in other products. For example, the growth of naturally occurring lactic acid bacteria in milk before processing results in defects attributable to the denaturation of casein and the production of lactic acid.

Pasteurization of milk destroys most lactic acid bacteria and pathogens, but there are some heat-resistant lactic streptococci and spore-forming organisms which survive this process. When pasteurized milk is stored for long periods it usually does not become sour as does raw milk, but goes putrid because of the proteolytic action of the heat-resistant organisms which, in raw milk, are suppressed by competition from the lactic acid bacteria that occur naturally.

In addition to organisms that can have both advantageous and disadvantageous properties, there are others which are solely spoilage organisms or are undesirable in other ways. After milk has been drawn from the cow, it usually becomes contaminated very quickly with many species of bacteria. Among these are a group of organisms known as psychrotrophs which grow at temperatures below 5°C and which have lipolytic and proteolytic activities in the milk. If these organisms are allowed to multiply in the milk before processing, the products of lipolysis and proteolysis are carried into the



manufactured product, causing off-flavours.

This problem has been accentuated in recent years by the introduction of refrigerated vats for storing milk on the farm and the use of refrigerated tankers for transporting the milk. Under these conditions the mesophilic organisms such as lactic streptococci cannot compete with the psychrotrophs and the increased time of holding allows them further opportunity for growth.

The anaerobic bacteria belonging to the genus *Clostridium* may be normally present in milk and cause no problem, except when the milk is used in the manufacture of cheese such as Gouda, in which they may cause gas production. The deep interior of the cheese is anaerobic and since the acid and salt levels in this region are initially low, conditions are favourable for the growth of these organisms. Clostridia may also be a problem in processed cheese.

While pasteurization destroys pathogens in the milk, there are opportunities during manufacture for these and spoilage organisms to enter dairy products and to grow during processing. It is the duty of the food processor and the food handler to take every precaution with hygienic handling of food, cleaning of equipment and personal hygiene. If coagulase-positive staphylococci have an opportunity to grow in food including dairy products, they may also have the opportunity to produce enterotoxins which are not destroyed by any further processing or cooking. As many people carry these organisms in their noses and on their hands, and because other organisms such as viruses and enteric pathogens are also carried by humans, strict hygiene is essential at all times to avoid transmission from food to the consumer.

*Escherichia coli* is an organism which is used as an index of direct faecal pollution of water, but it cannot be regarded in the same light for food because in the processing of milk and other foods it is possible that the *E. coli* remains on the equipment and is simply transferred from batch to batch of food. *E. coli* can, together with the coliform organisms, as a group, be an index of the cleanliness of the equipment and it is in this context that it is used in the food industry.

### Control of microorganisms

There are many microorganisms whose natural habitat is milk and others which

are always present in air. It is by utilizing knowledge of the growth requirements of microorganisms that the processor can prevent spoilage in food. Thus processing conditions can be controlled so that the products have a low water activity, a pH range unfavourable for microbial growth, or some other growth-limiting characteristics. For example, in butter, although the total water content is sufficient to support bacterial growth, a limit to growth is imposed by dispersing the water throughout the fat in droplets that are too small to accommodate a bacterial cell, let alone allow it to multiply. However, if the butter is stored at a temperature which allows migration and aggregation of the water droplets conditions favouring bacterial growth can arise. Addition of the salt to butter, while causing some enlargement of the water droplets, has a compensating inhibitory action on some bacteria.

Ripening of cream with aroma cultures lowers the pH of the cream and thus has a protective action against the growth of some spoilage organisms.

In cheese, bacteria multiply in the gel spaces containing water and on moist interfaces between the original curd particles. The more water that is removed by curd shrinkage during the cheese-making process the better is the keeping quality of the cheese. Soft cheeses, such as cottage cheese, have a short shelf life because of their high water content and, at the other end of the scale, cheeses such as hard, grating Parmesan may keep for several years. As discussed earlier bacterial growth is essential in the manufacture of cheese, but because of the unfavourable conditions most bacteria die relatively early in the life of the cheese. It is the residual bacterial and natural milk enzymes which play the major part in the maturation process. The growth of contaminant moulds is controlled not only by the anaerobic conditions, which are quickly established in the body of the cheese, but also by careful packaging with films of low oxygen permeability.

Low water activity of a product can be achieved in several other ways. The water may be removed by spray or roller drying as with milk powder. In sweetened condensed milk the addition of sugar combined with evaporation of water increases the percentage of total solids and gives rise to a high osmotic pressure, thereby lowering

the water activity. Unlike other canned foods, sweetened condensed milk is not sterilized in a retort. The air must be excluded from the can by filling it completely as any remaining head space could result in the growth of moulds and osmophilic bacteria.

Recognition of the habits of microorganisms, careful attention to the methods set down for manufacturing dairy products and observance of hygienic practices in food handling, packaging and storage are the factors which keep microorganisms under control.

## References

- Anon. (1964). On the manufacture of yoghurt. *Misset's Zuivel* **70**, 688–9, 691.
- Galesloot, T. E., Hassing, F., and Veringa, H. A. (1968). Symbiosis in yoghurt. I. Stimulation of *Lactobacillus bulgaricus* by a factor produced by *Streptococcus thermophilus*. *Neth. Milk Dairy J.* **22**, 50–63.
- Lang, F., and Lang, A. (1973). Cultured milk products. *Food Manuf.* **48**, 23–8, 54.
- Pette, J. W., and Lolkema, H. (1950a). Yoghurt. I. Symbiosis and antibiosis in mixed cultures of *Lactobacillus bulgaricus* and *Streptococcus thermophilus*. *Neth. Milk Dairy J.* **4**, 197–208.
- Pette, J. W., and Lolkema, H. (1950b). Yoghurt. II. Growth stimulating factors for *Streptococcus thermophilus*. *Neth. Milk Dairy J.* **4**, 209–24.
- Veringa, H. A., Galesloot, T. E., and Davelaar, H. (1968). Symbiosis in yoghurt. II. Isolation and identification of a growth factor for *Lactobacillus bulgaricus* produced by *Streptococcus thermophilus*. *Neth. Milk Dairy J.* **22**, 114–20.
- Veringa, H. A. (1973). The biochemistry of Yoghurt. Nizo-Mededelingen No. 7, pp. 88–96.

## General bibliography

- Anon. (1964). On the manufacture of yoghurt. *Misset's Zuivel* **70**, 676–78.
- Davis, J. G. (1952). Yoghurt. *Food* **21**, 249–54, 284–8.
- Davis, J. G. (1975). In 'Lactic Acid Bacteria in Beverages & Food', Proc. 4th Long Ashton Symp., Univ. Bristol, 1973. Eds. J. G. Carr, C. V. Cutting and G. C. Whiting. (Academic Press: London.)
- Davis, J. G. (1975). Yoghurt—recent developments. *Proc. Inst. Food Sci. Technol.* **8**, 50–66.
- Kosikowski, F. (1966). 'Cheese and Fermented Milk Foods.' (the Author: Ithaca, N. Y.)
- Siegenthaler, E. (1964). New developments in yoghurt manufacture. *Nord. Mejeri-Tidsskr.* **30**, 27–30.

# Time – temperature history recorders

By A. J. Carseldine and R. R. Weste

CSIRO Division of Food Research, Cannon Hill, Qld.

Two types of small, low-cost electronic devices have been developed to indicate the time-temperature history of frozen and chilled meat or other foodstuffs during storage and/or transport

Modern techniques of chilling and freezing can maintain food quality for a considerable time. However, quality deteriorates when the temperature during the subsequent storage and transport chain departs from the optimum. A simple, reliable, low-cost device to record the time-temperature

histories of food products would be most useful in detecting faults in the chain. Van't Root (1973) examined several devices and proposed methods but concluded, for either economic or practical reasons, that none was suitable.

At the Meat Research Laboratory of the



**Fig. 1.** Time-temperature integrator (left) and multi-temperature-band time recorder (right).

Division of Food Research, two devices have been developed that should prove suitable for investigations. One is a time-temperature integrator and the other a multi-temperature-band time recorder (Fig. 1).

The design of each instrument is based on the memory capability of a small microcoulometer known as an 'E'-cell,\* which consists of a silver case with a centre electrode of gold, the intervening space being filled with an electrolyte. When a current is passed through the cell in a forward direction, i.e. with the case positive and the centre electrode negative, silver is removed from the case and plated onto the centre electrode, the quantity transferred being directly proportional to the current flow per unit of time.

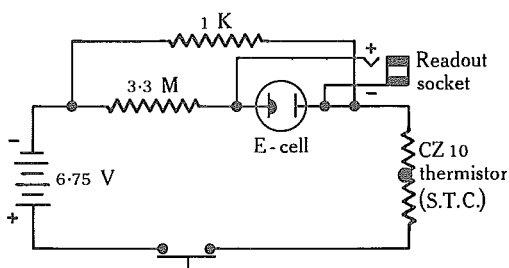
With a temperature-sensitive resistor (a thermistor) interposed in the circuit, the quantity of silver transferred is proportional both to temperature and to the duration of current flow. The 'E'-cell is read back by reversing the direction of current flow and measuring the time required, at a constant current flow, to plate the silver from the central electrode back to the case.

### Time-temperature integrator

The purpose of this device is to record the average temperature at which products are

held during the surveillance period. Reference to the circuit diagram (Fig. 2) will explain the operation.

The thermistor and the 1 k $\Omega$  resistor form a voltage divider across the battery supply. The 'E'-cell, with a 3.3 M $\Omega$  current-limiting resistor in series, is connected from the junction of the thermistor and the 1 k $\Omega$  resistor to the negative rail. Temperature variations change the resistance of the thermistor, thereby varying the voltage at this point. As the resistance of the 'E'-cell remains virtually constant, the voltage variation with temperature causes a change in current flow through the cell. Power is supplied by a small 6.75 V, 250 mAh mercury battery (in which the voltage remains practically constant throughout its life). A read-out socket is wired across the 'E'-cell in opposite polarity to the recording



**Fig. 2.** Circuit diagram of time-temperature integrator.

\*Plessey Ducon Pty Ltd.



by reference to the circuit diagram (Fig. 3).

Switching is performed by a C-MOS Schmitt Trigger integrated circuit. As there are two Schmitt Triggers in each IC package, two temperature detectors and 'E'-cells are assembled on one circuit board measuring 80 mm by 32 mm. Two circuit boards, battery, switch and read-out sockets are enclosed in a die-cast aluminium case (110 mm by 60 mm by 30 mm) having a total weight of 200 g.

The temperature detector is a thermistor situated in the lower leg of a voltage divider, the upper leg of which comprises a fixed resistor in series with a variable resistor, which is used to set the switching point. The junction of the upper and lower legs of the divider is connected to the input pin of the Schmitt Trigger. The output pin is connected through a current-limiting resistor to the input of the 'E'-cell, the output of which is taken to the negative rail.

As the temperature of the thermistor rises, its resistance falls, thereby reducing the voltage at the input to the Schmitt Trigger. When the set point is reached the Schmitt Trigger turns on and raises the corresponding output pin to full battery voltage causing current to flow through the 'E'-cell.

When the temperature decreases, the Schmitt Trigger turns off as the voltage rises through the set point and consequently

stops current flow through the 'E'-cell.

Power is supplied from a 2.7 V, 500 mAh mercury battery. A small push-button on-off switch is operated in the same way as in the time-temperature integrator. As the C-MOS integrated circuit has a very low current drain, it is estimated that the battery life would be more than 3 years (which is in excess of the normal shelf life of the battery). The capacity of each 'E'-cell under continuous operation is *c.* 75 days at temperatures down to  $-20^{\circ}\text{C}$ .

Each stage is calibrated by operating in a temperature-controlled bath at the mid-point of the temperature band for 24 hours. It is then read back and the time for the 'E'-cell to clear is noted. This figure is then divided by 24 to give the stage constant. When an instrument is read out at the end of a period of surveillance the total count for a stage is divided by the stage constant to give the operating time in hours with an accuracy of about  $\pm 3\%$ .

Patent applications have been lodged for both these devices with the aim of having them licensed for commercial production.

#### Read-out instrument

To read out the 'E'-cell, a constant current of known magnitude must be passed through the cell for a recorded time in the reverse, i.e. unplating, direction until the

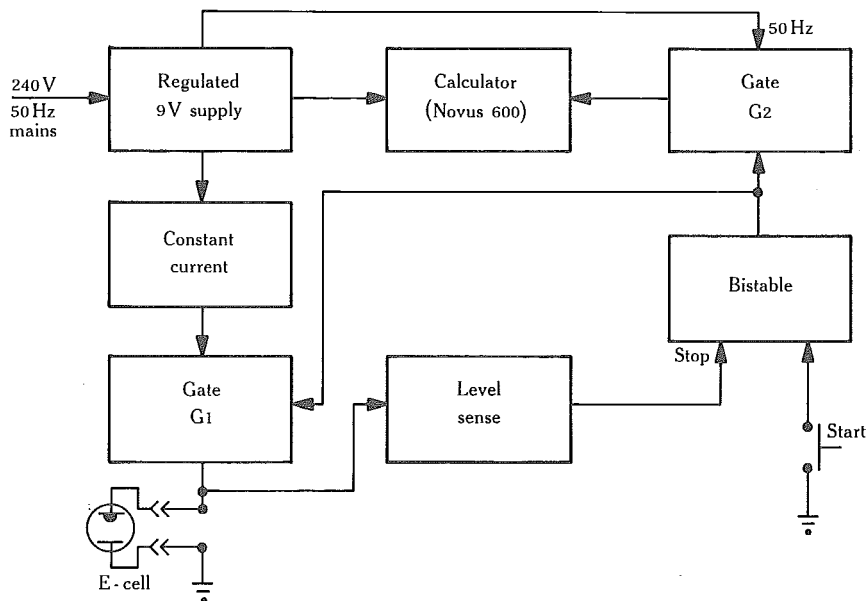


Fig. 4. Block diagram of read-out instrument.

'end point' is reached.

#### Timer system

A regulated 9-V power source (see Fig. 1) supplies a simple four function calculator, 1 mA constant-current generator and other associated circuitry (Fig. 4). The constant current is gated into the 'E'-cell via gate G1 when the bistable is in the RUN condition. At the same time, 50 Hz is gated by gate G2 to the ADD button of the calculator. If 2 had previously been selected on the calculator, it would then add 2 at the 50 Hz gate giving in this case a read-out of 100 per second. Owing to the low counting rate of this calculator it is not possible to run directly at 100 Hz.

When the 'end point' of the 'E'-cell unplating is reached, the voltage across the device rises, setting, via the level sense circuit, the bistable to the STOP condition. The two gates G1 and G2 are then opened, interrupting both constant current and the counting circuit. The display on the calculator then represents milliamp seconds to two decimal places.

#### Reference

Van't Root, M. J. M. (1973). Is it feasible to measure the time-temperature history of frozen foodstuffs? Proc. 13th Int. Congr. Refrig. (Washington), Vol. 4, 445-51.

## An apparatus for continuously monitoring the structural rigidity of a gel\*

By G. Vanderheiden

CSIRO Division of Food Research, Highett, Vic.

The development of an accurate robust apparatus for monitoring the gelation processes and its application to cheese manufacture are described. The welded stainless-steel sensing unit is rugged and easy to clean and maintain, and does not contaminate the product. Because the sensing unit does not damage the structure of the gel during measurements it has potential as a means of providing a standard objective method of measuring the firmness of a gel and reproducing a given end point.

An important operation in the manufacture of cheese is the addition of rennet and acid to the milk to form a coagulum resembling junket. A further operation is cutting this coagulum at exactly the right degree of firmness to allow separation of the curd and whey. All these operations have an

important influence on the yield and ultimate quality of the cheese. As reported by Richardson (1971), there have been many attempts to produce apparatus for monitoring the progress of coagulation brought about by acidification and enzyme treatment of milk. All of these instruments had some important limitation, the most common being damage to the structure of the curd under test. Other problems included stray electrical currents, gel build-up on cutting edges, limited test time due to restricted cutting-path length, lack of adhesion of the

\*This paper was presented at the Second Australian Conference of Science Technology organized by the Australian and New Zealand Association for the Advancement of Science at the University of Adelaide in 1975.

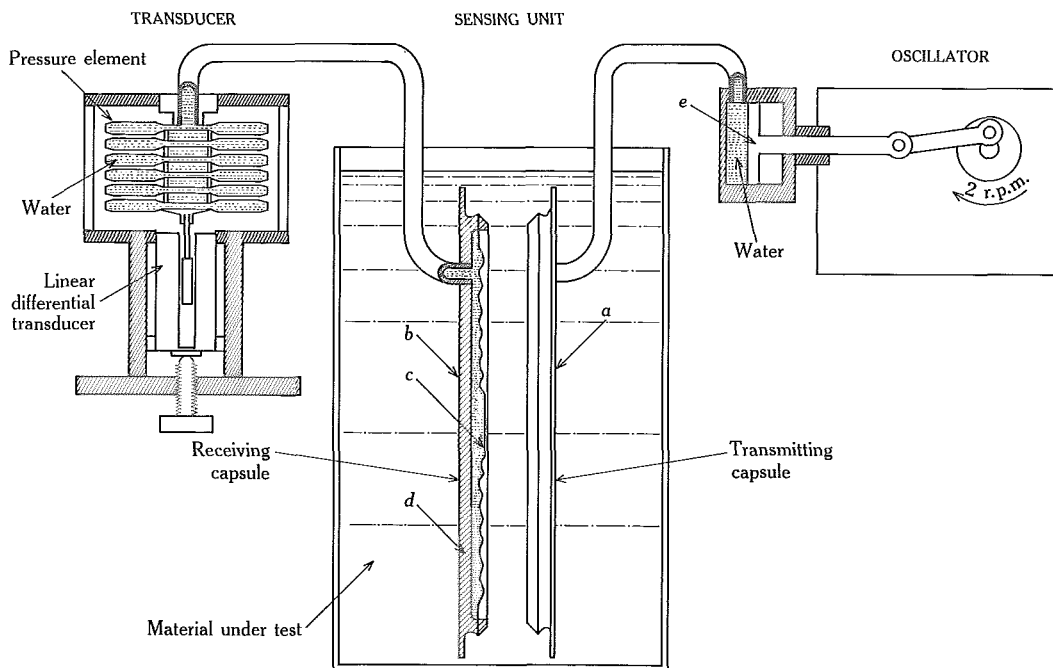


Fig. 1. Cross-section (not to scale) of the apparatus. *a*, transmitting capsule; *b*, receiving capsule; *c*, corrugated diaphragm; *d*, flat rigid plate; *e*, reciprocating piston.

gel to probe surfaces and cleaning difficulties. In addition, the services of a trained technician were mandatory. The apparatus (Australian Patent Application No. 55488/73) described in this article was developed for use in studies on milk coagulation and for objectively assessing the optimum cutting point in cheese manufacture. The limitations of earlier instruments have been avoided.

### Description of apparatus

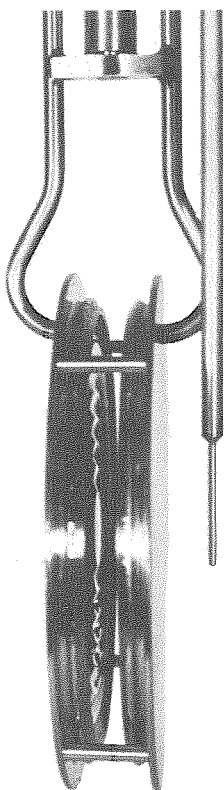
The apparatus comprises three separate components: a liquid-filled sensing unit employing a transmitting and receiving capsule, an oscillator containing an electric motor driving a reciprocating piston sealed with a Bellofram diaphragm, and a pressure transducer consisting of a pressure-sensitive element attached to the iron core of a linear differential transducer (LVDT) (Schaevitz 0-50 DC-B, range  $\pm 1.25$  mm, output 8.2 V/mm).

The transmitting and receiving capsules (*a* and *b*, Fig. 1; Fig. 2) of the sensing unit are constructed of stainless steel and are rigidly mounted in relation to one another 25 mm apart. Each capsule (130 mm in

diameter and 6 mm in width) is constructed with a 0.08-mm corrugated diaphragm forming one face (*c*, Fig. 1) and a flat rigid 4-mm plate (*d*, Fig. 1) forming the other with a gap of 2 mm between the two faces. In operation the transmitting diaphragm is hydraulically displaced  $\pm 0.25$  mm by the reciprocating piston (*e*, Fig. 1) in the oscillator forcing liquid (water) in and out of the capsule at a period of two pulses per minute through a connecting high-pressure nylon tube. The opposite receiving capsule (*b*, Fig. 1) is connected with a similar tubing to the pressure element of the linear differential transducer.

### Principle of operation

With the sensing unit submerged in the material under test and the transmitting diaphragm oscillating at a constant frequency (2 pulses/min) and displacement of  $\pm 0.25$  mm, the material while still in the liquid state produces minimum deflections of the receiver and consequently minimum output from the transducer. As coagulation progresses the increasing firmness of the material produces, with each oscillation of

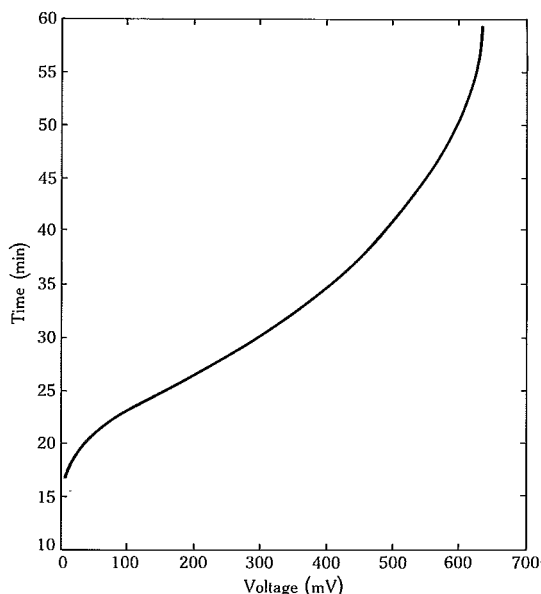


**Fig. 2.** The transmitting and receiving capsules.

the transmitting diaphragm, a greater deflection in the receiving diaphragm, and hence a greater output voltage from the transducer. This output voltage is related to the rigidity of the gel under test. A typical curve of voltage  $v$ . time for the coagulation of milk in cheesemaking is shown in Fig. 3.

### Discussion

The apparatus would provide a standard objective method of measuring the firmness of the coagulum in cheese manufacture and of detecting with a degree of reproducibility a given end point. For instance, the output



**Fig. 3.** Plot of the maximum output of the transducer against time as milk coagulates in making cheese.

voltage may be correlated with the point of cut as subjectively assessed by an experienced cheesemaker. The apparatus is therefore well suited for incorporation into an automated system of cheese manufacture. As a research tool it may be used to monitor the changes in firmness of the curd during the coagulation process and to study the influence of variables on coagulation. This apparatus, having been shown to be useful in cheese manufacture, clearly has a potential for monitoring gelation or coagulation processes in other media.

### Reference

Richardson, G. H., Gandi, N. R., Divatia, M. A., and Ernstom, C. A. (1971). Continuous curd tension measurements during milk coagulation. *J. Dairy Sci.* 54, 182-6.

## History of the Division of Food Research

The September and December issues of the *Quarterly* will comprise a two-part history of the Division of Food Research as our contribution to the jubilee celebrations of CSIRO.



# News from the Division

## Honours and awards

Mr M. V. Tracey, Chief of the Division, has been elected by Convocation to the Council of Macquarie University.

Drs B. V. Chandler and K. J. Nicol have been awarded the degree of M.Sc. (Biotech.) by the University of New South Wales. Dr R. L. Johnson has qualified for the Diploma of Education from the University of New England. All three are in FRL's Food Chemistry Section.

## General

Mr J. B. Davenport returned to FRL's Biochemistry Section in January, after completing a three-year term of secondment to the Reserve Bank of Australia. As Science Liaison Officer, he administered the Bank's Rural Credits Development Fund. Mr Davenport continues as a member of the Council of ANZAAS and as Honorary Editor of 'Search' magazine. He is Chairman-elect of ANZAAS for 1976.

*National Health & Medical Research Committee.* The following are members of the NH&MRC committees listed, for the triennium 1/1/76–31/12/78.

Mr M. V. Tracey: Food Standards (Standing) Committee;

Dr J. H. B. Christian: Food Microbiology (Reference) Subcommittee; and

Mr K. C. Richardson: Food Science and Technology (Reference) Subcommittee.

*Consumer liaison.* As part of its consumer liaison activities, the Division is producing a series of leaflets in non-technical language on a variety of food topics. Four of these leaflets have already enjoyed wide public acceptance and many thousands have been distributed; they are:

- ▶ Handling Food in the Home
- ▶ Don't Poison Your Family
- ▶ Citrus Juices—How to Preserve Your Own
- ▶ Prawns—Fresh and Frozen

By the time this issue of the *Quarterly* is printed, a fifth leaflet will be ready for distribution, called 'Milk and Cheese and All That—A Guide to Dairy Products'.

More consumer-orientated leaflets are in preparation and will deal *inter alia* with fish and other seafoods, fruits and vegetables.

*Australian Fishexpo '76.* The Division is involved in the first exposition of the Australian fishing industry, to be held in the Exhibition Buildings in Melbourne in September. Participation will be from fishermen, processing firms, marketing representatives, industry suppliers, government departments, consumer organizations, retailers and the public. The Fisheries Division of the Department of Primary Industry and the CSIRO Divisions of Fisheries and Oceanography, and Food Research are mounting an integrated stand for the Commonwealth Government.

The table below shows that Australians eat a remarkably small amount of fish and the exhibition organizers hope to stimulate a greater demand for seafoods from the general public. If, as seems probable, Australian territorial waters are to be extended to the 200-mile limit, the local fishing industry will be able to introduce several new species of fish to the market, but this will call for considerable expenditure on research, vessels and equipment.

Estimated consumption of foodstuffs per head of population during 1972–73 in Australia

Meat	96 kg
Poultry	13 kg
Fish	6 kg
Eggs	218
Milk, fluid, whole	124 litres
Milk, condensed and powdered	12 kg
Cheese	5 kg
Butter	8 kg
Margarine	6 kg
Sugar, refined	21 kg
Vegetables	119 kg
Fruit	92 kg
Breakfast foods	7 kg
Flour (largely as bread)	75 kg
Rice	3 kg

Source: Apparent Consumption of Foodstuffs & Nutrients 1972–73 Aust. Bur. Statistics, Canberra. Ref. No. 10.10, pp. 5–6, Jan. 1975.