

# NISIN in Food Technology—1

By H. B. Hawley\*

Nisin, an inhibitory metabolite produced by a cheese-starter organism, is the first antibiotic substance to be accepted by many countries for the control of certain types of bacterial spoilage. Below we publish the first part of a comprehensive survey of its discovery, its properties and its uses in a wide variety of foodstuffs. In Part 1 of this survey the author deals with the current use of nisin in natural and processed cheeses and discusses some of its potential applications in canned goods.

**N**ISIN is an inhibitory substance which is produced by a number of strains of the cheese-starter organism *Streptococcus lactis*, and cheeses in which it occurs naturally are not prone to certain bacterial defects. This property can be exploited by using these strains of *Str. lactis*, or the inhibitory principle itself in the manufacture of other types of cheese and in cheese processing.

Since this substance inhibits *Clostridia* (anaerobic spore-bearing bacteria) and certain other spoilage bacteria, it has applications in other fields of food handling, manufacture and processing, particularly in canning and pickling. It is valuable as a supplement to various established methods of food preservation, but it is not a substitute for them. Canned foods must always be rendered safe against *Clostridium botulinum* by the traditional methods of processing. Unfortunately, such treatment may not make the product sterile, and the use of nisin is therefore a valuable additional safeguard.

## Properties of nisin

The inhibitory effects of certain strains of *Streptococcus lactis* when used with *Lactobacillus bulgaricus* in the preparation of Yoghourt was first noted by Rogers in 1928.<sup>1</sup> Later Whitehead and Riddet<sup>2</sup> in New Zealand observed the existence of an inhibitory substance during an investigation of the behaviour of the mixed strains of lactic streptococci used in cheese starters. Whitehead<sup>3</sup> subsequently succeeded in concentrating and partially characterising it. In England, Meanwell<sup>4</sup> isolated

similar lactic streptococci from farm milk and dairy farm equipment. Mattick and Hirsch<sup>5</sup> eventually isolated and studied this inhibitory substance, which they named "nisin." More detailed investigations were made by Hirsch,<sup>6,7</sup> Berridge,<sup>8,9,10</sup> Berridge, Newton and Abraham<sup>11</sup> and Lilley.<sup>12</sup> The therapeutic potentialities of nisin have been investigated by Gowans, Smith and Florey,<sup>13</sup> who concluded that it was of little or no medical value, especially in view of the fact that adequate therapeutic levels of nisin are soluble only in aqueous solutions at pH 2 or lower.

Nisin is a metabolite of certain group-N lactic streptococci, the natural habitat of which is milk and the products of its fermentation such as cheese and Yoghourt-like drinks. It is a large polypeptide or small protein with a molecular weight of approximately 10,000, and consists of five polypeptide components with slightly different physical, chemical and biological properties.<sup>8,9,11</sup>

Nisin is inhibitory to various species and strains of the genera *Staphylococcus*, *Streptococcus*, *Neisseria*, *Bacillus*, *Clostridium*, *Corynebacterium*. Pure nisin was believed by Berridge<sup>8</sup> to contain approximately  $50 \times 10^6$  Reading units per gram, but recent work in the research division of Aplin & Barrett, Ltd., indicates an activity of  $40 \times 10^6$  R.u./g. On this basis the titre of nisin required to inhibit the organisms listed ranges from 0.25 to 500 units (0.006  $\mu\text{g.}$  to 12  $\mu\text{g.}$ ) per gram of substrate.

Precise quantitative data on the solubility of nisin is not yet available, since this depends on the pH and the nature of the substrate. At pH 7 in water the solubility is

in the neighbourhood of 75  $\mu\text{g.}/\text{ml.}$ , but it increases rapidly as the pH is lowered, being 1,000  $\mu\text{g.}/\text{ml.}$  at pH 5.6 and 12,000  $\mu\text{g.}/\text{ml.}$  at pH 4.2. The presence of protein in the substrate apparently increases the solubility on account of adsorption effects without any loss in activity. Crystalline nisin and high-titre preparations are most readily dissolved in 0.02 M. hydrochloric acid. In practice, it may safely be assumed that the solubility of nisin is adequate for all its potential applications to food technology.

In the dry state crystalline nisin and high-titre preparations may be stored at room temperature for at least several years, and probably indefinitely. In solution, however, the stability of nisin depends on the pH, temperature and nature of the substrate. Rapid inactivation occurs in alkaline solution, but under some conditions this is reversible. In most practical applications it is preferable not to exceed pH 6.5, although in view of the protective effect of many substrates, the stability is best determined under requisite conditions. As an example, an aqueous substrate containing 3% protein and 50  $\mu\text{g.}/\text{ml.}$  nisin when heated at 100°C. required for complete inactivation of the nisin, 2 hr. at pH 8, 1 hr. at pH 9 and 30 min. at pH 10. Between pH 2 and pH 6 there was no loss during 30 min. heating at 100°C., and only 25% loss at pH 6 after 1 hr.<sup>13</sup> Little loss of activity occurs on heating nisin preparations in processed cheese at 80-95°C. during the normal cooking time.

## Pharmacological and health aspects

Nisin is of a proteinaceous nature and does not contain any

\* Director and Director of Research, Aplin & Barrett, Ltd.

amino-acids of abnormal physiological configuration (D-series). It is partially inactivated by ptyalin and is digested by trypsin. Its maximum stability is in the range pH 2 to pH 4, and it is almost insoluble in water except in the neighbourhood of pH 2. Thus, nisin is not of medical interest and consequently problems of sensitisation and antibiotic resistance do not arise from its presence in foodstuffs.

A number of toxicity studies have been made, and recently a systematic investigation has been carried out on the acute and sub-acute toxicity of nisin and on a cheese containing nisin produced naturally by a strain of *Str. lactis* used as the starter organism. All these investigations show that nisin is harmless.<sup>13, 14, 15</sup> Tests made by Frazer and Hickman were carried out on albino rats, and they conclude:

"It has been shown that nisin is a harmless substance even when fed in large daily doses over a period of three months. It has been shown to have no lethal, or indeed any observable deleterious effects when given at 20,000 times and 40,000 times the standard dietary dose, assuming this to be of the order of 25 units/kg. body weight/day, of nisin."

Work in progress in the Research Division of Aplin & Barrett, Ltd., shows that the nisin-resistant bacteria examined are not cross-resistant to the medical antibiotics. Consequently, there is no danger that the use of nisin would militate against the therapeutic effectiveness of the medical antibiotics.

The sensitivity of nisin to ptyalin, its digestion by trypsin, its destruction by yeasts,<sup>16</sup> by certain lactobacilli<sup>17</sup> and by many lactic streptococci,<sup>18</sup> its relatively narrow spectrum and the insensitivity of most Gram-negative bacteria, and also its instability in the vicinity of pH 7 make it unlikely that it could have any effect on intestinal flora. This is borne out by experiments on pigs<sup>19</sup> and on poultry.<sup>20</sup>

There is also evidence that nisin is not foreign to the human

alimentary tract since nisin-producing strains of *Str. lactis* have been isolated from the human throat and from stools.<sup>21</sup> They are quite common in naturally soured milk and on milking equipment.<sup>4</sup>

## USES OF NISIN

### Processed cheese

At present nisin is being used in many countries in the manufacture of processed cheese and cheese spreads.

Processed cheese manufactured under properly controlled conditions contains only the spores of aerobic and anaerobic bacilli derived from the original cheese, but as a result of many circumstances outside the control of the cheese processor bacterial deterioration may follow germination of these spores.

The processing and packaging of the pasteurised cheese renders the product fairly anaerobic, and in consequence the Eh (oxidation-reduction potential) is usually too low for the aerobic spores to germinate and multiply. Unfortunately, these conditions are ideal for the germination of the anaerobic (clostridial) spores, and pasteurisation often provides the requisite degree of "heat-shock" to encourage germination. Arising from these circumstances, processed cheese may contain clostridia in practically pure culture.

The multiplication of clostridia in processed cheese often results in the formation of gas holes leading to blowing or inflation. This is a serious defect and heavy losses can be incurred. Blowing is not an inevitable consequence of clostridial growth since this depends on whether the metabolic reaction is homo- or hetero-fermentative. In some instances, putrefaction without gas production occurs, whilst in other cases there may be no very obvious organoleptic changes. Quite apart from the economic losses which may be incurred as a result of clostridia, their presence in processed cheese is objectionable on aesthetic and health grounds. It should also be borne in mind that many of the clostridia found in processed cheese are potentially toxigenic strains.<sup>22</sup>

Various attempts have been made to destroy the clostridial spores in processed cheese or to discourage clostridial proliferation. Cheese-cookers have been developed in which the processing can be carried out at temperatures of 105°C. or higher, but these temperatures have not proved satisfactory in destroying bacterial spores within practicable cooking times because of the protective effect of butterfat and protein. A serious objection to this process is the heat damage to the body and flavour of the cheese as well as damage to the milk protein, particularly the destruction of lysine, by the Maillard reaction.<sup>23</sup>

A reduction in pH and an increase in the salt content of processed cheese has been tried as a further means of control, but with little success.<sup>24</sup> A sufficiently low pH seriously impairs the quality and causes great processing difficulties; a high salt content makes the cheese unacceptable.

Attempts have been made to render conditions unfavourable for the clostridia by the addition of oxidising agents such as perphosphates, bromates, chlorates, chlorites, nitrates, in order to raise the Eh. Such additions are precluded by the preservatives regulations of most countries, and there is always the possible hazard of toxic complexes being formed

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(c.f. methionine sulphoximide in aged flour). The use of such additions may control clostridia, but the higher Eh encourages the growth of aerobic spore-bearers. In these circumstances, various undesirable changes can take place, such as proteolysis and "wheying-off," and occasionally blowing due to *Bacillus polymyxa* or related strains.<sup>25</sup> The aerobic sporeformers have also been implicated in various outbreaks of food poisoning. Suspicion particularly centres round *B. cereus*,<sup>26</sup> which is quite a common organism in processed cheese which has been rendered aerobic.

The clostridial deterioration of processed cheese and cheese spreads is a world-wide problem. The trouble is irregular and erratic, but in spite of numerous laboratory check procedures the manufacturer is frequently unable to ascertain the propensity to blow prior to distribution. Consequently, there is potential risk to the public, damage to the manufacturer's reputation and the liability to prosecution for purveying unwholesome food with the attendant risk of claims for damages.

Numerous workers have demonstrated that these troubles can be eliminated by the use of nisin, and wholesome, highly acceptable and biologically stable processed cheese is being manufactured by this method.<sup>16, 17, 27-31</sup> These observations have now received ample confirmation from manufacturing experience in the use of nisin during the past few years.

The use of nisin in processed cheese does not enable the manufacturer to process decomposed or unwholesome cheese, since it is not possible to produce a satisfactory product with such cheese; also if the processing and packaging are unhygienic, the nisin is often destroyed by the contaminating mesophilic bacteria.<sup>18, 32</sup>

### Original cheese

The potentialities of nisin are attracting much attention in the manufacture of many Continental types of cheese, particularly the low-acid sweet cheeses such as Emmental, Gruyère, Edam,

Gouda, Samsøe, Danish-Swiss, Tilsit, Limburger and also the hard cheeses, Parmesan and Grana.

All these types of cheese are very susceptible to clostridial blowing or other forms of clostridial deterioration, partly because of the lactose they retain and their relatively high pH. Heavy losses may be incurred, and these have been accepted in the past as inherent in the process. Numerous workers have shown that nisin can mitigate these defects,<sup>17, 18, 27, 30, 33-37</sup> and Edam containing nisin is now being produced commercially in Holland.

There is, as yet, no large-scale application because of various technical difficulties of varying magnitude depending on the type of cheese, but experimental work is in progress in France, Holland, Denmark, Austria, Norway, Sweden, Italy, Spain and Greece. Nevertheless, steady progress is being made as knowledge of the ecological inter-relationships of the micro-flora of cheese grows.

Parmesan cheese is particularly susceptible to clostridial deterioration,<sup>38</sup> and serious economic losses may be incurred, particularly in view of the long period of maturing and the relatively high incidence of defects occurring as late as twelve to eighteen months after manufacture. The addition of formaldehyde during manufacture appears to be widely practised as a means of mitigating some of these defects, but its use is widely regarded as undesirable if not positively dangerous. If nisin is capable of providing the answer to these problems, its adoption would make a vast difference to the Parmesan and Grana cheese industry. Work is in progress in the research division of Aplin & Barrett, Ltd., and the preliminary results are encouraging, but the long storage of these cheeses necessitates tests over protracted periods since it would be unwise to draw any hasty conclusions. Similar investigations have recently been commenced in Italy.

Recently, crystalline nisin and various high-titre preparations have become available commer-

cially and nisin-resistant strains of various lactobacilli concerned in the ripening of cheese have been developed in the research division of Aplin & Barrett, Ltd. These developments may obviate many of the disadvantages of nisin-producing starters by allowing the incorporation of nisin at later stages in the manufacture of the cheese. By this means it may prove possible to use ordinary starters in the early stages of production, the nisin being added when full acid development and curd-formation has occurred. The inclusion of additional cultures of nisin-resistant lactobacilli with the starters, or subsequently, should ensure continuity of acidification during ripening.

### Canned foods

Adequately heat-processed canned vegetables and meat packs which are regarded as commercially sterile may contain spore-bearing rods or their spores.<sup>39</sup> This is due to the extreme heat-resistance of certain of these organisms and the impossibility of using sufficiently high processing temperatures without incurring damage to the products. For example, Weinzirl<sup>40</sup> examined 782 normal cans of fruit, vegetables, meat and milk and found that 179, or 23%, contained viable organisms, of which spore-bearing rods formed almost the only type. Non-spore-bearing organisms were attributable to faulty seams. A similar investigation by Cheyney<sup>41</sup> revealed 8% non-sterile cans in his series of 725 cans after incubation at 37°C. for ten days. The only bacteria isolated were spore-bearing rods, except in the case of two cans with defective seams, which contained staphylococci.

The spore-bearing rods commonly occurring in such cans are *Bacillus stearothermophilus*<sup>42</sup> and related strains, which cause "flat-sour" spoilage by producing acid without gas, and *Clostridium thermosaccharolyticum*,<sup>43</sup> which gives rise to "hard swells" due to gaseous fermentations. Considerable spoilage may occur when packs infected with these organisms or related strains are exported

to hot climates or stored in the warm parts of cargo-ships. Spoilage may also arise if the cans are not cooled quickly after processing.

Damage and loss as a result of "flat-sours" and "hard swells" present a world-wide problem on which there is an extensive literature.<sup>44,52</sup> Such spoilage is particularly serious in wartime, since in the case of canned foodstuffs supplied to the armed forces no restrictions can be placed on storage, and soundness must be guaranteed under all climatic conditions.

Since nisin inhibits *Clostridia* and certain other spoilage bacteria, it has applications in many fields of food-processing, particularly in canning and pickling. It is proposed only as a supplement to certain established methods of food preservation and not as a substitute for them. It is not suggested that it should be used for the purpose of reducing the heat-treatment of canned foods which must be rendered safe against *Clostridium botulinum*.

Nisin has not yet been used on a commercial scale in canning, but encouraging laboratory data has been obtained. Its application in the canning of vegetables has been under experimental investigation at the Fruit and Vegetable Canning and Quick Freezing Research Association, Chipping Campden, Gloucestershire, England, during the past three years; work is also in progress at the Western Utilisation Research Branch, Agricultural Research Service of the United States Department of Agriculture.<sup>53, 54, 55</sup> A considerable amount of work is also going on in various commercial establishments.

It would appear that nisin will have the greatest application as a supplement to the normal processing schedules as a means of controlling spoilage by organisms resisting normal heat treatment. On the other hand, it may prove possible to reduce the thermal processing of certain heat-labile products, since it has been shown that nisin reduces the thermal resistance of certain spores.<sup>54, 55</sup> It should be noted that nisin is not active against most mesophiles en-

countered in canning, and spoilage would thus occur in the event of serious under processing. Bacteria may occasionally enter cans through faulty seams when they are cooled in contaminated water,<sup>56, 57, 58</sup> but nisin has no effect on the bacteria encountered under these circumstances and the manufacturer consequently obtains no protection against faulty techniques.

#### Vegetables and tomato products

The commonest forms of spoilage in canned vegetables are flat-souring caused by the facultative anaerobes typified by *Bacillus stearothermophilus* and the hard swells due to *Clostridium thermosaccharolyticum* and related strains. The work carried out at Chipping Campden, England, has clearly shown that nisin effects complete control of these microorganisms.<sup>56</sup> Its use should therefore eliminate a serious source of spoilage of many types of canned vegetables.

Canned beans with tomato sauce are fairly commonly spoiled by hard swells due to *Cl. thermosaccharolyticum*, which often comes from the tomato paste used.<sup>59</sup> Flat-sour spoilage is probably more common in canned peas although *B. stearothermophilus* may be present without causing spoilage in temperate climates.<sup>60</sup>

*Cl. thermofermentans* has been shown to be a cause of spoilage in canned egg-plant,<sup>61</sup> and *Cl. butyricum* is a cause of spoilage in canned beans, peas, potatoes and tomato products.<sup>62</sup> The effect of nisin in the control of these defects has not yet been investigated, but there is no reason to think that it would not prove effective.

Sugar is sometimes used in vegetable canning, for example in the brine with canned peas, and unless special grades are available it can be a source of mesophilic and thermophilic clostridia. High counts of thermophilic spores have been reported by British,<sup>63</sup> Dutch<sup>64</sup> and American<sup>65</sup> workers, who also found high counts of the spores of *B. stearothermophilus*. The mesophilic anaerobes, *Cl.*

*sporogenes* and *Cl. putrificum* were detected in 85% of the 33 samples of sugar examined by Weinzirl.<sup>66</sup>

Although, strictly speaking, tomatoes are classed as berries, it may be more appropriate to consider canned tomatoes in this section. Various types of spoilage bacteria have been isolated from canned tomatoes, tomato paste and tomato juice and different types of deterioration have been described. The spore-bearing rods isolated include *Cl. pasteurianum*, which will grow down to pH 3.55, *Cl. thermosaccharolyticum*,<sup>67, 68</sup> *B. thermoacidurans*<sup>59, 69</sup> and *Cl. butyricum*.<sup>70, 71</sup>

Canned tomato products can carry a heavy load of spore-bearing spoilage bacteria, which frequently cause trouble in packs containing tomato, such as beans with tomato sauce,<sup>59</sup> sardines or pilchards with tomato sauce and sandwich spreads. Tomato containing packs are also very susceptible to heat damage, so that it is desirable to keep heat-processing down to the absolute minimum consistent with safety. In view of this, nisin should have an important place in this field of food technology.

#### Fruit

Since yeasts and moulds are not sensitive to nisin, it has a somewhat more restricted application in fruit and fruit juice processing because the pH of these products is generally unfavourable to microbial spoilage. Canned pears and pineapple are, however, particularly susceptible to fermentation by butyric acid bacteria. *Cl. pasteurianum* will flourish at a pH below that desirable for high quality packs since too low a value causes maceration of the fruit and this organism will grow in fruit juices down to pH 3.55.<sup>67, 72</sup> Off-flavours and souring can be caused by the flat-sour aërobe, *Bacillus thermoacidurans*, which grows actively at pH 4 and shows slight growth at pH 3. Another spore-bearing aërobe causing spoilage in canned fruit is *Erwabacillus macerans* (*B. macerans*), which flourishes in the range pH 3.8-4.0.

## Meats

The application of nisin to the bacteriological problems of meat and meat-products processing opens up many attractive possibilities, since normal heat-treatment, although adequate to ensure a botulinum kill, may not be sufficient to achieve sterility. This is often the case with products incorporating spices, condiments, various cereal fillers and soya,<sup>73</sup> which may harbour large numbers of thermophilic spore-bearing bacteria. Meat extracts are also a prolific source of thermophiles.<sup>74</sup> Mossel<sup>19</sup> has drawn attention to the presence of clostridia in many of the ingredients used in meat canning as fillers and condiments; spices, starches and cereals may be prolific sources of mesophilic and thermophilic clostridia, milk powder may be a source of various clostridia. *Cl. perfringens*, *Cl. butyricum* and *Cl. sporogenes* were detected in 14% of a series of 671 samples of milk powder examined by Crossley and Johnson.<sup>75</sup> Milk powder may also be an abundant source of thermophiles.

Ruyle and Tanner<sup>76</sup> examined 559 normal cans and reported that 31, or 5.5%, were unsterile. The bacterial types isolated were as follows: obligate anaerobes, 2; sporing aërobes, 17; facultative thermophiles, 1; non-sporing rods, 9; and micrococci, 15. In a similar investigation Crossley<sup>77</sup> examined 14,365 apparently sound jars of potted meats and fish pastes from various sources. Spore-bearing aërobes or anaerobes were isolated from 12.1% of these samples. Spoilage in meat and fish pastes was generally caused by *Cl. sporogenes* and *Cl. putrificum*, but other organisms, including *Cl. welchii*, were isolated; in canned meats Crossley<sup>62</sup> found that *Cl. sporogenes* is probably the cause of the most serious spoilage.

Most of these defects are amenable to effective control by nisin, as in the case of processed cheese, although the problem is much simpler in the case of these products because the spoilage bacteria often occur in pure culture, or at least free from mesophilic

contaminants, whereas in processed cheese the flora after packaging may be predominantly mesophilic.

## Fish and crustaceans

The bacteriological picture is much the same as with canned meats, and spoilage is attributable mainly to thermophilic clostridia and facultative anaerobes. Nevertheless, mesophilic clostridia occur much more frequently in canned fish products than in canned meat products. A bacteriological investigation by Crossley<sup>77</sup> of the ingredients of potted pastes showed that a significant proportion of the canned tomalley, canned sardines or canned shellfish examined was not sterile and many contained clostridia, mainly *Cl. butyricum* and *Cl. welchii*, occasionally *Cl. putrificum* and *Cl. sporogenes*. This investigation, showed that 30% of all samples of canned salmon contained spores of aërobes and anaerobes. On the other hand, Fellers,<sup>78</sup> in his series of 5,276 cans of salmon of normal commercial quality, found that 3.4% were unsterile. It is interesting to note that more than 50% of the bacilli isolated were facultative anaerobes. Neither of these investigators make any reference to the presence of strict or facultative thermophiles, although Jarvis<sup>79</sup> states that the incidence of flat-sour spoilage in canned fishery products is high. In some cases this is attributable to the inclusion of such fillers as potato and dried egg powder, which may be heavily infected with *Cl. thermosaccharolyticum* or related strains.<sup>51</sup>

The judicious use of nisin would not only eliminate most of the bacterial defects discussed but it would also greatly diminish the spoilage and potential spoilage arising when these products are incorporated in other processed food packs.

## Soups

A great variety of materials may enter into the formulation of soups, and the spices and other condiments, cereals and milk may be heavily infected with mesophilic

and thermophilic spore-bearing bacteria. The bacteriological condition of some of the ingredients has already been discussed under the section on Meats. Thermophiles are particularly troublesome in soups and may cause "hard swells" or "flat-souring," often with protein coagulation.

Milk and milk powder may cause serious contamination of cream soups with thermophiles, and there is a very extensive literature on the subject<sup>80, 81, 82</sup> which testifies to the importance of this aspect and to the losses which can be incurred.

There is now evidence that nisin at a concentration of a few parts per million in canned soups should prove effective with almost every formulation of ingredients in eliminating spoilage due to "flat-sours" and "hard swells."

## Evaporated milk and milk foods

Properly manufactured evaporated milk may not be sterile, but it should contain only the spores of the more heat resistant bacilli and clostridia. Spoilage due to non-sporing bacteria has been reported, but in these cases defects were due either to faulty heat-treatment or defective cans and need not be considered further. According to published reports spoilage appears to be due commonly to aërobic spore-formers, the species being *B. subtilis*, *B. cereus*, *B. coagulans*, *B. megatherium*, *B. calidolactis*, with optimum growth temperatures ranging from 28°C. to 65°C.<sup>83</sup> Some of these organisms cause sweet curdling or bitterness, others cause flat-sours. At present, there is not sufficient information available concerning the sensitivity of all these bacilli to nisin, although there is little doubt that much of the flat-sour spoilage could be prevented. It is interesting, however, to note that Campbell and O'Brien<sup>84</sup> have demonstrated that nisin reduces the thermal resistance of certain spores, among them is *B. coagulans*. Nisin should also eliminate defects caused by anaerobes such as *Plectridium fætidum* (*Cl. sporogenes*) and *Cl. putrificum*,<sup>84</sup> the spores of which are resistant to the

heat-treatments normally used in evaporated milk manufacture.

Canned milk puddings such as creamed rice puddings are products which would benefit from the use of nisin, since both milk and rice may have high thermophile counts. Flat-souring and hard swells can be a serious source of trouble with larger packs of such products.

*In Part 2 of this survey, to be published later, an account will be given of the possible uses of nisin in the preparation of other classes of foodstuffs and in minimising the danger of outbreaks of food poisoning.*

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## Sensitivity of Cheese Starter Cultures to Nisin and Penicillin

Apart from bacteriophage, the principal inhibitors of acid production in a commercial cheese vat are penicillin and nisin according to Barbara P. Keogh, *Aust. J. Dairy Tech.*, 1956, **11** (3), 107. The sensitivity of cheese starter cultures to these two inhibitors has been studied by preliminary bacteriostatic serial dilutions followed by activity tests. At least 30% loss of activity was taken as the criterion of inhibition.

The results of these tests showed that sensitivity to penicillin is not dependent on species since *Str. diacetylactis* strains are distributed among the four groups, and *Str. cremoris* among three. There is not sufficient evidence to confirm that *Str. cremoris* strains are more sensitive than *Str. lactis*.

It is possible to develop by "training" sufficiently resistant starter cultures, but the acquisition of resistance is accompanied by the loss of some other property of the culture. A commercial starter culture claimed to be resistant to penicillin was found to tolerate at least 2 I.U. of penicillin but its rate of acid production was too slow to be of use in cheese manufacture.

The range of sensitivity of different strains is much greater for nisin than for penicillin. The extreme range of inhibitory dilutions of nisin varied by a factor of 200, whilst the factor for penicillin was only 10. It is interesting to note that all the *Str. lactis* and *Str. diacetylactis* strains were in the more resistant groups.

The results indicate that inhibition of starter activity by nisin can be overcome by the selection of the more resistant strains.

► The suitability of foods packed in aluminium foil for radiation sterilisation is one of the topics discussed by Dr. T. H. Angel of Venesta Ltd., in a paper on Aluminium Foil and Foil Laminates, reprinted from the *Journal* of the Institute of Packaging and issued by Venesta. He also covers many other aspects of foil laminates from their specification to their various uses.

► A new edition of *Bulk Flour Handling and Storage* has been published by Henry Simon. Its main feature is an article describing a bulk flour handling and storage plant built for the Wonder Baking Co., Ltd.