

# RECENT DAIRY RESEARCH IN THE NETHERLANDS: DAIRY SCIENCE 1953 - 1958

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## MILK COMPOSITION

The protein percentage of milk has received much attention. In Friesland it was found by Janse *et al.* (1953), that as the butterfat content of milk increased the protein content also tended to increase, but that the increase in protein content was less than the corresponding increase in butterfat content. No correlation could be found between total milk yield and protein content (Janse, 1953).

After Ketelaars (1956) had investigated the heredity of butterfat, protein and S.N.F., Politiek (1957*a,b*) concluded from an investigation of the protein content of milk of individual cows that it must be possible to increase the protein content to a considerable extent by selection in cattle breeding.

Methods for estimating the protein content of milk have been studied and adapted for routine analyses; the province of Friesland started milk payment according to protein content (Lolkema, 1956).

The iodine content of milk was studied by Binnerts (1956), with regard to human iodine requirements.

Holwerda (1954) found that the lowest percentage of citric acid in milk occurred in the months of November, December and January. According to Van Koetsveld (1955) 100 g. of cows' milk contains 10-14 mg. of inorganic sulphate, while colostrum contains 0-9 mg., blood serum 14.7 mg. and human milk 0.5-2.5 mg.

In a series of 6 papers Evenhuis and de Vries (1955, 1956, 1957*a*) discussed the condition of calcium phosphate in milk. According to these authors a colloidal calcium magnesium phosphate is present in raw milk [ratio (Ca + Mg) : P = 1.60]. The protein adsorbed on the phosphate may be assumed to be casein or some other milk protein. The amount of protein adsorbed is only small. On heating milk, a calcium phosphate with a (Ca + Mg) : P ratio of 1.6 (possibly an hydroxy apatite), is precipitated. In connection with this theory the stability of milk was discussed by Evenhuis (1957).

An extreme case of instability of milk, the so-called "Utrecht instability", was studied by Boogeardt (1955) who reported that it was caused

by an abnormal quantity of free calcium ions in the milk.

Van Krefeld and van Minnen (1955) made use of a cation exchange resin conditioned with a mixed salt solution to estimate the calcium ion activity in milk. Compared with total concentrations of 30 millimol of calcium and 5 millimol of magnesium in whole raw milk the calcium ion concentration was estimated to be 2.2 millimol and the magnesium ion concentration 0.8 millimol per l.

Mulder and Koppejan (1953) found that some days after calving, the copper content of the milk is at a maximum and may amount to  $>200\mu\text{g}/\text{kg}$ ., while some weeks later, when the copper content is normal it amounts to about  $40\mu\text{g}/\text{kg}$ .

A new method for studying the composition of the surface layers of milk fat globules in the natural state was introduced by Mulder (1957*b*) who calculated the composition from analyses of milk and the corresponding cream and skim-milk. Of the phospholipids present in milk, about 60% are in the surface layers, which consist of about 30% phospholipids (Mulder *et al.*, 1957).

Using the same method Mulder and Koppejan (1953) investigated the distribution of copper and iron in the milk; about 10-15% of the copper and 25% of the iron are contained in the surface layer of the fat globules.

Pasteurization of cream causes some transfer of phospholipids from the fat globules to the serum (Radema, 1956*b*, 1957).

## LIQUID MILK

In a symposium on the quality of liquid milk, bacteriological defects were discussed by Galesloot (1954), the influence of transport from the dairy to the consumer by Huisman (1954), the treatment of milk in the dairy by Stienstra (1954), and chemical defects by Kruisheer (1954).

General principles for adequate pasteurization of milk were mentioned by Posthumus (1953). After studying the influence of heat-treatment on the keeping quality of pasteurized milk, Galesloot (1953*a*) concluded that provided no reinfection takes place, the milk which was pasteurized at low or high temperatures had better keeping quality than milk

pasteurized at moderate temperatures. If however, post-pasteurization infection took place and the milk was kept at normal room temperature, the keeping quality was best after the milk had been pasteurized at very low temperatures. The post-pasteurization infection was studied in detail by Galesloot (1955*b*) who distinguished between three types of pasteurized milk: (i) milk with phosphatase test just negative, (ii) peroxidase test just negative, (iii) severer heating than (ii).

The same author (1955*c*) reported that if there is no reinfection, pasteurized milk usually deteriorates through sweet curdling. In most cases a re-infection takes place and this causes the milk to deteriorate through souring. The variations of the temperature during in-bottle-pasteurization in a steel cap pasteurizer were measured by Mulder and Broersma (1953).

Sterilized milk is important in the Netherlands and much attention has been paid to this product. Tierie (1953) discussed the bacteriological requirements for sterile milk. According to Galesloot (1956*b*), the U.H.T.S.T. method for sterilization of milk has a smaller effect on certain properties of the milk than other methods which employ lower temperatures.

Meyknecht and van Dam (1956) observed that the keeping quality of commercially low-temperature pasteurized milk was much better than that of high-temperature pasteurized milk (peroxidase reaction negative).

The advantages of U.H.T.S.T. sterilization can only be completely utilized by aseptic packing of milk in sterile containers, but no practical system has been developed. Therefore, after bottling, the U.H.T.S.T. sterilized milk is often subjected to further mild treatment in an "in-bottle sterilizer". For this treatment large continuous in-bottle sterilizers are used and the milk is often bottled at temperatures of 80°C.

Mossel and Drion (1954), recommending that commercial sterilized milk should be sterile, concluded from statistical considerations that for the safety of the consumer, without undue strain for the manufacturer at least 7 samples per batch of sterilized milk should be tested and found sterile. A biological method for the detection of preservatives in milk was devised by Galesloot (1953*c*).

Pette and Hassing (1951) isolated from sterilized milk, obligate thermophilic bacteria which survived heating at 120°C.

According to Pette (1953*b*) a bacteriolytic enzyme is produced by the action of phages on lactic acid bacteria. The symbiotic association between streptococci and phages can be very strong.

According to Galesloot (1956*a*) penicillin has a typical influence on the morphology of yoghurt and starter bacteria which can often be used as a means of discovering the presence of penicillin. Stolk (1955) described how strains of lactobacilli found

in yoghurt form typical globules due to the presence of a phage for the streptococci. Kooy (1951) described the production of yoghurt from cream.

Mulder *et al.* (1953) observed that buttermilk flavour is essentially an oxidized flavour; its development can be prevented by the use of antioxidants.

Kalshoven (1953) found that the results obtained using violet red bile agar plates were in good agreement with those obtained using Netherlands Standard N 955; the latter method is more time consuming.

Galesloot (1956*d*) found that some types of coliform bacteria cannot be counted by the bacto-strip method.

According to Posthumus (1954*a*) chloramin aerosol was not an adequate substitute for steam-sterilization of milk tanks.

The possibilities of providing adequate liquid milk supplies for Calcutta and Madras were discussed by van Dam (1956).

Van Ouwerkerk and van Dam (1956) studied the assessment of costs of milk distribution by retailers.

## BUTTER

Surveying his previous work on butterfat crystallization, Mulder (1953) mentioned that butterfat can solidify into a glass-like state; it can also exist in an unstable form, and it can show a double melting point. The great use which can be made of a basic hypothesis devised in 1938 was demonstrated; according to this hypothesis, butterfat can crystallize in the form of mixed crystals. The solidification temperature rather than the composition of the butterfat determines the temperature of melting. The absolute quantity of solid fat present at different temperatures was discussed by Mulder and Klomp (1956) and by Hannewijk and Haighton (1957). According to the last-mentioned authors, butterfat is not solidified completely before -30°C.

Mulder and Schols (1953) reported that it is possible to make butter with complete exclusion of air, providing the cream is moved with sufficient intensity.

According to Radema (1953) pasteurization of cream gives buttermilk with a lower fat content than pasteurization of milk with subsequent separation and churning.

Mulder with his co-workers Welle (1953) and den Braver (1956*a,b*) tried to fill a serious gap in the theoretical knowledge of buttermaking by devising a theory on the working of butter. During working, butter is forced to flow; a high velocity gradient will promote a breaking up of moisture droplets, a low gradient of velocity a fusion of droplets. In a second paper, the microscopical estimation of the dispersion of the moisture in butter was discussed; in a third the changes in moisture

dispersion caused by the working; and in a fourth paper the applications of the new theory to different working and packaging apparatus was discussed.

According to Mulder (1957a) we have to distinguish between at least four different types of moisture in butter; when discussing the moisture of buttergrains the internal and the external moisture of the grains must be distinguished. Starting from this theory Mulder discussed a number of conditions which influence the moisture content of butter; however, he pointed out that only very little really good experimental work had been carried out although the subject is of the greatest importance.

Several authors paid attention to the formation of aromatic substances in butter. Evenhuis (1953, 1954a,b) reported that the quantity of air present in the churn can be reduced considerably without influencing the formation of aromatic substances. The same author could not demonstrate a direct relationship between oxygen consumption and the formation of substances with a high oxidation-reduction potential. It was concluded that citric acid is probably the fundamental material for the production of diacetyl. De Man and Pette (1956) obtained diacetyl from citrate and pyruvate but not from glucose with suspensions of  $\beta$  Leuconostocs. The same author (De Man, 1956a,b) is of the opinion that in starters, butter, etc.,  $\alpha$ -acetolactic acid is formed by aroma bacteria and then oxidized on decarboxylation if oxygen is present. Kneteman (1952) described a method of enrichment and isolation of *Streptococcus citrophilus* based on the competition of this organism with other lactic streptococci at different temperatures. The growth of aroma bacilli and lactic acid bacteria in different starters was investigated by Holwerda (1955). Van Haften and Pette (1953) concluded that a "cod liver oil" flavour results from the oxidation of pure butterfat, while a fishy flavour is connected with lecithin. Summer butter is said to be much more susceptible to the development of a fishy flavour than winter butter. Koops (1955, 1957) studied cold storage defects in connection with the phospholipids present in butter and described a method for the isolation of the phospholipids from butter serum. The oxidation of phospholipids causes the formation of substances which can give butterfat a fatty, a cod liver oil or a chalky flavour, (Koops & Pette, 1956). Van Duin (1957), isolated from butter, aldehydes originating from phospholipids.

Kruisheer and Krol (1955) again drew attention to the fact that very small quantities of copper can have a detrimental effect on the keeping quality of butter, and concluded from large-scale experiments that it is difficult to fix a standard limit for the copper content of winter butter. According to Mulder and Menger (1957) the natural copper of the milk does not endanger the keeping quality of

butter, while added copper is detrimental even in very small quantities.

According to Tollenaar (1953) many antioxidants mentioned in the literature have little or no value in the prevention of tallowiness although some were found to give good results. As sweet-cream butter never shows cold store defects, large-scale experiments were carried out in which butter was made in winter from sweet summer cream. According to the report of Meyknecht (1956) the butter was of a very good quality. A cultured sweet-cream butter of high quality was made by cutting sweet butter into slices and churning it with ripened cream or skim-milk. The micro-structure of both products differed in some cases from that of normal butter, although the consistency was good.

In large-scale experiments on the retail packaging of butter, aluminium foil gave better results than vegetable parchment paper + carton under all the conditions studied. The advantages of aluminium foil were, however, doubtful unless packaging was carried out very carefully and the butter was of excellent quality (van den Berg, 1952; Grouwstra & van den Berg, 1954).

The carotene, vitamin A, cholesterol and tocopherol contents of Netherlands butter was studied by Kruisheer (1955, 1956) and Kruisheer and den Herder (1953a,b). No distinct antioxidant activity of tocopherol could be detected.

A method for the detection of adulteration of butter by margarine, based on the examination of sterols, was described by den Herder (1955).

Seasonal variations in the fatty acid composition of Netherlands butterfat were studied in detail by Stadhouders and Mulder (1955, 1956a) and Stadhouders *et al.* (1956), using a chromatographic method (rubber column) for the saturated fatty acids and a spectrophotometric method for the unsaturated acids.

Radema (1956a) described the use of carotene for colouring butter.

#### CONCENTRATED PRODUCTS

In a more general paper on U.H.T.S.T. sterilization methods, Galesloot (1957a) stresses that it is at present very difficult to come to a well-founded conclusion on the suitability of U.H.T.S.T. sterilization for evaporated milk.

Evenhuis and de Vries (1957b) concluded from their experiments that a continuous high vacuum cooler could be introduced in the manufacture of sweetened condensed milk by using a special lactose crystallization method.

Fat lenses on condensed milk were described by van Kreveld (1953).

The effect of storage temperature and moisture content on the keeping quality of dried whole milk was studied by Radema (1954), while differences in quality between roller-dried and spray-dried milk

were discussed by van den Berg (1956) who reported in another paper (1952) on practical experiments on packing full cream roller-dried milk in Multiwall paper bags.

The importance of gas inside the particles of spray-dried milk was discussed by Rutgers (1957) in relation to gas packing, and methods were mentioned for its estimation (residual gas). Gerlisma (1957) reported observations on the causes of insolubility of milk powder.

According to Galesloot (1955*d*) increased plate counts of spray-dried milk powder are caused by *Str. durans*, *Str. thermophilus*, *Bacillus cereus* and, to a smaller degree, by *Micrococcus caseolyticus*, *Str. bovis* and *Str. faecalis*.

### CHEESE

A general survey of the ripening of cheese was given by Pette (1953*a*). Stadhouders and Mulder (1953, 1957) made it clear why many investigators had had little or no success in trying to isolate "ripening bacteria" from well-ripened cheese. Most of the organisms occurring in milk die in the cheese; they leave behind enzymes which contribute to the ripening of the cheese. This conception resulted from experiments on fat hydrolysis in cheese, which is caused by milk lipase and enzymes from micro-organisms occurring in the milk. Active lipases are not produced in the cheese during ripening. The same authors stress the importance of a thorough study of the surface flora of hard cheese such as Dutch cheese, as the flora affects the ripening.

Raadsveld (1953) isolated from bitter tasting cheese a bitter substance which could not be identified.

The presence of lypolytic bacteria on the surface of Dutch cheese was studied by Stadhouders and Mulder (1956*b*).

From experiments with cheese made on an industrial scale, Raadsveld (1957*b*) concluded that Dutch-type cheese contain only traces of galactose and glucose and that these traces disappear within a few days.

Pette (1955) isolated bacteria producing hydrogen sulphide from cheese and proposed the name *Str. faecalis* var. *malodoratus*, on account of the smell. The defect is prevented by keeping the pH below 5.4.

The composition of rapidly souring cheese starters was studied by Kooy (1951); the strains used influenced each other. De Man (1956*d*) studied in detail the properties of 46 strains of lactobacilli isolated from cheese. He found no indication that lactate can be fermented by lactobacilli. The normal formation of gas in Dutch cheese is probably caused by betacocci and possibly lactobacilli fermenting citrate.

Raadsveld (1951) showed that the rheological properties of cheese change considerably on storage

at 30°C, the cheese becoming more brittle. It is supposed that an accelerated proteolysis causes this change in consistency.

Much work was done in the field of antibiotic-producing bacteria in cheese. Galesloot and Pette (1957) described a method for estimating the nisin content of antibiotic starters and cultures and of cheese made by means of antibiotic starters.

Galesloot (1957*b*) mentioned the production of a nisinase by certain streptococci. The same author (1957*c*) showed that it is of little use to use antibiotic starters if raw milk containing large numbers of group N streptococci has to be used. On the other hand it is possible to make starters which are not sensitive to nisin.

A co-operation between nitrate and xanthine oxidase is necessary, according to Galesloot, for the prevention of butyric acid fermentation in Dutch cheese (1956*f*).

Galesloot (1957*c*) and Galesloot and Pette (1956) investigated the influence of nisin produced by antibiotic starters on the growth of micro-organisms which may be involved in bacteriological processes in cheese and processed cheese. The same author reported that the nisin production of daily transferred antibiotic starters sometimes decreases rather suddenly. A starter used in practice in the dairy industry has to be checked periodically.

In Gouda and Edam cheese, made from pasteurized milk, no eyes are formed when antibiotic starters are used. This is caused, according to Galesloot and Pette (1957), by the sensitivity of betacocci as well as of propionic acid bacteria for antibiotic starters, but as is to be expected, it was quite possible to make a cheese of good quality using antibiotic starters. Antibiotic starters had some retarding action on the ripening of the cheese (Galesloot *et al.*, 1957). The cause of this retarding action was thought to be due to the fact that antibiotic starters do not contain aroma bacteria. Galesloot (1956*e*) reported on a taste defect of Gouda cheese which had not been described before.

At a symposium on the utilization of whey (1952) Blaauw discussed: the technology and economics of the processing of whey; van Dam (1952) the processing of whey at a co-operative dairy factory; van der Molen and Kapelle (1952) the use of whey paste and whey powder in the preparation of silage; Wegelin (1952) the proteins of milk whey; Wöstmann (1953) whey proteins in bread; Wieringa (1952) a general discussion on the technical utilization of whey; Dammers (1954) an investigation of the value of whey powder and dried whey albumin in rations for fattening pigs.

Experiments on the treatment of the surface of cheese were carried out by Blaauw (1957), while Radema and Raadsveld (1957) carried out experiments with emulsions of plastics.

The results of the analysis of an Edam cheese coming from the depot of the Scott expedition, were published by Raadsveld (1957c).

Blaauw (1956) made large-scale experiments on the cutting and stirring of the curd.

### NUTRITIONAL VALUE

The resorption of milk fat was studied by Ten Bokkel Huinink (1953), especially with children suffering from celiac disease.

Riboflavin, nicotinic acid, pantothenic acid and biotin in the milk of Netherlands cows were assayed by Hoeflake (1953) over one whole year. According to Koetsveld (1953) the vitamin  $B_{12}$  level of morning milk was appreciably lower than that of evening milk (resp. 10.98 and 9.37 g./l.). Engel (1955) estimated the vitamin *D* content of condensed, evaporated and irradiated milk and discussed the possibility of increasing the vitamin *D* content of milk by irradiation. Frens (1955) considered that low vitamin *A* potency of butter in winter is caused by a wrong feeding technique.

A growth retarding effect in rats fed rations containing large quantities of lactose (even if in the form of dried milk) was observed by Groot (1956), Groot and Engel (1957), Groot and Hoogendoorn (1957). This effect may be primarily ascribed to the action of the disaccharide as such; it is markedly decreased by substituting a mixture of glucose and galactose for the lactose. The detrimental effects of lactose consumption after weaning may be primarily ascribed to the action of the disaccharide as such in the intestine, from which it disappears extremely slowly.

Nutritional differences between butterfat and vitamin-enriched vegetable oils were investigated by Nieman (1954).

### ANALYSIS

Mulder and van der Have (1956), in a preliminary report compared a number of methods for estimating the protein content of milk. The method of Steinsholt for the estimation of the protein content was described by Raadsveld (1957a). In Friesland the method of Kófranyi was used as a routine method for estimating the protein content of very large numbers of milk samples (Lolkema, 1957).

The calcium and the magnesium content of milk were estimated by Van der Have (1954) in an accurate, simple and rapid way by a complexone titration method. Raadsveld (1956) described a more simple complexometric method for the estimation of calcium in milk.

A flame photometric method for the estimation of sodium, potassium and calcium in milk and cheese, which gave good results, was described by van der Have and Mulder (1957).

Van Kreveld (1954), described a method for estimating the calcium ion concentration in milk, based on the equilibration of milk with ion exchangers, while Smeets (1955) gave a number of methods for estimating the calcium ion concentration in milk; the purpurate method gave satisfactory results.

According to Radema (1955) the method of Jones for the determination of fat in dairy products is not very accurate, but for certain special cases in which the fat content is low, it may be suitable.

Mulder and Meyers (1953) described a method for estimating the butterfat content of milk and dairy products; in this method the milk is boiled in a Mojonnier tube with hydrochloric acid after which the fat is extracted with petrol ether. The SBR method gives results which are somewhat too high, according to Mulder and Hoogendijk (1956). A butyrometric method for the estimation of fat content of gruels and custards was devised by Brus and van den Berg (1955) who used a specially constructed syringe for measuring the product.

Nuyten (1953) composed tables giving the percentage of total solids and S.N.F. of milk, skim-milk, buttermilk, yoghurt and whey.

Kruisheer and Eisses (1956) reported on the use of the Meyhuizen apparatus for estimating the moisture content of dried milk.

A routine method for the estimation of copper in milk and in dried milk was described by Krol and den Herder (1955).

The air content of butter was calculated by Posthumus (1954b) from the specific gravity of the butter which was determined aerometrically.

Van Ginkel and Hamelink (1953) distinguished between normal butter and whey butter by making use of the renneting properties of whey butter serum. Den Herder (1955) described the method used by the Government Dairy Station at Leiden for the detection of adulteration of butter with margarine; the method is based on the examination of the sterols.

An almost complete analysis of the fatty acid composition of Netherlands butterfat was made by Stadhouders and Mulder (1956a) who used the spectrophotometric method of Brice and Swain for the estimation of unsaturated fatty acids, and the chromatographic method of Bolding (rubber column) for the saturated acids. One of the fraction collectors was described in detail by Stadhouders *et al.* (1956).

The specific rotation of lactose was studied by Ter Horst (1953).

A simple microbiological test for the detection of preservatives was reported by Mossel and Mandersloot (1953), who used as a medium milk enriched with 2.5% glucose and inoculated with bakers yeast. Mol and de Haan (1956) described a method for the estimation of hypochlorites in raw milk.

Galesloot and Pette (1957) described a method for estimating the nisin content of milk, cheese, etc., and expressed this content in Reading units. Various methods for the detection of penicillin in a number of dairy products were described by Galesloot (1955a). Adamse (1955) reported his experiments using lactic acid bacteria for the examination of milk containing penicillin.

Stoutjesdijk (1956) reported that the method of reconstitution influences the plate count of milk powder, while Labots (1956) mentioned the influence of traces of copper on the estimation of the plate count.

According to Galesloot (1953b) the use of rennet in the methylene blue reduction test has no advantages.

The following is a list of standards for the analysis of milk and dairy products which was published in English in the Netherlands Milk & Dairy Journal:

- N 958 (1953) The determination of the density of milk at 20°C with a lactodensimeter.  
 N 1616 (1953) The calculation of the percentage of total solids.  
 N 913 (1953) The determination of the titratable acidity (°N) of whole milk, "standardized liquid milk", partly or completely skimmed milk, buttermilk, acidified partly or completely skimmed milk, and whey.  
 N 1358 (1954) The determination of the density of buttermilk, acidified skimmed milk and "yoghurt milk" at 20°C with a lactodensimeter.  
 N 1381 (1954) The peroxidase test (Storch's test).  
 N 1507 (1954) The plate count of bacteria in milk.  
 N 1672 (1954) The short butter trier.  
 N 1673 (1954) The long butter trier.  
 N 1674 (1954) The broad cheese trier.

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