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The water agar test: a new test to measure the bacteriological quality of cream

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SUMMARY

A new test, the water agar test, is described that gives a qualitative index of the presence of bacteria that indicate contamination of the cream, poor storage conditions or both of these factors. The method is simple and requires little equipment. The bacteria grow in a film of diluted cream adsorbed on the surface of a non-nutrient base. After incubation at $30 \pm 0{\cdot}5°$ C. for 18–20 hr., a proteolytic and mucoid colony count is obtained which has the same percentage coefficient of variance as a standard plate count.

An examination of the effect of storage at different temperatures on the types of bacteria present in cream showed that of all the tests done initially, only the water agar test could predict subsequent bacterial growth with any consistency. The multiplication of presumptive coliform organisms occurred even at 3·5° C. Irrespective of the colony count, the methylene blue reduction time was not shorter than $7\frac{1}{2}$ hr. unless the bacteria were in the logarithmic phase of growth when sampled.

A survey was made of the bacterial flora of 188 retail samples of double cream of 15 different brands. The age of the samples varied from freshly separated cream to cream that had been kept in the shop for a day longer than that recommended for sale. The water agar test was compared with the colony count, the presumptive coliform test, a confirmatory coliform count in violet red-bile agar, a lipolytic colony count, a staphylococcal count and the methylene blue reduction test.

INTRODUCTION

There are, at present, statutory biological standards for raw and pasteurized milk but not for cream. This illogical situation with regard to cream has developed partly because of the relatively small demand for the product, but mainly because some authorities have failed to realize that a dye reduction test is far from ideal as an indication of the bacterial content of milk produced under modern conditions, and are not prepared to accept the time-consuming, relatively costly plate count or coliform test as standards. Contributory causes of this illogical situation have been the equating of the possible hazard to public health arising from post-pasteurization contamination with off-flavours and the fact that samples may vary considerably in age at testing. The relation between hygiene and off-flavours was probably true in the past, when dairy hygiene was poor, but is not so under

modern conditions of production. The relation between the initial bacterial content of cream and the development of off-flavours will be considered in another paper.

The demand for a rapid, simple bacteriological test for milk, resulted in the introduction of the methylene blue reduction test in England and Wales about 40 years ago (Milk (Special Designation) Order 1936). At that time it had been found that dye reduction gave more reproducible results than either the colony count or the presumptive coliform count. It had been shown that dye reduction was a measure of the number of metabolically active bacteria (Hobbs, 1939). Since, at that time, dairy products were rarely cooled below 10° C., the largely mesophilic bacterial population was able to remain in a metabolically active state, thus giving a good correlation between reduction time and the colony count. The use of 37° C. as an incubation temperature for the test, was based on the assumption that, since human pathogens grow well at this temperature, only that part of the bacterial flora capable of growth at 37° C. signified a hazard to public health. It was recognized that mesophilic organisms may have a longer lag phase of growth than normal after being held at 10° C. or lower and that this could cause anomalous results. Preliminary incubation has been used in an attempt to eliminate the long lag phase, as for example in the temperature-compensated resazurin test (Department of Health for Scotland Memo, 1955) although it had been shown that the results had little relation to the initial bacterial content (Higginbottom, 1941).

In 1958 a working party of the Public Health Laboratory Service (Report, 1958) recommended the use of the methylene blue reduction test only as an advisory test for cream, since a significant number of anomalous results was recognized. The introduction of chemical sterilization of dairy equipment and the increasing use of refrigerated storage on the farm, at the dairy and in the home, had so altered the bacterial environment that a selective change had been produced in the flora. Hence, contamination by mainly mesophilic, acid-forming bacteria capable of dye reduction was replaced to a large extent by psychrotrophic, proteolytic organisms that were poor dye reducers and often unable to grow at 37° C. (Thomas, Druce, Davis & Bear, 1966). Under certain circumstances it has been shown that the presence of very large numbers of psychrotrophs may cause significant dye reduction (Morris, 1941; Jezeski & Macy, 1946) but observations made over many years have shown that dye reduction tests are not suitable for the detection of psychrotrophs (Chilson & Collins, 1940; La Grange & Nelson, 1965). It follows that the reappraisal of methylene blue reduction as a test for cream, made by a second Public Health Laboratory Service working party (Report, 1971) also found anomalous results. These workers recommended that the test should be used for screening or advisory purposes only, but they did suggest that the anomalies might have been removed if a lower incubation temperature had been used, since the presence of pseudomonads (psychrotrophic organisms) was recognized.

The hygienic quality of cream may be defined as the measure of those bacteria that have been added directly by a human agent or indirectly either by failure to clean equipment adequately or by permitting bacterial contamination to occur.

Tests are only of use if they consistently indicate that contamination has occurred. Under modern dairy conditions, poor hygiene is most consistently demonstrated by the presence of psychrotrophs. These organisms may not grow at 37° C., do not form acid from lactose or reduce dyes strongly, but they usually grow at 30° C. and are often lipolytic or proteolytic. During the 1920s, Ayres & Mudge (1920) and Zoller (1923) used a proteolytic count to estimate the hygienic index of milk. The water agar test that is used in the present work is a refinement of this concept and, by using a film of milk as both growth medium and indicator, can give results within 24 hr.

MATERIALS & METHODS

Cartons or jars of double cream, either pasteurized or ultra-high temperature (U.H.T.) treated, were obtained from retail sources or directly from the dairy by Ayr Burgh Sanitary officers and delivered to the Institute by 10 a.m. with a history of the sample. The samples were examined immediately for bacterial content and the remainder subdivided into 4–5 oz. quantities, stored at various temperatures (3·5°, 5·0°, 10·0°, 15·0° and 22·0° C.) and examined periodically for changes in the bacterial composition. The age of the samples varied from freshly separated cream to cream that had been kept in the shop for a day longer than that recommended for sale.

A survey was made of the bacterial content of 188 retail samples from 15 brands of cream. All the creams were prepared from tanker milk.

The samples were examined by a plate count, presumptive coliform test, lipolytic and staphylococcal counts after incubation at $30 \pm 1°$ C. for 72 hr.; a confirmatory coliform count was done in violet red-bile agar (VRB) after incubation at $30 \pm 1°$ C. for 24 hr. using British Standard methods (British Standard 4285: 1968; Supplement No. 1 (1970)). The Statutory English methylene blue test was also done (Statutory Instrument 1571: 1963). A phosphatase test was done to confirm the efficiency of the pasteurizing process.

The medium used for the water agar test was 1·3 % Davis New Zealand agar in distilled water. The bacteria grow in the film of diluted cream adsorbed on the surface of this non-nutrient base (Taylor, 1967, 1971).

In this work 9 cm. plastic disposable petri dishes were used. A supply of water agar plates were poured and the agar allowed to solidify. About 10 g. of silica gel (coarse grade, self-indicating) was placed in the lid of a petri dish as a single layer of granules, and the agar plate inverted over it until the cobalt chloride indicator turned from blue to pink (usually 4–6 hr.). The silica gel was then removed and re-dried. The water agar plate was ready for use on the following day. In an emergency, the plates were used on the day of drying but the zones of clearing were always much smaller than usual. Any moisture of condensation on the lids of the plates to be inoculated was removed using one or more 9 cm. No. 1 Whatman filter papers. If left in the petri dishes, this moisture interfered with the test, proteolytic zones failing to develop. The filter papers did not have to be sterilized, but any box that was used for this work was used for no other purpose.

A 1/10 dilution of the cream was made using a sterile 10 % solution of Oxoid

skim milk powder. The dry water agar plates were inoculated by completely flooding the surface of the agar with 2–4 ml. of the well-mixed cream dilution. There was no need to measure the quantity of the dilution, provided the surface area of the plate was constant for all samples. After flooding, the plate was quickly inverted to allow the surplus fluid to drain into a beaker or other receptacle. The plate was then replaced in its lid, where any further surplus fluid was absorbed by a fresh 9 cm. filter paper in the lid. Duplicate plates were inoculated and incubated for 18–20 hr. at 30° C., at which time the number of colonies surrounded by a clear zone and mucoid colonies were counted. The presence of mucoid colonies was confirmed after incubation for 48 hr. For experimental purposes, further 1/10 dilutions were made so that counts greater than 300 could be obtained, but this was not necessary for a routine test.

The colonies surrounded by a clear zone are termed 'proteolytic' (Plate 1). This term is used descriptively rather than scientifically since lipolysis is probably also involved. Those enzymes that attack the casein can be distinguished from those that form alkali by placing a little acetic acid in the lid of the petri dish, and allowing the vapour to neutralize the alkali and reprecipitate the casein. Occasionally, an organism appears to attack the fat globule membrane and the colony is surrounded by a ring of free fat globules within the clear zone. The proteolytic rennet enzymes form rings of precipitation not clearing (Cheeseman, 1963). The mucoid colonies are quite characteristic (Plate 1) and flow when the plate is tilted.

The standard deviation and percentage coefficient of variance for the water agar test was calculated using the counts obtained by three workers (other than the author) who each prepared 30 plates from each of 3 samples.

RESULTS

The storage of freshly produced cream at different temperatures gave rise to two basic patterns of bacterial growth which were largely independant of the initial colony count within the range 10^2–10^5 colonies/ml. of cream. The pattern was most closely associated with the method for producing the cream, one being associated almost entirely with pasteurized cream and the other with cream that had been separated from pasteurized milk. Bacterial growth at 22° C. was too rapid for differences to be shown between the two patterns which could be most conveniently shown by comparing growth curves at 15 and 5° C. The growth curves obtained at 10 and 3·5° C. were similar to those obtained at 15 and 5° C. respectively but the changes were less rapid.

The first pattern (Fig. 1A) showed a prolonged lag phase at 5° C. which was rarely shorter than 10 days and even at 15° C. growth was relatively slow. The proteolytic count was less than 5, presumptive coliforms were normally absent in 1/10 ml. at the initial test, the methylene blue reduction time was greater than $7\frac{1}{2}$ hr. and the colony count varied between 10^2 and 10^5 colonies/g. With this pattern of growth, presumptive coliforms were rarely detected during incubation, sporeformers forming the major bacterial group.

The second pattern of growth (Figs. 1B, C, 2A) showed little or no lag phase at

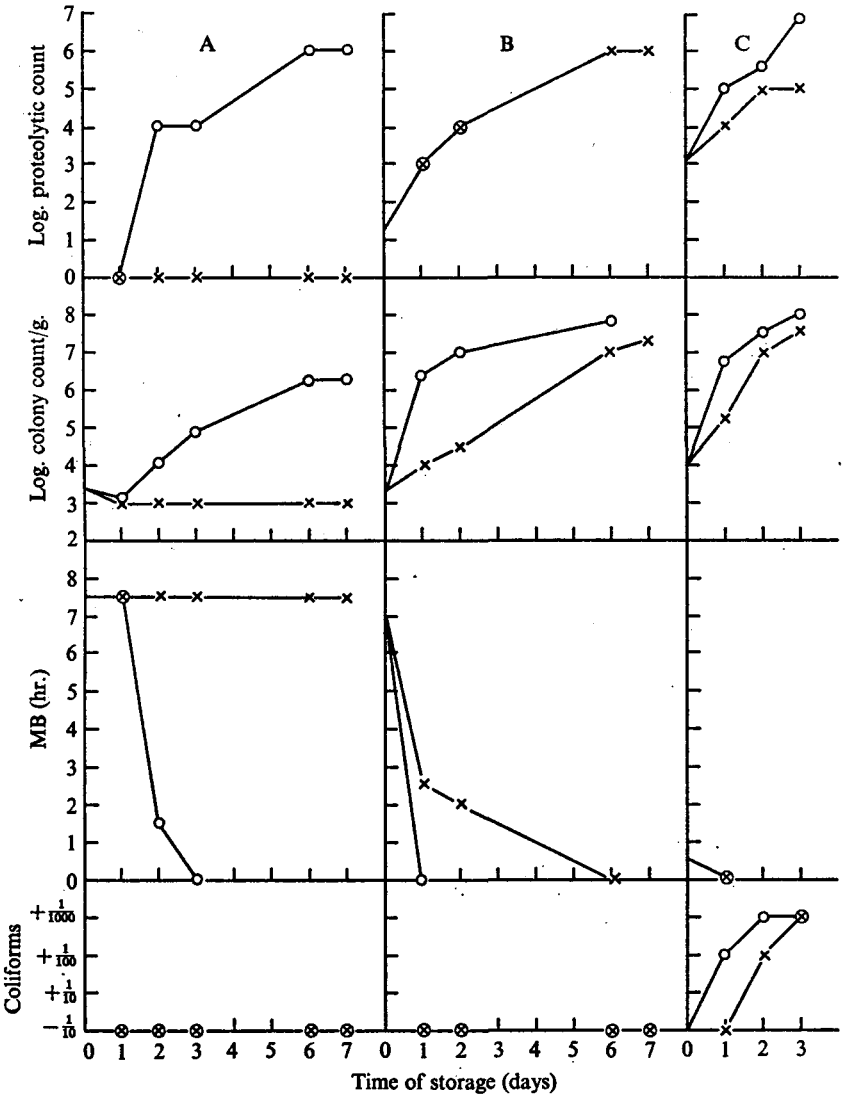

Fig. 1. The relation between the proteolytic count of the water agar test, the colony count, the methylene blue reduction test (MB) and the presence of presumptive coliforms in creams A, B and C when stored at 5° C. (× — ×) and 15 °C. (○—○).

5° C. and growth at 15° C. was more rapid than in the first pattern although the initial colony count varied between the same limits as for the first pattern. This pattern of growth occurred in the presence or absence of presumptive coliforms at the initial test; these organisms might develop during incubation (Figs. 1C, 2A) although apparently absent in the initial sample. If freshly separated cream was examined, a proteolytic count of less than 5 could be obtained (Fig. 2A) but when the sample was obtained at the time of delivery to the retail shop, proteolytic organisms could be detected even when the colony count was low (Figs. 1B, C, 2B). The methylene blue reduction time for the initial sample showed a very

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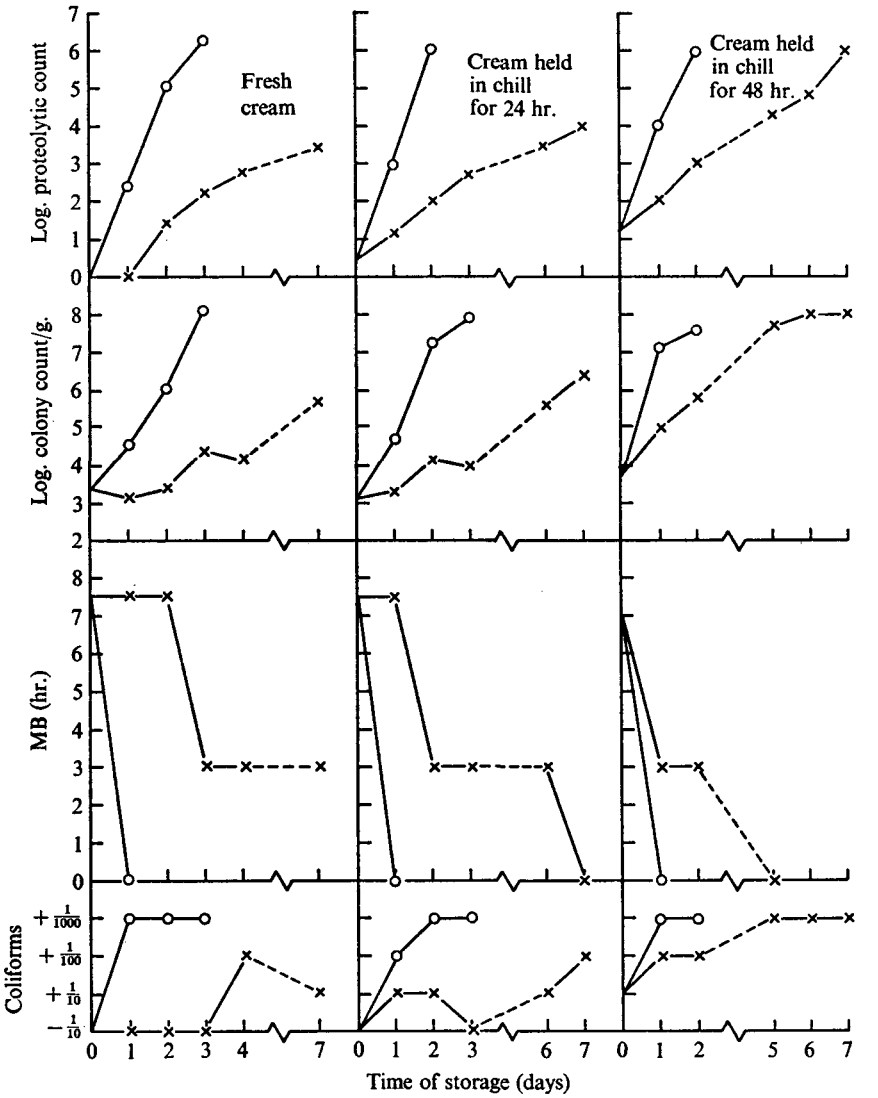

Fig. 2. The effect of holding cream in a commercial chill-room for 24 and 48 hr. on the bacterial flora during further storage at 5° C. (×—×) and 15° C. (○—○).

large variation which could not be related to the types of bacteria that were present, or predict the rate of bacterial growth. A long reduction time could be obtained in samples where sporeformers were the predominant organisms (Fig. 1A) or a very short time when these organisms comprised only a minor fraction of a relatively low colony count (Fig. 1C).

The reaction of the two types of creams towards refrigerated storage was also highly characteristic. If pasteurized cream was stored at 5° C. for 7 days before being incubated at other temperatures, the only difference from the growth curves observed for the fresh cream was that there was a very slow increase in the colony count at 5° C. However, if post-pasteurization contamination had occurred, the

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Table 1. *The distribution of the colony count, presumptive coliform test, violet red-bile agar and lipolytic counts, proteolytic and mucoid colony counts in the water agar test and the methylene blue reduction time for different brands of cream*

Brand	No. of samples	Colony count/g. $<10^3$	10^3–$<10^4$	10^4–$<10^5$	$\geqslant 10^5$	Coliforms absent in $\frac{1}{10}$ ml.	Violet red-bile agar <50/g.	Lipolytic <50/g.	Water agar proteolytic + mucoid <5/g.	Methylene blue (hr.) $4\frac{1}{2}$–7	Methylene blue (hr.) $\geqslant 7\frac{1}{2}$
1	18	0	1	7	10	8	11	2	5	2	4
2	27	0	12	6	9	12	12	4	8	4	9
3	6	6	0	0	0	6	6	6	6	0	6
4	22	5	14	1	2*	22	22	17	16	5	17
5	27	6	6	4	11	14	9	3	3	1	4
6	21	2	7	4	8	14	13	7	5	7	3
7	1	1	0	0	0	1	1	1	1	0	1
8	22	5	11	1	5	15	14	6	14	0	10
9	6	0	1	3	2	4	4	0	1	3	1
10	1	0	0	0	1	0	0	0	0	0	0
11	4	0	4	0	0	4	4	4	4	1	3
12	7	0	2	2	3	3	2	0	0	0	2
13	16	1	9	3	4	9	12	6	4	4	6
14	1	1	0	0	0	1	1	0	1	1	0
15	9	9	0	0	0	9	9	9	9	0	9

* Pre-incubated.

Table 2. *The relation of the proteolytic and mucoid colony counts of the water agar test to the colony count at 30° C., the presumptive coliform test, the coliform count in violet red-bile agar, the lipolytic count and the methylene blue reduction test for 188 cream samples*

Water agar counts/g.		No. of creams	Colony count/g.	Presumptive coliforms/ml.	Violet red-bile agar count/g.	Lipolytic count	Methylene blue (hr.)
Proteolytic	Mucoid						
<5	<5	79	<50–$110{,}000$	$-\frac{1}{10}$	<50	<50–$68{,}000$	$1->7\frac{1}{2}$
5–50	—	23	150–$1{\cdot}1\times10^6$	$-\frac{1}{10}$ to $+\frac{1}{10}$	<50–300	<50–$850{,}000$	$1->7\frac{1}{2}$
51–300	—	21	400–$32{\cdot}4\times10^6$	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–$4{,}900$	<50–$204{,}000$	$0->7\frac{1}{2}$
>300	—	56	2300–810×10^6	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–29×10^6	<50–114×10^6	$0->7\frac{1}{2}$
<5	>5	9	1550–103×10^6	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–75×10^6	<50–4×10^6	0–6

Table 3. *The relation of the methylene blue reduction test to the colony count at 30° C., the presumptive coliform test, the coliform count in violet red-bile agar, the lipolytic count and the proteolytic and mucoid colony counts of the water agar test for 188 cream samples*

Methylene blue (hr.)	No. of creams	Colony count/g.	Presumptive coliforms/ml.	Violet red-bile agar count/g.	Lipolytic count/g.	Water agar counts/g.	
						proteolytic	mucoid
0	51	6600–163×10^6	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–39×10^6	50–18×10^6	<5–10^7	<5–4×10^6
$\frac{1}{2}$–4	34	400–47×10^6	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–$6{\cdot}8\times10^6$	<50–$807{,}000$	<5–$550{,}000$	<5–$1{\cdot}4\times10^6$
$4\frac{1}{2}$–7	28	350–$1{\cdot}1\times10^6$	$-\frac{1}{10}$ to $+\frac{1}{1000}$	<50–$4{,}000$	<50–$620{,}000$	<5–$7{,}900$	<5–150
$\geqslant7\frac{1}{2}$	75	<50–$110{,}000$	$-\frac{1}{10}$ to $+\frac{1}{100}$	<50–$1{,}900$	<50–$57{,}000$	<5–$3{,}900$	<5–3000

second pattern of growth resulted (Fig. 1B). Cream that had been made from pasteurized milk was much more sensitive towards storage especially under commercial conditions where there was even a small cyclic variation in the storage temperature. In the example illustrated (Fig. 2A, B, C), the temperature of the chill-room was 4·4° C. (40° F.) when opened at 6·0 a.m. There was a constant rise in temperature of 1·1° C. (2° F.) per hr. until 2.0 p.m., when the temperature started to fall at the same rate to 6·7° C. (44° F.) when the chill-room was closed for the night at 6·0 p.m. Although there was no appreciable alteration in the colony count or the methylene blue reduction for cream that had been held in the chill-room for up to 48 hr., there was a progressive increase in the proteolytic count and presumptive coliforms were present in 1/10 ml. after 48 but not 24 hr. Incubation subsequent to storage for 24 and 48 hr. periods in the chill-room showed that the lag phase of growth at 5° C. was eliminated after the 48 hr. storage period. The proteolytic and colony counts and the incidence of presumptive coliforms increased more rapidly with the length of prior storage in the chill-room. The methylene blue reduction time showed no significant alteration until the logarithmic phase of growth was established, thereafter the decrease in reduction time was not usually linear.

A phosphatase test was done on all the samples that were examined. All the initial samples gave a zero reading. There was a progressive increase in the reading for samples of cream in which sporeformers were the major bacterial type when incubated at 15° C. but not at 5° C. A colony count of 10^4–10^6/ml. sporeformers gave a phosphatase reading of 6–10 μg., 10^7 sporeformers a reading of 10–18 μg. and more than 10^7 sporeformers a reading of 18–25 μg. *p*-nitrophenol/ml. milk. There was no such increase in the phosphatase reading when sporeformers were present as less than half of the colony count.

The survey of retail samples of pasteurized double cream showed that, with the exception of one sample of uncertain origin, all the producers were capable of producing fresh cream with a very good bacteriological standard from tanker milk. However, the frequency with which good samples were obtained depended mainly on the producer, but was also affected by the method of transport to the retailer and the conditions of retail storage (Table 1). The cream brands 4 and 6 were made by the same producer. Brand 4 was transported and stored under excellent conditions and the bacteriological quality of the cream was good even on the last day for sale. Less care was used during the transport of brand 6. This brand was sold at two shops only, one of which had good storage conditions and stock management. There was little difference in the bacteriological quality of the cream obtained from these two shops. Brand 11 was an U.H.T.-treated frozen cream.

A well-produced cream usually had a colony count of not more than 10^4 colonies/g., a presumptive coliform test negative in 1/10 ml., a violet red-bile agar (VRB) and lipolytic counts of less than 50/g., a proteolytic and mucoid colony count in the water agar test of less than 5 and a methylene blue reduction time of not less than 7½ hr. The variation between the results of these tests and the results of the water agar test is given in Table 2. There was a general deterioration in the bacteriological standard as the proteolytic colony count increased, and the mucoid

colony count indicated the probable presence of coliforms. Mucoid colonies were usually formed by Gram-negative rods, but occasionally also by sporeformers. However, there was a considerable variation between the other tests within any one class of the water agar test. For example, when the proteolytic and mucoid colony counts were both less than 5, the colony count was usually less than 10^4 but varied from less than 50–10^5. The presumptive coliform test was negative in 1/10 ml. and the VRB count was usually less than 50 but occasional low counts were obtained. The methylene blue test was usually not less than $7\frac{1}{2}$ hr. but two creams failed this test that were apparently of good bacteriological quality. The presence of lipolytic colonies had no obvious relationship to any of the other tests. There was some evidence of the suppression of gas formation in the presumptive coliform test in a few poor-quality creams.

The methylene blue reduction test showed a moderate agreement with the other tests that were investigated. In general, when the MB test was 0 hr. the colony count was raised, coliforms, lipolytic and proteolytic organisms were present; when the test was not less than $7\frac{1}{2}$ hr., the bacteriological quality of the cream was good. There was little difference in the distribution of results for creams which reduced methylene blue in $\frac{1}{2}$–4 hr., and those that reduced the dye in $4\frac{1}{2}$–7 hr. There was considerable overlap of results between groups when the creams were classified on the basis of the methylene blue reduction time.

Statistical analysis of the reproducibility of the water agar test

The method of analysis was to calculate a set of anovars removing the variance due to samples, readers and the sample × readers interaction. The five anovars in the set used the counts from (1) the first 2 plates inoculated by each worker from each sample, (2) the first 5 plates, (3) the first 10 plates, (4) the first 20 plates, and (5) all 30 plates. The standard deviations and coefficients of variation obtained were:

No. plates	S.D. (±)	C.V. (%)
2	22·2	12·6
5	20·0	11·6
10	20·8	11·7
20	19·4	10·7
30	20·5	11·5

The slight inconsistency of these results is probably due to the systematic selection of the plates used in each analysis. However, they clearly show a slight advantage in counting 5 plates rather than 2, but no additional improvement by counting more than 5 plates.

In all five anovars the mean differences between the three samples were highly significant. The mean counts for the samples were 72, 110 and 354. The mean differences between the three readers were not significant in the anovar of the two-plate data.

Using the results from the 30-plate anovar, an estimate can be obtained of the number of randomly selected plates to give a sample mean which is within a certain

limit of the true or population mean. Some estimates are given below, L being the deviation of the sample count from the true count and n the required number of plates. Thus a sample of two plates should, on average, give an estimated count within ± 30 of the true count.

L ($\pm$)	n
5	67
10	17
15	8
20	4
25	3
30	2

DISCUSSION

The original purpose of statutory bacteriological standards was to eliminate possible hazards to public health. Modern methods for food processing have improved so much that these tests are more usually employed to ensure that the product will remain palatable for a reasonable length of time after purchase. When cream is considered, there are two main difficulties in establishing a bacteriological standard. The first is that the cream may have been kept under very different conditions for varying lengths of time before testing. The second is that cream is an excellent bacteriological growth medium. Even when pasteurized cream is considered, it is not helpful that there are two definitions of pasteurization. Milk must be heated at 71·7–78·3° C. for more than 15 sec. before the cream is separated, but cream separated from milk which has not been heat-treated must either be heated at not less than 74° C. for more than 15 sec. or momentarily at not less than 80° C. As a result, pasteurized cream has usually been heated to a higher temperature than cream made from pasteurized milk.

In Great Britain, the colony count, the presumptive coliform test and methylene blue reduction are the three tests that are statutorily used for milk. Until more precise conditions are defined for the manufacture and storage of cream, the colony count even of the freshly produced product is only a very rough guide to the conditions under which the cream was produced and maintained. A well produced cream will have a colony count of 10^3 colonies/g. or less but the count may be considerably higher if the original milk has been contaminated with thermoduric organisms. Even under optimal conditions, a cream produced from a raw milk with a high bacterial content will have a higher colony count than a cream produced from clean milk. The presumptive coliform test was originally developed to detect faecal contamination in water (Report, 1939). *Escherichia coli* does not survive for long in water and therefore a positive test indicates recent contamination. However, when used to assess the bacteriological safety of foods, Mossel (1974) states that the presumptive coliform test underestimates the health risk which may be caused by organisms of other than faecal origin. The present work shows clearly that coliforms are capable of growth in cream from very small numbers at 5° C. Thus a positive coliform test need not indicate faecal contamination but only that the cream has been stored for a sufficient time to permit the growth of a

detectable number of organisms. The methylene blue reduction test is no more reliable if the currently recommended time of greater than 4 hr. is taken as a standard. It has been shown in this work, that unless test readings are taken for at least 7½ hr., it is not possible to identify those creams in which bacteria are not actively multiplying at the time of sampling. The plateau effect in the reduction time (Figs. 1B, 2A, B, C) does not necessarily occur at a reduction time of 4 hr. or less. The results in Table 3 suggest that the plateau effect may occur over a wide range of colony counts, presumably due to the very varied bacterial flora that may be present. It will be seen from Table 3, that 75 creams had methylene blue reduction times of not less than 7½ hr., but that in some of these creams coliform organisms were present in 1/1000 ml. A total of 103 samples passed the methylene blue reduction test, coliforms being present in 1/1000 ml. in some creams. When the water agar test is considered as a standard (Table 2), 79 samples would be classed as being of good bacterial quality with no coliforms present in 1/10 ml. If a standard of not more than 50 proteolytic colonies was taken, 102 samples would pass the test, all samples having no coliforms present in 1/1000 ml. Approximately the same number of samples pass both tests at the two levels. Since the presence of coliforms may indicate either contamination or bacterial growth, it is undesirable to use a test that is not capable of consistently detecting the presence of coliforms in 1/1000 ml.

There is an extensive list of papers (Thomas *et al.* 1966) which show that dye reduction tests only detect milk of very poor quality. Although rather better results are obtained with cream, this is no argument for retaining dye reduction as a test. Although the correlation is by no means perfect, the water agar test provides a better index of the bacterial quality of cream than do any of the other methods examined here. The test also predicts the rate at which bacterial growth is likely to occur during further storage. A statistical analysis of replicate counts shows that this test is as accurate as a standard plate count.

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MARGARET M. TAYLOR

(*Facing p.* 357)

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EXPLANATION OF PLATE

A water agar test plate showing numerous proteolytic colonies and two mucoid colonies.