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STAATSKOERANT VAN DIE REPUBLIEK VAN SUID-AFRIKA

REPUBLIC OF SOUTH AFRICA GOVERNMENT GAZETTE

REGULASIEKOERANT No. 2523

As 'n Nuusblad by die Poskantoor Geregistreer

PRYS 20c PRICE
OORSEE 30c OVERSEAS
POSVRY — POST FREE

REGULATION GAZETTE No. 2523

Registered at the Post Office as a Newspaper

VOL. 147]

PRETORIA, 2 SEPTEMBER 1977

[No. 5729

GOEWERMENTSKENNISGEWING

DEPARTEMENT VAN GESONDHEID

No. R. 1724

2 September 1977

WET OP VOEDINGSMIDDELS, SKOONHEIDS-MIDDELS EN ONTSMETTINGSMIDDELS, 1972 (WET 54 VAN 1972)

REGULASIE.—MELK EN MELKPRODUKTE

Die Minister van Gesondheid vaardig hierby, kragtens artikel 15 (1) van die Wet op Voedingsmiddels, Skoonheidsmiddels en Ontsmettingsmiddels, 1972 (Wet 54 van 1972), die volgende regulasie uit wat met ingang van die datum van afkondiging hiervan toegepas kan word maar waarvan die toepassing verpligtend word ses maande na genoemde datum van afkondiging:

Woordomskrywing

“Vir die toepassing van hierdie regulasie en tensy strydig met die samehang—

het die uitdrukkings ‘gerekonstitueerde melk’, ‘room’, ‘aangesuurde’, ‘melkpoeier’ en ‘afgeroomdemelkpoeier’ die betekenis aan hul geheg in die regulasies kragtens die Bemarkingswet, 1968 (Wet 59 van 1968), uitgevaardig; en beteken—

‘saamgestelde suiwelprodukt’ ’n voedingsmiddel wat nie-suiwelprodukte bevat, met of sonder additiewe soos in Aanhengsel B uiteengesit en met minstens 50 persent (m/m) melkbestanddele;

‘hermeties verseël’ verseël op sodanige wyse as wat ’n lugdigte sluiting vorm;

‘melk’ die vloeistof wat deur die melkliere van ’n koei van die beessoort afgeskei word;

‘melkprodukt’ ’n produk uitsluitlik van melk verkry, met of sonder additiewe in Aanhengsel B veroorloof;

‘pasteurisering’ die proses waardeur die betrokke voedingsmiddel onderwerp word aan die behandeling in paragraaf 14 van Aanhengsel A beskryf, en moet die woord ‘gepasteuriseer’ dienooreenkomsdig vertolk word;

‘sterilisering’ die proses waardeur die betrokke voedingsmiddel oor sodanige tydsduur heen en by sodanige temperatuur aan hitte blootgestel word dat die eindproduk, nadat dit drie dae lank by 37 °C geïnkubeer is, vry sal wees van lewensvatbare mikroorganismes, en moet die woord ‘gesteriliseerd’ dienooreenkomsdig vertolk word;

GOVERNMENT NOTICE

DEPARTMENT OF HEALTH

No. R. 1724

2 September 1977

FOODSTUFFS, COSMETICS AND DISINFECTANTS ACT, 1972 (ACT 54 OF 1972)

REGULATION.—MILK AND MILK PRODUCTS

The Minister of Health hereby, in terms of section 15 (1) of the Foodstuffs, Cosmetics and Disinfectants Act, 1972 (Act 54 of 1972), makes the following regulation which may be applied from the date of publication hereof but shall be applied with effect from a date six months after the date of publication:

Definitions

“For the purposes of this regulation and unless inconsistent with the context—

the terms ‘reconstituted milk’, ‘cream’, ‘cultured’, ‘milk powder’ and ‘skim-milk/skimmed milk powder’ have the meanings assigned to them in the regulations under the Marketing Act, 1968 (Act 59 of 1968);

‘composite dairy product’ means any foodstuff which contains non-dairy products, with or without additives as specified in Annex B and with not less than 50 per cent (m/m) of milk constituents;

‘hermetically sealed’ means sealed in such a manner as to form an air-tight closure;

‘milk’ means the fluid secreted from the mammary glands of a cow of the bovine genus;

‘milk product’ means any product derived exclusively from milk with or without additives permitted in Annex B;

‘pasteurisation’ means the process whereby the foodstuff concerned is subject to the treatment described in paragraph 14 of Annex A; and the term

‘pasteurised’ shall be construed accordingly;

‘sterilisation’ means the process whereby the foodstuff concerned is exposed to heat at such a temperature and for such a period that the finished product, after being incubated for three days at 37 °C, will be free from viable micro-organisms, and the term ‘sterilised’ shall be construed accordingly;

'U.H.T.'-behandeling of 'ultrahoëtemperatuurbehandeling' die proses waardeur die betrokke voedingsmiddel een of meer sekondes lank aan temperatuur bokant 130 °C blootgestel word sodat die eindproduk, nadat dit 10 dae en sewe dae lank teen onderskeidelik 30 °C en 55 °C geïnkubeer is, geen aanduiding van bakteriegroei toon nie, en ook nie andersins versleg nadat dit vir 'n tydsduur van minstens vier weke by temperatuur van 20 °C±2,0 °C geïnkubeer is nie.

(1) Iemand is aan 'n misdryf skuldig indien hy aan die verbruiker melk of gerekonstitueerde melk verkoop wat—

(a) nie voldoen aan die standaarde vir melkvet en vir vetylre vaste stowwe van melk soos vir melk voor-geskryf in die regulasies kragtens die Bemarkingswet, 1968 (Wet 59 van 1968), nie;

(b) die volgende bevat:

(i) Antibiotikum of ander antimikrobiese stowwe;
(ii) stowwe wat om die een of ander rede die melk ongeskik vir menslike gebruik sal maak;

(iii) patogene mikro-organismes;

(iv) kolostrum;

(v) enige vreemde stof of ontstekingsproduk wat met die blote oog sigbaar is of wat opgespoor kan word deur dit met 'n filtrerpluis of filterskyf deur te syg;

(c) 'n plaattelling van meer as 200 000 kolonievormende bakteriese eenhede per 1,0 ml oplewer by onderwerping aan die toets wat in paragraaf 12 van Aanhangel A beskryf word;

(d) afgeskei is 14 dae voor en vyf dae na kalwing of 'n positiewe resultaat tot gevolg het by onderwerping aan die stol-by-kook-toets wat in paragraaf 10 van Aanhangel A beskryf word;

(e) nie verpak is in 'n hermeties verseëldde houer wat vloeistofdig is nie;

(f) by onderwerping aan die metileenbloureduksietoets wat in paragraaf 3 van Aanhangel A beskryf word, metileenblou binne minder as vier uur ontkleur;

(g) by onderwerping aan die resasurienreduksietoets wat in paragraaf 4 van Aanhangel A beskryf word, 'n Lovibond-skyflesing van minder as 2 na twee uur of 4 na een uur gee;

(h) by uitvoering van die gewysigde Eijkmann-toets wat in paragraaf 5 van Aanhangel A beskryf word, blyk *Escherichia coli* in 0,1 ml te bevat;

(i) by onderwerping aan die toets vir kolivormige bakterieë wat in paragraaf 9 van Aanhangel A beskryf word, blyk meer as 10 kolonievormende eenhede kolivormige bakterieë in 1,0 ml vloeistof te bevat;

(j) by onderwerping aan die toets wat in paragraaf 11 van Aanhangel A beskryf word, blyk meer as 1 000 000 sometiese selle per ml te bevat of enige ander tekens toon van abnormale afskeidingsaktiwiteit van die melkklier(e) van die bees.

(2) Iemand is aan 'n misdryf skuldig indien hy—

(A) gepasteuriseerde melk of gepasteuriseerde room verkoop wat—

(a) 10 mikrogram (μg) of meer p-nitrofenol per ml lewer volgens die Aschaffenburg-en-Mullen-fosfatase-toets wat in paragraaf 6 van Aanhangel A beskryf word of volgens enige ander toets, mits ten opsigte van akkuraatheid laasgenoemde toets aan eersgenoemde gelykwaardig is;

(b) metileenblou binne minder as twee uur ontkleur nadat die monster 18 uur lank by 18 °C±0,5 °C geïnkubeer is op die wyse wat in paragrawe 7 en 8 van Aanhangel A beskryf word;

(c) by die uitvoering van die toets wat in paragraaf 9 van Aanhangel A beskryf word, blyk 10 of meer kolonievormende eenhede kolivormige bakterieë per 1,0 ml vloeistof of 1,0 g halfvaste stof te bevat;

'U.H.T.' treatment or 'ultra-high temperature treatment' means the process whereby the foodstuff concerned has been exposed for one or more seconds to temperatures above 130 °C so that the finished product, after being incubated for periods of 10 days and seven days at 30 °C and 55 °C respectively, shows no indication of bacterial growth and does not otherwise deteriorate after being incubated for a period of at least four weeks at temperatures of 20 °C±2,0 °C.

(1) Any person shall be guilty of an offence if he sells to the consumer milk or reconstituted milk which—

(a) does not meet the standards for milk fat and milk-solids-not-fat as laid down for milk in the regulations under the Marketing Act, 1968 (Act 59 of 1968);

(b) contains—

(i) antibiotics or other antimicrobial substances;
(ii) substances which for any reason will render the milk unfit for human consumption;
(iii) pathogenic micro-organisms;
(iv) colostrum;
(v) any foreign matter or inflammatory product which is visible to the naked eye or which can be detected by straining through a filter wad or disc;

(c) gives a total plate count of more than 200 000 colony-forming bacterial units per 1,0 ml when subjected to the test described in paragraph 12 of Annex A;

(d) was secreted between 14 days before and five days after parturition or gives a positive result when subjected to the clot-on-boiling test described in paragraph 10 of Annex A;

(e) is not packed in a hermetically sealed package impermeable to liquid;

(f) when subjected to the methylene-blue reduction test described in paragraph 3 of Annex A, decolourises methylene blue in less than four hours;

(g) when subjected to the resazurin reduction test described in paragraph 4 of Annex A gives a Lovibond disc reading of less than 2 after two hours or 4 after one hour;

(h) on application of the modified Eijkmann test described in paragraph 5 of Annex A is found to contain any *Escherichia coli* in 0,1 ml;

(i) when subjected to the coliform test described in paragraph 9 of Annex A is found to contain more than 10 colony-forming units of coliform bacteria in 1,0 ml of fluid;

(j) when subjected to the test described in paragraph 11 of Annex A is found to contain more than 1 000 000 somatic cells per ml or shows any other signs of abnormal secretory activity of the bovine mammary gland(s).

(2) Any person shall be guilty of an offence if he sells—

(A) pasteurised milk or pasteurised cream which—

(a) has been shown by the Aschaffenburg and Mullen phosphatase test described in paragraph 6 of Annex A or any other test, provided its accuracy equals that of the afore-mentioned test, to yield 10 micrograms (μg) or more of p-nitrophenol per ml;

(b) decolourises methylene blue in less than two hours after the sample has been incubated at 18 °C±0,5 °C for 18 hours, as described in paragraphs 7 and 8 of Annex A;

(c) on application of the test described in paragraph 9 of Annex A, is found to contain 10 or more colony-forming units of coliform bacteria per 1,0 ml of fluid or 1,0 g of semi-solid;

(d) by uitvoering van die gewysigde Eijkmann-toets wat in paragraaf 5 van Aanhangsel A beskryf word, blyk *E.coli* in 1,0 ml vloeistof of 1,0 g halfvaste stof te bevat;

(e) enige van die stowwe of organismes bevat wat in subregulasie (1) (b) bedoel word; of

(B) gesteriliseerde room of melk of U.H.T.-room of melk verkoop wat—

(a) die volgende etanolstabiliteitstoets nie deurstaan nie:

Meng een volume 68 persent (m/m)-waterige etanol met een volume melk. Indien geen neerslag gevorm word nie, voldoen die melk aan die etanolstabiliteitstoets.

(b) indien die toets beskryf in paragraaf 13 van Aanhangsel A uitgevoer word sowel voor as na 'n inkubasie van of 14 dae by $30^{\circ}\text{C} \pm 1^{\circ}\text{C}$ of sewe dae by $55^{\circ}\text{C} \pm 1^{\circ}\text{C}$, 'n toename in titreerbare suur toon van meer as 0,02, uitgedruk as g melksuur per 100 g melk.

(3) Behoudens die ander bepalings van die Wet, is iemand aan 'n misdryf skuldig indien hy enige van die melkprodukte verkoop wat genoem word in die regulasies uitgevaardig ingevolge die Bemarkingswet, 1968 (Wet 59 van 1968), of ingevolge die Wet op Suiwelnywerhede, 1961 (Wet 30 van 1961), en indien sodanige melkprodukt—

(a) die volgende bevat:

(i) Antibiotika of enige ander antimikrobiiese stowwe;
(ii) stowwe wat om die een of ander rede sodanige produk ongeskik vir menslike gebruik sal maak;
(iii) patogene mikro-organismes;

(iv) ontstekingsprodukte wat met die blote oog sigbaar is; of

(v) additiewe wat nie in Aanhangsel B veroorloof word nie;

(b) andersins ongeskik is vir menslike gebruik;
(c) in die geval van—

(i) 'n aangesuurde melkproduk, nie van gepasteuriseerde melk afkomstig nie;

(ii) melkpoeier of afgeroomdemelkpoeier, meer as 200 000 kolonievormende eenhede lewensvatbare bakterieë per gram blyk te bevat by toepassing van die Standaard-agar-plaatkolonietellingstoets wat in paragraaf 12 van Aanhangsel A beskryf word;

(d) nie verpak is in hermities verseëlde houers wat vloeistofdig is nie;

(e) by uitvoering van die toets wat in paragraaf 9 van Aanhangsel A beskryf word, blyk meer as 10 koliforme bakterieë per 1,0 ml vloeistof of per 1,0 g droë stowwe te bevat;

(f) by uitvoering van die gewysigde Eijkmann-toets wat in paragraaf 5 van Aanhangsel A beskryf word, blyk enige *E.coli* in onderskeidelik 1,0 ml vloeistof of 1,0 g halfvaste stowwe of 0,1 g melkpoeier te bevat; of

(g) met uitsondering van aangesuurde melk en joghurt, metileenblou binne minder as twee uur ontkleur nadat die monster 18 uur lank by $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ geïnkubeer is op die wyse wat in paragrawe 7 en 8 van Aanhangsel A beskryf word.

(4) Ter beslissing van die vraag of melk en melkprodukte voldoen aan die vereistes in subregulasies (1), (2) en (3) vervat, word die toetse uitgevoer wat in Aanhangsel A beskryf word, en genoemde toetse is vir genoemde doel afdoen.

(5) Iemand is aan 'n misdryf skuldig indien hy melk of 'n melkproduk verkoop wat 'n additief bevat, behalwe dat 'n voedingsmiddel in kolom I van Aanhangsel B vermeld enige van die additiewe daarteenoor in kolom II genoem, kan bevat volgens die toleransie, en op die voorwaardes, wat in kolom III aangedui word.

(d) on application of the modified Eijkmann test described in paragraph 5 of Annexure A is found to contain any *E. coli* in 1,0 ml of fluid or 1,0 g of semi-solid;

(e) contains any of the substances or organisms referred to in subregulation (1) (b); or

(B) sterilised cream or milk or U.H.T. cream or milk which—

(a) fails the following stability test with ethanol:

Mix one volume of 68 per cent (m/m) aqueous ethanol with one volume of milk. If no precipitate is formed the milk conforms to the ethanol stability test.

(b) shows an increase in titratable acid greater than 0,02 expressed as g of lactic acid per 100 g of milk on application of the test described in paragraph 13 of Annex A before and after incubation at $30^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for 14 days or $55^{\circ}\text{C} \pm 1^{\circ}\text{C}$ for seven days.

(3) Subject to other provisions of the Act, any person shall be guilty of an offence if he sells any milk product referred to in the Regulations framed under the Marketing Act, 1968 (Act 59 of 1968), or the Dairy Industries Act, 1961 (Act 30 of 1961), which—

(a) contains—

(i) antibiotics or any other antimicrobial substances;

(ii) substances which for any reason will render such product unfit for human consumption;

(iii) pathogenic micro-organisms;

(iv) inflammatory products visible to the naked eye; or

(v) additives not permitted in Annex B;

(b) is otherwise unfit for human consumption;

(c) in the case of—

(i) a cultured milk product is not derived from pasteurised milk;

(ii) milk powder or skim-milk/skimmed milk powder found to contain more than 200 000 colony-forming units of viable bacteria per gram on application of the Standard Agar Plate Colony Count Test described in paragraph 12 of Annex A;

(d) is not packed in hermetically sealed packages impermeable to liquid;

(e) on application of the test described in paragraph 9 of Annexure A is found to contain more than 10 coliform bacteria per 1,0 ml of fluid or 1,0 g of dry matter;

(f) on application of the modified Eijkmann test described in paragraph 5 of Annex A is found to contain any *E. coli* in 1,0 ml of fluid or 1,0 g of semi-solid or 0,1 g of milk powder respectively; or

(g) with the exception of cultured milk and yogurt decolourises methylene blue in less than two hours after the sample has been incubated at $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ for 18 hours, as described in paragraphs 7 and 8 of Annex A.

(4) For the purpose of determining whether milk and milk products meet the requirements laid down in sub-regulations (1), (2) and (3) the tests set out in Annex A shall be applied and shall be conclusive.

(5) Any person shall be guilty of an offence if he sells milk or any milk product containing any additive, except that any of the foodstuffs mentioned in column I of Annex B may contain any of the additives mentioned in the corresponding line of column II to a tolerance and under the conditions indicated in column III.

(6) Iemand is aan 'n misdryf skuldig indien hy melk of 'n melkproduk verkoop wat meer as een keer aan pasteurisering, sterilisering, of U.H.T.-behandeling onderwerp is of wat aan meer as een sodanige proses onderwerp is."

Regulasie 7 van die regulasies wat kragtens die herroep Wet op Voedingsmiddels, Medisyne en Ontsmettingsmiddels, 1929 (Wet 13 van 1929), by Goewermentskennisgewing 575 van 28 Maart 1930 afgekondig is soos gewysig, word hierby herroep met ingang van die datum van inwerkingtreding van die bepalings van hierdie kennisgewing.

AANHANGSEL A

METODES VIR DIE TOETSING VAN MELK, ROOM EN MELKPRODUKTE

1. (1) Die toets wat in hierdie Aanhangle beskryf word, is dié wat in toepaslike gevalle uitgevoer moet word ten einde die suiwerheid of onsuwerheid van melk, room en melkprodukte te bepaal. Alle monsters word, totdat die toets uitgevoer word, by 'n temperatuur van hoogstens 10 °C gehou.

(2) Vir die toepassing van hierdie Aanhangle beteken "melk" ook melk wat reeds onderwerp is aan pasteurisering, sterilisering of ultrahoëtemperatuur behandeling, asook room, hetsy wel of nie in 'n hitteverseëld houer, maar nie ook room of gekondenseerde melk in 'n hermetiese verseëld houer nie.

MIKROBIOLOGIESE ONDERSOEK

2. (1) Erkende bakteriologiese tegnieke vir die handhawing van aseptiese toestande moet tydens die volle duur van elke mikrobiologiese toets toegepas word.

(2) Al die gedistilleerde water wat vir die bereiding van media gebruik word, moet of glasgedistilleerde water wees of water van 'n gelykwaardige kwaliteit.

(3) Al die glasware wat gebruik word vir die toets wat in hierdie Aanhangle voorgeskryf word, moet steriel wees.

(4) Die steriliteit van glasware, media en verdunningsmiddels moet gekontroleer word deur—

(a) met elke toets verteenwoordigende kontrolebuise en -bakkies te inkubeer; of

(b) die een of ander vorm van groeimedium te gebruik.

(5) Al die pipette wat gebruik word, moet ten minste graad B-pipette van die uitblaastipe wees met 'n gesikte nie-absorberende watteprop.

(6) Al die glasware wat vir volumetriese meting gebruik word, moet 'n akkuraatheidsgraad hê wat ten minste gelykstaande is met Graad B van die Nasionale Fisiiese Navorsingslaboratorium.

(7) Al die chemikalieë wat gebruik word by die bereiding van oplossings en media wat hierin genoem word, moet, tensy dit anders voorgeskryf word, van 'n analitiesereagensgraad wees of van 'n graad wat geskik is vir die bereiding van bakteriologiese media.

(8) In plaas van die voorgeskrewe media kan gesikte ontwaterde kultuurmedia gebruik word indien dit beskikbaar is: Met dien verstande dat sodanige ontwaterde media aan die gegewe beschrywing voldoen en gelykwaardige resultate oplewer. Die standaard van die pepton, gal-soute, triptoone, gisektrak en beesgal wat gebruik word, moet gelykstaande wees met die verwysingstandaard wat gehou word deur die Suid-Afrikaanse Buro vir Standaarde, Privaatsak X191, Pretoria.

(9) Die verteenwoordigende monsters melk moet met steriele toerusting geneem word en in steriele monsterhouers geplaas word, en voorsorgmaatreëls moet getref word om kontaminasie van die monsters te voorkom. Elke sodanige monsterhouer moet toegemaak word, en

(6) Any person shall be guilty of an offence if he sells milk or a milk product which has been subjected to pasteurisation, sterilisation or U.H.T. treatment more than once or which has been subjected to more than one such treatment."

Regulation 7 of the regulations under the repealed Food, Drugs and Disinfectants Act, 1929 (Act 13 of 1929), published under Government Notice 575 of 28 March 1930, as amended, is hereby rescinded with effect from the date of coming-into-effect of the provisions of this notice.

ANNEX A

METHODS FOR THE TESTING OF MILK, CREAM AND MILK PRODUCTS

1. (1) The tests described in this Annex are those to be applied in appropriate cases in order to ascertain the purity or impurity of milk, cream and milk products. All samples shall be kept at a temperature not exceeding 10 °C until the tests are applied.

(2) For the purposes of this Annex "milk" includes milk that has been subjected to pasteurisation or sterilisation or ultra-high temperature treatment, and also cream, whether or not contained in a heat-sealed container, but does not include cream or condensed milk in a hermetically sealed container.

MICROBIOLOGICAL EXAMINATIONS

2. (1) Recognised bacteriological techniques for the maintenance of aseptic conditions shall be applied throughout all microbiological testing.

(2) All distilled water used in the preparation of media shall be glass-distilled water or water of a similar quality.

(3) All glassware used in the tests prescribed in terms of this Annex shall be sterile.

(4) The sterility of all glassware, media and diluents shall be checked by—

(a) incubating representative control tubes and dishes with each test; or

(b) using some form of growth medium.

(5) All pipettes used shall be at least Grade B pipettes of the blow-out type, suitably plugged with non-absorbent cotton wool.

(6) All glassware used for volumetric measurement shall be of an accuracy at least equal to National Physical Research Laboratory Grade B.

(7) All the chemicals used in the preparation of solutions and media mentioned herein shall, except where otherwise prescribed, be of analytical reagent grade or a grade acceptable for the preparation of bacteriological media.

(8) Appropriate dehydrated culture media, where such preparations are available, may be used in lieu of the media prescribed: Provided that such dehydrated preparations conform to the description given and yield equivalent results. The peptone, bile salts, tryptone, yeast extract and ox-bile used shall be equivalent to the reference standard held by the South African Bureau of Standards, Private Bag X191, Pretoria.

(9) Representative samples of milk shall be taken with sterile equipment and transferred to sterile sample containers, precautions being taken to prevent the contamination of the samples. Each sample container shall be closed and within 15 minutes of the sample being taken, the

binne 15 minute nadat 'n monster genoem is, moet die betrokke monsterhouer omring word met gebreekte ys of 'n ander gesikte koel-middel in blikke of plastiekhouders wat in regstreekse aanraking met die monsterhouer geplaas moet word. Genoemde koelmiddel moet in staat wees om die temperatuur van die monster af te bring tot tussen 5 °C en 10 °C en dit daar te hou.

DIE METILEENBLOU-REDUKSIE-TOETS

3. (1) Die metileenblou-reduksietoets vir melk word uitgevoer op die wyse wat in onderstaande subparagraawe beskryf word.

(2) Los een Britsestandaard-metileenbloutablet van 15,8 mg of die ekwivalent daarvan op in 800 ml koue, steriele, gedistilleerde water sodat die eindkonsentrasie metileenblou in die stamoplossing 1:50 000 is.

(3) Plaas die oplossing in 'n ligwerende houer met 'n prop op en bêre op 'n koel, donker plek.

(4) Moenie die metileenblou-oplossing vir 'n toets kragtens hierdie paragraaf gebruik nie indien—

(a) die oplossing aan sonlig blootgestel was, of

(b) daar twee maande verloop het sedert die bereidingsdatum van die oplossing.

(5) Giet in 'n glashouer soveel van die stamoplossing as wat vir 'n dag se werk nodig is.

(6) Meng die monster melk deeglik en pipetteer 10 ml daarvan in 'n steriele proefbuis wat nominaal 'n lengte van 150 mm en 'n buitemiddellyn van 16 mm het en met 'n prop toegemaak is.

(7) Voeg, met behulp van 'n steriele 1-ml-pipet, 1 ml metileenblou-oplossing by die melk in die proefbuis. Sorg dat die pipet nie met die melk in aanraking kom nie. Sorg ook dat al die nodige aseptiese voorsorgmaatreëls nagekom word.

(8) Maak die buis met 'n steriele rubberprop toe en meng die inhoud deur twee keer die buis stadiig om te keer.

(9) Plaas die buis, binne vyf minute nadat die inhoud daarvan gemeng is, in 'n termostatis beheerde, bedekte waterbad waarvan die temperatuur op $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ geregeer word.

(10) Berei vir elke toets, as kontrole, een buis met melk waarby gevoeg is 1 ml kraanwater wat drie minute lank gekoos is, en een buis met melk wat drie minute lank gekook is en waarby 1 ml metileenblou-oplossing gevoeg is.

(11) Die waterhoogte in die waterbad moet hoër wees as die hoogte van die inhoud van die buise.

(12) Ondersoek die inhoud al om die 30 minute met die oog op kleurreduksie. Genoemde proses van kleurreduksie word geag afgeloop te wees wanneer die kolom melk tot binne 5 mm van die oppervlak daarvan ontkleur is.

(13) Indien na verloop van 30 minute die ontkleuring wel begin het maar nog nie afgeloop is nie, plaas dan die buis in die waterbad terug totdat die proses afgeloop is. Indien die oplossing egter na verloop van 30 minute nog geen ontkleuring toon nie, keer die buis dan een maal om voordat u dit in die waterbad terugplaas.

(14) Veronagsaam 'n moontlike skynseltjie kleur wat op die bodem van die buis sigbaar word maar wat hoogstens 5 mm opwaarts strek.

(15) 'n Monster melk wat, indien dit by $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ geïnkubeer word, die metileenblou binne minder as vier uur ontkleur, word geag nie die toets te deurstaan het nie.

RESASURIENREDUKSIE-TOETS

4. (1) Die resasuriendreduksietoets vir melk word toegepas ooreenkomsdig onderstaande subparagraawe.

sample container shall be surrounded by crushed ice or other suitable refrigerant in tins or plastic containers. Such refrigerant shall be in direct contact with the sample container and shall be capable of reducing the temperature of the sample to and maintaining it at between 5 °C and 10 °C.

METHYLENE BLUE REDUCTION TEST

3. (1) The methylene blue reduction test for milk shall be carried out in accordance with the succeeding sub-paragraphs.

(2) Dissolve one British Standard methylene blue tablet of 15,8 mg or its equivalent in 800 ml of cold, sterile distilled water so that the final concentration of methylene blue in the stock solution is 1:50 000.

(3) Place this solution in a light-resistant stoppered vessel and store in a cool, dark place.

(4) Do not use the methylene blue solution to make a test in terms of this paragraph if—

(a) it has been exposed to sunlight; or

(b) a period of two months has elapsed since the date of preparation of the solution.

(5) Pour off as much of the stock solution as is required for a day's work into a glass container.

(6) Thoroughly mix the sample of milk and pipette 10 ml of the sample into a sterile stoppered test tube nominally measuring 150 mm in length by 16 mm in outer diameter.

(7) Add to the milk in the test tube 1 ml of methylene blue solution using a sterile 1 ml pipette. This pipette must not come into contact with the milk and all the necessary aseptic precautions should be observed.

(8) Close the tube with a sterile rubber stopper and mix the contents by slowly inverting the tube twice.

(9) Within five minutes of the mixing, place the tube in a thermostatically controlled covered water bath regulated to a temperature of $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$.

(10) Set up as a control with each test one tube containing milk to which has been added 1 ml of tap water boiled for three minutes and one tube containing milk which has been boiled for three minutes, to which has been added 1 ml of methylene blue solution.

(11) The level of the water in the water bath shall exceed the level of the contents of the tubes.

(12) Examine the contents every 30 minutes for dye reduction. The process shall be taken as complete when the column of milk is decolourised up to within 5 mm of the surface.

(13) If at the end of 30 minutes decolourisation has started and is not yet complete, replace the tube in the water bath until the process is complete, but if at the end of 30 minutes the contents show no decolourisation, invert the tube once before replacing it in the water bath.

(14) Ignore any trace of colour at the bottom of the tube extending upwards for not more than 5 mm.

(15) Samples of milk which decolourise the methylene blue in less than four hours' incubation at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ shall be deemed to have failed the test.

RESAZURIN REDUCTION TEST

4. (1) The resazurin reduction test for milk shall be carried out in accordance with the succeeding sub-paragraphs.

(2) Die resasurienoplossing van 0,005 persent (m/v) wat vir die toets gebruik word, moet—

(a) van gestandaardiseerde resasurientablette berei word;

(b) met steriele, gedistilleerde water aangemaak word; en

(c) elke dag waarop daar 'n toets plaasvind, vars aangemaak word.

(3) Meng die monster melk deeglik en gooi 10 ml daarvan in 'n steriele proefbuis wat nominaal 'n lengte van 150 mm en 'n buitemiddellyn van 16 mm het en op 10 ml afgemerk is.

(4) Voeg, met behulp van 'n steriel 1-ml-pipet, 1 ml van 'n resasurienoplossing van 0,005 persent (m/v) by die monster in die proefbuis.

(5) Sorg dat die pipet nie met die melk in die buis in aanraking kom nie.

(6) Tref al die nodige voorsorgmaatreëls om kontaminasie van monster te voorkom.

(7) Maak die buis met 'n steriele rubberprop toe en meng die inhoud deur tweeker die buis stadig om te keer.

(8) Plaas die buis, binne vyf minute nadat die inhoud daarvan gemeng is, in 'n termostatis beheerde, bedekte waterbad waarvan die temperatuur op $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ geregeer word.

(9) Die waterhoogte in die waterbad moet slegs effens hoër wees as die hoogte van die inhoud van die buise.

(10) Neem, na verloop die een- of twee-uurtydperk, 'n Lovibond-skyflesing ten einde die kleurreduksie te bepaal.

(11) Gebruik vir die Lovibond-skyflesing 'n spesiale resasurientoetsskyf, No. 4/9, en gebruik daarby as kontrole 'n blanko buis met dieselfde melk, maar sonder resasurien.

(12) Neem die toetslesing in goeie weerkaatse lig maar nie in regstreekse sonlig nie.

DIE GEWYSIGDE EIJKMANN-TOETS

5. (1) Die gewysigde Eijkmann-toets vir melk gepasteuriseerde melk, gepasteuriseerde room, gerekonstitueerde melk, afgeroomde melkpoeier, melkpoeier en aangesuurde melkprodukte word uitgevoer op die wyse wat in onderstaande subparagrafe beskryf word. Vir die toepassing van hierdie paragraaf beteken die uitdrukking "*Escherichia coli*" daardie mikro-organisme wat in die aanwesigheid van galsoute laktose by $44^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ binne 48 uur laat fermenteer met die produksie van suur en gas en wat in triptoonwater by dieselfde temperatuur, na 'n inkubasietydperk van 48 uur, indool vorm binne 'n verdere 48 uur.

(2) Meng die monster melk of room deeglik. Indien die room te dik is om maklik te hanteer, verwarm dit tot 'n temperatuur van hoogstens 37°C .

(3) Nadat al die nodige voorsorgmaatreëls getref is om kontaminasie te voorkom, inokuleer met behulp van 'n 1-ml-pipet die inhoud van die buise wat briljante groen 2-persent(m/v)-galboeljon bevat en wat voorsien is van 'n omgekeerde Durhamfermentasiebuisie vir gasopsporing, met 0,01 ml in die geval van melk, of 1 ml in die geval van gepasteuriseerde melk en gepasteuriseerde room.

(4) Vir die meet van die hoeveelhede van 0,01 ml wat in die geval van melk getoets moet word, berei desimale verdunde oplossings voor volgens die Standaard-agar-plaatkolonietellingmetode wat in paragraaf 12 (2) beskryf word.

(5) Inokuleer die inhoud van drie buise vir elke monster wat getoets word.

(6) Inkubeer die geïnokuleerde brillante groen galboeljon 48 uur lank in 'n waterbad waarvan die temperatuur spesial op $44^{\circ}\text{C} \pm 0,25^{\circ}\text{C}$ gehou word.

(2) The 0,005 per cent (m/v) resazurin solution to be used in making the said test shall be—

(a) prepared from standardised resazurin tablets;

(b) made up in sterile, distilled water; and

(c) freshly prepared each day on which a test is made.

(3) Thoroughly mix the sample of milk and pour 10 ml of it into a sterile test tube nominally measuring 150 mm in length by 16 mm in outer diameter and a mark at the 10 ml level.

(4) To the sample in the test tube add 1 ml of a 0,005 per cent (m/v) resazurin solution by means of a sterile 1 ml pipette.

(5) Do not allow the pipette to come into contact with the milk in the tube.

(6) Take all necessary precautions to prevent contamination of the sample.

(7) Close the test tube with a sterile rubber stopper and mix the contents by slowly inverting the tube twice.

(8) Within five minutes of the mixing, place the tube in a thermostatically controlled, covered water bath regulated to a temperature of $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$.

(9) The level of the water in the water bath shall just exceed the level of the contents in the tubes.

(10) At the end of the one or two hour period take a Lovibond disc reading to assess the dye reduction.

(11) For the Lovibond disc reading use a special resazurin test disc, No. 4/9, and a control consisting of a blank tube of the milk being tested.

(12) Carry out the reading in good reflected light, but not in direct sunlight.

MODIFIED EIJKMANN TEST

5. (1) The modified Eijkmann test for milk, pasteurised milk, pasteurised cream, reconstituted milk, skim-milk powder, milk powder and cultured milk products shall be carried out in accordance with the succeeding subparagraphs. For the purposes of this paragraph the term "*Escherichia coli*" means that micro-organism which ferments lactose with the production of acid and gas within 48 hours at $44^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ in the presence of bile salts and produces indole in tryptone water within 48 hours at the same temperature after an incubation period of 48 hours.

(2) Thoroughly mix the sample of milk or cream, and if the cream is too thick for convenient handling, warm it to a temperature not higher than 37°C .

(3) All necessary precautions having been taken to prevent contamination, the tubes containing 2 per cent (m/v) brilliant green bile broth and fitted with an inverted Durham fermentation tube for the detection of gas are inoculated, by means of a 1 ml pipette, with 0,01 ml in the case of milk, or 1 ml in the case of pasteurised milk and pasteurised cream.

(4) For the measurement of the 0,01 ml quantities to be tested in the case of milk, prepare decimal dilutions in accordance with the Standard Agar Plate Colony Count method described in paragraph 12 (2).

(5) Inoculate the contents of three tubes for each sample tested.

(6) Incubate the inoculated brilliant green bile broth for 48 hours in a water bath specially controlled at a temperature of $44^{\circ}\text{C} \pm 0,25^{\circ}\text{C}$.

(7) Indien blykens die Durhambuisie die inkubasie by subparagraaf (6) voorgeskryf, tot die vorming van gas lei, bring dan uit iedere buis wat briljante groen galboeljon bevat en waarin gas aldus gevorm is, 'n inoculum van 0,2 ml oor na 'n afsonderlike buis triptoont water.

(8) Inkubeer die buise triptoont water wat in subparagraaf (7) genoem word, 24 uur lank by $44^{\circ}\text{C} \pm 0,25^{\circ}\text{C}$. in die waterbad in subparagraaf (6) vermeld.

(9) Om te bepaal of daar indool ontstaan het, toets die triptoont water in die buise na verloop van genoemde 24 uur deur 0,5 ml Kovac-reagens daarby te voeg.

(10) Die vorming van 'n rooskleurige ring by die tussenvlak van die twee vloeistowwe word geag die aanwesigheid van indool aan te dui.

(11) Indien daar gas en indool gevorm word in een, twee of drie van die buise wat melk van dieselfde monster bevat, word sodanige resultaat geag aan te dui dat daar *Escherichia coli* aanwesig is.

(12) Berei die briljante groen 2-percen(m/v)-galboeljon, die triptoont water en die Kovac-reagens soos volg:

(a) (i) die samestelling van die briljante groen 2-percen(m/v)-galboeljon moet soos volg wees:

Beesgal: 20 g.

Peptoont: 10 g.

Laktose: 10 g.

1 per cent (m/v) wateroplossing van briljante groen: 1,3 ml.

Gedistilleerde water tot: 1 l.

(ii) Los die bestanddele in die gedistilleerde water op.

(iii) Reguleer die pH-waarde sodat dit tussen 7,2 en 7,4 is.

(iv) Verdeel die medium in hoeveelhede van 10 ml tussen proefbuise wat elkeen 'n omgekeerde Durham fermentasiebuise bevat en steriliseer die proefbuise dan 15 minute lank by 121°C in 'n outoklaaf.

(b) (i) Die samestelling van die triptoont water moet soos volg wees:

Triptoont: 10 g.

Natriumchloried: 5 g.

Gedistilleerde water tot 1 l.

(ii) Los die bestanddele in die gedistilleerde water of deur dit effens te verwarm.

(iii) Reguleer die pH met natriumhidroksiedoplossing of soutsuuroplossing sodat dit tussen 7,4 en 7,5 is.

(iv) verdeel die medium in hoeveelhede van 5 ml tussen die proefbuise. Verhit die verdeelde medium 15 minute lank in 'n outoklaaf by 121°C .

(c) (i) Die samestelling van die Kovac-reagens moet soos volg wees:

Paradimetylaminobensaldehyd: 5 g.

Gekonsentreerde soutsuur: 25 ml.

Amielalkohol (piridienvry): 75 ml.

(ii) Los die paradimetylaminobensaldehyd in die amielalkohol op en voeg dan soutsuur by.

(iii) Die reagens moet, nadat dit klaar berei is, geel van kleur wees.

(iv) Plaas die reagens in 'n houer van amberkleurige glas met 'n prop op en bêre genoemde houer op 'n koel donker plek.

(v) Die reagens moenie binne die eerste 24 uur nadat dit voorberei is, gebruik word nie.

DIE ASCHAFFENBURG-EN-MULLEN-FOSFATASE-TOETS

(Gepasteuriseerde melk, gerekonstitueerde melk, gepasteuriseerde room, gesteriliseerde melk, gesteriliseerde room, gepasteuriseerde aangesuurde melkprodukte en gerekonstitueerde aangesuurde melkprodukte)

6. (1) Bogenoemde fosfatase-toets word toegepas op die wyse wat in onderstaande subparagrafe beskryf word.

(7) If the incubation prescribed in terms of subparagraph (6) leads to the formation of gas as seen in the Durham tube, an inoculum of 0,2 ml from each brilliant green bile broth tube showing gas shall be transferred to a separate tube of tryptone water.

(8) Incubate the tryptone water tubes referred to in subparagraph (7) in the water bath mentioned in subparagraph (6) at $44^{\circ}\text{C} \pm 0,25^{\circ}\text{C}$ for 24 hours.

(9) At the end of the said 24 hours test the tryptone water tubes for indole production by the addition of 0,5 ml of Kovac's reagent.

(10) The development of a rose-coloured ring at the interface of the two liquids shall be taken as indicating the presence of indole.

(11) A positive result for gas and indole in one, two or three of the tubes from the same sample of milk shall be taken as indicating the presence of *Escherichia coli*.

(12) Prepare the 2 per cent (m/v) brilliant green bile broth, the tryptone water and the Kovac's reagent as follows:

(a) (i) The composition of the 2 per cent (m/v) brilliant green bile broth shall be as follows:

Ox-bile: 20 g.

Peptone: 10 g.

Lactose: 10 g.

1 per cent (m/v) aqueous solution of brilliant green: 1,3 ml.

Distilled water to: 1 l.

(ii) Dissolve the constituents in the distilled water.

(iii) Adjust the pH to a value of 7,2 to 7,4.

(iv) Distribute the medium in 10 ml quantities among test tubes containing an inverted Durham fermentation tube and then sterilise them in an autoclave at 121°C for 15 minutes.

(b) (i) The composition of the tryptone water shall be as follows:

Tryptone: 10 g.

Sodium chloride: 5 g.

Distilled water to: 1 l.

(ii) Dissolve the constituents in the distilled water by warming it slightly.

(iii) Adjust the pH to 7,4-7,5 with the aid of sodium hydroxide solution or hydrochloric acid solution.

(iv) Dispense the medium in 5 ml aliquot in test tubes. Autoclave the dispensed medium at 121°C for 15 minutes.

(c) (i) The composition of the Kovac's reagent shall be as follows:

Paradimethylaminobenzaldehyde: 5 g.

Concentrated hydrochloric acid: 25 ml.

Amyl alcohol (pyridine-free): 75 ml.

(ii) Dissolve the paradimethylaminobenzaldehyde in the amyl alcohol, and then add hydrochloric acid.

(iii) After preparation the reagent should be yellow in colour.

(iv) Place the reagent in an amber-coloured, glass-stoppered vessel and store in a cool dark place.

(v) The reagent shall not be used within 24 hours of preparation.

ASCHAFFENBURG AND MULLEN PHOSPHATASE TEST

(Pasteurised milk, reconstituted milk, pasteurised cream, sterilised milk, sterilised cream, pasteurised cultured milk products and reconstituted cultured milk products)

6. The phosphatase test shall be carried out in accordance with succeeding subparagraphs.

(2) Ondersoek iedere monster wat getoets moet word, so gou doenlik nadat dit in die toetslaboratorium aangekom het.

(3) Indien 'n monster nie onmiddellik nadat dit in die toetslaboratorium aangekom het, ondersoek word nie, hou dit by 'n temperatuur van tussen 0 °C en 5 °C totdat dit ondersoek word.

(4) Verhoog die temperatuur van die monster tot kamertemperatuur net voordat dit ondersoek word.

(5) Tref die volgende voorsorgmaatreëls gedurende of in verband met die ondersoek van 'n monster:

(a) Moenie 'n monster toets wat tekens van bederf of suurheid toon nie.

(b) Gebruik 'n skoon pipet vir iedere monster melk of room en sorg dat geen pipet met speeksel gekontamineer word nie.

(c) Moenie die ondersoek in regstreekse sonlig uitvoer nie.

(d) Gebruik deurgaans slegs gedistilleerde water.

(6) Gebruik oral waar doenlik reagense van analitiese gehalte vir hierdie toets en berei die buffersubstraatoplossing soos volg:

(a) Die bufferoplossing: Los, in 'n standaardmaatfles, 3,5 g watervrye natriumkarbonaat en 1,5 g natriumbikarbonaat in gedistilleerde water op en voeg gedistilleerde water by die oplossing tot by die 1-l-merk.

(b) Hou die soliede substraat, dinatrium-p-nitrofenylfosfaat, in 'n koelkas.

(c) Die buffersubstraatoplossing:

(i) Plaas 150 mg van die substraat in 'n standaardmaatfles van 100 ml en vul die fles met die bufferoplossing tot by die 100-ml-merk.

(ii) Hou die oplossing in 'n koelkas en beskerm dit teen lig.

(iii) Die lesing vir die oplossing blykens 'n veeldoelvergelyker moet laer as die standaard van 10 op die vergelykerskyf A.P.T.W. of A.P.T.W. 7 wees wanneer daar vir die vergelyking gedistilleerde water gebruik word en die oplossing deur 'n sel van 25 mm in deurgelate lig beskou word.

(iv) Moenie die oplossing langer as een week gebruik nie.

(7) Gebruik die ondergenoemde apparaat vir die ondersoek:

(a) 'n Lovibond-veeldoelvergelyker met 'n staander vir werk in weerkaatse lig.

(b) 'n Lovibond-vergelykerskyf A.P.T.W. of A.P.T.W. 7.

(c) Twee selle van saamgesmelte glas, 25 mm diep, of proefbuise van kleurlose glas, met 'n binneleysnee van 13,5 mm, ooreenkomsdig B.S. 625, met rubberstoppers, vir gebruik in die Lovibond 1 000-veeldoelvergelyker.

(d) 'n Waterbad of broeikas waarvan die temperatuur op $37,0^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ gehandhaaf kan word.

(e) 'n Pipet wat 5,0 ml kan lewer.

(f) 'n Voorraad regaf pipette met 'n houvermoë van 1,0 ml.

(g) Standaardmaatflesse met 'n houvermoë van 1 l.

(h) 'n Standaardmaatfles met 'n houvermoë van 100 ml.

(8) (a) Maak elke proefbuis leeg nadat dit gebruik is, spoel dit in water af, was dit deeglik in warm water wat soda bevat, spoel dit in warm water en dan in gedistilleerde water af en maak dit droog; of, so nie, volg 'n ander skoonmaakmetode wat ewe doeltreffend is.

(b) Indien 'n proefbuis, nadat dit volgens (a) van hierdie subparagraaf behandel is, nie skoon lyk nie, herhaal genoemde behandeling, maar voeg die volgende twee stappe by nadat die proefbuis in warm water afgespoel is: naamlik, laat die proefbuis in soutsuur week en spoel dit

(2) Examine every sample to be tested as soon as possible after its arrival at the testing laboratory.

(3) If the sample is not examined immediately on its arrival at the testing laboratory, keep it at a temperature of between 0 °C and 5 °C until examined.

(4) Raise the temperature of the sample to room temperature immediately before the examination.

(5) Take the following precautionary measures during or in connection with the examination of a sample:

(a) Do not test a sample which shows evidence of taint or souring.

(b) Use a different pipette for each sample of milk or cream and ensure that no pipette is contaminated with saliva.

(c) Do not carry out the examination in direct sunlight.

(d) Use distilled water throughout the examination.

(6) Whenever practicable, use reagents of analytical quality for this test, and prepare the buffer-substrate solution as follows:

(a) Buffer solution: Dissolve 3,5 g of anhydrous sodium carbonate and 1,5 g of sodium bicarbonate in distilled water and make up to 1 l in a standard flask.

(b) Keep the solid substrate, disodium p-nitrophenyl phosphate, in a refrigerator.

(c) Buffer-substrate solution:

(i) Place 150 mg of the substrate in a 100 ml standard flask and make up to 100 ml with the buffer solution.

(ii) Store the solution in a refrigerator and protect from light.

(iii) When distilled water is used for comparison, the solution should give a reading of less than the standard marked 10 on the comparator disc A.P.T.W. or A.P.T.W. 7 when viewed in transmitted light through a 25 mm cell in the all-purpose comparator.

(iv) Do not use the solution for more than one week.

(7) Use the following apparatus for the examination:

(a) A Lovibond all-purpose comparator with a stand for work in reflected light.

(b) A Lovibond comparator disc A.P.T.W. or A.P.T.W. 7.

(c) Two fused glass cells, 25 mm deep, or test tubes of colourless glass, with an internal diameter of 13,5 mm, conforming to B.S. 625, fitted with rubber stoppers, for use in the Lovibond all-purpose 1 000 comparator.

(d) A water bath or incubator capable of being maintained at $37^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(e) A pipette to deliver 5,0 ml.

(f) A supply of 1,0 ml straight-sided pipettes.

(g) 1 l standard flask.

(h) A 100 ml standard flask.

(8) (a) After use, empty each test tube, rinse it in water, wash well in hot water containing soda, rinse in warm water, rinse in distilled water and dry, or clean it by some other equally effective method.

(b) If after treatment in accordance with (a) of this subparagraph a test tube does not appear to be clean, repeat the treatment, but, in addition after rinsing it in warm water, soak it in hydrochloric acid and then rinse

dan weer in warm water af voordat u dit in gedistilleerde water afspoel en dit droogmaak of voordat u dit volgens 'n ander, ewe doeltreffende, metode skoonmaak.

(c) Reinig nuwe glasware deur dit te dompel in 'n oplossing chroomsuur wat bestaan uit vyf volumes kalium-dichromaat van 8 persent (m/v) en uit vier volumes gekoncentreerde swaelsuur wat stadig en versigtig by die mengsel dichromaat en water gevoeg is.

(d) Sorg dat die oplossing in (c) van hierdie subparagraaf bedoel, bedek bly en dat, wanneer dit groen verkleur, die wegemaak word.

(e) Nadat die nuwe glasware gereinig is op die wyse in (c) hierbo beskryf, spoel dit dan eers in warm water en daarna in gedistilleerde water af en maak dit dan droog.

(f) wat pipette betref, spoel dit deeglik in koue water af, reinig dit daarna deur dit 24 uur lank te laat lê in 'n oplossing chroomsuur in 'n glassilinder of ander geskikte houer wat 250 ml hou, spoel die pipette daarna deeglik in warm water en in gedistilleerde water af en maak dit droog; of, so nie, reinig die pipette volgens 'n ander metode wat ewe doeltreffend is.

(g) Moenie glasware wat vir die ondersoek gebruik word, vir welke ander doel ook al gebruik nie en hou sodanige glasware afsonderlik van alle ander apparaat in die laboratorium.

(9) (a) Voer die ondersoek uit op die wyse wat in (b) tot en met (k) van hierdie subparagraaf beskryf word.

(b) Plaas, met behulp van 'n pipet, 5 ml van die buffer-substraatoplossing in 'n proefbuis; maak die proefbuis met 'n prop toe en verhit dit tot by 'n temperatuur van $37^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(c) Voeg by 1 ml van die melk of room wat getoets gaan word; sit weer die prop van die proefbuis op en meng die inhoud daarvan deeglik deur dit te skud.

(d) Inkubeer die proefbuis daarna 2 uur \pm een minuut lank by $37^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(e) Inkubeer saam met elke reeks monsters een kontrolemonster bestaande uit 5 ml buffersubstraatoplossing en 1 ml gekookte melk of room van dieselfde tipe as dié wat getoets word.

(f) Haal die proefbuis na die inkubasie uit die waterbad en meng die inhoud daarvan deeglik.

(g) Plaas die kontrolemonster op die linkerkantse kompartiment van die staander en die toetsmonster op die regterkantse een.

(h) Neem die lesings in weerkaatste lig deur op die twee openings af te kyk, met die vergelyker in die rigting van toereikende daglig gekeer.

(i) Indien daar vir die doel van vergelyking kunsmatige lig nodig is, gebruik dan beligting van 'n daglighetipe.

(j) Draai die skyf totdat die kleur van die toetsmonster met dié van die kontrolemonster klop.

(k) Wat betref die lesings wat tussen twee standaardstande val, teken elke sodanige lesing aan deur by die syfer vir die naaste standaardstand, na gelang van die geval, 'n plusteken of 'n minusteken te trek.

DIE METILEENBLOU-REDUKSIE-TOETS

(Gepasteuriseerde of gesteriliseerde of U.H.T.-melk)

7. (1) Die metileenblou-reduksietoets vir gepasteuriseerde, gesteriliseerde of U.H.T.-melk word uitgevoer op die wyse wat in onderstaande subparagrawe beskryf word.

(2) Berei die metileenblou-oplossing wat vir hierdie toets nodig is, op die wyse wat in paragraaf 3 (2) tot (6) beskryf word.

(3) (a) Meng die bottel of karton melk wat getoets moet word, deeglik; gooi, op aseptiese wyse, 'n monster van ongeveer 100 ml daarvan in 'n steriele wyebekbottel met 'n glasprop en met 'n houvermoë van ongeveer 150 ml.

(b) Hou die monster in 'n koelkas by 'n temperatuur van hoogstens 5°C totdat die toets moet begin.

it again in warm water before rinsing it in distilled water and drying it, or cleaning it by some other equally effective method.

(c) Clean new glassware by soaking it in a solution of chromic acid consisting of five volumes of 8 per cent (m/v) potassium dichromate and four volumes of concentrated sulphuric acid added slowly and carefully to the mixture of dichromate and water.

(d) Keep the solution referred to in (c) of this subparagraph covered and discard it when it turns green.

(e) After cleaning new glassware in the manner described in (c) above, rinse it in warm water, then rinse it in distilled water and then dry it.

(f) Pipettes should be well rinsed in cold water and then cleaned by soaking for 24 hours in a solution of chromic acid in a 250 ml glass cylinder or other suitable container, and then well rinsed in warm water, rinsed in distilled water and dried, or cleaned by some other equally effective method.

(g) Glassware used for the examination shall not be used for any other purpose and shall be kept separate from all other apparatus in the laboratory.

(9) (a) Carry out the examination in the manner described in (b) to (k) inclusive of this subparagraph.

(b) Transfer 5 ml of the buffer-substrate solution to a test tube by means of a pipette; stopper the test tube and bring it to a temperature of $37^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(c) Add 1 ml of the milk or cream to be tested, replace the stopper of the test tube and mix the contents well by shaking.

(d) Incubate the test tube for 2 hours \pm one minute at $37^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(e) With each series of samples incubate one blank prepared from 5 ml of buffersubstrate solution and 1 ml of boiled milk or cream of the same type as that undergoing the test.

(f) After incubation, remove the test tube from the water bath and mix its contents well.

(g) Place the blank on the left-hand ramp of the stand and the test sample on the right.

(h) Take readings in reflected light by looking down onto the two apertures, with the comparator facing a good source of daylight.

(i) If artificial light is needed for matching, use a daylight type of illumination.

(j) Revolve the disc until the colour of the test sample matches that of the control sample.

(k) Record readings falling between two standards by affixing a plus or minus sign to the figure for the nearest standard.

METHYLENE BLUE REDUCTION TEST

(Pasteurised or sterilised or U.H.T. milk)

7. (1) The methylene blue reduction test for pasteurised, sterilised or U.H.T. milk shall be carried out in accordance with the succeeding subparagraphs.

(2) Prepare the methylene blue solution required for this test in the manner described in paragraph 3 (2) to (6).

(3) (a) Thoroughly mix the bottle or carton of milk to be tested and pour off a sample of approximately 100 ml aseptically into a sterile, wide-mouthed, glass-stoppered bottle of approximately 150 ml capacity.

(b) Store this sample in a refrigerator at a temperature not exceeding 5°C until commencement of the test.

(4) Nadat u die temperatuur van die monster melk op $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ gebring het, inkubeer die monster dan 18 uur ± 15 minute lank in 'n broeikas of waterbad by $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(5) Pas die res van die toets toe op die wyse wat in paragraaf 3 (7) tot (15) beskryf word.

(6) 'n Monster melk wat, indien dit by $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ geïnkubeer word, die metileenblou binne minder as 4 uur ontkleur, word geag nie die toets te deurstaan het nie.

METILEENBLOU-REDUKSIE-TOETS

(Gepasteuriseerde of gesteriliseerde of U.H.T.-room)

8. (1) Die metileenblou-reduksie-toets vir gepasteuriseerde, gesteriliseerde of U.H.T.-room word uitgevoer op die wyse wat in onderstaande subparagrawe beskryf word.

(2) Berei die metileenblou-oplossing wat vir hierdie toets nodig is op die wyse wat in paragraaf 3 (2) tot (6) beskryf word.

(3) Stel die toets op om ongeveer 16h00 op die dag waarop die monster geneem word.

(4) Hou, totdat die toets op die punt is om te begin, die monster in 'n koelkas by 'n temperatuur van hoogstens 5°C .

(5) Plaas, met behulp van 'n steriele pipet, 7 ml Ringer-oplossing van kwartsterkte in elk van 3 steriele proefbuise wat 'n nominale grootte van 150 mm en 'n buitemiddellijn van 16 mm het en wat op 10 ml afgemerk is.

(6) Meng die monsterroom deeglik en pipetteer dit in elk van genoemde drie proefbuise oor tot by die 10-ml-mark. Een van genoemde drie buise word gebruik as die toetsmonsterbuis, een as 'n kontrolebuis en een as 'n blanko buis.

(7) Maak die proefbuise met steriele rubberproppe toe en meng die inhoud deur elke buis om te keer.

(8) Verhit die kontrolebuis 3 minute lank by kookpunt.

(9) Inkubeer die drie buise 18 uur lank ± 15 minute by $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ in 'n broeikas of waterbad.

(10) Voeg, na afloop van voornoemde 18 uur, 1 ml metileenblou by die toetsmonsterbuis en 1 ml metileenblou by die kontrolebuis. Plaas die drie buise oor in 'n waterbad teen $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$. Die blanko buis, wat geen metileenblou bevat nie, word gebruik om algehele ontkleurung aan te dui.

(11) Die waterhoogte in die waterbad moet slegs effens hoër wees as die hoogte van die inhoud van die buise.

(12) Ondersoek die geïnkubeerde buise room al om die 30 minute met die oog op kleurreduksie. Genoemde proses van kleurreduksie word geag afgeloop te wees wanneer, by vergelyking met die kontrolebuis, die hele kolom room tot binne 5 mm van die oppervlak daarvan ontkleur is.

(13) Indien die ontkleurung nie volledig is nie, keer die buis om en plaas dit in die waterbad terug.

(14) 'n Monster room wat, indien dit by $36^{\circ}\text{C} \pm 1,0^{\circ}\text{C}$ geïnkubeer word, die metileenblou binne minder as twee uur ontkleur, word geag nie die toets te deurstaan nie het nie.

(15) Berei die onderstaande wyse die Ringer-oplossing van kwartsterkte wat gebruik word vir die toets in hierdie paragraaf beskryf:

(a) Berei 'n Ringer-oplossing van volle sterkte deur—

natriumchloried B.P.: 9 g;

kaliumchloried: 0,42 g;

anhidriese kalsiumchloried: 240 mg;

natriumbikarbonaat: 200 mg;

in 1 l gedistilleerde water op te los.

(b) Voeg een deel van die Ringer-oplossing van volle sterkte by drie dele gedistilleerde water.

(c) Giet die oplossing wat in (b) van hierdie subparagraaf genoem word, in bottels met 'n houvermoë van 100 ml en verhit die bottels 15 minute lank in 'n outoklaaf by 121°C .

(4) Incubate the sample of milk in an incubator or a water bath at $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ for 18 hours ± 15 minutes after adjusting the temperature of the milk to $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$.

(5) Carry out the remainder of the test as specified in paragraph 3 (7) to (15).

(6) Samples of milk which decolourise the methylene blue in less than four hours' incubation at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$ shall be deemed to have failed the test.

METHYLENE BLUE REDUCTION TEST

(Pasteurised, sterilised or U.H.T. cream)

8. (1) The methylene blue reduction test for pasteurised, sterilised or U.H.T. cream shall be carried out in accordance with succeeding subparagraphs.

(2) Prepare the methylene blue solution required for this test in the manner described in paragraph 3 (2) to (6).

(3) Set up the test at approximately 16h00 on the day on which the sample is taken.

(4) Store the sample in a refrigerator at a temperature not exceeding 5°C until the test is about to be set up.

(5) Into each of three sterile test-tubes of a nominal size of 150 mm, with a 16 mm outer diameter, and a mark at the 10 ml level, introduce 7 ml of one quarter strength Ringer's Solution by means of a sterile pipette.

(6) Thoroughly mix the sample of cream and pipette it into each of the said three test tubes up to the 10 ml mark. One of the said three tubes shall be used as the test sample tube, one as a control tube, and one as a blank tube.

(7) Close the test tubes with sterile rubber stoppers and mix by inverting the tubes.

(8) Heat the tube used as a control at boiling point for three minutes.

(9) Incubate the three tubes in an incubator or water bath at $18^{\circ}\text{C} \pm 0,5^{\circ}\text{C}$ for 18 hours ± 15 minutes.

(10) At the end of the aforesaid 18 hours add 1 ml of methylene blue to the test sample tube and 1 ml to the control tube. Transfer the three tubes to a water bath at $36^{\circ}\text{C} \pm 1^{\circ}\text{C}$. The blank tube, containing no methylene blue, will serve to indicate complete decolourisation.

(11) The level of the water in the water bath should just exceed the level of the contents in the tubes.

(12) Examine the incubated tubes of cream every 30 minutes for dye reduction, which process shall be taken to be complete when the whole column of cream is decolourised up to within 5 mm of the surface when compared with the control tube.

(13) If decolourisation is not complete, invert the tube and replace it in the water bath.

(14) Samples of cream which decolourise the methylene blue in less than two hours' incubation at $36^{\circ}\text{C} \pm 1,0^{\circ}\text{C}$ shall be deemed to have failed the test.

(15) The quarter-strength Ringer's Solution used for the test described in this paragraph shall be prepared as follows:

(a) Prepare full-strength Ringer's Solution by dissolving—

sodium chloride B.P.: 9 g;

potassium chloride: 0,42 g;

anhydrous calcium chloride: 240 mg;

sodium bicarbonate: 200 mg;

in 1 l of distilled water.

(b) Add one part of full-strength Ringer's Solution to three parts of distilled water.

(c) Pour the solution referred to in (b) of this sub-paragraph into 100 ml bottles and autoclave them at 121°C for 15 minutes.

DIE TOETS VIR KOLIVORMIGE BAKTERIEË

9. (1) Die toets vir kolivormige bakterieë vir gepasteuriseerde melk, gepasteuriseerde room en melkprodukte moet toegepas word op die wyse wat in onderstaande subparagrafe beskryf word, en vir die toepassing van hierdie paragraaf beteken die uitdrukking "kolivormige bakterieë" aërobiese en fakultatief anaërobiese, Gram-negatiewe, nie-spoorvormende staafbakterieë wat in staat is om laktose in die aanwesigheid van galsoute te laat fermenteer en wat indien dit by 30 °C of 37 °C geïnkubeer word, binne onderskeidelik 12 of 48 uur suur en gas produseer.

(2) Meng die melkvoorraad deeglik alvorens 'n monster daarvan te neem.

(3) Berei die monsters soos volg:

(a) Meng die monsters melk, afgeroomde melk, karrimelk of room deeglik. Indien die room te dik is om maklik te hanteer, verwarm dit tot by 'n temperatuur van hoogsens 37 °C.

(b) Meng die viskeuse of halfvaste aangesuurde melkprodukte deeglik en plaas 11 g van die produk in 'n steriele wyebekhouer. Voeg dan 89 ml verwarmde (40 °C) steriele 2-percen(m/v)-natriumsitraatoplossing by en skud die mengsel totdat dit egalig vermeng is.

(4) Berei violetrooi gal-agar soos volg:

Gisekstrak: 3 g.

Pepton: 7 g.

Galsoute: 1,5 g.

Laktose: 10 g.

Natriumchloried: 5 g.

Neutraalrooi: 30 mg.

Kristalviolet: 2 mg.

Agar: 15 g.

Gedistilleerde water tot: 1 l.

Meng die bestanddele deeglik en reguleer die pH op 7,4; verwarm die mengsel terwyl u dit roer, en kook dit dan twee minute lank; laat dit tot ongeveer 45 °C afkoel en gebruik dit as 'n plaatmedium.

(5) Bring 1 ml van die monster, of 'n desimale volume van genoemde hoeveelheid, oor in elk van twee steriele bakkies. Voeg by die inhoud van iedere bakkie 10 tot 15 ml violetrooi gal-agar of McConkey-agar waarvan die temperatuur tussen 44 °C en 46 °C is.

(6) Meng die inhoud van die bakkies deeglik deur die bakkies te kantel en in die rondte te draai. Laat die mengsel vinnig stol, en plaas dan nog 3 tot 4 ml van die plaatmedium bo-oor die gestolde medium sodat die gestolde medium heeltemal daarmee bedek is.

(7) Keer die bakkies om en inkubeer hulle 24±2 uur lank by 32 °C±1 °C.

(8) Donkerrooi kolonies met 'n diameter van 0,5 mm of meer in ylbevolkte bakkies word geag kolivormige bakterieë te wees. Tel slegs sodanige kolonies, in bakkies wat verkiesslik uiters 150 kolonies bevat. Druk die resulataat, na gelang van die gevall, as die getal kolivormige bakterieë per ml of per g uit.

(9) Produkte waarby suur ontwikkel het, moet getoets word binne 48 uur nadat hulle vervaardig is.

STOL-BY-KOOK-TOETS

10. (1) Meng die melk deeglik alvorens 'n monster te neem.

(2) Meet 5 ml melk in 'n proefbuis af.

(3) Plaas die proefbuis in kookwater.

(4) Die vlak van die kookwater moet hoër wees as die vlak van die melk.

(5) Laat die melk vyf minute lank in die kookwater staan.

(6) Haal die proefbuis uit die water en kantel dit tot in 'n bykans horizontale posisie sonder om die melk in die buis te skud.

(7) Wag totdat daar 'n dun vlies op die melk gevorm is.

THE COLIFORM TEST

9. (1) The coliform test for pasteurised milk, pasteurised cream, and milk products shall be carried out in accordance with the succeeding subparagraphs, and for the purposes of this paragraph the term 'coliform bacteria' shall mean aerobic and facultatively anaerobic, Gram-negative, non-sporeforming rods capable of fermenting lactose in the presence of bile salts and producing acid and gas at 30 °C or 37 °C within 12 or 48 hours of incubation respectively.

(2) Mix milk thoroughly before sampling from bulk.

(3) Prepare samples as follows:

(a) Thoroughly mix samples of milk, skimmed milk, buttermilk or cream. If the cream is too thick for convenient handling it may be warmed to a temperature not exceeding 37 °C.

(b) Thoroughly mix viscous or semi-solid cultured milk products, and place 11 g of the product in a sterile wide-mouthed container. Then add 89 ml of heated (40 °C) sterile 2 per cent (m/v) sodium citrate solution and shake the mixture until a homogeneous dispersion is obtained.

(4) Prepare violet red bile agar as follows:

Yeast extract: 3 g.

Peptone: 7 g.

Bile salts: 1,5 g.

Lactose: 10 g.

Sodium chloride: 5 g.

Neutral red: 30 mg.

Crystal violet: 2 mg.

Agar: 15 g.

Distilled water to: 1 l.

Mix thoroughly and adjust to pH 7,4; heat with agitation and boil for two minutes; cool to about 45 °C and use as a plating medium.

(5) Transfer 1 ml of the sample or a decimal volume of the afore-mentioned quantity in duplicate into sterile dishes. To each dish add 10 to 15 ml of violet red bile agar or McConkey's agar the temperature of which shall be from 44 °C to 46 °C.

(6) Mix the contents of the dishes thoroughly by tilting and rotating. Allow the mixture to solidify promptly, then pour an additional 3 to 4 ml of the plating medium as an overlay, completely covering the surface of the solidified medium.

(7) Invert and incubate the dishes for 24±2 hours at 32 °C±1 °C.

(8) Dark red colonies measuring 0,5 mm or more in diameter on uncrowded dishes are considered to be coliform bacteria. Count such colonies only, preferably in dishes containing not more than 150 per dish. Express the result as the number of coliform bacteria per ml or per gram, as the case may be.

(9) Products with developed acidity shall be tested within 48 hours of their manufacture.

THE CLOT-ON-BOILING TEST

10. (1) Thoroughly mix milk before sampling.

(2) Measure 5 ml of milk into a test tube.

(3) Place tube in boiling water.

(4) The water level must be higher than the milk level.

(5) Leave milk for five minutes in boiling water.

(6) Remove test tube from water and tilt almost horizontally without shaking the milk in the tube.

(7) Observe the thin milk film exposed.

(8) Die resultaat is positief indien al die melk in die buis stol of indien vlokkies teen die kante van die proefbuis waargeneem word wanneer die buis na 'n vertikale posisie teruggebring word. Weens die invloed wat die toenemende suurheid op die hittestabiliteit van die kaseïen uitoefen, lever die toets by 'n pH van 5,89 (gelykwaardig met ongeveer 0,24 persent melksuur) 'n positiewe resulataat. Kolostrum in die melk sal eweneens 'n positiewe stolby-kook-toets tot gevolg hê. Ook ander faktore beïnvloed die stabiliteit van kaseïen teenoor hitte.

SOMATIESE SELTELLING

11. Ondergenoemde twee metodes is algemeen in gebruik:

A. *Direkte mikroskopiese bepaling van somatiese selinhoud van melk.*

(Breed-Prescott-smeermetode.)

B. *Elektroniese seltelling.*

("Coulter Counter".)

(A) *Direkte mikroskopiese bepaling van somatiese selinhoud van melk*

(1) Spoel smeerglasies in alkohol af en laat hulle droog word. Trek elke glasie deur 'n vlam voor gebruik.

(2) Meng die monster deur die houer 10 keer deur 180° te beweeg. Moenie die houer skud nie.

(3) Stryk 0,01 ml melk oor 1 cm² uit. Gebruik of 'n gegradeerde platinalussie of 'n mikropipet (Breed Smear Syringe, 0,01 ml, Applied Research Institute, New York, U.S.A.). Maak vier smere van 1×1 cm of agt smere van 0,5×2 cm per glasie.

(4) Droog die smere in 'n horizontale posisie in 'n partikelvrye, warm atmosfeer (maksimum temperatuur 45 °C).

(5) Kleur en ontvet die gedroogte smere 2–3 minute lank in "Single Stain Solution".*

(6) Maak die smere vinnig en deeglik droog, bv. met behulp van 'n haardroëer.

(7) Spoel die smere in kraanwater by 37 °C af, en herhaal (6).

(8) Plaas elke smeer onder die olie-immersielens van 'n mikroskoop waarvan die mikroskopiese faktor (MF) bekend is.

$$M.F. = \frac{10\ 000}{\pi r^2}, \text{ waar } r = \text{straal van mikroskopiese veld.}$$

Bv.

Middellyn (2×r)

M.F.

0,206 mm.....	300 000
0,170.....	400 000
0,160.....	500 000
0,146.....	600 000

(9) Beweeg die platform van die mikroskoop van links na regs en tel die somatiese selle in elke derde veld totdat die selle in 10 velde getel is.

(10) Herhaal die telling van bo na onder totdat 10 velde getel is.

(11) Bereken die gemiddelde totale getal selle in 10 velde (T) en vermenigvuldig T met WF, waar $WF = \frac{MF}{10}$

$T \times WF = \text{getal selle/ml melk}$

"Single Stain Solution"*

Etielalkohol (96%): 520 ml.

Tetrachloretaan: 440 ml.

Metileenblouchloried: 6 g.

Ysasynsuur: 40 ml.

(8) When the whole of the milk clots or when flocules are seen to be adhering to the side of the tube when it is returned to the vertical position, the result is positive. At pH 5,89 (equivalent to some 0,24 per cent lactic acid) the test will give a positive result owing to the influence of increased acidity on the heat stability of casein. Colostrum in milk will also cause milk to test COB+. Other factors also influence the stability of casein to heating.

SOMATIC CELL COUNT

11. The following two methods are generally used:

A. *Direct microscopic determination of somatic cell content of milk.*

(Breedt-Prescott smear method.)

B. *Electronic cell count.*

("Coulter Counter".)

(A) *Direct microscopic determination of the somatic cell content of milk*

(1) Rinse the smearslides in alcohol and leave to dry. Pass each slide through flame before use.

(2) Mix the sample by inverting the container 10 times through 180°. Do not shake.

(3) Spread 0,01 ml of milk over 1 cm². Use a graded platinum loop or a micro-pipette (Breed Smear Syringe, 0,01 ml, Applied Research Institute, New York, U.S.A.). Prepare four 1×1 cm smears or eight 0,5×2 cm smears per slide.

(4) Dry smears in a horizontal position in a particle-free, warm atmosphere (maximum temperature 45 °C).

(5) Kleur en ontvet die gedroogde smere 2–3 minute in Single Stain Solution*.

(6) Dry rapidly and thoroughly, e.g. by using a hair drier.

(7) Rinse in tap water at 37 °C and repeat (6).

(8) Place each smear under the oil-immersion lens of a microscope of which the microscopic factor (M.F.) is known.

$$M.F. = \frac{10\ 000}{\pi r^2}, \text{ where } r = \text{radius of microscopic field.}$$

e.g.

Diameter (2×r)	M.F.
0,206 mm.....	300 000
0,170.....	400 000
0,160.....	500 000
0,146.....	600 000

(9) Move the platform of the microscope from left to right and count the number of somatic cells in every third field until the cells in 10 fields have been counted.

(10) Repeat counting from top to bottom until 10 fields have been counted.

(11) Calculate the average total number of cells per 10 fields (T) and multiply T by WF, where $WF = \frac{MF}{10}$

$T \times WF = \text{number of cells/ml of milk.}$

*Single Stain Solution**

Ethyl alcohol (96%): 520 ml.

Tetrachlorethane: 440 ml.

Methylene blue chloride: 6 g.

Glacial acetic acid: 40 ml.

Meng eers die etielalkohol en tetracholoretaan in 'n 2-l-fles. Voeg die kleurstof by, meng goed en bêre die mengsel 24 uur lank in 'n yskas by 4–5 °C. Voeg dan ysasynsuur by, filtreer (Whatman No. 40 of soortgelyke filter), verdeel en hou in yskas in digte flesse.

(B) Elektroniese somatiese melkseltelling

Verskeie metodes word teenswoordig gebruik. Al die metodes berus op die elektroniese telling van partikels of van werklike somatiese selle. Ten einde slegs sodanige partikels te tel as waarom die grootte met dié van somatiese selle gelykwaardig is, is die partikelsteller afhanglik van noukeurige instelling. Partikelvrye monsters, verdunningsmiddels, additiewe en glasware is noodsaklik. Die volgende metode is algemeen in gebruik:

(1) Meng die melkvoorraad deeglik alvorens die monster te neem.

(2) Plaas ongeveer 10 ml van die monster in 'n partikelvrye houer met 'n houvermoë van ongeveer 20–30 ml. Verkoel totdat stap (3) begin.

(3) Voeg fikseer middel by die monster en laat die mengsel 18–24 uur lank by 20 °C staan. Die mengsel kan nou 5–7 dae lank by kamertemperatuur gehou word.

(4) Berei die monster in 'n verdunningskonsentrasie van 1:100 deur 0,1 ml van die monster in (1), (2) en (3) bedoel, by 10 ml van 'n vetoplosmiddel-elektrolytmengsel te voeg, meng dit deur 10 maal om te keer en laat dit 10 minute lank in 'n waterbad by 'n temperatuur van 56–60 °C staan. Sorg dat die vlak in die proefbuis onderkant die watervlak in die bad is.

Haal die monster uit die bad, meng deeglik (drie sekondes lank met die "whirlmix") en laat teen kamertemperatuur afkoel.

(5) Meng die inhoud nogeens deur die houer saggies om te keer, en neem dan 'n telling in die partikelsteller ("Coulter Teller" of soortgelyke een). Voer, met dieselfde monster, die telprosedure 'n tweede keer uit. Indien die tweede lesings aldus verkry van mekaar verskil met meer as 10 persent, herhaal dan die telprosedure totdat twee lesings verkry word wat nagenoeg aan mekaar is—bereken die gemiddelde van sodanige twee lesings en teken dit aan as telling/cm³ melk.

(6) Die tellers met behulp waarvan die somatiese selteling uitgevoer word, moet toegerus wees sowel met 'n monsternemingsbuis waarvan die opening 'n deursnee van 100 µ het as met 'n manometer wat die toetssuspensie in hoeveelhede van 0,1 of 0,5 cm³ kan meet. 'n Gesikte drempelwaarde vir die partikelgroote berus op die soort of model teller, en moet bepaal en ingestel word deur die verskaffer van die teller. Gewoonlik wissel genoemde drempelwaarde tussen 4,7 en 5,2 m. Slegs gestandaardiseerde en gekalibreerde tellers moet gebruik word.

STANDAARD-AGAR-PLAATKOLONIETELLING

12. (1) Meng die melkvoorraad deeglik alvorens 'n monster daarvan te neem.

(2) Beweeg die monsterhouer 10 keer deur 180°. Trek 1 ml van die melk af, bring oor in 'n proefbuis wat 9 ml steriele verdunningsmiddel, naamlik fosfaatbuffer* by pH 7,2, bevat, en berei vier tienvoudige verdunnings ooreenkomsdig die onderskeie proporsies 1:10, 1:100, 1:1 000, en 1:10 000.

(3) Giet, met behulp van 'n skoon pipet, 1 ml van elk van genoemde verdunnings in 'n afsonderlike steriele petribakkie deur met die hoogste konsentrasie te begin en met die laagste te eindig.

(4) Giet in elk van die vier petribakkies in (3) bedoel, 10 ml Standaard-plaattelling-agar,** wat vooraf gesmelt en tot by 45 °C afgekoel is.

First mix the ethyl alcohol and tetrachlorethane in a 2 l flask. Add the colourant, mix well and keep in the refrigerator at 4–5 °C for 24 hours. Then add the glacial acetic acid, filter (Whatman No. 40 or similar), divide and keep in refrigerator in sealed flasks.

(B) Electronic somatic milk cell counting

Various methods are used at present. All depend on electronic counting of particles or of actual somatic cells. The particle counter depends on proper setting to count only particles of a size equivalent to somatic cells. Particle-free samples, diluents, additives and glassware are imperative. The following is a commonly used method:

(1) Thoroughly mix milk supply to be sampled.

(2) Draw about 10 ml of the sample into a particle-free container of about 20–30 ml capacity. Keep refrigerated until step (3).

(3) Add fixative and stand at 20 °C for 18–24 hours. The mixture may then stand at room temperature for 5–7 days.

(4) Prepare a 1:100 dilution of the sample by adding 0,1 ml of the sample referred to in (1), (2) and (3) to 10 ml of a fatsolvent-electrolyte mixture, invert 10 times to mix and stand in a water bath at 56–60 °C for 10 minutes. Ensure that the level in the tube is *below* the level of water in the bath. Remove the sample from the bath, mix thoroughly (three seconds on "Whirlmix") and allow to cool at room temperature.

(5) Mix the contents again by gentle inversion of the container and proceed to count the cells in the particle counter (Coulter Counter or similar). Repeat cell count on sample. If readings differ by more than 10 per cent, repeat until two similar readings are obtained—calculate the average of the two readings and record count/cm³ of milk.

(6) Counters for somatic cell counting should be fitted with a 100µ orifice sampling tube and a manometer capable of metering 0,1 or 0,5 cm³ volumes of test suspension. An appropriate threshold value for the particle size depends on the type or model of counter, and should be determined and set by the supplier of the counter. This threshold value will usually vary from 4,7 to 5,2µm. Only standardised and calibrated counters should be used.

STANDARD AGAR PLATE COLONY COUNT

12. (1) Mix milk thoroughly before sampling from bulk.

(2) Invert sample container 10 times through 180°. Remove 1 ml of the milk, transfer to test tube containing 9 ml of sterile diluent, viz phosphate buffer* (pH 7,2) and make the following four tenfold dilutions: 1:10, 1:100, 1:1 000, 1:10 000.

(3) Using a fresh pipette, transfer 1 ml of each of the dilutions to each of four sterile Petri dishes, beginning with the highest concentration, and finishing with the lowest.

(4) To each Petri dish add 10 ml of Standard Plate Count Agar** previously melted and cooled to 45 °C.

(5) Meng die inhoud van elke bakkie deeglik deur middel van horisontale draaibewegings terwyl die medium nog vloeibaar is.

(6) Sodra die medium stol, keer die bakkies om en inkubeer hulle 72 uur lank by 30 °C – 32 °C of 48 uur lank by 37 °C.

(7) Verwyder die bakkies uit die broeikas by verstryking van die inkubasietydperk en tel die kolonies onder egale kunsmatige beligting met behulp van vergroting.

(8) Kies, om die kolonies te tel, spreiervrye bakkies uit wat 30–300 kolonies bevat; tel al die kolonies en bereken die aantal lewensvatbare bakterieë.

(9) Indien daar aansienlik meer as 300 kolonies per bakkie is, tel die kolonies op daardie gedeeltes van die bakkies wat verteenwoordigend is vir die kolonieververspreiding, en beraam die totale aantal vir elke bakkie dienoordeekomstig.

*Fosfaatbufferoplossing

KH_2PO_4 : 5,08 g.
 Na_2HPO_4 : 13,63 g.
 in 2 l gedistilleerde water.

**Plaattelling-agar

Triptoon (pankreasverteerde Kasseien): 5 g.
 Gisekstrak: 2,5 g.
 Glukose: 1 g.
 Agar (bakteriegraad): 15 g.
 Gedistilleerde water tot: 1 l.

Finale pH van gesterilliseerde medium: 7,0±0,1.
 Steriliseer 15 minute lank by 121 °C.

Titreerbare suurtegraad

13. (1) Pipetteer 20 ml melk in 'n keëlformige fles met 'n houvermoë van 250 ml.

(2) Voeg 40 ml koolstofdioksiedvrye gedistilleerde water by.

(3) Voeg dan 2 ml fenolftaleien-indikator by.

(4) Titreer met 'n 0,1N NAOH-oplossing totdat die eerste pienk tint verskyn.

(5) Druk die resultaat van die toets as g melksuur per 100 g melk uit met behulp van die faktor dat 1 ml 0,1N NAOH gelykwaardig is met 0,0090 g melksuur.

PASTEURISERING

14. (1) Die pasteurisering van melk word uitgevoer of—

(a) deur elke deeltjie van die melk tot 'n temperatuur van minstens 63 °C te verhit en oor 'n tydsduur van minstens 30 minute by genoemde temperatuur te hou; hierdie proses word hieronder die "houproses" genoem; of

(b) deur elke deeltjie van die melk tot minstens 72 °C te verhit en oor 'n tydsduur van minstens 15 sekondes by genoemde temperatuur te hou; hierdie proses word hieronder "die hoëtemperatuur-snelproses" genoem; of

(c) volgens sodanige ander metode as wat by regulasie voorgeskryf word:

Met dien verstande dat melk in geen geval as gepasteuriseer beskou word nie tensy dit die Aschaffenburg-en-Mullen-fosfatase-toets wat in paragraaf 6 van hierdie Aanhangsel beskryf word, deurstaan.

(2) In die geval van melk, room, ander melkprodukte of saamgestelde suiwelprodukte wat bygevoegde versooeters bevat, word die pasteurisering uitgevoer of—

(a) deur elke deeltjie van die produk tot 'n temperatuur van minstens 66 °C te verhit en oor 'n tydsduur van minstens 30 minute by genoemde temperatuur te hou; of

(5) Mix the contents of each dish thoroughly by horizontal rotational movements, while the medium is still liquid.

(6) When the medium is set invert the dishes and incubate at 30 °C–32 °C for 72 hours or at 37 °C for 48 hours.

(7) At the end of the incubation period remove the dishes from the incubator and count the colonies with the aid of magnification under uniform artificial illumination.

(8) For counting, select spreader-free dishes with 30–300 colonies, count all the colonies and calculate the number of viable bacteria.

(9) If the colonies appreciably exceed 300 per dish, count the colonies in portions of the dish that are representative of colony distribution and estimate the total number per plate accordingly.

*Phosphate buffer:

KH_2PO_4 : 5,08 g.
 Na_2HPO_4 : 13,63 g.
 in 2 l of distilled water.

**Plate count agar

Tryptone (pancreatic-digestive casein): 5 g.
 Yeast extract: 2,5 g.
 Glucose: 1 g.
 Agar (bacterial grade): 15 g.
 Distilled water: 1 l.

Final pH of sterilised medium 7,0±0,1.
 Sterilise for 15 minutes at 121 °C.

Titratable acidity

13. (1) Pipette 20 ml of milk into a 250 ml conical flask.

(2) Add 40 ml of carbon dioxide-free distilled water.

(3) Add 2 ml of phenolphthalein indicator.

(4) Titrate with a 0,1N NAOH solution until the first tinge of pink colour appears.

(5) Using the factor that 1 ml of 0,1N NAOH is equivalent to 0,0090 g of lactic acid, express the result of the test as g of lactic acid per 100 g of milk.

PASTEURISATION

14. (1) The pasteurisation of milk shall be performed by—

(a) heating every particle of the milk to a temperature of at least 63 °C and holding it at that temperature for not less than 30 minutes (a process hereinafter referred to as the "holder method" or the "batch method");

(b) heating every particle of milk to and holding it at a temperature of at least 72 °C for at least 15 seconds (a process hereinafter referred to as the "high-temperature short-time method"); or

(c) such other method as may be prescribed by regulation:

Provided that no milk shall in any instance be deemed to have been pasteurised if it fails to pass the Aschaffenburg and Mullen phosphatase test described in paragraph 6 of this Annex.

(2) In the case of cream or milk, milk products or composite dairy products containing added sweeteners, pasteurisation shall be performed by—

(a) heating every particle of the product to a temperature not lower than 66 °C and holding it at that temperature for not less than 30 minutes;

(b) deur elke deeltjie van die produk tot 'n temperatuur van minstens 74 °C te verhit en oor 'n tydsduur van minstens 15 sekondes by genoemde temperatuur te hou; of

(c) volgens sodanige ander metode as wat by regulasie voorgeskryf word:

Met dien verstaande dat sodanige produk in geen geval as gepasteuriseer beskou word nie tensy dit die Aschaffenburg-en-Mulen-fosfatasetoets wat in paragraaf 6 van hierdie Bylae beskryf word, deurstaan.

(3) Alle gepasteuriseerde melk, melkprodukte of saamgestelde suiwelprodukte, uitgesonderd dié wat vir kuituurenting bestem is, word, onmiddellik nadat dit gepasteuriseer is, in goedgekeurde toerusting verkoel en word, totdat dit die melkwinkel verlaat, by 'n temperatuur van hoogstens 7 °C gehou.

(4) Die pasteurisering moet met behulp van aanvaarde apparaat en instrumente uitgevoer en beheer word en sowel die apparaat en as die instrumente moet behoorlik gebruik en in 'n goeie toestand gehou word.

(5) Die pasteuriseerproses moet, indien dit volgens die hoëtemperatuur-snelproses metode uitgevoer word, meganies beheer word wat betref sowel die temperatuurbestek van die melk as die tydsuur waaroorheen die melk by die betrokke temperatuur gehou word.

(6) Die apparaat waarmee melk gepasteuriseer word, moet so ontwerp wees en so gebruik word, en moet met sodanige beheertoestelle toegerus wees, dat elke deeltjie van die melk oor die voorgeskrewe tydsuur heen aan die voorgeskrewe temperatuur blootgestel word.

AANHANGSEL B

I	II	III Voorwaarde en toleransies mg/kg
Melk en melkprodukte	Veroorloofde additiewe	
Aangesuurde melk	Stabiliseerders Natrium-, kalium-, en kalsiumsoute van: Soutsuur Siroensuur Koolsuur Ortofosfousuur Polifosfousuur Kleurstowwe Betakaroteen Koperkompleks van chlorofiel Riboflavin Kleurstowwe Annatto-ekstrakte Betakaroteen Kurkumien of borrie Neutraliseersoute Kalsiumhidroksied Natriumortofosfaat Natriumkarbonaat Natriumbikarbonaat Natriumhidroksied Antiöksideermiddels Butielhidroksie-anisoel (BHA) Butielhidroksie-tolueen (BHT) Propiel-, oktiel- en dodesielgallate Tertiële butielhidroki-noon (TBHQ)	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe. 600. GVP.
Blouaarkaassoorte		
Botter en weibotter	GVP.	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.
Botterolie...	200, afsonderlik of in kombinasie; maar in produkte wat nie bedoel is vir direkte gebruik of vir gebruik in gerekonstitueerde melk of gerekonstitueerde melkprodukte nie, moet die gallate nie 100 oorskry nie.	
Cheddar-kaas	Bederfwerende middel Sorbiensuur Pimaricin	1 000. 2, in die skil sonder plastiekbedekking. 500 in die plastiekbedekking. 10 binne-in.

(b) heating every particle of the product to a temperature not lower than 74 °C and holding it at that temperature for at least 15 seconds; or

(c) such other method as may be prescribed by regulation:

Provided that no such product shall in any instance be deemed to have been pasteurised if it fails to pass the Aschaffenburg and Mullen phosphatase test described in paragraph 6 of this Annex.

(3) All pasteurised milk, milk products or composite dairy products, except those to be cultured, shall, immediately after pasteurisation, be cooled in approved equipment and shall be maintained at a temperature not exceeding 7 °C until they leave the milk shop.

(4) Pasteurisation shall be carried out and controlled by means of accepted apparatus and instruments, and both apparatus and instruments shall be properly operated and maintained in good order and repair.

(5) The process of pasteurisation, if carried out according to the high-temperature short-time method, shall be mechanically controlled in respect of the temperature range of the milk and in respect of the period for which the milk is held at that temperature.

(6) Apparatus used for pasteurising milk shall be so designed and operated that every particle of the milk is subject to the prescribed temperature range for the prescribed period, and shall be provided with controls adequate to ensure this.

ANNEX B

I	II Permitted additives	III Conditions and tolerances mg/kg
Milk and milk products		
Butter and whey butter	Colourants Anatto extracts Beta-Carotene Curcumin or turmeric Neutralising salts Sodium orthophosphate Sodium carbonate Sodium bicarbonate Sodium hydroxide Calcium hydroxide Buttermilk..	GMP. 2 000, singly or in combination, expressed as anhydrous substances.
Butter oil...	Neutralising substances Sodium orthophosphate Sodium carbonate Sodium bicarbonate Sodium hydroxide Calcium hydroxide Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid Anti-oxidants Butylated hydroxyanisole (BHA) Butylated hydroxytoluene (BHT) Propyl, octyl and decyl gallates Tertiary butylhydroquinone (TBHQ)	2 000, singly or in combination, expressed as anhydrous substances.
Cheese, Cheddar	Preservatives Sorbic acid Pimaricin	2 000, singly or in combination, but gallates not to exceed 100 in products not intended for direct consumption or for use in reconstituted milk or reconstituted milk products.
	Colourants Anatto extracts Beta-Carotene	1 000. 2 in the rind without plastic coating. 500 in the plastic coating. 10 in the interior. 600.

I	II	III	I	II	III
Melk en melkprodukte	Veroorloofde additiewe	Voorwaardes en toleransies mg/kg	Milk and milk products	Permitted additives	Conditions and tolerances mg/kg
Cheddarkaas (vervolg)	Kleurstowwe Anatto-ekstrakte Betakaroteen	600.	Cheeses, various (Edam, Gouda, Tilsiter, Limburger)	Colourants Anatto extracts Beta-Carotene Preservatives Potassium nitrate, sodium nitrate Pimaricin	600.
Cheshire-kaas	Bederfwerende middel Sorbiensuur Pimarisenien	1 000. 2, in die skil sonder plastiekbedekking. 500 in die plastiekbedekking. 10 binne-in.	Cheese, Cheshire	Preservatives Sorbic acid Pimaricin	200. 2 in the rind without plastic coating. 500 in the plastic coating. 10 in the interior.
Hardekaas..	Bederfwerende middel Sorbiensuur Pimarisenien	1 000. 2, in die skil sonder plastiekbedekking. 500 in die plastiekbedekking. 10 binne-in.	Cheese, blue-veined	Colourants Beta-Carotene Chlorophyll copper complex Riboflavin Preservatives Sorbic acid Pimaricin	1 000. 2 in the rind without plastic coating. 500 in the plastic coating. 10 in the interior.
Joghurt.....	Kleurstowwe Kyk melk, gegeurde Bederfwerende middels Sorbiensuur Bansoësuur Swaweldioksied Pimarisenien Stabiliseerders Natrium, kalium-, en kalsiumsoute van: Koolsuur Siroensuur Soutsuur Ortofosforsuur Polifosforsuur Versoeters Veroorloofde natuurlike en/of kunsmatige versoeters	Kyk melk, gegeurde. 1 000. 50 in die eindproduk. 10. 5 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Hard grating cheese	Colourants Annatto extracts Beta-Carotene Carotenes (except betacarotene) Chlorophyll Chlorophyll copper complex Iron oxides Pigment Rubine Lithol Rubine BK Harmless flavouring substances not derived from milk Preservatives Nisin Potassium and sodium nitrate	3 000. 2 in the rind without plastic coating. 500 in the plastic coating. 10 in the interior.
Kaas (behalwe waar anders aangedui)	Kleurstowwe Anatto-ekstrakte Betakaroteen Chlorofil Karotene (uitgesonderd betakaroteen) Koperkompleks van chlorofil Litolrubien BK Pigmentrubien Ysteroksiede Skadelose geurmiddels nie van melk afkomstig nie Bederfwerende middels Nisien Kalium- en natriumnitraat Waterstofperoksied	600. GVP.	Soos in regulasie voorgeskrif.	Hydrogen peroxide Hexamethylene-tetramine Sulphur dioxide Pimaricin	On the rind only.
	Heksametileentetramien Swaweldioksied Pimaricin	Slegs aan die skil. GVP.	Cottage cheese	Propionic acid Calcium sorbate Benzoic acid	GMP.
	Propioonsuur Kalsiumsorbaat	12,5 is gelyk aan 500 i.e.g. 200 mg/kg van die melk wat gebruik word, afsonderlik of in kombinasie, bereken as natriumnitraat. Nie gespesifieer nie (residu word deur middel van katalase vernietig).		Harmless flavouring substances not derived from milk Preservatives Sorbic acid and Na and K salts Pimaricin Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid	12,5 equals 500 i.u./g. 200 mg/kg of the milk used, singly or in combination, calculated as sodium nitrate.
	Bensoësuur	2 000. 2, in die skil sonder plastiekbedekking. 500, in die plastiekbedekking. 10 binne-in. 2 000. Vanuit ensiempreparatie oorgedra. Vanuit ensiempreparatie oorgedra.			Not specified (residue destroyed using catalase). GMP.
Karringmelk	Neutraliseerstowwe Kalsiumhidroksied Natriumortofosfaat Natriumkarbonaat Natriumbikarbonaat Natriumhidroksied	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Cheese, cream	Harmless flavouring substances not derived from milk Preservatives Sorbic acid and Na and K salts Pimaricin	2 000, singly or in combination, expressed as anhydrous substances.
					GMP.
					1 000 calculated as sorbic acid. 10.

I	II	III	I	II	III
Melk en melkprodukte	Veroorloofde additiewe	Voorwaardes en toleransies mg/kg	Milk and milk products	Permitted additives	Conditions and tolerances mg/kg
Karringmelk (vervolg)	Stabiliseerders Natrium-, kalium-, en kalsiumsoute van: Koolsuur Ortofosfousuur Polifosfousuur Sitroensuur Soutsuur	2 000, afsonderlike of in kombinasie, uitgedruk as watervrye stowwe.	Cheese, cream (cont.)	Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid	2 000, singly or in combination, expressed as anhydrous substances.
Kondensmelk (a) versoet	Stabiliseerders Natrium-, kalium- en kalsiumsoute van: Soutsuur Sitroensuur Koolsuur Ortofosfousuur Polifosfousuur Versoeters Veroorloofde natuurlike en kunsmatige soeters	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe. Soos in regulasie voorgeskryf.	Cheese preparations, process(ed)	Acidifiers Acetic acid Citric acid dl-Lactic acid Phosphoric acid Vinegar Colourants Annatto extracts Beta-Carotene Chlorophyll Circumin or turmeric Riboflavin Copper complex of chlorophyll	40 g/kg, singly or in combination, calculated as anhydrous substances. GMP.
(b) onversoet	Stabiliseerders Natrium-, kalium, en kalsiumsoute van: Soutsuur Sitroensuur Koolsuur Ortofosfousuur Polifosfousuur Carragenien	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe. 150. GVP.		Emulsifiers Sodium, sodium-aluminum, potassium and calcium salts of the mono-, di- and polyphosphoric acids Sodium, potassium and calcium salts of citric acid Citric acid and/or phosphoric acid with sodium hydrogen carbonate and/or calcium carbonate	40, singly or in combination, calculated as anhydrous substances, but mono-, di- and polyphosphates not to exceed 30.
Maaskaas...	Skadelose geurmiddels nie van melk afkomstig nie Bederfwerende middel Sorbiensuur en Na- en K-soute Pimarisien Stabiliseerders Natrium-, kalium- en kalsiumsoute van: Soutsuur Sitroensuur Koolsuur Ortofosfousuur Polifosfousuur	1 000, bereken as sorbien-suur. 10. 2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.		Preservatives Benzoinic acid Nisin Propionic acid and its K, Na and Ca salts Sorbic acid and its Na and K salts Sodium diacetate Pimarin	600. 12.5 equals 500 i.u./g. 2 000.
Melk en afgeroomde melk, gevurde	Kleurstowwe Asoberanien Beetrooi Cochenille Eritrosien BS Glanswart BN Groen S Indigotien Karamel Karmoisien Ponceau 4R Sonsonderganggeel FCF Tartrasien Helderblou FCF Skadelose geurmiddels nie van melk afkomstig nie	30. 250. 20. 27. 12. 2. 6. 150. 57. 48. 12. 18. GVP. GVP.		Other additives Calcium chloride Calcium carbonate and/or Sodium hydrogen carbonate Agar-agar Arabic gum Carrageenan Gelatine Guar gum Karaya gum Locust (carob) bean gum Oat gum Pectin Propylene glycol ester of alginic acid Sodium carboxymethylcellulose (cellulose gum) Sodium, potassium, calcium and ammonium salts of alginic acid Tragacanth gum Acidifiers Acetic acid Citric acid dl-Lactic acid Phosphoric acid Vinegar	1 500. 10. 40, singly or in combination, calculated as anhydrous substances. 8 000, singly or in combination.
Melkpoeier..	Antikoekmiddels in melkpoeiers wat in voedselverkoopmasjiene gedispenseer word Kalsiumkarbonaat Magnesiumkarbonaat Magnesiumoksied Magnesiumfosfaat, driebasisies Aluminium-, kalsium-, magnesium- en natrium-aluminiumsilikate Silikondioksied (amorf) Trikalsiumfosfaat Emulgeermiddels Lesitiën Mono- en di-giseriede	10, afsonderlik of in kombinasie. 5 000. 2 500.	Cheese, spreadable, processed		40, singly or in combination, calculated as anhydrous substances.

I	II	III	I	II	III
Melk en melkprodukte	Veroorloofde additiewe	Voorwaardes en toleransies mg/kg	Milk and milk products	Permitted additives	Conditions and tolerances mg/kg
Melkpoeier (vervolg)	Stabiliseerders Kaliwm- en kalsium-soute van: Koolsuur Sitroensuur Soutsuur Ortofosforsuur Polifosforsuur	5 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Cheese, spreadable, processed (continued)	Colourants Annatto extracts Beta-Carotene Chlorophyll Curcumin or turmeric Riboflavin	GMP.
Proseskaas-preparate	Aansuurmiddels Aynsuur Sitroensuur dl-melksuur Fosforsuur Asyn. Kleurstowwe Annatto-ekstrakte Betakaroteen Chlorofil Koperkompleks van chlorofil Kurkumien of borrie Riboflavien Emulgeermiddels Natrium-, natrium-aluminium-, kaliwm- en kalsiumsoute van die mono-, di- en polifosfure, natrium-, kaliwm- en kalsiumsoute van sitroensuur Sitroensuur en/of fosforsuur met natrium-waterstofkarbonaat en/of kalsiumkarbonaat Bederfwerende middels Bensoësuur Nisien Propoontsuur en K, Na en Ca soute of Sorbiensuur en Na en K soute Natriumdiasetaat Pimaricin Ander additiewe Kalsiumchloried Kalsiumkarbonaat en/of Natriumwaterstofkarbonaat Agar-Agar Arabiekgom Carragenien Gelatien Guargom Karaagom Lokus (karob) boontjiegom Hawergom Pektien Propileenglikolester van algiensuur Natriumkarboksime-tielsellulose (sellulose gom) Natrium-, kaliwm-, kaliwm- en ammonium-soute van algiensuur Tragasantgom	40 g/kg afsonderlik of in kombinasie, bereken as watervrye stowwe. GVP.		Emulsifiers Sodium, sodium-aluminium, potassium and calcium salts of the mono-, di- and polyphosphoric acids Sodium, potassium and calcium salts of citric acid Citric acid and/or phosphoric acid with sodium hydrogen carbonate and/or calcium carbonate Preservatives Sorbic acid and Na and K salts Propionic acid and its Ca, K and Na salts Benzoic acid Nisin Sodium diacetate Pimaricin Other additives Calcium chloride Sodium hydrogen carbonate and/or calcium carbonate	40, singly or in combination, calculated as anhydrous substances; mono-, di- and polyphosphates not to exceed 30. 1 000. 1 000. 600. 12,5 equals 500 i.u./g. 1 500. 10. 40, singly or in combination, calculated as anhydrous substances.
	600. 12,5 is gelyk aan 500 i.e./g. 2 000. 1 500. 10.		Condensed milk (a) sweetened	Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid Sweeteners Permitted natural and artificial sweeteners	2 000, singly or in combination, expressed as anhydrous substances. As laid down by regulation.
	40, afsonderlik of in kombinasie, bereken as watervrye stowwe. 8 000, afsonderlik of in kombinasie.		(b) unsweetened	Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid Carrageenan	2 000, singly or in combination, expressed as anhydrous substances. 150.
Proseskaas, smeerkas	Aansuurmiddels Aynsuur Asynsuur Fosforsuur dl-melksuur Sitroensuur Kleurstowwe Annatto-ekstrakte Betakaroteen Chlorofil Kurkumien of borrie Riboflavien	40, afsonderlik of in kombinasie, bereken as watervrye stowwe. GVP.	Cream..... Cultured milk	Stabilisers Sodium, potassium and calcium salts of: Carbonic acid Citric acid Hydrochloric acid Orthophosphoric acid Polyphosphoric acid Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid	5 000, singly or in combination, expressed as anhydrous substances. 2 000, singly or in combination, expressed as anhydrous substances.

I	II	III	I	II	III
Melk en melkprodukte	Veroorloofde additiewe	Voorwaardes en toleransies mg/kg	Milk and milk products	Permitted additives	Conditions and tolerances mg/kg
Proseskaas, smeerkas (vervolg)	Emulgeermiddels Natrium, natriumaluminium, kalium en kalsium soute van die mono-, di- en polifosfoure, natrium-, kalium- en kalsiumsoute van sitroensuur Sitroensuur en/of fosfoursuur met natriumwaterstofkarbonaat en/of kalsiumkarbonaat Bederfwerende middels Sorbiensuur en Na- en K-soute Propioonsuur en die Ca-, K- en Na-soute daarvan Bensoësuur Nisien Natriumdiacetaat Pimarisien Ander additiewe Kalsiumchloried Natriumwaterstofkarbonaat en/of kalsiumkarbonaat	40, afsonderlik of in kombinasie, bereken as watervrye stowwe; mono-di- en polifosfate moenie 30 oorskry nie. 1 000. 1 000. 600. 12,5 is gelyk aan 500 i.e./g. 1 500. 10. 40, afsonderlik of in kombinasie, bereken as watervrye stowwe.	Milk and skim(med) milk, flavoured	Colourants Azogeranine Beetroot red Brilliant black BN Brilliant blue FCF Caramel Carmoisine Cochineal Erythrosine BS Green S Indigotine Ponceau 4R Sunset yellow FCF Tartrazine Harmless flavouring substances not derived from milk	30. 250. 12. GMP. 150. 57. 20. 27. 2. 6. 48. 12. 18. GMP.
Room.....	Stabiliseerders Natrium-, kalium-, en kalsiumsoute van: Koolsuur Sitroensuur Soutsuur Ortofosfoursuur Polifosfoursuur	5 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Milk powder	Anti-caking agents in milk powders intended to be dispensed in food-vending machines Calcium carbonate Magnesium carbonate Magnesium oxide Magnesium phosphate, tribasic Silicates of aluminium, calcium, magnesium and sodium-aluminium Silicon dioxide (amorphous) Tricalcium phosphate Emulsifiers Lecithin Mono- and diglycerides	10, singly or in combination. 5 000. 2 500.
Roomkaas..	Skadelose geurmiddels nie van melk afkomstig nie Bederfwerende middels Sorbiensuur en Na- en K-soute Pimarisien Stabiliseerders Natrium-, Kalium-, en Kalsiumsoute van: Soutsuur Sitroensuur Koolsuur Ortofosfoursuur Polifosfoursuur	GVP. 1 000, bereken as sorbiensuur. 10. 2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Sour milk..	Stabilisers Potassium and calcium salts of: Carbonic acid Citric acid Hydrochloric acid Orthophosphoric acid Polyphosphoric acid Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid	5 000, singly or in combination, expressed as anhydrous substances. 2 000, singly or in combination, expressed as anhydrous substances.
Suurmelk...	Stabiliseerders Natrium-, kalium-, en kalsiumsoute van: Soutsuur Sitroensuur Koolsuur Ortofosfoursuur Polifosfoursuur	2 000, afsonderlik of in kombinasie, uitgedruk as watervrye stowwe.	Yogurt, flavoured	Colourants See milk, flavoured Preservatives Sorbic acid Benzoic acid Sulphur dioxide Pimaricin Stabilisers Sodium, potassium and calcium salts of: Hydrochloric acid Citric acid Carbonic acid Orthophosphoric acid Polyphosphoric acid	See milk, flavoured. 1 000. 50 in the final product. 10.
Verskeidenheid kaas (Edam, Gouda, Tilsiter, Limburger)	Kleurstowwe Annatto-ekstrakte Betakaroteen Bederfwerende middels Kalium- en natriumnitraat Pimarisien	600. 200. 2, in die skil sonder plastiekbedekking. 500 in die plastiekbedekking. 10 binne-in.		Sweeteners Permitted natural and/or artificial sweeteners	5 000, singly or in combination, expressed as anhydrous substances. As laid down by regulation.

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Bosman Street, Private Bag X85,
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Bosman Street, Private Bag X85, Pretoria, 0001.