AFRICAN STANDARD

DARS 1474

First Edition 2018

Antibacterial bathing bars — Specification

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This African Standard was prepared by the ARSO Technical Harmonization Committee Number 5 on Chemistry and Chemical Engineering (ARSO/THC 05).

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Introduction

Performance of soaps, for a long time, has been primarily based on Total Fatty Matter (TFM).

Whereas the above is a fact, technological trends have shown that performance of soap can be enhanced by acceptable and safe surface active agents where TFM levels have been reduced.

In this standard the TFM levels have been reduced from that of antibacterial toilet soap with introduction of surface active agents whilst serving the same purpose. However this does not necessarily imply substitution of antibacterial toilet soap but rather an alternative and affordable antibacterial soap product.

This standard therefore sets minimum requirements for performance and safety characteristics of antibacterial bathing bar.

Antibacterial bathing bars — Specification

1 Scope

This African Standard specifies the requirements and methods of sampling and test for solid antibacterial bathing bars used for personal care.

This Standard applies to antibacterial bathing bars supplied in the form of bars and produced from vegetable or animal oils or fats, fatty acids, or from a blend of all or part of these materials, with or without the addition of rosins or non-soapy/synthetic surfactants.

2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

CD-ARS 1462:2018, *Powder detergents — Biodegradability*

CD-ARS 1468:2018, *Anti-bacterial liquid toilet soap — Specification*

ISO 456, *Surface active agents — Analysis of soaps — Determination of free caustic alkali*

ISO 685, *Analysis of soap — Determination of alkali content and total fatty matter content*

ISO 4315, *Surface active agents -- Determination of alkalinity -- Titrimetric method*

Regulation (EC) No 1223/2009 of the European Parliament and of the Council of 30 November 2009 on Cosmetic Products

3 Terms and definitions

For the purpose of this standard the following definitions apply.

antibacterial bathing bar
is a product in the form of a bar containing an antibacterial agent and soap of fatty acids and synthetic surface active agents listed in 5.4.1 as active ingredients and which could be used for bathing purposes in soft and hard water

4 Requirements

4.1 General requirements

4.1.1 Antibacterial bathing bars shall be in the form of bars.

4.1.2 The colour of the bar shall generally be uniform, except for multi-coloured bars.

4.1.3 The product shall not be harmful to skin.

4.1.4 The bar shall not have an unpleasant odour.

4.1.5 The antibacterial bathing bars shall be firm.

4.1.6 They may contain suitable quantities of colouring matter, perfume, opacifiers and optical brightening agents.

4.1.7 Antibacterial bathing bars shall not contain any ingredients in amounts that are harmful to the human body and environment

4.1.8 Synthetic surface active agents, may be used and when used shall not be more than 4% by mass when tested in accordance to Annex F

4.1.9 The antibacterial bathing bar shall contain permitted antibacterial agent. The label shall clearly state the antibacterial agent used and its level.

4.1.10 The antibacterial bathing bar shall pass the antibacterial activity test when determined by the method given in Annex J.

4.2 Specific requirements

4.2.1 Ingredients

The antibacterial bathing bar shall have one or more of the following surfactants:

4.2.1.1 There is no restriction on the use of soap of fatty acids, fatty acid ester sulphonates, fatty alkalonamide, fatty alcohol ethoxylates, sarcosinates, taurides, fatty isothionates, alpha olefin sulphonates, alcohol sulphates and amphoterics such as betaines and fatty alcohol ethoxy sulphate, linear alkyl benzene sulphonates (LAS) and alkyl poly glycosides (APG) and fatty alcohol sulphosuccinate and fatty alkanol amido sulphosuccinate.

4.2.1.2 In addition to the surfactants and perfume, the antibacterial bathing bar may contain other Ingredients such as electrolytes, bar structuring and processing aids, colouring matter, permitted antioxidants, preservatives, permissible germicides, super fatting agents, humectants and plant extracts.

4.2.1.3 Rosins, as % of total fatty matter, shall not exceed 2 % m/m.

4.2.1.4 The synthetic surface active agents shall pass the biodegradability test as given in CD-ARS 1462:2017

4.2.2 All ingredients shall be declared on the label following descending order in terms of quantity.

4.2.3 The bathing bar shall not contain any materials prohibited by regulations such as Regulation (EC) No 1223.

4.2.4 Performance and safety requirements

Antibacterial bathing bars shall also comply with the requirements given in Table 1 when tested against the methods prescribed therein.

Table 1 — Performance and safety requirements

No.	Characteristic	Requirement	Test method
(ii)	Lather, ml, min.	200	Annex A
(iii)	Mush (loss in mass due to mushing on a wet surface), g/30 cm^2, max.)	10	Annex B
(iv)	Freedom from grittiness	Pass test	Annex C
(v)	Total alkalinity (as NaOH), % by mass, max.	1.0	ISO 685/ ISO 4315
(vi)	Free caustic alkali (as NaOH), % by mass, max.	0.1	ISO 456
(vii)	Triclosan (TCN) and Trichlorocarbanilide (TCC) [a)]	≤1% by mass	Annex D
(viii)	Chloroaniline content [a)]	≤10 ppm	Annex E

NOTE Trichlorocarbanilide (TCC) is not heat stable and decomposes into chloroanilines on prolonged heating above 60°C. If TCC is used in soap, the manufacturer should take care that such soap is not subjected to temperature above 60°C during the entire manufacturing process or during storage
[a] Delete. On the label you specify what actives you have used. Alternative active agents to be researched and to replace the two.

5 Sampling

Sampling shall be done in accordance to with CD-ARS 1468:2017, Annex D.

6 Packaging and labelling

6.1 Packaging

Each bar shall be wrapped and packed in suitable boxes, packages or cartons to avoid contamination or damage during transportation.

6.2 Labelling

Each antibacterial bathing bar shall be marked legibly and indelibly with the following particulars:

a) the words “Antibacterial Bathing Bar”;

b) manufacturer’s name and physical address;

NOTE The name, physical address of the distributor/supplier and trade mark may be added as required.

c) nominal weight of each bar at the time of packaging;

d) number of bars contained in the package;

e) batch number or code number; (Check bathing bar)

f) all ingredients;

g) date of manufacture and expiry date;

h) antibacterial agents used and their levels;

i) country of origin.

Annex A
(normative)

Test for lather volume

A.1 General

Strict attention shall be paid to all details of the procedure in order to ensure concordant results. Particular care should be taken to invert the cylinder exactly as described.

A.2 Outline of the method

A suspension of the material in standard hard water is taken in a graduated cylinder and given 12 inversions under prescribed conditions. The volume of the foam formed is observed after keeping the cylinder for 5 minutes.

A.3 Reagents

A.3.1 Calcium chloride $CaCl_2.2H_2O$, AR

A.3.2 Magnesium sulphate $MgSO_4.7H_2O$, AR

A.3.3 Distilled water

A.4 Apparatus

A.4.1 Graduated cylinder — Glass stoppered with graduation from 0 to 250 mL, with 2 mL divisions. Overall height about 35 cm and the height of the graduated portion about 20 cm.

A.4.2 100-mL glass beaker

A.4.3 Thermometer of range 0 – 110 °C

A.5 Preparation of standard hard water

Dissolve 0.220 g of calcium chloride dihydrate and 0.246 g of magnesium sulphate heptahydrate in distilled water. Dilute to 5 L with distilled water.

NOTE This standard hard water has a hardness of approximately 50 ppm calculated as calcium carbonate.

A.6 Sample preparation

Cut away the outer edges of bathing bar using a knife

Using a stand up type of grater, grate up to 10 g – 15 g of the bathing bar into small chips.

A.7 Procedure

Weigh 1 g of the grated chips antibacterial bathing bar accurately in a 100-mL glass beaker. Add 10 mL of the standard hard water. Cover the beaker with a watch glass and allow to stand for 30 min. The operation is carried out to disperse the antibacterial bathing bar.

Stir the contents of the beaker with a glass rod and transfer the slurry to a 250-mL graduated cylinder ensuring that not more than 2 mL foam is produced. Repeat the transfer of the residue left in the beaker with further portions of 20 mL of standard hard water ensuring that all the matter in the beaker is transferred to the cylinder.

Adjust the contents in the cylinder to 100 mL by adding sufficient standard hard water. Bring the contents of the cylinder to 30 °C. Stir the contents of the cylinder with a glass rod or thermometer to ensure a uniform suspension.

As soon as the temperature of the contents of the cylinder reach 30 °C, stopper the cylinder and give it 12 complete inversions, each inversion comprising movements in a vertical plane, upside down and vice versa. After the 12 inversions, let the cylinder stand for 5 min. Take the following readings as shown in Figure A.1:

a) foam plus water (V1 mL).

b) water only (V2mL).

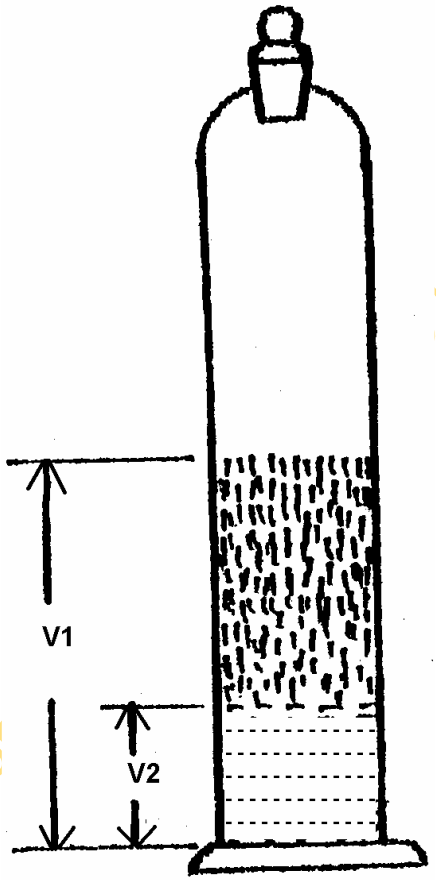

Figure A.1 — Measurement of foam

A.8 Calculation

Lather volume = V_1 - V_2

where

V_1 = Volume, in mL of foam + water;

V_2 = Volume, in mL of water only.

Annex B
(normative)

Evaluation of the mushing properties of a bathing bar

B.1 Principle

A test piece of defined size is cut from the sample bar to remove harder outer layers. The test piece is preconditioned by giving 18 x 180 degree twists under running water at 25 °C or in a bowl of water at 25 °C. The bar is left for six hours on a piece of fabric that has been wetted and drained of excess water. During the six hours the soap/ cloth are covered to prevent drying. At the end of the test period mush is removed from the test piece face in contact with the cloth. Weight loss from the test piece is expressed as mush per 30 cm^2 of original surface area in contact with the cloth.

B.2 Equipment

B.2.1 For sample preparation

B.2.1.1 Coarse kitchen cheese grater

B.2.1.2 Sharp thin blade knife or carpenters plane

B.2.1.3 Callipers or ruler, to ensure the sample dimensions

B.2.1.4 Other equipment/ materials for the test

Plastic or non-corrodible trays which are suitable sized for the test piece. Plastic soap dishes 7 cm x 11 cm x 2 cm are quite suitable.

Cotton cloth pieces cut and folded to fit as a triple layer inside the trays. Normal, flat weave, cotton sheeting as used for bed sheets will be quite suitable.

B.3 Bar preparation

B.3.1 Three (3) individual bars of a type should be tested. A test piece is cut from each bar. The test piece should if possible have a working face (to be applied to the fabric) of 6 cm ±1 cm x 4 cm ±1 cm.

All bars in a set shall be cut to have the same face size. If the smallest of the range of bars to be tested at a given time is too small to allow a working face within these limits, then all bars should be cut to the maximum size possible from the smallest bar.

The longest axis of the test piece (6 ±1) cm should be from a direction parallel to the longest axis of the original bar sample.

The working face should be a fresh surface from the interior of the bar sample. The face opposite the working face should be identified by making a small hole with a sharp object. This enables the working face to be identified after the preconditioning step.

B.3.2 To cut the bar it is convenient to first trim it to the approximate size using a coarse kitchen cheese grater and then to make the final adjustments to a smooth surface with a sharp thin-bladed knife or carpenters plane. If a plane is used, it is better to move the bar over the plane blade.

B.4 Test procedure

For each test piece

B.4.1 The tray plus triple thickness of cloth is filled with demineralised water. The tray is then held vertically to drain the water from the cloth. The vertical position is maintained until water ceases to run from the dish in a continuous stream i.e. starts to drip.

B.4.2 The area of the working face of the test piece is measured (A).

B.4.3 The working face of the bar is placed onto the damp fabric and then the tray plus soap are covered e.g. with a sealed plastic bag, to prevent water loss.

B.4.4 The covered test piece and holder are maintained at 25 °C for 6 h.

B.4.5 The mushed soap test piece is removed from the tray and is weighed (W_1).

B.4.6 Mush is removed from the working face of the soap test piece by scraping with the edge of a blunt sided spatula or plastic ruler.

B.4.7 The test piece is reweighed (W_2) and the amount of mush removed is calculated as in D.5. The mush is expressed as grams per 30 cm^2 of original test piece surface area.

NOTE The procedure for weighing the bar and removing the mush will take some minutes. During that time the remaining soaps will continue to form mush. While this time is not critical for a set of three test pieces from a given product, if more than one product is under test it is advised to stagger the start of the test for the second product. This will give adequate time to complete work on the first set before the 6-hour storage time of the subsequent set is completed.

B.5 Calculation

Weight of mush (grams) $W = W_1 - W_2$

Surface area of bar (cm^2) A = (width x breadth)

$$\text{Mush} = \frac{W \times 30g}{A} \text{ per } 30\ cm^2$$

B.6 Criteria for conformity

The test is done with three (3) separate samples of each product type, and the mean value from three samples is quoted (X). The range of values (R) is quoted as the difference between the highest and lowest values obtained for a given product type.

The sample lot of products shall be declared as conforming to the requirements for this standard if X + 0.6R is less than the maximum value given in Table 1.

Annex C
(normative)

Determination of grittiness in antibacterial bathing bar

C.1 Procedure

Either
Hold the antibacterial bathing bar under a smooth stream of running water at a temperature of 30 °C and gently rub the two sides of the bar on the palm of one hand for one minute each side.

or

Immerse the soap in a bowl containing 5 L of water at 30 °C and gently rub two opposite bar faces with the palm of one hand for 30 s (15 s per bar face). Remove the bar from the water and continue to gently rub the two opposite bar faces for a further 30 s (15 s per face).

Allow the used bar to dry in the open for 4 hours and examine the surface.

A set of 3 samples will be tested for each product.

NOTE 1 Hands will become hydrated and insensitive with prolonged immersion in water. Testers should wait 15 min between testing every 3 sets of products (9 grit tests).

NOTE 2 If using a bowl rather than running water use fresh water after testing every set of 3 samples.

C.2 Criteria for conformity

The performance criteria are:

During manipulation under running water the washing bar will not have a visibly rough surface and will feel smooth to the touch. No gritty particles will be observed on the surface of the dried bar 4 h after the washing test.

Annex D
(normative)

Determination of TCC and TCN in antibacterial bathing bar by HPLC

D.1 Principle

TCC and TCN are antibacterial agents, which are separated from other components in soap by normal phase or reverse phase liquid chromatography, detected spectrophotometrically and quantified by comparison with standard TCC and TCN. The method can estimate as low as 1 ppm of the above compounds:

Procedures for both normal and reverse HPLC has been described and provide the option to use either method whichever is available to the users. Both methods are comparable.

D.2 Normal phase HPLC

D.2.1 Reagents

D.2.1.1 Iso-octane, HPLC grade.

D.2.1.2 Iso-propanol (2-propanol), HPLC grade

D.2.1.3 Hexane, HPLC grade.

D.2.1.4 Standard TCC, 99 % pure

D.2.1.5 Standard TCN, 99 % pure

D.2.2 Apparatus

D.2.2.1 High Performance Liquid Chromatograph consisting of a pump, a sample injector of fixed volume with UV detector having variable wavelengths and a recorder.

D.2.2.2 Standard volumetric flasks

D.2.2.3 Pipettes

D.2.2.4 Magnetic stirrer

D.2.2.5 Millipore filter apparatus with 0.5 µ filter

D.2.2.6 Column

D.2.2.6.1 Silica column, stainless steel 25 cm x 0.46 cm packed with Normal phase-silica 5µ (Lichrosorb Si-60)

D.2.2.6.2 Cyano column, stainless steel 25 cm x 0.40 cm packed with (Lichrospher 100) cyano 5µ.

NOTE Either, of the above columns can be used depending on the availability.

D.2.2.7 Mobile phase

D.2.2.7.1 For silica column — Transfer 20 ml of iso-propanol into a 500 ml volumetric flash and make upto mark with iso-octane and mix well. Assemble millipore filter apparatus and filter the solvent system prior to use.

D.2.2.7.2 For cyano column — Transfer 50 ml of HPLC grade iso-propanol (2-propanol) into a 500 ml volumetric flask, fill upto the mark with hexane and mix well assemble milllipore filter apparatus and filter the solvent system prior to use.

D.2.2.7.1 For silica column — Transfer 20 ml of iso-propanol into a 500-ml volumetric flask and make up to mark with iso-octane and mix well. Assemble millipore filter apparatus and filter the solvent system prior to use.

D.2.2.7.2 For cyano column — Transfer 50 ml of HPLC grade iso-propanol (2-propanol) into a 500-mL volumetric flask, fill up to the mark with hexane and mix well. Assemble millipore filter apparatus and filter the solvent system prior to use.

D.2.2.8 HPLC conditions

Detector wavelength flow rate: 280 nm

Flow rate: 0.5 ml/min

Injection volume: 20 µl

Retention time

Silica column
TCN - 7.5 min
TCC - 19.2 min

Cyano column
TCN - 4.0 min
TCC - 7.5 min

D.2.3 Procedure

D.2.3.1 Standard preparation (see note under D.3.4)

Weigh accurately 25 mg of triclosan (TCN) and 25 mg of TCC into a 100-ml volumetric flask and make up to volume with the mobile phase and mix well. Pipette 1.0 ml of this solution in a 50 ml volumetric flask and dilute with mobile phase. Final concentration of TCC and TCN is 250 µg/50 ml (5.0 ppm).

D.2.3.2 Sample preparation

Weigh accurately 1 g of homogenized sample into a 100-ml standard flask, and dilute to the mark with mobile phase. Pipette 10 ml of the supernatant liquid to a 50-ml volumetric flask, dilute with mobile phase, to the mark, and filter through 0.45 µm filter.

D.2.3.3 Chromatography

Equilibrate the column, maintained at a temperature of 30 °C, with the mobile phase with a flow rate of 0.5 ml /min for iso-octane - iso-propanol mobile phase and 1.0 ml/min for Hexane - iso-propanol mobile phase for 30 min. Set the wavelength at 280 nm. Inject 20 µl of standard solution and then sample solutions.

Measure area of the peaks of respective retention time for standard and sample.

D.2.4 Calculation

$$\text{TCN, \% by mass} = \frac{\text{Area of sample for TCN} \times \text{Concentration of standard TCN}}{\text{Area of standard TCN} \times \text{Concentration of sample}} \times 100$$

$$\text{TCC, \% by mass} = \frac{\text{Area of sample for TCC} \times \text{Concentration of standard TCC}}{\text{Area of standard TCC} \times \text{Concentration of sample}} \times 100$$

D.3 Reverse phase

D.3.1 Reagents

D.3.1.1 *Methanol* — HPLC grade.

D.3.1.2 *Sodium Dihydrogen Phosphate Monohydrate* — Chemical grade.

D.3.1.3 Standard TCC

D.3.1.4 Standard TCN (TCS)

D.3 Reverse phase

D.3.1 Reagents

D.3.1.1 Methanol, HPLC grade.

D.3.1.2 Sodium Dihydrogen Phosphate Monohydrate, Chemical grade.

D.3.1.3 Standard TCC

D.3.1.4 Standard TCN (TCS)

D.3.2 Apparatus

D.3.2.1 Column

Octyldimethylsilyl (C-DB)

Supercosil LC-8-DB - 15 cm x 4.6 mm. 5µ

D.3.2.2 Mobile phase

MeOH/0.01 M Phosphate buffer 62:38 v/v

0.01 M Phosphate buffer: Dissolve 1.38 g sodium dihydrogen phosphate monohydrate in 1 000 ml of distilled water. Prepare to pH 3.0 by 10 % phosphate solutions.

D.3.3 Procedure

D.3.3.1 Standard preparation (see Note under D.3.4)

D.3.3.1.1 Weigh accurately about 90 mg of TCN. Dissolve in methanol and make up to 1 000 ml volumetric flask with methanol.

D.3.3.1.2 Weigh about 110 mg of TCC, dissolve well with methanol, and make up the volume to 1 000 ml.

D.3.3.1.3 Accurately pipette 10 ml of the solution prepared in (D.3.3.1.1) into the (D.3.3.1.2) volumetric flask containing TCC. And make up to the volume with methanol. Then accurately pipette 5 ml of the solution into a 50-ml volumetric flask. Make up to the volume with methanol. Filter this standard solution through 0.45 µm filter.

D.3.3.2 Sample preparation

Weigh accurately about 1.0 g of product, dissolve in methanol and make up to 100 ml in a volumetric flask with methanol. Filter this sample solution through 0.45 μm filter.

D.3.3.3 HPLC conditions

Detector wavelength	280 nm
Column temperature	35 ºC
Flow rate	1.0 ml/min
Injection volume	10 μl

Prepare the standard solution and the sample solution at the same time. Inject the standard solution three times and calculate the average of each ingredients peak count. Inject 10 μg the sample solution and determine each ingredients percentage by the calculation shown.

D.3.4 Calculations

$$\text{TCN, \% by mass} = \frac{M_s \times A_r \times F}{A_s \times M_t \times 100}$$

$$\text{TCC, \% by mass} = \frac{M_s \times A_r \times F}{A_s \times M_t \times 100}$$

where

A_r is the peak area of the test sample,

A_s is the averaged peak area of the standard,

F is the purity of standard (percent).

M_s is the mass, in grams, of the standard, and

M_t is the mass, in grams, of the test sample,

NOTE Both TCC and TCN are photosensitive, hence standards should be freshly prepared.

Annex E
(normative)

Determination of chloroaniline

E.1 Principle

The chloroanilines are extracted from soap with dimethyl sulfoxide and diazotized with nitrous acid. The reaction products are then coupled with N-1-(naphthyl) ethylenediamine hydrochloride to produce coloured compounds which are estimated spectrophotometrically.

E.2 Safety precautions

Dimethyl sulfoxide (DMSO) is readily absorbed into the skin. Inhalation or skin penetration shall be avoided.

DMSO should never be pipette using mouth. Always use pipette bulb. The standard chloroanilines and N-1-(naphthyl) – ethylenediamine hydrochloride shall not be allowed to come into contact with the skin. If they should, then wash the contaminated parts thoroughly with soap and water.

A supply of diluted sodium hypochlorite should be at hand at all times to deal with accidental spillages of chloraniline solution. Spillage on laboratory surface should be treated immediately with the sodium hypochlorite solution, followed by water.

E.3 Reagents

E.3.1 Dimethyl Sulphoxide(DMSO), AR grade.

E.3.2 Hydrochloric Acid – Concentrated(specific gravity — 1.18).

E.3.3 Sodium Nitrite – 0.4 percent w/v analytical grade, freshly prepared (aqueous).

E.3.4 Ammonium Sulphamate, 2 % w/v solution freshly prepared, (aqueous).

E.3.5 N-1-(naphthyl) Ethylene, 0.1 % w/v solution diamine hydrochloride freshly prepared (aqueous).

E.3.6 n-Butanol, AR grade

E.3.7 Sand, acid purified 40 — 100 micron mesh.

E.3.8 Solvent mixture

DMSO	5 volumes
n-Butanol	2 volumes
distilled water	2 volumes
hydrochloric acid	1 volume

Mix n-butanol, water and HCL, cool the mixture and add DMSO.

E.3.9 4-Chroroaniline and 3, 4-Dichloroaniline, AR grade.

E.4 Apparatus

E.4.1 Spectrophotometer, suitable for use at 554 nm

E.4.2 Cuvettes — Glass (matched pair) 10 mm

E.4.3 Water bath — Thermostatically controlled at 25 °C

E.4.4 Stop watch

E.4.5 Standard laboratory glassware

E.4.6 Filter Paper, Whatman No. 541

E.5 Procedure

E.5.1 Preparation of Calibration Curve

E.5.1.1 Dissolve 0.349 8 g of 3, 4-dichloroaniline and 0.2753 g of 4-chloroaniline in solvent mixture (see C.2.8) in a 250 ml amber volumetric flask.

Dilute to mark with solvent mixture. 1 ml = 2.5 mg mixed chloroanilines (stock solution).

E.5.1.2 Dilute this stock solution with solvent mixture as given below:

a) Take 5 ml of stock solution and dilute it to 250 ml with solvent mixture

1 ml = 50 µg mixed chloroanilines.

b) Take 5 ml of the above solution [see E.5.1.2(a)] and further dilute to 250 ml with solvent mixture.

1 ml = 1 µg mixed chloroanilines.

Use this solution for preparation of calibration curve.

Transfer using a burette 0, 1 ml, 2 ml, 5 ml, 10 ml, 20 ml, 40 ml into 50 ml amber volumetric flasks.

E.5.1.3 From a burette, add sufficient solvent mixture to make total volume to 40-mL in each flask. The flasks are incubated in a water bath at 25 °C for 20 min: After exactly 20 min, add 2-mL of reagent (see E.3.3) into each flask and return them to the water bath for exactly 10 min (measure with a stop watch).

Then add 2-mL of reagent (see C.3.4) into each flask and return them to the water bath for exactly 10 min. Swirl the flask occasionally.

Then add 2-ml of reagent (see E.3.5) into each flask and remove them from the water bath. Dilute to volume with distilled water, mix and allow to stand for 30 min. Measure absorbance at 554 nm against the blank solution as prepared in E.5.1.4.

E.5.1.4 In preparing the blank solution, take 40 ml of solvent mixture in a 50 ml amber volumetric flask. Incubate the flask in a water bath at 25 °C for 20 min. After exactly 20 min, add 2 ml of reagent (see E.3.3) into the flask and return it to the water bath for exactly 10 min. Then add 2 ml of reagent (see E.3.4) into the flask and return it to the water bath for exactly 10 min (swirl the flask occasionally). Then add 2 ml of reagent (see E.3.5) into the flask and remove it from the water bath. Dilute to volume with distilled water, mix and allow to stand for 30 min. Use this blank solution for preparation of calibration curve only.

E.5.1.5 Prepare a graph by plotting weight (µg) of chloroanilines contained in each 50 ml flask against absorbance. The linear calibration will pass through the origin/or determine the average absorbance (*AA*) of 1 µg of mixed chloroanilines by dividing sum of absorbances of all different aliquots of the standard by sum of µg of chloroanilines in all different aliquots of standard.

E.6 Determination of chloroanilines

E.6.1 Weigh to the nearest mg 3.0 - 15 g of finely grated soap add 10.0 g - 15.0 g of acid purified sand. Transfer quantitatively the sample and the sand into a mortar and grind the mixture thoroughly with a pestle to give a homogenous mass. Transfer the mass to a previously weighed 250 ml flat

bottom flask quantitatively and reweigh. Add DMSO (100 ml), stopper firmly and attach the flask to an automatic shaker. Shake for 1 h. Filter the DMSO extract through Whatman No. 541 into a 250 ml amber volumetric flask. Wash the flask and filter paper with small aliquots of DMSO. Allow the filtrate to drain completely, dilute to volume with DMSO and mix. Transfer 20 ml DMSO extract into a 50 ml amber volumetric flask. Add 20 ml of solvent mixture. The flask is incubated in a water bath at 25 °C for 20 min. After exactly 20 min, add 2 ml of reagent (see E.3.3) into the flask and return it to the water bath for exactly 10 min (measure with a stop watch). Then add 2 ml of reagent (see E.3.4) into the flask and return it to the water bath for exactly 10 min (swirl the flask occasionally). Then add 2 ml of reagent (see E.3.5) into the flask and remove it from the water bath. Dilute to volume with distilled water, mix and allow to stand for 30 min. Read the absorbance at 554 nm against blank (prepared as below).

E.6.2 Prepare the blank solution by mixing 20 ml of DMSO extract of sample and 20 ml of solvent mixture in a 50 ml amber volumetric flask. Incubate the flask in a water bath at 25 °C for 20 min.

After exactly 20 min, add 2 ml of distilled water into the flask and return it to the water bath for exactly 10 min. Then add 2 ml of reagent (see E.3.4) into the flask and return it to the water bath for exactly 10 min (swirl the flask occasionally). Then add 2 ml of reagent (see E.3.5) into the flask and remove it from the water bath. Dilute to volume with distilled water, mix and allow to stand for 30 min. Use this solution as a blank for reading sample only.

E.6.3 Deduce the amount of chloroanilines (μg) from the calibration graph curve.

NOTE The determination should be completed in one day.

E.7 Calculations

Determine the amount of mixed chloroanilines in the aliquot of test solution from the calibration graph.

$$\text{Chloroaniline content (in ppm)} = \frac{250(M + M_1)M_3}{20M_2M}$$

where

M is the mass, in grams, of soap

M_1 is the mass, in grams, of sand

M_2 is the mass, in grams, of soap and sand transferred to the flask

M_3 is the mass, in micrograms, (μg) of mixed chloroanilines found from calibration graph/or it can be calculated as given below:

$$M_3 = \frac{\text{Mass of the sample}}{\text{Average absorbance of 1}\mu\text{g mixed chloroaniline (AA)}}$$

where

$$M_3 = \frac{\text{Sum of the OD of the standards}}{\text{Sum of concentration of standard chloroanilines in }\mu\text{g}}$$

$$\text{Weight of soap actually used, in g} = \frac{M_2M}{(M + M_1)}$$

Annex F
(normative)

Determination of active detergent content

F.1 Outline of the method

When equivalent amounts of cationic and anionic detergents are present in a two-phase mixture of water and chloroform, methylene blue will colour the two phases to the same degree. Sodium alkyl benzene sulphonate and sodium lauryl sulphate or any other detergent can be titrated with a standard solution of cetyl trimethyl ammonium bromide.

F.2 Reagents

Weigh 1.5 ± 0.001 g of cetyl trimethyl ammonium bromide into a 250 mL beaker. Add 100 mL of distilled water and stir until dissolved. Transfer quantitatively to a 1 litre volumetric flask and make to volume. Mix thoroughly and standardize against solution B. (See F.2.1).

F.2.1 Anionic solution (Solution B)

Weigh accurately such amount of standard alkyl sulphate of known combined SO_3 or active content so as to give exactly 0.320 g of combined SO_3 into a 250 mL beaker. Dissolve in 100 mL to 200 mL of warm water. Transfer quantitatively to 1-litre volumetric flask and make to volume with water at room temperature. Mix thoroughly. This is the primary standard against which solution A, is standardized. Solution B is 0.004 N.

F.2.2 Methylene blue indicator

Dissolve 0.1 g of methylene blue in 100 mL of water. Transfer 30 mL of this solution to a 1-L flask. Add 500 mL of water, 6.8 mL of concentrated sulphuric acid, 50 g of sodium dihydrogen phosphate monohydrate ($NaH_2PO_4H_2O$) and shake until solution is complete. Dilute to the mark.

F.2.3 Chloroform

Analytical reagent grade

F.3 Procedure

F.3.1 Weigh accurately a sample of sufficient size to give approximately 0.320 g of combined SO3 into a 250 mL beaker. Sample size is crucial (see Note). Use 700 mL to 800 mL of warm water to transfer quantitatively to a 1-L volumetric flask. Warm on steam bath and shake gently until the sample is dissolved and solution is clear. Cool, dilute to the mark and mix thoroughly.

NOTE The titration value V should be as near as to 10 mL as possible, say between 8 mL and 12 mL but never outside 5 mL and 15 mL.

F.3.2 Pipette 10.0 mL of the sample solution into a 100 mL glass stoppered cylinder (25 x 300 mm). Add 25.0 ± 0.5 mL of methylene blue solution and 10 ± 0.5 ml of chloroform (see Note). Titrate with solution A to the correct end point, shaking the cylinder carefully after such addition to avoid emulsion and maintaining temperature within prescribed limits of 20 °C - 30 °C by immersion in water bath, if necessary. As the end point is approached, the rate of transfer of colour increases and solution A shall be added dropwise with vigorous shaking after each addition. If the approximate titration is known, before shaking since this avoids emulsion formation.

Application of vacuum to the titration cylinder may help to break some emulsions, if formed. The end point is reached when both layers have same colour intensity. The end point is very sharp and 0.05 mL will cause a distinct change in colour distribution at or near the equivalence point.

NOTE The titration value V should be as near to 10 mL as possible, say between 8 mL and 12 mL but never outside 5 mL and 15 mL.

F.3.3 Pipette 10.0 mL of the sample solution into a 100-mL glass stoppered cylinder (25 x 300 mm). Add 25.0 mL ± 0.5 mL of chloroform (see Note). Titrate with solution A to the correct end point, shaking the cylinder carefully after such addition to avoid emulsion and maintaining temperature within prescribed limits of 20 °C to 30 °C by immersion in water bath if necessary. As the end point solution A shall be added dropwise with vigorous shaking after each addition. If the approximate titration is known, 80 % of the required titrating solution should be added before shaking since this avoids emulsion formation.

Application of vacuum to the titration cylinder may help to break some emulsions, if formed. The end point is reached when both layers have same colour intensity. The end point is very sharp and 0.05 mL will cause a distinct change in colour distribution at or near equivalence point.

NOTE The volume of methylene blue solution and chloroform may be changed if found advantageous provided the same volumes are used in standardizing solutions A and B.

F.3.4 Calculation

F.3.4.1 The percent combined SO_3 shall be expressed as follows:

$$\% \text{ combined } SO_3 = \frac{V \times N \times 8.0}{M}$$

where:

V volume, in millilitres, of solution A used in the titration;

N normality of solution A; and

M mass, in grams, of the sample in the aliquot.

F.3.4.2 The percent active detergent content shall be expressed as follows:
Percent active detergent content = percent combined SO_3 × Mol. weight of active detergent.

NOTE The molecular weight of active detergent should be supplied by the manufacturer on request.

F.4 Alternative method for determination of active detergent content

(To be used only if the first method (Clause H.1) fails to work on the product).

F.4.1 Field of application

This method is applicable to the analysis of alkylbenzene sulphonates, alkyl sulphonate, sulphates and hydroxy-sulphates, alkylphenol and fatty alcohol ethoxysulphates and dialkyl sulphosuccinates and to the determination of active materials containing one hydrophilic group per molecule.

F.4.2 Principle

Determination of anionic-active matter in a medium consisting of an aqueous and chloroform phase, by volumetric titration with a standard cationic-active solution (benzethonium chloride), in the presence of an indicator which consists of a mixture of a cationic dye (dimidium bromide) and an anionic dye (acid blue 1).

F.4.3 Reagents

F.4.3.1 The water used shall be of distilled quality.

F.4.3.2 Chloroform, (sp. gravity = 1.48 g/m, distilling between 59.5 ºC and 61.5 ºC).

F.4.3.3 Sulphuric acid, 2.5 M solution.

F.4.3.4 Sulphuric acid, 0.5 M solution.

F.4.3.5 Sodium hydroxide, 1.0 M standard volumetric solution.

F.4.3.6 Sodium lauryl sulphate (sodium dodecyl sulphate) ($CH_3(CH_2)$ (11 OSO_3Na), 0.004 M standard volumetric solution.

Check the purity of the sodium lauryl sulphate and simultaneously prepare the standard solution.

F.4.3.6.1 Determination of purity of sodium lauryl sulphate — Weigh to the nearest 1 mg, 5 g ± 0.2 g of the product into a 250-mL round bottom flask with ground glass neck. Add exactly 25 mL of the sulphuric acid solution (H.4.3.4) and reflux into a water condenser.

During the first 5 min - 10 min, the solution will thicken and tend to foam strongly; control this by removing the source of heat and swirling the contents of the flask.

In order to avoid excessive foaming, instead of refluxing the solution may be left on a boiling water bath for 60 min.

After a further 10 min the solution clarifies and foaming ceases. Reflux for further 90 min. Remove the source of heat, cool the flask and carefully rinse the condenser with 30 mL of ethanol followed by water.

Add a few drops of the phenolphthalein solution (F.4.3.8) and titrate the solution with the sodium hydroxide solution (F.4.3.5).

Carry out a blank test by titrating 25 mL of the sulphuric acid solution (F.4.3.4) with the sodium hydroxide solution (F.4.3.5).

The purity of the sodium lauryl sulphate, expressed as a percentage,

$$= \frac{28.84(V_1 - V_0)M_0}{M_1}$$

where,

V_0 volume, in millilitres, of sodium hydroxide solution used for the blank test;

V_1 volume, in millilitres, of sodium hydroxide solution used for the sample;

M_1 mass, in grams, of the sodium lauryl sulphate to be checked; and

M_0 exact molarity of the sodium hydroxide solution.

F.4.3.6.2 Weigh 0.004 M sodium lauryl sulphate standard volumetric solutions. Weigh, to the nearest 1 mg between 1.14 g and 1.16 g of sodium lauryl sulphate and dissolve in 200 mL of water. Transfer to a ground glass stoppered 1-L one-mark volumetric flask and dilute to the mark with water.
Calculate the molarity, M1, of the solution by means of the solution by means of the formula:

$$M_1 = \frac{m_2 \times \text{purity (\%)}}{288.4 \times 100}$$

where,

m_2 mass in grams of sodium lauryl sulphate.

F4.3.7 Benzethonium chloride 0.004 M standard volumetric solution

Weigh, to the nearest 1 mg, between 1.75 g and 1.85 g benzethonium chloride and dissolve in water. Transfer to a ground glass-stoppered 1-L one-mark volumetric flask and dilute to the mark with water.

NOTE In order to prepare a 0.004 M solution, dry the benzethonium chloride at 105EC, weigh 1.792 g, to the nearest 1 mg, dissolve in water and dilute to 1 L.

F.4.3.8 Phenolphthalein, ethanolic solution containing 10 g/L. Dissolve 1 g of phenolphthalein in 100 mL of 95 % (v/v) ethanol.

F.4.3.9 Mixed indicator

F.4.3.9.1 Stock solution

Weigh to the nearest 1 mg 0.5 g ± 0.005 g dimidium bromide into a 50-mL beaker, and 0.025 g ± 0.005 g of acid blue 1 into a second 50-mL beaker.

Add between 20 mL and 30 mL of hot 10 %. (v/v) ethanol to each beaker. Stir until dissolved and transfer the solutions to a 250-mL one mark volumetric flask. Rinse the beakers into the volumetric flask with ethanol and dilute to the mark with 10 % (v/v) ethanol.

F.4.3.9.2 Mixed acid indicator solution

Take 20 mL of the stock solution prepared above, put it in a 500-mL one-mark volumetric flask. Add 200 mL of water, and 20 mL of 2.5 M sulphuric acid (F.4.3.3) mix and dilute to the mark with water. Store away from direct sunlight.

F.4.4 Apparatus

Ordinary Laboratory apparatus, and

a) bottles, 200-mL, glass stoppered, or measuring cylinders, glass stoppered.

b) burettes, 25-mL and 50-mL.

c) one-mark volumetric flask, 1-L capacity glass stoppered.

d) one-mark pipette, 25-mL.

F.4.5 Procedure

F.4.5.1 Standardization of benzethonium chloride solution

By means of the pipette transfer 25 mL of the 0.004 M sodium lauryl sulphate solution to a bottle or measuring cylinder, add 10 mL of water, 15 mL of the chloroform and 10 mL of the mixed indicator solution.

Titrate with the 0.004 M benzethonium chloride solution. Stopper the bottle or measuring cylinder after each addition and shake well. The lower layer will be coloured pink. Continue the titration with repeated vigorous shaking. As the end point approaches, the emulsions formed during shaking tend to break easily continue the titration drop by drop. Shaking after each addition of titrant, until the end point is reached. This is at the moment when the pink colour is completely discharged from the chloroform layer, which becomes a faint greyish blue.

The molarity, M, of the benzethonium chloride solution is given by the formula:

$$M = \frac{M_1 \times 25}{V_2}$$

where,

M_1 molarity of the sodium lauryl sulphate solution; and

V_2 Volume, in millilitres, of benzethonium chloride added.

F.4.5.2 Determination

Weigh to the nearest 1 mg a sample of 30 g; dissolve the test portion in water. Add a few drops of the phenolphthalein solution and neutralize to a faint pink colour with the sodium hydroxide solution or sulphuric acid solution as required.

Transfer to a 1-L one-mark volumetric flask and dilute to the mark with water. Mix thoroughly and, by means of the pipette transfer 25 mL of this solution to a bottle or measuring cylinder, , and add 10 mL of water, and 15 mL of chloroform. Titrate with the benzethonium chloride solution as described in F.4.5.1.

F.4.6 Expression of results

The content as a percentage by mass, of anionic-active matter

$$= \frac{V_3 \times M \times 1000 \times M_0 \times 100}{25 \times 1000 \times M_0}$$

$$= 4V_3M$$

The amount of active matter, expressed in milliequivalents per gram,

$$= \frac{40 \times V_3 \times M_1}{M_0}$$

where,

M_0 mass, in grams, of the test portion;

M relative molar mass of anionic-active matter;

M_1 molarity of the benzethonium chloride solution;

V_3 volume, in millilitres, of benzethonium chloride solution used for the titration of a 25-mL aliquot of anionic-active matter solution.

Annex G
(informative)

Permitted structuring and processing aids

The following is the list of structuring and processing aids used generally in bathing bars.

1) Starch and derivatives
2) Cellulose and derivatives
3) Propylene glycol
4) Sorbitol
5) Glycerol
6) Dextrin
7) Kaolin
8) Talc
9) Bentonite
10) Calcite
11) Sodium lactate
12) Soda ash
13) Vegetable/animal oil fatty acids and salts
14) Phosphates
15) Sodium chloride
16) Sodium sulphate
17) Dolomite
18) Fatty alcohol
19) Rosin and rosin salts
20) Fatty acid ethanolamide
21) Diethylene glycol monostearate
22) Paraffin
23) Polyoxyethylene glycol
24) Glycerol monostearates
25) Silicates
26) Sodium citrate
27) Chelating agents
28) Any other internationally accepted builder

Annex H
(informative)

Permitted antibacterial agents

The following is the list of antibacterial agents used generally in antibacterial bathing bars.

a) Triclosan (TCN)

b) Trichlorocarbanilide (TCC)

c) Zinc oxide

d) Hexa chlorophene

e) Chloro xylenols

f) Plant extracts

g) Any other internationally accepted antibacterial agent.

Annex J
(normative)

Determination of microbial inhibition of cosmetic soap bars and liquid hand and body washes

J.1 Scope

This annex specifies a method for testing and comparing the microbial inhibition properties of cosmetic soap bars and liquid hand and body washes.

J.2 Application

This method applies to the verification of efficacy claimed by antibacterial soaps in the inhibition of microorganisms.

J.3 Principle

When soap bars and liquid hand and body washes that have antimicrobial properties are inoculated into medium No.1 (see J.4.3.1.1), specific active ingredients diffuse into the surrounding agar thereby creating a zone of inhibition around the product, which is measured as an indication of the microbial inhibition properties of the product.

NOTE The organisms referenced in this standard are the indicator organisms used when testing for hygiene purposes.

J.4 Test method

J.4.1 General

Sampling and testing shall be carried out by personnel familiar with microbiological procedures.

J.4.2 Accuracy

Except where otherwise specified, all the following tolerances:

temperatures	± 2 °C;
masses	± 1.0 %;
volumes	± 1.0 %; and
pH values	± 0.1 pH unit.

J.4.3 Culture medium, reagents, reference cultures and controls

J.4.3.1 Culture medium

NOTE 1 The culture medium listed in J.4.3.1.1 is commercially available in dehydrated form and is made up in accordance with the manufacturer's instructions.

NOTE 2 Glass distilled water or deionised water should be used

J.4.3.1.1 Medium No. 1

Ingredients

agar	15.0 g
beef extract	15.0 g
glucose	1.0 g
pancreatic digest of casein	4.0 g
peptone	6.0 g
Yeast extract	3.0 g
Water	1 000 mL

J.4.3.1.2 Preparation

J.4.3.1.2.1 Dissolve the ingredients in approximately 900 mL of water and mix.

J.4.3.1.2.2 Make up to 1 L.

J.4.3.1.2.3 Adjust the pH so that after sterilization the pH value is 6.6 ± 0.05.

J.4.3.1.2.4 Dispense 20 mL ± 0.2 mL volumes into suitable containers and sterilize in an autoclave for 15 min ± 0.5 min at 121 °C.

J.4.3.2 Horse serum

Sterile inactivated horse serum that is free from preservatives.

J.4.3.3 Reference cultures

J.4.3.3.1 Test organisms

Use the following test organisms:

Staphylococcus aureus	Sta 10 ATCC 6538;
Escherichia coli	Esc 20 ATCC 8739; and
Pseudomonas aeruginosa	Pse 16 ATCC 15442.

NOTE Other reference organisms may also be used in addition to the ones specified in J.4.3.3.1.

J.4.3.3.2 Preparation of test organism suspensions

From a newly opened freeze-dried culture or recently received agar culture, subculture the test organisms into bottles of 10 mL nutrient medium (J.4.3.3.2.1)

J.4.3.3.2.1 Nutrient medium

J.4.3.3.2.1.1 Ingredients

peptone	5.0 g
sodium chloride	5.0 g
yeast extract	2.0 g
beef extract	1.0 g
water	1 000 mL

J.4.3.3.2.1.2 Preparation

Dissolve the ingredients in the water and adjust the pH value to 7.1. Dispense in 10 mL volumes into suitable bottles and sterilize by autoclaving at 121 ±2 °C for 15 minutes.

J.4.3.3.3 Incubate the bottles at 37 °C for 24 h. Subculture onto nutrient agar slopes (see J.4.3.3.3.1). Incubate the slopes at 37 °C for 24 h.

J.4.3.3.3.1 Nutrient agar

J.4.3.3.3.1.1 Ingredients

Agar	15.0 g
peptone	5.0 g
sodium chloride	5.0 g
yeast extract	2.0 g
beef extract	1.0 g
water	1 000 mL

J.4.3.3.3.1.2 Preparation

Dissolve the ingredients in the water and adjust the pH value to 7.1. Dispense 10 mL and 15 mL volumes into suitable bottles and sterilize by autoclaving at 121 ±2 °C for 15 minutes. Allow only the 10 mL volumes to solidify in a sloped position.

J.4.3.3.4 From each of these slope cultures, prepare four subcultures (stock cultures) of each test organism onto 10 mL nutrient agar slopes (see J.4.3.3.3.1). Incubate the stock cultures at 37 °C for 24 h and then store in a refrigerator maintained at 4 °C, except for *Pseudomonas aeruginosa* which is stored at ambient temperature.

J.4.3.3.5 Use the stock cultures to prepare further subcultures for the test, but do not make more than six serial subcultures from each stock culture. After the sixth serial subculture, resort to n new freeze-dried culture.

J.4.3.3.6 Preparation of cultures for test suspensions

J.4.3.3.6.1 For each of the test organisms, inoculate a nutrient agar slope (see J.4.3.3.3.1) from a stock culture (see J.4.3.3.4) and incubate at 37 °C for 24 h.

J.4.3.3.6.2 For the test, use a 24h culture that has been subcultured for two successive days. After six subcultures, restart the process using a fresh stock culture (see J.4.3.3.4).

NOTE The physiological condition of the test organisms is important and might influence inter-laboratory and intra-laboratory variations in test results.

J.4.3.3.6.3 After incubation, wash the bacterial growth from the slope using 10 mL sterile water and, if necessary, scrape the agar surface. Carefully decant the suspension into a sterile Erlenmeyer flask and shake vigorously to suspend all growth in the water. Standardize the suspension, by using a spectrophotometer in conjunction with a standard curve, a haemocytometer, Petroff-Hausser counting chamber or any other suitable means, so that it contains 10^5 cfu/mL ± 10^4 cfu/mL. Use the suspension within 3 h of preparation.

J.4.3.4 Control

J.4.3.4.1 Positive control

Natural honey with no additives, e.g. colourants

J.4.3.4.2 Negative control

Sterile deionised water.

J.4.4 Procedure

J.4.4.1 Melt the contents of a bottle of the medium No. 1 (see J.4.3.1.1) and cool to 45 ± 2 °C. Add 1 mL of the sterile inactivated horse serum (see J.4.3.2) and 1 mL of S. aureus test organism suspension prepared in accordance with J.4.3.3.2. Mix well and avoid the formation of air bubbles, then pour the mixture into a Petri dish of 90 mm diameter and allow to solidify.

J.4.4.2 Using as sterile cutter that produces cylindrical wells (holes) of 8 mm diameter, make five evenly spaced straight cylindrical wells in the solidified medium. Remove and discard the plugs of medium. Seal the bottom of each well using one or two drops of the molten medium in J.4.3.1.1 and allow to solidify.

J.4.4.3 Introduce the test sample into the wells in such a way that each well is completely filled with the sample, taking care not to form bubbles. When the test sample is a soap bar, grind a sufficient amount to a powder, using a sterile pestle and mortar, and introduce the powder into the wells. Ensure that the surface of the agar remains free from the sample.

J.4.4.4 Incubate the petri dish at 37 ± 1 °C for 18 h to 24 h.

J.4.4.5 At the end of this period (see J.4.4.4), remove the Petri dish from the incubator. Measure the diameter of the zone of inhibition diagonally across the well to the nearest millimeter

J.4.4.6 Take the average of the five diameters and record this to the nearest millimeters**.**

J.4.4.7 Repeat the procedure described in J.4.4.1 to J.4.4.6 (inclusive) using the *E. coli* and *P. aeruginosa test* suspensions successively.

J.4.4.8 Repeat the procedure described in J.4.4.1 to J.4.4.6 (inclusive) for the positive control and the negative control

J.5 Interpretation of results

J.5.1 For the product to pass the test, the average diameter of the zone of inhibition for each of the test organisms shall be at least 10 mm.

J.5.2 If the zones are not clearly defined for one of the reference organism (e.g. hazy, incomplete zone, or distorted shape), the test shall be repeated for the specified reference organism.

Bibliography

Working Group to identify and acknowledge useful literature used in the preparation of this standard.

EAS 794:2013, *Kenya Standard — Determination of the microbial inhibition of cosmetic soap bars and liquid hand and body washes — Test method*

EAS 878:2016, *Antibacterial bathing bars — Specification*

