IS : 7874 (Part II) - 1975
(Reaffirmed 2004)

Indian Standard

METHODS OF TESTS FOR ANIMAL FEEDS AND FEEDING STUFFS

PART II MINERALS AND TRACE ELEMENTS

(Second Reprint DECEMBER 1992)

UDC 636.085.12

BUREAU OF INDIAN STANDARDS
MANAK BHAVAN, 9 BAHADUR SHAH ZAFAR MARG
NEW DELHI 110002

Gr 5 *April* 1976

Indian Standard

METHODS OF TESTS FOR ANIMAL FEEDS AND FEEDING STUFFS

PART II MINERALS AND TRACE ELEMENTS

Animal Feeds Sectional Committee, AFDC 15

Chairman	*Representing*
Dr B. K. Soni	Indian Council of Agricultural Research, New Delhi
Members	
Agricultural Marketing Adviser to the Government of India	Directorate of Marketing and Inspection (Ministry of Agriculture & Irrigation), Faridabad
Shri S. Jayaraman (*Alternate*)	
Dr S. L. Anaokar	Godrej Soaps Private Limited, Bombay
Dr S. P. Arora	National Dairy Research Institute (ICAR), Karnal
Dr B. N. Gupta (*Alternate*)	
Dr C. S. Barsaul	U. P. College of Veterinary Science & Animal Husbandry, Mathura
Dr D. C. Joshi (*Alternate*)	
Brig R. C. Datta	Directorate of Military Farms, Army Headquarters
Shri R. K. Tripathi (*Alternate*)	
Director (Vanaspati)	Directorate of Sugar and Vanaspati (Ministry of Agriculture & Irrigation), New Delhi
Deputy Director (Vanaspati) (*Alternate*)	
Dr N. S. Dronawat	The Compound Livestock Feeds Manufacturers' Association of India, Bombay
Shri V. Virmani (*Alternate*)	
Shri R. L. Gajwani	Voltas Limited, Bombay
Shri K. R. Francis (*Alternate*)	
Dr A. N. Ghosh	Animal Husbandry Commissioner, Ministry of Agriculture & Irrigation, New Delhi
Shri S. S. Chhibbar (*Alternate*)	
Head, Animal Nutrition Division	Indian Veterinary Research Institute (ICAR), Izatnagar
Dr J. S. Ichhponani	Punjab Agricultural University, Ludhiana
Dr L. S. Hundal (*Alternate*)	

(*Continued on page* 2)

(*Continued from page* 1)

Members	*Representing*
DR M. G. JACKSON	Govind Ballabh Pant University of Agriculture & Technology, Pantnagar (Dist Nainital)
SHRI N. S. MAINI	Department of Agriculture, Ministry of Agriculture & Irrigation, New Delhi
SHRI S. S. CHAUHAN (*Alternate*)	
DR V. M. PATIL	Dairy Development Commissioner, Government of Maharashtra, Bombay
DR B. SAHAI (*Alternate*)	
DR D. V. RANGNEKAR	Bhartiya Agro-Industries Foundation, Uruli-Kanchan (Dist Pune)
DR D. V. R. PRAKASH RAO	Shaw Wallace & Company Limited, Madras
SHRI B. LAKSHMANAN (*Alternate*)	
DR A. P. SACHDEV	All India Association of Poultry Industry, Bombay
DR G. F. MITHUJI (*Alternate*)	
DR A. SAIKIA	College of Veterinary Science, Assam Agricultural University, Gauhati
DR P. K. SHARMA (*Alternate*)	
DR S. K. SARKAR	Roller Flour Millers' Federation of India, New Delhi
SHRI S. M. MAZUMDAR (*Alternate*)	
DR (SMT) SAROJINI CHANDRASHEKHARAN	Kaira District Co-operative Milk Producers' Union Limited, Anand
DR A. S. DAVE (*Alternate*)	
DR N. SATPATHY	The Tata Oil Mills Company Limited, Bombay
DR K. C. DANDONA (*Alternate*)	
DR H. C. SAXENA	Hindustan Lever Limited, Bombay
DR S. YAMDAGNI (*Alternate*)	
DR K. C. SEN	In personal capacity (*403, Jodhpur Park, Calcutta 31*)
SHRI K. N. R. SHARMA	Directorate General of Technical Development (Ministry of Industries & Civil Supplies), New Delhi
DR P. C. SHUKLA	Institute of Agriculture, Anand
SHRI G. V. SIRUR	All India Cottonseed Crushers' Association, Bombay
SHRI R. V. BIJUR (*Alternate*)	
SHRI SURESH CHANDRA	General Molasses Co (P) Ltd, Sahibabad (Ghaziabad)
SHRI T. S. TAMBOLIA	The Maharashtra Agro-Industries Development Corporation Limited, Bombay
DR D. N. RANADE (*Alternate*)	
SHRI L. J. TANNA	Solvent Extractors' Association of India, Bombay
SHRI M. K. RAHMAN (*Alternate*)	
SHRI VINEET VIRMANI	Jawala Flour Mills, New Delhi
SHRI ANAND VIRMANI (*Alternate*)	
SHRI T. PURNANANDAM, Deputy Director (Agri & Food)	Director General, ISI (*Ex-officio Member*)

Secretary

SHRI G. S. VILKHU

Deputy Director (Agri & Food), ISI

(*Continued on page* 20)

Indian Standard

METHODS OF TESTS FOR ANIMAL FEEDS AND FEEDING STUFFS

PART II MINERALS AND TRACE ELEMENTS

0. FOREWORD

0.1 This Indian Standard (Part II) was adopted by the Indian Standards Institution on 28 November 1975, after the draft finalized by the Animal Feeds Sectional Committee had been approved by the Agricultural and Food Products Division Council.

0.2 The importance of adoption of standard and uniform methods of analysis for quality control purposes need no emphasis. Such methods not only help in reducing divergence in the analytical results but also ensure and enable a proper comparison and correct interpretation of the test results.

0.3 During the past decade a large number of Indian Standards covering a wide range of animal feeds, feeding stuffs and feed supplements have been issued and many more are envisaged to be formulated. All these standards include methods of tests that are common. It was, therefore, considered desirable to have all test methods in a consolidated form as applicable to the whole range of animal feeds, feeding stuffs and supplements. It was felt that such a step would not only facilitate easy reference but also prevent undue repetition. Accordingly, this standard is being issued.

0.4 It is intended to issue this standard in three parts. Part I covers general methods such as determination of moisture, crude protein, crude fat, crude fibre, total ash, acid insoluble ash, castor husk and *MAHUA* cake. This standard will cover methods for determining minerals and trace elements, and Part III covers the microbiological methods.

0.5 In reporting the result of a test or analysis made in accordance with this standard, if the final value, observed or calculated, is to be rounded off, it shall be done in accordance with IS : 2-1960*.

---

1. SCOPE

1.1 This standard (Part II) prescribes methods for determination of minerals and trace elements in animal feeds, feeding stuffs and feed supplements.

*Rules for rounding off numerical values (*revised*).

2. QUALITY OF REAGENTS

2.1 Unless specified otherwise, pure chemicals and distilled water (*see* IS : 1070-1960*) shall be employed in tests.

NOTE — ' Pure chemicals ' shall mean chemicals that do not contain impurities which affect the test results.

3. PREPARATION OF THE SAMPLE

3.1 If necessary, the test sample should be ground so that it passes through 850-micron IS Sieve (*see* IS : 460-1962†). Alternatively, ASTM Sieve 20 or BS Sieve 18 or Tyler Sieve 20 may be used.

4. DETERMINATION OF SALT (CHLORINE AS SODIUM CHLORIDE)

4.1 Reagents

4.1.1 *Ferric Sulphate Solution* — Dissolve 60 g of ferric sulphate [$Fe_2 (SO_4)_3$] in one litre of water.

4.1.2 *Ammonium Hydroxide Solution* — prepared by mixing one volume of ammonium hydroxide with 12 volumes of water.

4.1.3 *Concentrated Nitric Acid* — r.d. 1·42 (*see* IS : 264-1968‡).

4.1.4 *Ferric Sulphate Indicator Solution* — Prepare as 25 percent (*m/v*) solution. Filter and add equal volume of nitric acid.

4.1.5 *Standard Silver Nitrate Solution* — 0·1 N.

4.1.6 *Standard Potassium Thiocyanate Solution* — 0·1 N.

4.2 Procedure

4.2.1 Weigh accurately about 1 g of the dried material and transfer to a 250-ml graduated flask. Add 50 ml of the ferric sulphate solution with a pipette, gently swirling the flask. Add 100 ml of the ammonium hydroxide solution to bring it to the mark. Swirl the flask enough to ensure thorough mixing but avoid vigorous shaking. Allow to settle for 10 minutes. Filter through 11-cm Whatman filter paper No. 41 or its equivalent. Transfer from the filtrate, an aliquot of 25 ml to a 250-ml beaker. Add 10 ml of nitric acid and 10 ml of the ferric sulphate indicator solution. Then add with constant stirring, known quantity of the

*Specification for water, distilled quality (*revised*).
†Specification for test sieves (*revised*).
‡Specification for nitric acid (*first revision*).

standard silver nitrate solution in slight excess. Heat the solution to the boil and cool to room temperature and stir to coagulate the precipitate. Titrate the excess of silver nitrate with the standard potassium thiocyanate solution. The end point is indicated by the first appearance of reddish tint that persists for 15 seconds.

4.3 Calculation

4.3.1 Chlorine as sodium chloride (on moisture-free basis), percent by mass $= \frac{5{\cdot}845\ (AN_1 - BN_2)}{m\ (100 - M)}$

where

A = volume in ml of the standard silver nitrate solution used,

N_1 = normality of the standard silver nitrate solution,

B = volume in ml of the standard potassium thiocyanate solution used up by the excess silver nitrate solution,

N_2 = normality of the standard potassium thiocyanate solution,

m = mass in g of the dried material taken for the (*see* **4.2.1**), and

M = percent moisture content.

5. DETERMINATION OF CALCIUM

5.1 Reagents

5.1.1 *Hydrochloric Acid* — 25 ml of concentrated hydrochloric acid (*see* IS : 265-1962*) diluted to 100 ml.

5.1.2 *Methyl Red Indicator* — Dissolve 0·15 g of methyl red in 500 ml of water.

5.1.3 *Ammonium Hydroxide Solution* — 50 percent (*v/v*).

5.1.4 *Dilute Ammonium Hydroxide Solution* — 2 percent (*v/v*).

5.1.5 *Ammonium Oxalate Solution* — saturated.

5.1.6 *Concentrated Sulphuric Acid* — r.d. 1·84 (*see* IS : 266-1961†).

5.1.7 *Standard Potassium Permanganate Solution* — 0·1 N (*see* IS : 2316-1968‡).

---

*Specification for hydrochloric acid (*revised*).

†Specification for sulphuric acid (*revised*).

‡Methods of preparation of standard solutions for colorimetric and volumetric analysis (*first revision*).

5.2 Procedure

5.2.1 *Ashing and Extraction* — Accurately weigh about 3 g of the material into a silica dish. Char carefully and continue the ashing in a muffle furnace at a temperature not above 450°C until the ash is white or almost so. Cool the ash, moisten with a few millilitres of distilled water and add 3 to 5 ml of concentrated hydrochloric acid drop by drop. Evaporate to dryness on a water-bath and continue heating on the water-bath for one hour to render silica insoluble. Moisten the residue with 20 ml distilled water and add about 2 to 3 ml of concentrated hydrochloric acid. Heat on a water-bath for a few minutes and filter through medium filter paper into a 250-ml volumetric flask. Wash the filter paper thoroughly with hot water, cool the filtrate and make it up to volume, shake thoroughly.

5.2.2 Transfer a 25-ml aliquot of the solution prepared as in **5.2.1** to a 400-ml beaker, dilute to about 100 ml with water and add two drops of methyl red indicator solution. Add ammonium hydroxide solution dropwise till a brownish-orange colour is obtained (*p*H 5·6). Add two drops of hydrochloric acid so that the colour of solution is pink (*p*H 2·5 to 3·0). Dilute to about 150 ml, bring to the boil and add slowly, with constant stirring, 10 ml of hot ammonium oxalate solution. If the red colour of the solution changes to orange or yellow, add hydrochloric acid dropwise until the colour again changes to pink. Leave overnight to allow the precipitate to settle. Filter the supernatant liquid through ashless filter paper and wash the precipitate thoroughly with dilute ammonium hydroxide solution. Place the paper with the precipitate in the beaker in which precipitation was carried out and add a mixture of 125 ml of water and 5 ml of concentrated sulphuric acid. Heat to 70 to 90°C and titrate with the standard potassium permanganate solution until the first slight pink colour is obtained.

5.3 Calculation

5.3.1 Calcium (as Ca) (on moisture-free basis), percent by mass $= \frac{2\,000\,AN}{m\,(\,100 - M\,)}$

where

A = volume in ml of the standard potassium permanganate solution required in the titration,

N = normality of the standard potassium permanganate solution,

m = mass in g of the material taken for the test (*see* **5.2.1**), and

M = percent moisture content.

6. DETERMINATION OF PHOSPHORUS

6.1 Reagents

6.1.1 *Concentrated Nitric Acid* — r.d. 1·42 (*see* IS : 264-1968*).

*Specification for nitric acid (*first revision*).

6.1.2 *Nitric Acid (1 : 1)* — A mixture of equal volumes of concentrated nitric acid and water.

6.1.3 *Ammonium Molybdate Stock Solution* — Take 200 g of powdered ammonium molybdate in a stoppered graduated cylinder of 1 000 ml capacity, add to it 800 ml of water and shake well for 25 minutes to dissolve the ammonium molybdate. Add gradually 25 percent (*m/v*) ammonium hydroxide solution till the solution is clear (about 100 to 140 ml of ammonium hydroxide may be required). Avoid adding excess of ammonia. Make up the volume to one litre. If necessary, filter the solution through a fluted filter paper and stock this solution.

6.1.4 *Nitric Acid Solution* — 2 percent (*m/v*).

6.1.5 *Potassium Nitrate Solution* — 3 percent (*m/v*).

6.1.6 *Standard Sodium Hydroxide Solution* — 0·1 N.

6.1.7 *Standard Nitric Acid Solution* — 0·1 N.

6.1.8 *Phenolphthalein Indicator Solution* — Dissolve 0·1 g of phenolphthalein in 100 ml of 60 percent (*m/v*) rectified spirit (*see* IS : 323-1959*).

6.2 Procedure

6.2.1 *Precipitation* — Take a 10-ml aliquot of the prepared solution (*see* **5.2.1**) in a 150-ml beaker. In a dry beaker, prepare ammonium molybdate solution by pouring into it, quickly and simultaneously 10 ml of the ammonium molybdate stock solution (*see* **6.1.3**) and 10 ml of concentrated nitric acid; or take 10 ml of concentrated nitric acid first in the beaker and into this pour quickly 10 ml of the ammonium molybdate stock solution, whirling the beaker during addition. Pour this freshly prepared clear liquid quickly into the beaker containing the aliquot and stir.

NOTE — The temperature developed in the molybdate solution is sufficient to precipitate all the phosphorus present in the aliquot. Under no circumstances the phosphomolybdate precipitate should be heated either on a water-bath or directly over a burner so as to avoid precipitation of molybdic anhydride.

6.2.2 *Filtration and Washing* — Allow the precipitate to stand overnight and then filter through a disc of Whatman filter paper No. 42 in a Gooch crucible by suction or through a 9-cm Whatman filter paper No. 42 over an ordinary funnel. As far as possible only the supernatant liquid is passed through the filter paper, retaining the precipitate in the beaker. When the supernatant liquid is decanted off, the precipitate is washed twice with dilute nitric acid and then with potassium nitrate solution until the washings are free from acid. If ordinary funnel and filter paper are used, freedom from acidity may be tested by collecting sufficient filtrate in a test-

*Specification for rectified spirit (*revised*).

tube to which a few drops of phenolphthalein indicator solution and one drop of the standard sodium hydroxide solution are added. If the pink colour appears with one drop of the standard alkali, the precipitate is free from acid.

6.2.3 *Titration* — Transfer the precipitate with the filter paper back to the beaker in which precipitation was carried out. When Gooch crucible is used for filtration, transfer the whole crucible along with the filter paper to the beaker in which precipitation was carried out. Add sufficient quantity of the standard sodium hydroxide solution from a burette just sufficient to dissolve the precipitate and then add 5 ml in excess. See that no yellow precipitate sticks to the filter paper. Note the total volume of the standard sodium hydroxide solution added. Add about 10 drops of phenolphthalein indicator solution and titrate the excess of alkali with the standard nitric acid.

6.3 Calculation

6.3.1 Phosphorus (on moisture-free basis), percent by mass $= \frac{336{\cdot}75\ (AN_1 - BN_2)}{m\ (100 - M)}$

where

A = volume in ml of the standard sodium hydroxide solution used (*see* **6.2.3**),

N_1 = normality of the standard sodium hydroxide solution,

B = volume in ml of the standard nitric acid used in to neutralize the excess alkali (*see* **6.2.3**),

N_2 = normality of the standard nitric acid,

m = mass in g of the material taken for the test (*see* **5.2.1**), and

M = percent moisture content.

7. DETERMINATION OF IRON

7.1 Apparatus

7.1.1 *Heat Resistant Glass Tube* — of 50 ml capacity, marked at 30 ml.

7.1.2 *Centrifuge* — suitable for clarifying the isoamyl alcohol phase.

7.1.3 *Photoelectric Colorimeter* — capable of measuring optical density at 495 nm.

7.2 Reagents

7.2.1 *Distilled Water* — redistilled.

7.2.2 *Concentrated Sulphuric Acid* — r.d. 1·84 (*see* IS : 266-1961*).

7.2.3 *Perchloric Acid* — 60 percent (*m/m*) solution.

7.2.4 *Concentrated Nitric Acid* — 60 percent (*m/m*).

7.2.5 *Ammonium Hydroxide Solution* — 25 percent (*m/m*).

7.2.6 *Concentrated Hydrochloric Acid* — 35 percent (*m/m*).

7.2.7 *Hydrogen Peroxide Solution* — 0·1 percent (*m/m*) solution in water stored in a brown bottle in a refrigerator.

7.2.8 *Isoamyl Alcohol* — of boiling point 129 to 132°C.

7.2.9 *Potassium Thiocyanate Solution* — Dissolve 50 g of potassium thiocyanate (KSCN) in 100 ml of water.

7.2.10 *Standard Iron Solution* — Dissolve 0·702 2 g of ferrous ammonium sulphate [$FeSO_4(NH_4)_2SO_4, 6H_2O$] in 100 ml of water, add 5 ml of concentrated sulphuric acid, warm slightly and add potassium permanganate solution (approximately 0·1 N) drop by drop until the solution shows a slight pink colouration. Make up the volume to one litre in a graduated flask. Pipette 10 ml of this solution into a one-litre graduated flask, add 10 ml of hydrogen peroxide solution and make up the volume with water. This solution contains 1 μg of iron per millilitre.

7.3 Procedure

7.3.1 *Preparation of the Test Solution* — Weigh accurately about 2·0 g of the material and transfer to a 200-ml Erlenmeyer flask. Add 2 ml of concentrated sulphuric acid, 3 ml of perchloric acid and 5 ml of concentrated nitric acid. Digest until a clear solution is obtained and white fumes of sulphuric acid are evolved. Dilute with 10 ml of water and make up the volume to 200 ml with water in a graduated flask. Preserve this solution for the determination of copper (*see* **9.3.1**) and cobalt (*see* **11.3.1**).

7.3.2 Take a suitable aliquot of the test solution containing about 10 μg of iron and transfer to the heat resistant glass tube. Add ammonium hydroxide solution until the solution is just alkaline to phenolphthalein. Add 1 ml of concentrated hydrochloric acid and 1 ml of hydrogen peroxide solution and make up the volume in the tube to 30 ml with distilled water. Add 10 ml of isoamyl alcohol, accurately measured, and 2 ml of potassium thiocyanate solution, stopper the tube and shake for 20 seconds. Transfer enough of the isoamyl alcohol phase meant for colour measurement to the centrifuge tubes, and centrifuge for 5 minutes at about 3 000 rev/min. Measure the absorption of the solution in a suitable photo-electric colorimeter at 495 nm setting the reading of the blank at zero absorption. The blank is prepared simultaneously by using the same quantities

*Specification for sulphuric acid (*revised*).

of acid employed in the digestion, making up the volume and developing the colour in the same size aliquot and in the same manner as in the case of the test solution.

7.3.3 Prepare a series of standards by treating aliquots of the standard iron solution (*see* **7.2.10**) in the same manner as the test solution. From the absorption of the standard solutions, prepare a standard curve plotting absorption values against concentrations. From this curve, obtain the mass of iron present in the test solution and calculate the quantity of iron present in 100 g of the material on moisture-free basis.

8. DETERMINATION OF IODINE (as KI)

8.1 Reagents

8.1.1 *Takadiastase*

8.1.2 *Methyl Orange Indicator* — Dissolve 0·50 g of methyl orange in water and dilute to one litre.

8.1.3 *Dilute Sulphuric Acid* — approximately 2 N.

8.1.4 *Bromine Water* — Saturated aqueous solution. Determine the approximate concentration (mg/ml) by adding (from a burette) a measured volume to a flask containing 5 ml of 10 percent potassium iodide solution, adding 5 ml of dilute sulphuric acid and titrating the liberated iodine with 0·1 N sodium thiosulphate solution.

8.1.5 *Sodium Sulphite Solution* — approximately one percent (*m/v*).

8.1.6 *Phenol Solution* — approximately 5 percent (*m/v*).

8.1.7 *Potassium Iodide Solution* — approximately 10 percent (*m/v*).

8.1.8 *Standard Sodium Thiosulphate Solution* — 0·005 N (freshly standardized).

8.1.9 *Starch Solution* — 1 percent (freshly prepared) (*m/v*).

8.1.10 *Sodium Chloride Solution* — Dissolve 10 g of sodium chloride in water and make up the volume to 100 ml.

8.1.11 *Potassium Iodide Control Solution* — Dissolve 0·328 0 g of potassium iodide in water and then make up the volume to 250 ml. Dilute 50 ml of this solution to 250 ml, and use 5-ml control (that is, 1·0 mg iodine or 0·308 mg potassium iodide).

8.1.12 *Concentrated Sulphuric Acid* — r.d. 1·84 (*see* IS : 266-1961*).

8.2 Preparation of Sample Solution — Weigh accurately about 50 g of the material and suspend in 100 ml of water. Add 2 g of takadiastase and allow to stand at 37°C for 2 hours. Filter the solution and wash the residue with water. Collect the filtrate and washings and make up the volume to 250 ml in a graduated flask.

*Specification for sulphuric acid (*revised*).

8.3 Procedure — Pipette 50 ml of the prepared sample solution (*see* **8.2.1**) into a 200-ml Erlenmeyer flask. Neutralize to methyl orange indicator with dilute sulphuric acid. Add bromine water dropwise from burette in a quantity equivalent to 20 mg of bromine. After a few minutes, destroy most of the remaining free bromine by adding sodium sulphite solution dropwise with stirring. Wash down the neck and sides of the flask with water and completely remove free bromine by addition of a drop or two of phenol solution. Add 1 ml of dilute sulphuric acid and 5 ml of potassium iodide solution and titrate the liberated iodine with the standard sodium thiosulphate solution adding 1 ml of the starch indicator near the end of the titration. Carry out a blank determination on reagents and make one or more control determinations, using 50 ml of sodium chloride solution to which have been added appropriate quantities of the potassium iodide control solution.

8.4 Calculation

$$\text{Iodine (as KI), on moisture-free basis, percent by mass} = \frac{1\,384\,(V_1 - V_2)\,N}{m\,(100 - M)}$$

where

V_1 = volume in ml of the standard sodium thiosulphate solution required for the test with the prepared sample solution,

V_2 = volume in ml of the standard sodium thiosulphate solution required for the blank determination,

N = normality of the standard sodium thiosulphate solution,

m = mass in g of the material taken (*see* **8.2**), and

M = percent moisture content.

9. DETERMINATION OF COPPER

9.1 Apparatus

9.1.0 *General* — The glassware used shall be free from copper contamination.

9.1.1 *Heat Resistant Glass Tube* — of 50 ml capacity and marked at 30 ml.

9.1.2 *Centrifuge* — capable of clarifying the isoamyl alcohol phase.

9.1.3 *Photoelectric Colorimeter* — capable of measuring the optical density at 430 nm.

9.2 Reagents

9.2.1 *Distilled Water* — redistilled.

9.2.2 *Sodium Citrate Solution* — saturated.

9.2.3 *Ammonium Hydroxide Solution* — 20 percent (*m/v*).

9.2.4 *Isoamyl Alcohol* — boiling point 129 to 132°C.

9.2.5 *Sodium Diethyldithiocarbamate Solution* — 0·1 percent (*m/v*) aqueous.

9.2.6 *Standard Copper Solution* — Dissolve 0·393 g cupric sulphate pentahydrate ($CuSO_4$, 5 H_2O) of analytical grade in distilled water, add a few drops of concentrated sulphuric acid and make up the volume to one litre in a graduated flask. Shake well. Pipette out 10 ml of this solution into a one-litre graduated flask and make up the volume. This solution contains 1 µg of copper per millilitre.

9.3 Procedure

9.3.1 Pipette a suitable aliquot of the test solution as prepared in **7.3.1**, containing about 10 µg of copper, in the glass tube marked at 30 ml. Add 3 ml of sodium citrate solution and ammonium hydroxide solution until just alkaline to phenolphthalein, followed by 3 ml of ammonium hydroxide solution. Make up the volume to 30 ml with distilled water. Add 10 ml of isoamyl alcohol accurately measured and 1 ml of sodium diethyldithiocarbamate solution. Stopper the tube and shake vigorously for 20 seconds. Transfer enough of the isoamyl alcohol phase meant for colour measurement to centrifuge tubes and centrifuge for 2 minutes at about 3 000 rev/min. Measure the absorption of the solution in a suitable photo-electric colorimeter at 430 nm, setting the reading of the blank at zero absorption. The blank is prepared simultaneously by using the same quantities of acid employed in the digestion, making up the volume and developing the colour in the same size aliquot and in the same manner as in the case of the test solution.

9.3.2 Prepare a series of standards by treating aliquots of the standard copper solution (*see* **9.2.6**) in the same manner as the test solution. From the absorption of the standard solutions prepare a standard curve plotting absorption values against concentrations. From this curve, obtain the mass of copper present in the test solution and calculate the quantity of copper present in 100 g of the material on moisture-free basis.

10. DETERMINATION OF MANGANESE

10.1 Apparatus

10.1.1 *Photoelectric Colorimeter* — capable of measuring optical density of 520 nm.

10.2 Reagents

10.2.1 *Concentrated Sulphuric Acid* — r.d. 1·84 (*see* IS : 266-1961*).

*Specification for sulphuric acid (*revised*).

10.2.2 *Sulphurous Acid* — saturated solution stored in an amber bottle in a cool place.

10.2.3 *Solution A* — Mix 42 ml of water, 2 ml of sulphuric acid, 5 ml of sulphurous acid solution and 1 ml of phosphoric acid. This solution should be freshly prepared before use.

10.2.4 *Potassium Periodate*

10.2.5 *Sodium Metabisulphite Solution* — 10 percent (*m/v*), aqueous.

10.2.6 *Phosphoric Acid* — r.d. 1·70.

10.2.7 *Concentrated Nitric Acid* — r.d. 1·42 (*see* IS : 264-1968*).

10.2.8 *Standard Manganese Solution* — Dissolve 0·575 6 g of dry potassium permanganate in about 50 ml of water in a beaker of suitable size. Add 40 ml of concentrated sulphuric acid and reduce the permanganate by careful addition of sodium metabisulphite solution until the manganese solution just becomes colourless. Oxidize the excess sulphurous acid in the hot solution by the addition of a little nitric acid. Cool and transfer the solution quantitatively to a 2-litre graduated flask. Make up the volume and store the solution in a glass-stoppered reagent bottle. This solution contains 0·1 mg of manganese per millilitre.

10.3 Procedure

10.3.1 Weigh accurately about 5 g of the material into a silica dish, char carefully and ash it in a muffle furnace at 600 to 700°C. Cool, extract the ash with 10 ml of solution A (*see* **10.2.3**) for 2 minutes and transfer to a 150-ml beaker. Rinse the dish first with 40 ml of solution A and then with distilled water, collecting the rinsings until the volume is 100 ml. Heat to the boil on a hot-plate and evaporate the solution, using a boiling tube until the volume is reduced to 20 ml. Care should be taken not to allow the solution to bump. Allow the solution to stand overnight. Filter through a small disc of ashless filter paper under slight suction into a 150-ml beaker Wash the filter paper and dilute the filtrate with water to about 100 ml. Add 2 ml of phosphoric acid and 0·3 g of potassium periodate. Boil to oxidize the manganese and continue boiling for about 15 minutes after the colour has been apparently fully developed. The final volume should not be less than 50 ml. (If necessary, boiling water may be added to the solution while boiling.) Cool and dilute to 100 ml. Measure the absorption of the solution at 520 nm by means of a suitable photo-electric colorimeter.

10.3.2 Simultaneously carry out a control determination under the same conditions as in **10.3.1** adding 5 ml of the standard manganese solution, 2 ml of sulphuric acid and 2 ml of phosphoric acid to 100 ml of water and oxidizing with potassium periodate as described in **10.3.1**.

*Specification for nitric acid (*first revision*).

10.3.3 Measure the absorption at 520 nm of a series of aliquots of the standard manganese solution (*see* **10.2.8**) treated in the same manner as the test solution. Plot a curve of these absorption values against concentration. From this curve, obtain the mass of manganese in the test solution and calculate the quantity of manganese present in 100 g of the material on moisture-free basis.

11. DETERMINATION OF COBALT

11.1 Apparatus

11.1.1 *Spectrophotometer or Photoelectric Colorimeter* — of a suitable type.

11.2 Reagents

11.2.1 *Distilled Water* — redistilled.

11.2.2 *Citric Acid* — 0·2 M. Prepare by dissolving 42 g of citric acid in 100 ml of water and standardize against standard sodium hydroxide solution using phenolphthalein as indicator.

11.2.3 *Bromophenol Blue Indicator Solution* — Dissolve 40 mg of bromophenol blue in 100 ml of water containing 5·7 ml of sodium hydroxide (0·01 N).

11.2.4 *Methyl Red Indicator Solution* — Dissolve 25 mg of methyl red in 100 ml of ethyl alcohol (60 percent *v/v*).

11.2.5 *Dithizone Solution in Chloroform* — 0·2 percent (*m/v*). Keep in a dark bottle in a refrigerator.

11.2.6 *Dithizone Solution in Carbon Tetrachloride* — 0·05 percent (*m/v*). Keep in a dark bottle in a refrigerator.

11.2.7 *Phenolphthalein Indicator Solution* — Dissolve 50 mg of phenolphthalein in 100 ml ethyl alcohol (50 percent *m/v*).

11.2.8 *Buffer Solution* — Dissolve 6·184 g of boric acid and 25·62 g of Sorensen's salt ($Na_2HPO_4,2H_2O$) in 500 ml of standard sodium hydroxide (1·0 N) and make up the volume to one litre with distilled water.

11.2.9 *Concentrated Nitric Acid* — 60 percent (*m/v*).

11.2.10 *Perchloric Acid* — 60 percent.

11.2.11 *Concentrated Sulphuric Acid* — r.d. 1·84 (*see* IS : 266-1961*).

11.2.12 *Cresol Red Indicator Solution* — Dissolve 40 mg of cresol red in 100 ml of water containing 10·5 ml of sodium hydroxide (0·01 N).

11.2.13 *Nitroso-R Salt* — 0·2 percent (*m/v*) aqueous solution stored in dark.

*Specification for sulphuric acid (*revised*).

11.2.14 *Standard Cobalt Solution* — Dissolve 4·769 4 g of cobalt sulphate ($CoSO_4, 7H_2O$) in distilled water, add one millilitre of concentrated sulphuric acid and make up the volume to one litre. Take one millilitre of this solution and make up volume to one litre with water in a graduated flask. This solution contains 1 μg of cobalt per millilitre.

11.3 Procedure

11.3.1 Take an aliquot of 5 ml of the test solution as prepared in **7.3.1**. Evaporate the water cautiously until all but a trace of sulphuric acid is removed. Add 7·5 ml of nitric acid to the residue and wash the solution into a 100-ml separating funnel. Dilute to about 30 ml with water.

11.3.2 *Extraction with Dithizone* — Add 5 drops of bromophenol blue to the solution. Run in sodium hydroxide solution (1·0 N) until a distinct greenish-blue colour appears through the yellowish tint due to the ferric citrate. The solution should still be acid to methyl red. Dilute the solution to 50 ml. Extract the solution with successive 20 ml portions of dithizone solution in chloroform. Shake vigorously and run off the chloroform layer. When the chloroform layer retains the original green colour of the dithizone solution, the test solution is washed once with pure chloroform.

Adjust the *p*H of the aqueous phase to approximately 8·3 by adding a few drops of phenolphthalein and cautiously titrating with the buffer solution until the first sign of a purplish-pink colour appears. Extract the cobalt with successive 10 ml portions of dithizone solution in carbon tetrachlorideu ntil the carbon tetrachloride phase retains the green colour of the original dithizone solution. Boil off the carbon tetrachloride from a heat resistant boiling tube. Add to the residue 1 ml of nitric acid, 0·5 ml of perchloric acid and 0·2 ml of sulphuric acid and heat till it becomes colourless. Heat the boiling tube for a few minutes in a muffle furnace at a temperature not above 350°C to ensure complete removal of sulphuric acid.

11.3.3 *Production of the Cobalt-Nitroso-R Salt Complex* — Dissolve the residue in 1 ml of citric acid and dilute with a little water so that the total volume is not more than 5 ml. Add accurately 1·2 ml of the buffer solution to adjust the *p*H. The *p*H is checked with cresol red by withdrawing a small drop of the solution. Develop the cobalt-nitroso-R salt complex by introducing 0·5 ml of the cobalt-nitroso-R salt solution while shaking. Boil for one minute. Cool and dilute to 10 ml. Measure the absorption of the solution in a suitable spectrophotometer or photoelectric colorimeter at 510 nm setting the reading of the blank at zero absorption. The blank is prepared simultaneously by using the same quantities of the reagents employed in the digestion and in the subsequent procedure. Make up the volume to 100 ml and develop the colour in the same size aliquot and in the same manner as in the case of the test solution.

11.3.4 Prepare a series of standards by treating aliquots of the standard cobalt solution in the same manner as the test solution. From the absorption of the standard solutions, prepare a standard curve plotting absorption values against concentrations. From this curve, obtain the mass of cobalt present in the test solution and calculate the quantity of cobalt present in 100 g of the material on moisture-free basis.

12. DETERMINATION OF FLUORINE

12.1 Apparatus

12.1.1 *Distillation Flasks*

12.1.2 *Nessler Tubes* — of 50 ml capacity.

12.1.3 *Microburette*

12.2 Reagents

12.2.1 *Lime Water Freed from Fluorine* — Dissolve lime in an excess of perchloric acid. Boil for 15 minutes. Dilute, cool and neutralize with flourine-free sodium hydroxide. Filter through a Buchner funnel and wash. Make a saturated solution in distilled water using the lime thus freed from fluorine.

12.2.2 *Perchloric Acid Solution* — 60 to 70 percent (*m/v*). Heat some quantity for an hour or longer at 140 to 150°C.

12.2.3 *Silver Perchlorate Solution* — 1 percent (*m/v*). Prepare by adding sufficient sodium hydroxide solution to a solution of silver nitrate to cause precipitation. Filter and wash the precipitate with water. Dissolve the precipitate in perchloric acid and dilute.

12.2.4 *Sodium Hydroxide Solution* — 0·5 N.

12.2.5 *Alizarin Indicator Solution* — Dissolve 0·02 g of sodium alizarin sulphonate in water and make up the volume to 100 ml.

12.2.6 *Dilute Hydrochloric Acid* — 0·05 N.

12.2.7 *Buffer Solution* — Dissolve 0·1 g of hydroxylamine hydrochloride in water and make up the volume to 100 ml.

12.2.8 *Thorium Nitrate Solution* — Dissolve 0·5 g of hydrated thorium nitrate [$Th(NO_3)_4,12\ H_2O$] in distilled water and make up the volume to one litre.

12.2.9 *Standard Fluorine Solution* — Dissolve 2·211 g of sodium fluoride in water and make up the volume to one litre in a graduated flask. Pipette out 10 ml of this solution into a one-litre graduated flask and make up the volume. This solution contains 0·01 mg of fluorine per millilitre.

12.3 Procedure

12.3.1 Weigh accurately about 5 g of the material. Moisten with lime water. Dry on a water-bath and ignite in a muffle furnace at about 550 ± 20°C. When the ashing is complete, cool and transfer to distillation flask, washing it with water. Dissolve the residual ash in 10 to 15 ml of perchloric acid and transfer to the same distillation flask. Add sufficient silver perchlorate to bring about complete precipitation. Steam-distil at 132 ± 3°C into another flask containing 2 ml of the sodium hydroxide solution. Collect about 150 ml of the distillate. Transfer the distillate to a 200-ml graduated flask and make up the volume to the mark with water.

12.3.2 Transfer a suitable aliquot of the test solution containing 10 to 30 μg of fluorine to a Nessler tube. Add 1 ml of alizarin indicator solution. Take the same quantity of the indicator solution in another Nessler tube. If necessary, the alkali in the test solution is neutralized with a drop or two of hydrochloric acid. To each tube, add 1 ml of the buffer solution, 2·0 ml of hydrochloric acid and dilute to about 45 ml. The colour of the solutions should be straw-yellow. Add the thorium nitrate solution from a microburette to the test solution until a permanent slight pink colour appears (0·5 to 2·5 ml of thorium nitrate solution would be required). Add an equal volume of the thorium nitrate to the other Nessler tube. Adjust the colour of the solution in this Nessler tube by adding the standard fluorine solution from a micro-burette to the same intensity as the colour of the test solution. When the colour in both the tubes matches, the amount of flourine in the standard flourine solution added is equal to the amount of flourine present in the aliquot of the test solution. From this, calculate the amount of fluorine present in 100 g of the material on moisture-free basis.

13. DETERMINATION OF ZINC

13.1 Apparatus

13.1.1 *Colorimeter* — A photo-electric colorimeter or suitable spectrophotometer capable of measuring optical density at a wavelength of 540 nm.

13.2 Reagents

13.2.1 *Copper Sulphate Solution* — Prepared by dissolving 8·0 g copper sulphate ($CuSO_4$, $5H_2O$) in distilled water and diluted to one litre.

13.2.2 *Ammonium Citrate Solution* — Dissolve 225 g ammonium citrate [$(NH_4)_3$ $C_6H_5O_7$] in distilled water, make alkaline to phenol red (*p*H ± 7·4) with concentrated ammonium hydroxide (±25 percent) and add a further 75 ml. Dilute to 2 litres. Before use, purify by adding a slight excess of dithizone solution (*see* **13.2.5**) and extract with successive portions of carbon tetrachloride until the solvent layer has a clear bright green colour. Remove the dithizone remaining in the solution by means of successive extractions with chloroform followed by a final extraction with carbon tetrachloride. (The dithizone shall be entirely removed to prevent loss of zinc during the removal of cobalt.)

13.2.3 *α-Nitroso-β-Naphthol Solution* — Dissolve 0·25 g α-nitroso-β-naphthol in chloroform and make the volume up to 500 ml with chloroform.

13.2.4 *Chloroform* — redistilled. Store in an amber bottle.

13.2.5 *Dithizone Solution* — Dissolve 0·050 g dithizone in 2 ml of 25 percent ammonia solution and make up to 100 ml with water. Extract with successive portions of carbon tetrachloride until the solvent layer has clear bright green colour. Discard the solvent and filter the aqueous solution through washed ashless filter paper.

13.2.6 *Carbon Tetrachloride* — redistilled.

13.2.7 *Standard Hydrochloric Acid Solution* — 0·04 N.

13.2.8 *Standard Zinc Solution* — Dissolve 0·500 g of pure granulated zinc in a slight excess of hydrochloric acid. Dilute to 1 litre with double distilled water. One millilitre of this solution contains 0·5 mg of zinc.

13.2.9 *Standard Zinc Working Solution* — Dilute 10 ml of standard zinc solution (*see* **13.2.8**) with 0·04 N hydrochloric acid to make 1 litre. One millilitre of this solution contains 5 μg of zinc.

13.2.10 *Ammonium Hydroxide Solution* — An aqueous solution of ammonia containing approximately 5 percent ammonia (*m/m*).

13.2.11 *Hydrochloric Acid Solution* — Concentrated hydrocloric acid diluted with distilled water in the proportion 1 : 6.

13.2.12 *Bromine Water* — A saturated solution of bromine in water.

13.2.13 *Hydrogen Sulphide*

13.2.14 *Methyl Red Indicator Solution* — Dissolve 25 mg methyl red in 100 ml 60 percent ethyl alcohol.

13.2.15 *Phenol Red Indicator Solution* — Dissolve 100 mg phenol red sodium salt in 100 ml distilled water.

13.3 Procedure — Dilute 10 ml of the test solution to about 40 ml. Add 2 drops of methyl red indicator and 1 ml copper sulphate solution and neutralize with ammonia. Add sufficient hydrochloric acid solution (**13.2.11**) to bring the concentration of this acid to 0·15 N. The *p*H value of the solution should now be between 1·9 and 2·1. Pass a stream of hydrogen sulphide through the solution until precipitation is complete. Filter through a fine filter paper (previously washed with hydrochloric acid solution and with water) into a 250-ml beaker. Wash the precipitate and filter paper with three or four small portions of water, adding the washings to the filtrate. Boil the solution until all trace of hydrogen sulphide has been removed, add 5 ml of bromine water and continue boiling until free from bromine. Cool,

neutralize to phenol red with ammonium hydroxide solution and add 0·6 ml hydrochloric acid solution. Make up to a suitable volume and take an aliquot containing 4 to 20 μg of zinc for the determination. Adjust the volume of the aliquot to about 20 ml by the addition of distilled water and transfer to a 125-ml separating funnel. Add 5 ml ammonium citrate solution and 10 ml α-nitroso-β-naphthol solution. Shake for 2 minutes, allow the phases to separate and discard the solvent layer.

13.3.1 Wash the aqueous layer with small portions of chloroform to remove residual α-nitroso-β-naphthol. If necessary, adjust the *p*H value of the aqueous solution to 8·0 to 8·2 by the addition of ammonium hydroxide or hydrochloric acid solutions and add 2 ml dithizone solution and 10 ml carbon tetrachloride. Shake for 2 minutes, allow the phases to separate and, using a pipette, withdraw the aqueous phase as completely as possible and discard it. Wash down the sides of the separating funnel with 25 ml distilled water and again withdraw the aqueous phase and discard it. Add 25 ml of 0·04 N hydrochloric acid to the contents of the separating funnel, shake for 1 minute to transfer zinc to the aqueous phase, allow the phases to separate and discard the solvent layer. To the aqueous solution remaining in the separating funnel add 5 ml ammonium citrate solution and adjust the *p*H value, if necessary, to 8·8 to 9·0. Add 10·0 ml carbon tetrachloride accurately measured. Determine the quantity of dithizone solution to be added as follows.

13.3.1.1 To a separating funnel containing 4·0 ml of the standard zinc working solution (20 μg zinc) made up to 25 ml with 0·04 N hydrochloric acid add 5·0 ml ammonium citrate solution and 10 ml carbon tetrachloride; then add dithizone solution from a burette in 0·1 ml increments, shaking after each addition, until a faint yellow colour in the aqueous phase indicates a slight excess of reagent. Multiply the volume of dithizone solution used by 1·5 and add this quantity to the test solution. Shake for 2 minutes and allow the phases to separate. Pipette 5 ml of the solvent phase into a test tube, dilute with 10 ml carbon tetrachloride and use for the determination of the absorption. Prepare a series of standards containing 0·5, 10, 15 and 20 μg zinc diluted in each case to 25 ml with 0·04 N hydrochloric acid solution and treated in the same manner as the test solution. Prepare a blank simultaneously by using the same quantities of reagents as were used in the digestion of the test sample and in the subsequent procedure, making up to 200 ml and developing the colour in the same size aliquot and in the same manner as in the actual determination. Measure the absorptions of the standard and test solutions at 540 nm in the colorimeter. From the absorptions of the standard solutions prepare a graph by plotting absorptions against concentrations and from it determine the concentration of zinc in the test solution. Express the result as percentage of zinc in the test sample.

(*Continued from page* 2)

Animal Feeds and Mineral Mixtures Subcommittee, AFDC 15 : 2

Convener	*Representing*
DR M. L. MATHUR	National Dairy Research Institute (ICAR), Karnal
Members	
DR V. D. MUDGAL (*Alternate* to Dr M. L. Mathur)	
AGRICULTURAL MARKETING ADVISER TO THE GOVERNMENT OF INDIA	Directorate of Marketing and Inspection (Ministry of Agriculture & Irrigation), Faridabad
SHRI S. JAYARAMAN (*Alternate*)	
SHRI M. K. DATTARAJ	Mysore Feeds Private Limited, Bangalore
SHRI M. K. PANDURANGA SETTY (*Alternate*)	
HEAD, ANIMAL NUTRITION DIVISION	Indian Veterinary Research Institute (ICAR), Izatnagar
DR P. V. RAO (*Alternate*)	
DR J. S. ICHHPONANI	Punjab Agricultural University, Ludhiana
DR M. G. JACKSON	Govind Ballabh Pant University of Agriculture & Technology, Pantnagar (Dist Nainital)
DR T. B. MIRCHANDANI	Aries Agro-Vet Industries Private Ltd, Bombay
DR R. S. ADAVI (*Alternate*)	
DR V. M. PATIL	Dairy Development Commissioner, Government of Maharashtra, Bombay
DR B. SAHAI (*Alternate*)	
DR B. C. PATNAYAK	Central Sheep & Wool Research Institute (ICAR), Avikanagar (Rajasthan)
DR D. V. R. PRAKASH RAO	Shaw Wallace & Company Limited, Madras
SHRI B. LAKSHMANAN (*Alternate*)	
SHRI K. V. R. RAO	The Boots Company (India) Ltd, Bombay
DR T. R. NATARAJAN (*Alternate*)	
DR (SMT) SAROJINI CHANDRASHEKHARAN	Kaira District Co-operative Milk Producers' Union Limited, Anand
DR A. S. DAVE (*Alternate*)	
DR H. C. SAXENA	Hindustan Lever Limited, Bombay
DR S. YAMDAGNI (*Alternate*)	
DR P. C. SHUKLA	Institute of Agriculture, Anand
SHRI VINEET VIRMANI	Nandi Provender Mills, New Delhi
SHRI ANAND VIRMANI (*Alternate*)	

BUREAU OF INDIAN STANDARDS

Headquarters :

Manak Bhavan, 9 Bahadur Shah Zafar Marg, NEW DELHI 110002

Telephones : 331 01 31
331 13 75

Telegrams : Manaksanstha
(Common to all Offices)

Regional Offices :		*Telephone*
Central	: Manak Bhavan, 9, Bahadur Shah Zafar Marg, NEW DELHI 110002	331 01 31 331 13 75
* Eastern	: 1/14 C.I.T. Scheme VII M, V.I.P. Road, Maniktola, CALCUTTA 700054	37 86 62
Northern	: SCO 445-446, Sector 35-C, CHANDIGARH 160036	2 18 43
Southern	: C.I.T. Campus, IV Cross Road, MADRAS 600113	41 29 16
† Western	: Manakalaya, E9 MIDC, Marol, Andheri (East), BOMBAY 400093	6 32 92 95

Branch Offices :	
'Pushpak', Nurmohamed Shaikh Marg, Khanpur, AHMADABAD 380001	2 63 48
‡ Peenya Industrial Area, 1st Stage, Bangalore-Tumkur Road, BANGALORE 560058	39 49 55
Gangotri Complex, 5th Floor, Bhadbhada Road, T.T. Nagar, BHOPAL 462003	55 40 21
Plot No. 82/83, Lewis Road, BHUBANESHWAR 751002	5 36 27
Kalai Kathir Building, 6/48-A Avanasi Road, COIMBATORE 641037	2 67 05
Quality Marking Centre, N.H. IV, N.I.T., FARIDABAD 121001	—
Savitri Complex, 116 G.T. Road, GHAZIABAD 201001	8-71 19 96
53/5 Ward No. 29, R.G. Barua Road, 5th By-lane, GUWAHATI 781003	3 31 77
5-8-56C L. N. Gupta Marg, (Nampally Station Road) HYDERABAD 500001	23 10 83
R14 Yudhister Marg, C Scheme, JAIPUR 302005	6 34 71
117/418 B Sarvodaya Nagar, KANPUR 208005	21 68 76
Plot No. A-9, House No. 561/63, Sindhu Nagar, Kanpur Road, LUCKNOW 226005	5 55 07
Patliputra Industrial Estate, PATNA 800013	6 23 05
District Industries Centre Complex, Bagh-e-Ali Maidan, SRINAGAR 190011	—
T. C. No. 14/1421, University P. O., Palayam, THIRUVANANTHAPURAM 695034	6 21 04
Inspection Offices (With Sale Point) :	
Pushpanjali, First Floor, 205-A West High Court Road, Shankar Nagar Square, NAGPUR 440010	52 51 71
Institution of Engineers (India) Building, 1332 Shivaji Nagar, PUNE 411005	5 24 35

*Sales Office Calcutta is at 5 Chowringhee Approach, P. O. Princep Street, CALCUTTA	27 68 00
† Sales Office is at Novelty Chambers, Grant Road, BOMBAY	89 65 28
‡ Sales Office is at Unity Building, Narasimharaja Square, BANGALORE	22 39 71

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 1 MARCH 1993
TO
IS 7874 (Part 2) : 1975 METHODS OF TESTS FOR ANIMAL FEEDS AND FEEDING STUFFS

PART 2 MINERALS AND TRACE ELEMENTS

(*Page* 5, *clause* **5**, *Determination of Calcium*) — This test method is withdrawn and replaced by:

'IS 13433 (Part 1) : 1992	Animal feeds and feeding stuffs — Determination of calcium : Part 1 Titrimetric method
IS 13433 (Part 2) 1992	Animal feeds and feeding stuffs — Determination of calcium : Part 2 Atomic absorption spectrometric method
IS 13574 : 1992	Animal feeds and feeding stuffs — Determination of calcium and magnesium in mineral supplements.'

(FAD 5)

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 2 MARCH 2006
TO
IS 7874 (PART 2) : 1975 METHODS OF TESTS FOR ANIMAL FEEDS AND FEEDING STUFFS
PART 2 MINERALS AND TRACE ELEMENTS

(*Page* 4, *clause* **4.2.1**) — Substitute the following for the existing text:

'Weigh accurately about 1 g of the material and transfer to 250 ml graduated flask. Add 50 ml of the ferric sulphate solution with a pipette, gently swirling the flask add 100 ml of the ammonium hydroxide solution. Add sufficient quantity of glass distilled water to bring it to the mark. Swirl the flask enough to ensure thorough mixing but avoid vigorous shaking. Allow to settle for 10 minutes. Filter through 11-cm Whatman filter paper No. 41 or its equivalent. Transfer from the filtrate, an aliquot of 25 ml to a 250-ml beaker. Add 10 ml of nitric acid and 10 ml of the ferric sulphate indicator solution. Then add with constant stirring, known quantity of the standard silver nitrate solution in slight excess. Heat the solution to the boil and cool to room temperature and stir to coagulate the precipitate.Titrate the excess of silver nitrate with the standard potassium thiocyanate solution. The end point is indicated by the first appearance of reddish tint that persists for 15 seconds.'

(*Page* 5, *clause* **4.3.1**) — Substitute the following for the existing text:

'Chlorine as sodium chloride (on moisture free basis), percent by mass $= \dfrac{58.45 \times (AN_1 - BN_2)}{m\,(100 - M)} \times 100$

where

A = volume in ml of the standard silver nitrate solution used,
N_1 = normality of the standard silver nitrate solution,
B = volume in ml of the standard potassium thiocyanate solution used up by the excess silver nitrate solution,
N_2, = normality of the standard potassium thiocyanate solution,
m = mass in g of the material taken for the test (see **4.2.1**), and
M = percent moisture content."

(FAD 5

Reprography Unit, BIS, New Delhi, India