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Indian Standard

METHODS OF SAMPLING AND TEST (PHYSICAL AND CHEMICAL) FOR WATER AND WASTEWATER

PART 53 IRON

(First Revision)

ICS 13.060.50

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

This Indian Standard (First Revision) was adopted by the Bureau of Indian Standards, after the draft finalized by the Environment Protection and Waste Management Sectional Committee had been approved by the Chemical Division Council.

Pollution caused by substances, on which biotic and abiotic agencies of decomposition are ineffective, is a unique type of pollution. Toxic trace elements and heavy metals come under the category of non-degradable pollutants. The problem caused by these elements is in fact due to their concentration in the environment in the bio-available state and above a certain concentration become harmful to the living organism.

Iron (Fe) is naturally abundant in earth's crust. Amount of iron available in soluble form depends upon the concentration of the complex forming ions, pH and oxidation conditions. In the absence of the complex forming ions, ferric iron is not significantly soluble unless the pH is very low. Oxygenated surface waters seldom contain more than 1 mg/l of iron. Ground waters and the surface waters which are acidic, may, on the other hand, contain considerably more iron. In water samples, iron may be present, as free hydrated ions, in the form of organic/inorganic complex ions, in a colloidal state or as relatively coarse suspended particles. Iron in water can cause staining of laundry and porcelain. A bitter sweet astringent taste is imparted to the drinking water at levels above 1 mg/l of iron. Iron appears to be an essential element for all organisms - both plant and animals. In animals iron is found in many important proteins, major functions of these proteins are in oxygen storage and transport.

As per IS 10500 : 1991 'Drinking water specification (first revision)', the permissible limits of iron in drinking water is 0.3 mg/l, Max. Beyond this limit, taste/appearance are affected, has adverse effect on domestic uses and water supply structures, and promotes iron bacteria.

In view of the environmentally prevalent nature of iron (Fe) compounds in levels should be closely monitored to avoid any likelihood of toxicity caused by this element.

The technical committee responsible for the formulation of IS 3025 : 1964 had decided to revise the standard and publish it as separate parts. This standard supersedes 32 of IS 3025 : 1964.

In the preparation of this standard, considerable assistance is derived from 'Standard Methods for the Examination of Water and Wastewater', 19th Edition-1995, published by the American Public Health Association, Washington, U.S.A.

The spectrometric method for determination of total dissolved iron and total (both dissolved and undissolved) iron using 1,10 phenanthroline are technically equivalent to ISO 6632 : 1988 'Water quality — Determination of iron — Spectrometric method using 1,10 phenanthroline'.

The composition of the Committee responsible for formulation of this standard is given at Annex A.

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 'Rules for rounding off numerical values (revised)'.
Indian Standard

METHODS OF SAMPLING AND TEST (PHYSICAL AND CHEMICAL) FOR WATER AND WASTEWATER

PART 53 IRON

( First Revision )

1 SCOPE

1.1 This standard prescribes two methods for the determination of iron:
   a) 1,10 phenanthroline method, and
   b) Atomic absorption method.

1.2 Depending upon the concentration range and interference levels, choice of the method is made.

2 REFERENCES

The standards listed below contain provisions which through reference in this text, constitute provisions of this standard. At the time of publication, the editions indicated were valid. All standards are subject to revision and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards.

<table>
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<td>Methods of sampling and test (physical and chemical) for water and wastewater : Part 1 Sampling (first revision)</td>
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<td>7022 (Part 1)</td>
<td>Glossary of terms relating to water, sewage and industrial effluents, Part 1</td>
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<tr>
<td>7022 (Part 2)</td>
<td>Glossary of terms relating to water, sewage and industrial effluents, Part 2</td>
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3 TERMINOLOGY

For the purpose of this standard, definitions given in IS 7022 (Part 1) and IS 7022 (Part 2) shall apply.

4 SAMPLING AND STORAGE

The sampling shall be done as prescribed in IS 3025 (Part 1). The sampling bottles shall be cleaned thoroughly with dilute nitric acid (6N), prior to the final rinsing with water. The water samples should be collected and stored preferably in polypropylene bottle or chemically resistant glass containers. For the determination of dissolved iron content, filtration through 0.45 μm membrane filter, at the time of sampling, is required. The analysis of such samples is to be carried out within 24 h of sampling. For preservation, the samples should be acidified with concentrated nitric acid (2 ml of conc nitric acid in 1 litre sample, just to bring down the pH below 2). The acidified samples can be stored for a few days (up to 5 days) in a refrigerator.

Iron in water may vary in concentration and form with duration and degree of flushing when taking a sample. Shake bottle vigorously to obtain uniform suspension of precipitated iron. Use particular care for when colloidal iron adheres to the sampling bottle. The problem is more acute with plastic bottle.

5 PURITY OF THE REAGENTS

Unless specified otherwise, only pure chemicals and iron free distilled water shall be used in tests.

NOTE — ‘Pure chemicals’ shall mean chemicals that do not contain impurities which affect the result of analysis.

6 1,10 PHENANTHROLINE METHOD

6.1 Principle

Iron in the solution is reduced to the ferrous state by boiling with hydrochloric acid and hydroxylamine, and treated with 1,10 phenanthroline at pH to 3.3. Three molecules of phenanthroline chelate each atom of ferrous iron to form an orange-red complex. The coloured solution obeys Beer's law. Its colour intensity is independent of pH from 3 to 9. A pH between 2.9 and 3.5 insures rapid colour development in the presence of an excess of phenanthroline. This method is applicable in the range of 75 to 500 μg/l of iron.

6.2 Interference

Among the interfering substances are strong oxidizing agents, cyanide, nitrite, and phosphates (polyphosphates more so than orthophosphates), chromium, zinc in concentrations exceeding 10 times that of iron, cobalt and copper in excess of 5 mg/l, and nickel in excess of 2 mg/l. Bismuth, cadmium, mercury, molybdate, and silver precipitate phenanthroline. The initial boiling with acid converts polyphosphates to orthophosphate and removes cyanide and nitrite that otherwise would interfere. Adding excess hydroxylamine eliminates errors caused by excessive concentrations of strong oxidizing reagents. In the presence of interfering metals, if noticeable amount of
colour or organic matter is present evaporate the sample and dissolve the residue in acid. Ashing is carried out in silicon, porcelain or platinum crucible that have been boiled for several hours in 6 N HCl. Use a larger excess of phenanthroline to replace that complexed by the interfering metals. The presence of excessive amounts of organic matter may necessitate digestion of sample before extraction of iron.

6.3 Apparatus

6.3.1 Spectrophotometer — For use at 510 nm, providing a light path of 1 cm or longer.

6.3.2 Acid-washed Standard Volumetric Glassware

NOTE — Wash all glassware with concentrated hydrochloric acid (HCl) and rinse with distilled water before use to remove deposits of iron oxide.

6.4 Reagents

6.4.1 Hydrochloric Acid — concentrated (11N) containing less than 0.000 05 percent iron.

6.4.2 Hydroxylamine Solution

Dissolve 10 g of NH₄OH.HCl in 100 ml water. This solution is stable for at least one week.

6.4.3 Ammonium Acetate Buffer Solution

Dissolve 250 g NH₄C₂H₃O₂ in 150 ml water. Add 700 ml glacial acetic acid. Prepare new reference standards with each buffer preparation.

6.4.4 Sodium Acetate Solution

Dissolve 200 g NaC₂H₃O₂.3H₂O in 800 ml water.

6.4.5 1.10 Phenanthroline Solution

Dissolve 100 mg 1.10 phenanthroline monohydrate, C₁₂H₈N₂H₂O, in 100 ml water, by stirring and heating to 80°C. Do not boil. Discard the solution, if it darkens. Heating is not necessary if 2 drops conc HCl are added to the water. This solution is stable for at least one week.

NOTE — One millilitre of this reagent is sufficient up to 100 µg of Fe.

6.4.6 Stock Iron Solution

Add carefully and with constant stirring 20 ml conc H₂SO₄ to 50 ml water contained in 250 ml beaker (cooled in ice water) and dissolve 1.404 g Fe (NH₄)₂(SO₄)₆H₂O. Add 0.1N potassium permanganate (KMnO₄) drop wise until a faint pink colour persists. Transfer quantitatively to a 1 000 ml volumetric flask and make up to the mark with water and mix (1.0 ml = 200 µg of Fe).

6.4.7 Standard Iron Solutions

Pipette out 50.0 ml stock solution into a 1 000 ml volumetric flask and make up to the mark with water and mix well (1.0 ml = 10.0 mg of Fe). Pipette 5.0 ml stock solution into a 1 000 ml volumetric flask and make up to the mark with water and mix well (1.0 ml = 1.0 µg of Fe).

NOTE — Prepare standard iron solutions daily for use.

6.4.8 Diisopropyl Ether

Caution — Diisopropyl ether is highly inflammable.

6.5 Procedure

6.5.1 Calibration Curve

Pipette out appropriate portions of standard iron solution into 125 ml conical flasks to contain from 10 to 100 µg of Fe. For the reagent blank, pipette out 10 ml of water to a separate conical flask. Dilute the contents of each conical flask to about 50 ml by adding water. To each flask, add 1 ml NH₄OH.HCl solution and 2 ml conc HCl. Add a few boiling chips and boil the solution until the volume is reduced to about 20 ml. Cool to room temperature and quantitatively transfer to 100 ml volumetric flasks. Add 10 ml ammonium acetate buffer solution first and add 10 ml 1,10 phenanthroline solution to each flask. Dilute to 100 ml with water and mix thoroughly and allow to stand for 10 to 15 min. Measure the absorbance of the iron complexes at 510 nm against the reagent blank. Construct a calibration curve by plotting absorbance values against micrograms of iron in 100 ml of the final solution.

6.5.2 Determination of Total Iron

If the sample contains an excessive amount of organic matter, then sample is digested with HNO₃ and H₂SO₄ and the iron present is separated by extraction process as described under. (In the absence of organic matter total iron contents, is determined as described in 6.5.5.)

6.5.3 HNO₃·H₂SO₄, Digestion

Transfer a suitable volume of the homogenized sample to a beaker. (Sample volume depends upon the expected Fe content. If the expected Fe concentration is of the order of 1 mg/l, the sample volume will be about 800 ml. If Fe concentration is between 10 to 100 mg/l, about 100 ml sample will be required.) Add 5 ml conc HNO₃ and a few boiling chips to both the solutions. Heat to boil the solutions, and concentrate carefully on a hot plate to lowest possible volume, if required cool the solutions and transfer quantitatively, to two separate beakers of smaller size (say 150 ml beaker). Add 5 ml conc HNO₃ and 10 ml conc H₂SO₄ to each of these. Heat up to the evolution of dense white fumes of SO₃. If the solution is not clear add 5 ml HNO₃ and re-heat until a clear solution (no evolution of brown fumes) results. Cool, transfer
quantitatively to a 100 ml volumetric flask. Dilute up to the mark with water, and mix well. Use this solution for extraction of iron as described in 6.5.3.

6.5.4 Extraction of Iron

Pipette out a portion of the solution obtained from acid digestion, containing 10 to 100 µg of iron in a separatory funnel. Add about 25 ml of conc HCl to this. Mix and allow to cool to room temperature. Extract the iron from the HCl solution in the separatory funnels by shaking with 25 ml of diisopropyl ether. Draw off the lower HCl layer into another separatory funnel; extract both again with 25 ml of diisopropyl ether. Continue the extraction process three times. Separate the acid layer and discard it. Transfer the combined ether layer to the original separatory funnel. Add about 25 ml of cone HCl to this. Mix and allow to stand for 15 min. Measure the absorbance of the iron complexes at 510 nm against the reagent blank.

6.5.5 Colour Development and Measurement

6.5.5.1 Soluble iron (for filtered sample direct determination)

Pipette out a portion of filtered sample (filtered through 0.45 µm membrane filter), containing 50-60 µg of iron in 150 ml conical flask. Dilute the contents of conical flask to about 50 ml by adding water. Add 1 ml NH₄OH.HCl solution and 2 ml conc HCl. Add a few boiling chips and boil the solution until the volume is reduced to about 20 ml. Cool to room temperature and quantitatively transfer to 100 ml volumetric flasks. Add 10 ml ammonium acetate buffer solution first and add 10 ml 1,10 phenanthroline solution to each flask. Dilute to 100 ml with water and mix thoroughly and allow to stand for 10 to 15 min. Measure the absorbance of the iron complexes at 510 nm against the reagent blank.

6.5.5.2 For samples after digestion and extraction

Add 1 ml of NH₄OH.HCl solution, add 10 ml ammonium acetate buffer solution and then add 10 ml 1,10 phenanthroline solution to each flask. Ten ml of phenanthroline solution in to the volumetric flasks containing extracted iron (see 6.5.2 and 6.5.3). Dilute to 100 ml with water, mix thoroughly and allow to stand for 10 min. Measure the absorbance of the iron-complex at 510 nm against the reagent blank prepared in an identical manner (using water instead of the sample solution). Determine micrograms of iron in the solution from the absorbance reading, by referring to the calibration curve.

6.6 Calculation

6.6.1 Soluble Iron (Direct Determination without the Digestion Step)

\[
\text{mg of Fe} = \frac{\mu g \text{ of Fe (in 100 ml of the final solution)}}{V}
\]

where

\[
V = \text{Volume in ml of the sample used.}
\]

6.6.2 Total Iron (When the Digestion is Carried Out)

\[
\text{mg of Fe/l} = \frac{\mu g \text{ of Fe (in 100 ml of the final solution) \times 100}}{V_1 \times V_2}
\]

where

\[
V_1 = \text{volume in ml of the sample used, and}
\]

\[
V_2 = \text{total volume in ml of digested solution used for Fe determination.}
\]

6.6.3 Precision and Accuracy

Precision and accuracy of this method depends upon the methods of sampling, methods of colour measurements (visual colourimetry or spectrophotometry), the Fe concentration and the presence of interfering colour, turbidity and foreign ions. The relative standard deviation reported in the literature for iron in the 300 µg/l concentration range is 25.5 percent.

7 ATOMIC ABSORPTION METHOD (DIRECT)

7.1 Principle

The iron content of the sample is determined by atomic absorption spectrophotometry. For dissolved iron, the filtered sample is directly aspirated to the atomizer. For total recoverable iron, HNO₃-H₂SO₄ digestion is to be carried out prior to aspiration of the sample. This method is applicable in the 0.1 to 10 mg/l range. However, the concentration range will vary with the sensitivity of the instrument used.

7.2 Interferences

Interferences of cobalt, copper, nickel can be controlled by using a very lean (hot) flame. Silicons depressed the iron response, and can be overcome by the addition of 0.2 percent calcium chloride.

7.3 Apparatus

7.3.1 Atomic absorption spectrophotometer with air-acetylene flames. Multi-element hollow-cathode lamp or electrodeless discharge lamp for use at 248.3 nm.

7.4 Reagents

7.4.1 Hydrochloric Acid — concentrated (11 N) and dilute to 1 : 1.
7.4.2 Nitric Acid — concentrated (16 N) and dilute (1 : 499).

7.4.3 Sulphuric Acid — concentrated (36 N).

7.4.4 Calcium Chloride Solution
Dissolve 630 mg CaCO₃ in 50 ml of 20 percent v/v HCl. If required, boil gently to obtain complete solution. Cool and dilute to 1000 ml with water.

7.4.5 Stock Iron Solution
Dissolve 0.100 g of iron wire in a mixture of 10 ml 1:1 HCl and 3 ml of conc HNO₃ and dilute to 1000 ml with water in a volumetric flask (1.0 ml = 100 μg of Fe).

7.5 Procedure

7.5.1 Calibration Curve
Prepare sufficient standard solutions containing 0 to 60 μg/l of iron by diluting suitable volumes of standard iron solution with nitric acid (1 : 499) to 100 ml in volumetric flasks. Transfer the contents of the volumetric flasks to 150 ml beakers. Add 25 ml of CaCl₂ solution to each of the volumetric flasks. Prepare a reagent blank using 10 ml of water in a similar manner. Aspirate the reagent blank and carry out zero adjustment. Aspirate sequentially the standard solutions and measure the absorbance at 248.3 nm. Construct a calibration curve by plotting absorbance values against micrograms of Fe in 100 ml final volume. Ensure that the calibration curve is linear, make the necessary changes in the volume of the standard solution used. (Increase in the final volume due to the CaCl₂ solution is not to be considered for calculation of micrograms of Fe.)

7.5.2 Determination of Iron
For the determination of dissolved iron content, filter the sample through 0.45 μm membrane filter. For total recoverable iron, digest the sample with HNO₃·H₂SO₄ (see 6.5.2).

Add 0.5 ml of conc nitric acid to a suitable volume of the sample taken (or the solution obtained after digestion, which contains 50 to 60 μg iron) in a 100 ml volumetric flask. Make up to the mark. Transfer the contents of the volumetric flask to a 150 ml beaker. Add 25 ml CaCl₂ solution to this. Prepare a reagent blank with 100 ml water. Rinse the nebulizer by aspirating water containing 1.5 ml conc HNO₃/l. Aspirate the reagent blank and carry out zero adjustment. Aspirate the sample solution and measure the absorbance at 248.3 nm. Determine micrograms of iron in the solution from the absorbance reading, by referring to the calibration curve.

7.6 Calculation

7.6.1 Soluble Iron (Direct Determination without the Digestion Step)

\[
\text{mg Fe/l} = \frac{\mu g \text{ of Fe (in 100 ml of the final solution)}}{V}
\]

where

\[V = \text{volume in ml, of the sample used.}\]

7.6.2 Total Iron (When the Digestion is Carried Out)

\[
\text{mg Fe/l} = \frac{\mu g \text{ of Fe (in 100 ml of the final solution)}}{V_1 \times V_2} \times 100
\]

where

\[V_1 = \text{volume in ml of the sample used, and}\]
\[V_2 = \text{total volume in ml of the digested solution used for Fe estimation.}\]

7.6.3 Precision and Accuracy
The relative standard deviation reported in the literature for Fe in the 300 μg/l concentration range is 16.5 percent.
# ANNEX A

**(Foreword)**

## COMMITTEE COMPOSITION

**Environment Protection and Waste Management Sectional Committee, CHD 32**

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</tr>
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<tr>
<td>Central Pollution Control Board, Delhi</td>
<td>Shri Dilip Biswas (Chairman)</td>
</tr>
<tr>
<td>Automotive Research Association of India, Pune</td>
<td>Representative</td>
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<td>Dr. S. Sadashivan</td>
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<tr>
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<tr>
<td>Central Fuel Research Institute, Dhanbad</td>
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<tr>
<td>Central Leather Research Institute, Chennai</td>
<td>Dr. Gulab Singh</td>
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<td>Central Mechanical Engineering Research Institute, Durgapur</td>
<td>Dr. L. C. Rakhe (Alternate)</td>
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<tr>
<td>Central Mining Research Institute, Dhanbad</td>
<td>Dr. S. Rajaman</td>
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<tr>
<td>Central Pollution Control Board, New Delhi</td>
<td>Representative</td>
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<tr>
<td>Confederation of Indian Industries (CII), New Delhi</td>
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<td>Department of Industrial Policy &amp; Promotion, New Delhi</td>
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<td>Department of Science &amp; Technology (TIFAC), New Delhi</td>
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<td>Directorate General Factory Advice Service and Labour Institute, Mumbai</td>
<td>Shri S. S. Gautam</td>
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<td>Directorate General of Health Services, New Delhi</td>
<td>Shri M. R. Raoput (Alternate)</td>
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<td>Engineers India Limited, New Delhi</td>
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<td>Envirotech Instruments Private Limited, New Delhi</td>
<td>Shri B. B. Lal</td>
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<td>Food Research &amp; Analysis Centre, New Delhi</td>
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<td>Dr. R. Prabhakaran (Alternate)</td>
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<td>Indian Council of Agricultural Research, New Delhi</td>
<td>Smt. P. S. Shah</td>
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<td>Indian Council of Medical Research, New Delhi</td>
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<tr>
<td>Municipal Corporation of Greater Mumbai, Mumbai</td>
<td>Shri H. K. Dubey (Alternate II)</td>
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<td>National Institute of Occupational Health (ICMR), Ahmedabad</td>
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Organization

National Environmental Engineering Research Institute, Nagpur

National Organic Chemical Industries Limited, Mumbai

National Productivity Council, New Delhi

National Thermal Power Corporation Limited, New Delhi

NCCBM, New Delhi

Pesticides Association of India, New Delhi

Shriram Institute of Industrial Research, New Delhi

SGS India Limited, Chennai

Standardization Testing and Quality Certification Directorate, New Delhi

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The Fertilizer Association of India, New Delhi

Thermax Limited, Pune

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Dr A. V. Shexdar (Alternate II)

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Shri B. V. Bapat (Alternate)
Shri R. C. Monga
Dr A. K. Saxena (Alternate)

Representative

Representative

Shri P. N. Parmeshwaran
Representative

Shri S. Ravi
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Shri J. Kumar (Alternate I)
Dr V. M. Shaheti (Alternate II)

Dr (Shrikrishna) B. Swaminathan
Dr S. Nand (Alternate)

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(Representing Director General (Ex-officio))

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Shri N. K. Pal
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This Indian Standard has been developed from Doc: No. CHD 32 (1012).

Amendments Issued Since Publication

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