Appendix

Analytical Methods

B

WARNING!

Aqua Ammonia sample containers and contents need to be cooled to between 5°C and 10°C and then maintained at that temperature to prevent loss of ammonia gas (NH_3) from the sample when opening the container and transferring for tests! All transferring shall be done as quickly as possible!

Failure to follow this requirement is likely to result in erroneous results when testing for ammonia concentrations!

When utilizing hydrometer measurements, realize that a 1 Fahrenheit degree error in the temperature observation results in a concentration measurement error on the order of 0.1%. Similarly, a 0.1 Baumé degree error results in a concentration measurement error on the order of 0.2%. Note than these errors can be additive, e.g., a Baumé reading 0.1° too low concurrent with a temperature reading 1 Fahrenheit degree too high will cause an error in the concentration calculation on the order of 0.3% below the true ammonia concentration!

The following information is taken from Federal Specification OA-451-F Ammonium Hydroxide Technical, available from the GSA Specification Section, Room 6654, 7th and D Sts., S.W., Washington, D.C. 20407.

Warning - Containers and contents shall be cooled to between 5°C and 10°C and maintained at this temperature to prevent the loss of ammonia gas (NH3) when operating and transferring for tests. All transferring shall be done as quickly as possible.

Determine by visual examination.

Evaporate 100 milliliters of the sample to dryness in a tared platinum crucible or other suitable dish, ignite, and weigh. Calculate the gain in weight of the crucible as percent total solids.

Determine specific gravity by method ASTM Designation D1122.

Introduce a glass-stoppered weighing bottle containing about 10g of the sample into an 800ml. Erlenmeyer flask containing about 200ml. Of distilled water and sufficient 0.5N sulfuric acid to combine with the ammonia and about 10ml excess. Stopper the flask and warm gently until the stopper in the weighing bottle is forced out and the ammonia combines with the acid. Upon thorough mixing, allow the solution to cool and titrate the excess acid with 0.5N sodium hydroxide using methyl red as indicator. Calculate the percentage of ammonia in the sample.

$\% NH_3 = [(AB-CD) \times 0.017 \times 100] / s$

A = ml. Of sulfuric acid

B = Normality of sulfuric acid

C = ml. Of sodium hydroxide solution to titrate excess acid

D = Normality of sodium hydroxide

S = Weight of sample in grams

Analytical Procedure
Ammonium Hydroxide (Aqua Ammonia)
Technical Grade

Procedure For Specific Gravity Test

Equipment and Apparatus:

473ml flint glass sample container with cap (Fisher Scientific #03-326-2F or equivalent) 0-120°F thermometer (Taylor Red Line 21413-1 or equivalent)

Vertical cylinder hydrometer flask (Fisher Scientific #08-530K or equivalent) Hydrometer (Fisher Scientific #11-55E or equivalent)

Appendix A, pages 15-18, graphs and charts relating aqua ammonia concentration, degrees Baumé, specific gravity and temperature.

Assay:

1. Cool the first four items mentions above to at least 5-10°C (40-50°F).

2. Open sample valve and flush lines. It is recommended that the sample line is routed through a chiller or ice bath to ensure that the sample being withdrawn is cooled to at least 5-10°C (40-50°F) before it is exposed to the atmosphere. Place the thermometer inside a cooled sample container. Fill about 3/4 full of aqua ammonia. Recap sample container. Allow for air bubbles to settle. Record temperature.

3. Place the hydrometer inside the vertical cylinder hydrometer flask. Pour sample into the hydrometer flask. Maintain flask and contents at 5-10°C (40-50°F) if the specific gravity reading requires more than 20 seconds to complete. Allow the hydrometer to stabilize. Record the specific gravity or degrees Baumé reading.

4. Refer to the aqua ammonia concentration conversion graph and additional tables as necessary in Appendix A, pages 15-28. Calculate the final concentration.

5. Compare readings from hydrometers in service with new stand-by hydrometers on a monthly basis. Hydrometers shall be calibrated by an external test laboratory on an annual basis or as often as desired. Calibration shall be done with certified hydrometers or by comparison to titration results.

Analytical Procedure
Ammonium Hydroxide (Aqua Ammonia)
Procedure For Assay By Titration

Principle:

A sample of aqua ammonia is added to an excess of 3N hydrochloric acid. The excess is then backtitrated with 2N sodium hydroxide.

Equipment and Apparatus:

Bottle, pressure, 200ml flask shape, with stopper and spring clamp.

Bottle, weighing, high-form, glass stopper, 40 x 80 mm (70ml) or equivalent. A glass-stoppered 250 ml Erlenmeyer flask is suitable.

Pipet, 50ml calibrated.

Pipet sampling device. This consists of a two-hole #2 rubber stopper through which are inserted a 10ml pipet and a short piece of glass tubing bent at a right angle. A 2oz rubber bulb is used to force aqua ammonia from the sample bottle into the pipet. Since the glass tube is small, a short piece of tubing is needed as an adapter between the glass tube and the bulb.

Buret, 25ml calibrated.

Reagents:

Hydrochloric acid, 3N solution. Purchase standardized solution of dilute 250ml of concentrated, reagent grade hydrochloric acid to 1L with distilled water and standardize against standard 2N sodium hydroxide.

Sodium hydroxide, standard 2N solution. Purchase standardized or dilute 160g of 50% sodium hydroxide solution to 1L with recently boiled, distilled water. Standardize against primary standard benzoic acid or potassium acid phthalate.

Methyl red indicator, 0.1% solution in water. Dissolve 0.10g of the sodium salt of methyl red in 100ml of distilled water. If necessary, adjust the pH to 7.

Procedure:

1. The sample is received in a pressure bottle. Cool the sample to 50°F or lower as a safety measure and to prevent losses of ammonia.

2. Pipet exactly 50ml of standard 3N hydrochloric acid into the weighing bottle, and obtain the tare weight. By means of the sampling device, and allow it to drain into the weighing bottle with stirring until about 1/3 remains. Lift the pipet, touch off the last drop of drainage, and remove. Obtain the weight of the bottle containing add acid sample.

3. Carefully pour the contents of the weighing bottle into a 250ml Erlenmeyer flask. Add 3 drops of methyl red indicator solution and titrate the excess acid with standard 2N sodium hydroxide until the indicator color changes to yellow. Rinse the weighing bottle into the flask and complete the titration.

4. Calculation:

$[(3A-2C)/W] \times 0.01703 \times 100 = \%$ ammonia by weight where A=ml 3N HCI, C=ml 2N NaOH, and w=weight of sample in grams.

Analytical Procedure
Ammonium Hydroxide (Aqua Ammonia)
FCC Grade

The following test procedures are reprinted with permission from the FOOD Chemical Codex. Fourth Edition, Copyright 1996 by the National Academy of Sciences. Courtesy of the National Academy Press, Washington, D.C.

Assay:

Tare accurately a 125ml glass-stoppered Erlenmeyer flask containing 35.0 ml of 1 N sulfuric acid. Partially fill a 10ml graduated pipet from near the bottom of a sample, previously cooled in the original sample bottle to 10°C or lower. (Do not use a vacuum for drawing up the sample.) Wipe off any liquid adhering to the outside of the pipet, and discard the first ml. Hold the pipet just above the surface of the acid, and transfer 2ml into the flask, mix, and weigh again to obtain the weight of the sample. Add methyl red TS, and titrate the excess acid with 1N sodium hydroxide. Each ml of 1N sulfuric acid is equivalent to 17.03mg of NH_3.

Methyl Red TS:

Dissolve 100mg of methyl red in 100ml of alcohol, and filter if necessary.

Arsenic:

Evaporate 11ml (10g sample) to about 2ml on a steam bath, dilute to 50ml with water, and mix. A 5 ml portion of this solution meets the requirements o the Arsenic Test, page 47.

[For Arsenic Test, see following pages.]

Heavy Metal:

Transfer 22ml (20g sample) to a beaker, add about 5mg of sodium chloride, evaporate to dryness on a steam bath, and dissolve the residue in 2ml of diluted acetic acid TS and sufficient water to make 50ml. A 10ml portion of this solution, diluted to 25ml with water, meets the requirements of the Heavy Metals Test, page 512, using 20µg of lead ion (Pb) in the control (Solution A).

Acetic Acid TS, Diluted:

A solution containing about 6% (w/v) of CH_3COOH Prepare by diluting 60.0ml of glacial acetic acid, or 166.6ml of 36% acetic acid (6N), with sufficient water to make 1000ml.

Nonvolatile Residue:

Evaporate 11ml (10g sample) in a tared platinum or porcelain dish to dryness, dry at 105°C for 1 h, cool, and weigh.

Readily Oxidizable Substances:

Dilute 4ml with 6 ml of water, and add a slight excess of diluted sulfuric acid TS and 0.1ml of 0.1N potassium permanganate. The pink color does not completely disappear within 10min.

Sulfuric Acid TS, Diluted:

A solution containing 10% (w/v) of H_2SO_4 Prepare by cautiously adding 57ml of sulfuric acid (95% to 98%) or sulfuric acid TS to about 100 ml of water, then cool to room temperature, and dilute with water to 1000ml.

Packaging and Storage:

Store in tight containers, preferably at a temperature not exceeding 25°C.

Functional Use In Food:

Alkali

Silver Diethyldithiocarbamate Colorimetric Method

Note: All reagents used in this test should be very low in arsenic content.

The general apparatus shown in Fig. 2 is to be used unless otherwise specified in an individual monograph. It consists of a 125ml arsine generator flask (a) fitted with a scrubber unit (e) and an absorber tube (e), with a 24/40 standard-taper joint (b) and a ball-and-socket joint (d), secured with a No. 12 clamp, connecting the units. The tubing between d and e and between d and c is a capillary diameter of 8mm. Alternatively, an apparatus embodying the principal of the general assembly described and illustrated may be used.

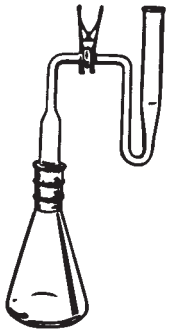

FIGURE 2 General Apparatus for Arsenic Test (Courtesy of the Fisher Scientific Co., Pittsburgh, Pa.)

Weigh accurately 132.0mg of arsenic trioxide that has been previously dried at 105° for 1h, and dissolve it in 5ml of sodium hydroxide solution (1 in 5). Neutralize the solution with diluted sulfuric acid TS, add 10ml in excess and dilute to 1000.0ml with recently boiled water. Transfer 10.0ml of this solution into a 1000-ml volumetric flask, add 10ml of diluted sulfuric acid TS, dilute to volume with recently boiled water, and mix. Use this final solution which contains 1 µg of arsenic (As) in each ml, within 3 days.

Dissolve 1g of recrystallized Silver Diethyldithiocarbamate, $(C_2H_5)2NCSSAg$, in 200ml of reagent-grade pyridine. Store this solution in a light-resistant container and use within 1 month.

Silver Diethyldithiocarbamate is available commercially or may be prepared as follows: Dissolve 1.7g of reagent grade silver nitrate in 100ml of water. In a separate container, dissolve 2.3 g of sodium diethyldithiocarbamate $(C_2H_5)2NCSSNa \bullet 3H_2O$, in 100ml of water and filter. Cool both solutions to about 15°, mix the two solutions, while stirring, collect the yellow precipitate in a medium-porosity sintered glass crucible or funnel, and wash with about 200ml of cold water.

Recrystallize the reagent, whether prepared as directed above or obtained commercially, as follows. Dissolve in freshly distilled pyridine, using about 100ml of solvent for each g of reagent, and filter. Add an equal volume of cold water to the pyridine solution, while stirring. Filter off the precipitate, using suction, wash with cold water, and dry in vacuum at room temperature for 2 to 3 h. The dry salt is pure yellow in color and should show no change in character after one month when stored in a light-resistant container. Discard any material that changes in color or develops a strong odor.

Dissolve 40 g of reagent-grade stannous chloride dihydrate, $SnCl_2 \bullet 2H_2O$, in 100ml of hydrochloric acid. Store the solution in glass containers and use within 3 months.

Soak cotton in a saturated solution of reagent-grade lead acetate, squeeze out the excess solution, and dry in a vacuum at room temperature.

The solution obtained by treating the sample as directed in an individual monograph is use directly as the Sample Solution in the Procedure. Sample solutions of organic compounds are prepared in the generator flask (a), unless otherwise directed. According to the following general procedure:

Caution: Some substances may react unexpectedly with explosive violence when digested with hydrogen peroxide.

Appropriate safety precautions must be employed at all times.

NOTE: If halogen-containing compounds are present, use a lower temperature while heating the sample with sulfuric acid, do not boil the mixture, and add the peroxide with caution, before charring begins, to prevent loss of trivalent arsenic.

Transfer 1.0g of the sample into the generator flask, add 5ml of sulfuric acid and a few glass beads, and digest at a temperature not exceeding 120° until charring begins, using preferably a hot plate in a fume hood. (Additional sulfuric acid may be necessary to completely wet some samples, but the total volume added should not exceed about 10ml.) After the sample has been initially decomposed by the acid, add with caution, dropwise, 30% hydrogen peroxide, allowing the reaction to subside and reheating between drops. The first few drops must be added very slowly with sufficient mixing to prevent a rapid reaction, and heating should be discontinued if foaming becomes excessive. Swirl the solution in the flask to prevent unreacted substance from caking on the walls, or bottom of the flask during digestion. Maintain oxidizing conditions at all times during the digestion by adding small quantities of the peroxide whenever the mixture turns brown or darkens. Continue the digestion until the organic matter is destroyed, gradually raising the temperature of the hot plate to 250°-300° until fumes of sulfur trioxide are copiously evolved and the solution becomes colorless or retains only a slight straw color. Cool, add cautiously 10ml of water, again evaporate to strong fuming, and cool. Add cautiously 10ml of water mix, wash the sides of the flask with a few ml of water, and dilute to 35ml.

Procedure

If the Sample Solution was not prepared in the generator flask, transfer to the flask a volume of the solution, prepared as directed, equivalent to 1.0g of the substance being tested, and water to make 35ml. Add 20ml of dilute sulfuric acid (1 in 5), 2 ml of potassium iodide TS, and 0.5 ml of Stannous Chloride Solution, and mix. Allow the mixture to stand for 30 min at room temperature. Pack the Impregnated Cotton, leaving a small air space between the two plugs, lubricate joints b and d with stopcock grease, if necessary, and connect the 3.0ml of Silver Diethyldithiocarbamate Solution to the absorber tube, add 3.0g of granular zinc (20-mesh) to the mixture in the flask, and immediately insert the standard-taper joint in the flask. Allow the evolution of hydrogen and color development to proceed at room temperature (25° ±3°) for 45 minutes, swirling the flask gently at 10min intervals. (The addition pf a small amount of isopropanol to the generator flask may improve the uniformity of the rate of gas evolution.) Disconnect the absorber tube from the generator and scrubber units and transfer the Silver Diethyldithiocarbamate Solution to a 1cm absorption cell. Determine the absorbance at the wavelength of maximum absorption between 535nm and 540nm, with a suitable spectromotometer or colorimeter, using Silver Diethyldithiocarbamate Solution as the blank. The absorbance due to any red color from the solution of the sample does not exceed that produced by 3.0ml of Standard Arsenic Solution (3 µg As) when treated in the same manner and under the same conditions as the sample. The room temperature during the generation of arsine from the standard should be held to with ±2° of that observed during the determination of the sample.

Interferences

Metals, or salts of metals such as chromium, cobalt, copper, mercury, molybdenum, nickel, palladium and silver are said to interfere with the evolution of arsine. Antimony, which forms stibine, is the only metal likely to produce a positive interference in the color development with the silver diethyldithiocarbamate. Stibine forms a red color that has a maximum absorbance at 510nm, but at 535 to 540 the absorbance of the antimony complex is so diminished that the results of the determination would not be altered significantly.

Heavy Metal Test

This test is designed to limit the content of common metallic impurities that are colored by sulfide ion (Ag, As, Bi, Cd, Cu, Hg, Pb, Sb, Sn) under the specified test conditions. It demonstrates that the test substance is not grossly contaminated by such heavy metals and, within the precision of the test, that it does not exceed the Heavy Metals limit given in the individual monograph, as determined by concomitant visual comparison with a control solution. It has been found that, in the specified pH range, the optimum concentration of lead ion (Pb) for matching purposes by this method is 20 µg in 50 ml of solution.

Special Reagents

Ammonia TS - dilute 400ml of ACS reagent-grade ammonium hydroxide to 1000ml with water.

Hydrochloric Acid - Sulfuric Acid, Nitric Acid. 30% Hydrogen, Peroxide Use ACS reagent-grade chemicals.

Lead Nitrate Stock Solution - Dissolve 159.8mg of ACS reagent-grade lead nitrate, Pb(NO3)2, in 100ml of water containing 1 ml of nitric acid, dilute with water to 1000.0 ml, and mix. Prepare and store this solution in glass containers that are free from lead salts.

Standard Lead Solution - On the day of use dilute 10.0ml of Lead Nitrate Stock solution with water to 100.0 ml. Each ml of Standard Lead Solution contains the equivalent of 10µg of lead ion (Pb).

Procedure

NOTE: In the following procedures, failure to adjust accurately the pH of the solution within the specified limits may result in a significant loss of test sensitivity.

Solution A - Pipet 2.0ml of Standard Lead Solution (20µg of Pb) into a 50ml color-comparison tube, and add water to make 25ml. Adjust the pH to between 3.0 and 4.0 (using short-range pH indicator paper) by addition of diluted acetic acid TS or ammonia TS, dilute with water to 40ml, and mix.

Solution B - Place 25ml of the solution prepared as directed in the individual monograph in a 50ml color-comparison tube that matches the one used for Solution A, adjust the pH to between 3.0 and 4.0 (using short-range pH indicator paper) by addition of diluted acetic acid TS or ammonia TS, dilute with water to 40ml, and mix.

Solution C - Into a third color comparison tube that matches those used for Solutions A and B, place 25ml of the solution prepared as directed in the individual monograph, and add 2.0ml of Standard Lead Solution. Adjust the pH to between 3.0 and 4.0 (using short-range pH indicator paper) by addition of diluted acetic acid TS or ammonia TS, dilute with water to 40ml, and mix.

To each tube add 10ml of freshly prepared hydrogen sulfide TS, mix, allow to stand for 5min, and view downward over a white surface. The color of Solution B is not darker than that of Solution A, and the intensity of the color of Solution C is equal to or greater than that of Solution A. If the color of Solution C is lighter than that of Solution A, the test substance is providing an interference with the test procedure and Method II must be used for the substance under examination.