CHAPTER 4

Precipitation, Complex Formation, and Oxidation–Reduction Methods

Joseph A. Zapotocky
COLLEGE OF PHARMACY
UNIVERSITY OF ARIZONA
TUCSON, ARIZONA

Although great advances in instrumentation have been made to date, many classical noninstrumental procedures continue to be relied upon for the assay of a substantial number of pharmaceuticals and reagent chemicals. This chapter is devoted to a review of the noninstrumental methods which utilize (1) precipitation or complex formation and (2) oxidation-reduction, as part of the titration procedure.

In the area of pharmaceutical analysis, there is no richer and more readily available source of assay procedures for study than those found in the various official compendia. Since the author is better acquainted with the *United States Pharmacopeia* (USP)[1] and the *National Formulary* (NF),[2] relatively complete lists of the drugs and chemicals whose assay procedures fall in the realm of the subheadings of this chapter are provided from these. In some instances, except for minor variations, corresponding procedures will be available in the *British Pharmacopoeia* (BP),[3] the *British Pharmaceutical Codex* (BPC),[4] and the *International Pharmacopoeia* (IP).[5–8] Assay procedures for the same product which differ noticeably in different compendia should be of special interest in the teaching laboratory and should provide material for group study projects and subsequent classroom discussions. The procedures of the various compendia are not reproduced; rather it is expected that the student will be encouraged to go to these or to other cited analytical references for his procedures and supplemental reading.

4.1 TITRATIONS INVOLVING PRECIPITATION AND COMPLEX FORMATION

Titrations which result in the formation of a very slightly soluble precipitate may be classified on the basis of the method used to identify the equivalence point.

1. *Titration to No Further Turbidity (Gay–Lussac Method).* The titration of chloride with silver nitrate solution to the point where no additional turbidity is produced is slow and there is the tendency for overtitration, but a fair amount of precision can be achieved with routine practice. The removal of a very small sample of the supernatant liquid for testing as the end point is approached is an aid to noting turbidity.

2. *Titration to the Appearance of Turbidity or Precipitation.* During the titration of cyanide with silver nitrate solution, a soluble complex ion $Ag(CN)_2^-$ is formed as long as $(CN)^-$ is in excess. Precipitation of $Ag[Ag(CN_2]$, silver argentocyanide, occurs when Ag^+ ions appear in excess.

3. *Titration to the Appearance of a Precipitate That Differs in Color from the Original Precipitate Formed during Titration (Mohr Method).* Of the two slightly soluble precipitates formed, the indicator precipitate must have the greater solubility. The greater the difference between the K_{SP} of the two precipitates the less the titration error.

4. *Titration to Change in Color of the Precipitate Produced by the Adsorption (or Desorption) of Colored Indicator Ions (Fajans Method).* The change in the colloidal-particle charge which occurs at the end point causes dye ions to be withdrawn from solution or returned to solution.

5. *Titration to the Appearance of a Soluble Colored Complex (Volhard Method).* Silver ions in excess are usually titrated with a thiocyanate solution. With the first appearance of SCN^- in excess, the Fe^{3+} ions of the indicator react to form $Fe(SCN)^{2-}$ (rust red).

6. *Modifications of the Above Procedures.* Of the above, methods (3) through (6) are most commonly used in the assay of pharmaceuticals. Method (2) is used in the assay of sodium cyanide and potassium cyanide, official reagents in most compendia. Turbidity as a result of silver argentocyanide is usually slow in forming, and high results are obtained unless a trace of potassium iodide is present to indicate excess Ag^+ by the formation of a pale yellow opalescence due to AgI. The titration is carried out in a weakly ammoniacal solution, which avoids the possibility of HCN formation.

Numerous improvements of method (1) have made it a highly accurate and precise method, used chiefly in government mint laboratories. In some instances results accurate to 0.01% are reported. Kolthoff and Stenger[9] present a comprehensive review of these.

A. MOHR METHOD

This procedure has been used since 1856 and makes use of potassium chromate as the indicator in the form of a 5 or 10% solution. Potassium chromate-dichromate combinations are sometimes used to approximate the pH conditions considered ideal for the titration (5% potassium chromate $\simeq$ pH 9; 1% potassium dichromate $\simeq$ pH 4).

In the direct titration of Cl^- with $AgNO_3$, AgCl is precipitated until the end point, when red silver chromate (Ag_2CrO_4) is formed permanently for the first time. Excessive indicator causes undertitration, and insufficient indicator causes overtitration. The use of an indicator blank and the standardization of the silver nitrate solution under the conditions of the assay help overcome some of the deficiencies of the procedure. The addition of an insoluble white substance such as chloride-free calcium carbonate makes the end point of the indicator blank simulate the conditions of the assay. The pH of the reaction mixture is somewhat critical. Increased acidity enhances solubility of the silver chromate and the conversion of indicator chromate ions to dichromate. At the other extreme, precipitation of silver hydroxide must be avoided. Various studies[10–12] recommend as optimum for the titration, a pH of not under 5.5, 5.5 to 7, and 7 to 10.5. Unless the titration error for the assay conditions is calculated, the safest policy is to run the titration close to neutrality. Alkaline solutions may be neutralized with dilute nitric acid (1 in 20); solutions which are too acidic may be neutralized with sodium bicarbonate, magnesium oxide, or calcium carbonate; or acetate buffers may be employed.

In addition to the above, the Mohr method has other disadvantages: (1) ions such as sulfides, phosphates, and arsenates will also be precipitated; (2) the end point with thiocyanates and iodides is less sensitive than with chlorides, owing to adsorption of colored chromate; (3) the end point is less sensitive when the titration involves dilute solutions; and (4) adsorbed ions of the sample become trapped and cause low results. Vigorous shaking near the end point is required to free trapped ions.

The direct titration of iodides with silver nitrate may be accomplished with the use of starch and a very small amount of oxidizing agent. The blue color is discharged at the end point, and the bright yellow AgI↓ is displayed.

Pharmaceutical Applications

Sodium Lauryl Sulfate (for Sodium Chloride Content); USP. The solution of the sample (usually slightly alkaline) is neutralized previous to titration.

Potassium Perchlorate, Potassium Perchlorate Tablets; BP. The sample is heated with volatile NH_4Cl (without melting) twice to reduce safely to chloride previous to solution and titration.

Titration with Silver Nitrate Solution using Starch Indicator: Iodoalphionic Acid, Iodopyracet Injection (for Iodine in a 3,5-Diiodo-4-pyridone-N-acetic acid), Sodium Iodomethamate (for Iodine), Sodium Iodomethamate Injection, NF. These organic iodine derivatives are digested in boiling alkaline permanganate solution which literally "chews up" the molecule and leaves the iodine as iodide following acidification and addition of sodium bisulfite. Permanganate and bisulfite are again added, and finally permanganate is added dropwise to faint yellow color (free iodine). After addition of starch the titration with silver nitrate is to the disappearance of blue color, leaving the yellow AgI visible. Better control of excess permanganate is possible if the starch is added before the final permanganate addition is made dropwise. The titer value in milligrams of organic compound per milliliters of designated silver nitrate normality will be $N \times$ mol. wt./n, where n is the number of iodine atoms present. For iodoalphionic acid, in terms of 0.05 N $AgNO_3$, this would be $0.05 \times 494.07/2$.

B. ADSORPTION METHODS

Adsorption, a problem in many precipitation procedures, was used to advantage by Fajans, in 1923. Very slightly soluble colloidal halides, such as those formed during titration of a soluble halide with silver nitrate, will have a negative charge resulting from an adsorbed layer of halide ions, the common ion in excess. A secondary layer of positive ions will surround the particle. When the halide has been completely precipitated, the colloidal particles will acquire a positive charge because of the presence of Ag^+, the common ion now in excess. A secondary layer of negative indicator ions will be adsorbed

from solution causing an immediate change in the color of the precipitate (Fig. 4.1).

Certain ionizable dyes, either weakly basic or acidic, such as the fluorescein dyes, are especially suitable as indicators. A number of excellent reviews are available.[13,14] Kodama[15] provides an up-to-date list of indicators with their applications. The quantity of indicator used must be kept at a minimum to avoid exceeding the solubility product of the silver dye compound. If indicator ions are adsorbed in preference to the common ion, premature end points

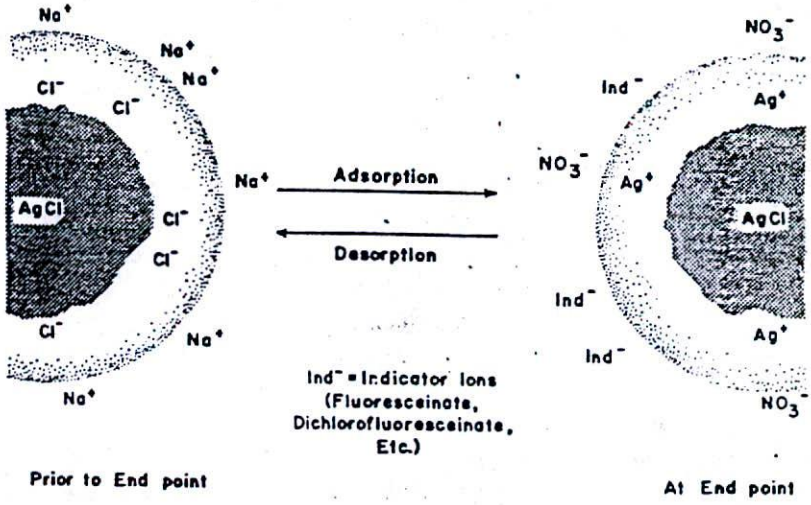


FIGURE 4.1: Indicator adsorption principle.

result. The indicator is limited to pH conditions which will keep it dissociated. Thus, fluorescein is limited to neutral or weakly basic solutions, whereas dichlorofluorescein can be used in weakly acidic media to about pH 4, and tetrabromofluorescein (eosin) to about pH 2 or 3. The latter is usually too strongly adsorbed to be used for chlorides, whereas tetraiodofluorescein is too strongly adsorbed for both bromides and chlorides. Solvent conditions will affect the choice of indicator.

Since the sensitivity of the adsorption method is dependent on the particles remaining in the colloidal state and not coagulating, agents such as gelatin, agar, and dextrin are used as protective colloids especially for silver chloride. One of the most recent agents recommended for this purpose is polyethylene glycol 400.[16]

Most commonly, titrations are performed by adding silver nitrate solution to the halide being estimated. By withholding the addition of the indicator until the end point is approached, the possibility of premature adsorption of indicator ions is decreased. Titration to desorption is a less common practice

and usually not as reliable. Adsorption of a cationic indicator occurs when the particles of the precipitate acquire a negative charge, owing to adsorption of anions present in excess.

Micromethods[17,18] for the determination of chloride or bromide in organic combinations are based on sample fusion with potassium or sodium and titration with silver nitrate, using an adsorption indicator.

Pharmaceutical Applications

Iopanoic Acid, Sodium Diatrizoate Injection, Meglumine Diatrizoate Injection, Meglumine Iodipamide Injection, Sodium Iodipamide Injection, USP; Iodopyracet Injection, Sodium Methiodal, Sodium Methiodal Injection, NF First Supplement. The iodine in the above organic iodine derivatives is converted to iodide by refluxing the sample with sodium hydroxide solution and zinc powder. The filtered acidified solution is titrated with silver nitrate using tetrabromophenolphthalein ethyl ester in glacial acetic acid (1 in 1000) as the indicator. The yellow AgI turns green at the end point.

Dichlorofluorescein (R) (sensitiveness), USP. The volume of 0.1 *N* $AgNO_3$ used to titrate 100 mg of KI, with this indicator present, is not more than 0.1 ml greater than the calculated volume.

Table 4.1 summarizes the conditions designated for some of the adsorption methods of the USP and NF.

TABLE 4.1: Adsorption Methods

Product	Compendium	Indicator	Solvent system*	Use
Betazole hydrochloride	USP	Eosin Y	Methanol-water	Cl content
Mecamylamine hydrochloride	USP	Eosin Y	Methanol-water	Cl content
Methoxamine hydrochloride	USP	Eosin Y	Methanol-water	Cl content
Phenylephrine hydrochloride	USP	Eosin Y	Methanol-water	Cl content
Pyridoxine hydrochloride	USP	Eosin Y	Methanol	Cl content
Succinylcholine chloride	USP	Eosin Y	Methanol-water	Cl content
Tubocurarine chloride	USP	Eosin Y	Methanol-water	Cl content
Hydroxyamphetamine bromide	USP	Eosin Y	Methanol-water	Br content
Propantheline bromide	USP	Eosin Y	Methanol-water	Br content
4-Aminoantipyrine hydrochloride (R)	USP	Dichloro-fluorescein	Neutralized aqueous	Assay
Meperidine hydrochloride	NF	Eosin	Methanol-water	Cl content
Azacyclanol hydrochloride	NF	Dichloro-fluorescein	Hydroalcoholic	Assay

* Methanol and methanol-water solvent systems are acidified with glacial acetic acid.

C. VOLHARD METHOD

This is the most common method used to determine halides or halogen content. It usually involves a residual titration of excess silver nitrate with a standard thiocyanate solution.

$$Ag^+ + X^- \rightarrow AgX\downarrow$$
$$Ag^+ + SCN^- \rightarrow AgSCN\downarrow$$

AgX must be removed before residual titration if it is more soluble than AgSCN. Filtration is sometimes used, or an immiscible liquid which coats and adheres to the insoluble particles may be added. Nitrobenzene, introduced by Caldwell and Moyer,[19] in 1935, is the most common agent of this type used today. AgCl is more soluble, whereas AgBr and AgI are less soluble than AgSCN.

Solutions of ferric ammonium sulfate or ferric nitrate serve as the indicator. The end point is the appearance of the red ferric thiocyanate ($Fe^{3+} + SCN^- \rightleftharpoons Fe(SCN)^{2+}$, which does not form permanently until the AgSCN precipitation is completed. Although the volume of indicator is not as critical as with potassium chromate or adsorption indicators, Swift et al.[20] report the optimum ferric and acid concentrations for the titration.

Ions which would normally interfere with the Mohr titration will not precipitate in the acid media (about 1 *N* HNO_3) used in the Volhard method. The nitric acid should be free of nitrite to avoid the nitrite-thiocyanate reaction which also causes a red color in a nitric acid medium.

Adequate shaking or boiling should precede filtration of AgX to ensure completeness of precipitation and elimination of adsorbed ions. As the end point is neared, adequate shaking should accompany each addition of thiocyanate. Adsorption and inadequate mixing result in premature end points. Titrations performed much above 25°C may result in over-titration. It is usually good practice to limit the excess silver nitrate to not more than a few ml. For the blank, an equal volume of silver nitrate is titrated under assay conditions but with the sample omitted.

When iodides are being titrated, the ferric indicator should not be added until the iodide has been precipitated to avoid the following oxidation-reduction reaction:

$$Fe^{3+} + I^- \rightarrow Fe^{2+} + I^0$$

Table 4.2 summarizes the pharmaceutical applications of the Volhard method and the handling of the precipitate following addition of excess silver nitrate.

In addition to the assay of the products listed in Table 4.2, the Volhard method is used to determine chloride. chloride content, or bromide content in the products listed at the top of p. 137.

TABLE 4.2: Volhard Methods

Product	Heat	Filtration	Nitrobenzene
USP			
Aminophylline (for theophylline);* injection;* suppositories;† tablets*	+	+	−
Ammonium chloride	−	−	+
Dextrose and sodium chloride injection	−	−	+
Dimenhydrinate (for 8-chlorotheophylline)*	+	+	−
Dimenhydrinate tablets*	+	+	−
Potassium chloride; injection; tablets	−	−	+
Ringer's injection (for sodium chloride)	−	−	+
Lactated Ringer's injection (for NaCl)	−	−	+
Silver nitrate; ophthalmic solution	−	−	−
Toughened silver nitrate	−	−	−
Sodium chloride; injection; solution; tablets	−	−	+
Ammonium sulfide solution (R)*	−	+	−
Calcium chloride (R)	−	+	−
Hydriodic acid (R)	+	−	−
Tetramethylammonium bromide (R)	−	−	−
Tetramethylammonium chloride (R)	−	−	+
Thiourea (R)*	+	+	−
Zinc chloride (R)	−	+	−
NF			
Ammonium bromide	−	−	−
Arecoline hydrobromide tablets	−	−	−
Three bromides elixir	−	−	−
Three bromides tablets (assay for bromide)	−	−	−
Fructose and sodium chloride injection	−	−	+
Diluted hydriodic acid	+	−	−
Hydriodic acid syrup	+	−	−
Methantheline bromide tablets	−	+	−
Potassium bromide	−	−	−
Ringer's solution (for NaCl)	−	−	+
Ammoniacal silver nitrate solution (for Ag)	−	−	−
Mild silver protein	−	−	−
Sodium bromide; elixir	−	−	−
Sodium chloride and dextrose tablets	−	−	+
Theophylline tablets*	+	+	−
Theophylline sodium acetate* tablets*	+	+	−
Theophylline sodium glycinate* tablets*	+	+	−
Gallamine triethiodide; injection	+	−	−

* Precipitation with silver nitrate in alkaline media (ammoniacal).
† Silver from precipitate and not from filtrate titrated.

Ammonium Chloride Injection, Bethanechol Chloride, Dibucaine Hydrochloride, Sodium Suramin, Trihexyphenidyl Hydrochloride, Acetylcholine Chloride (R), USP. Ammonium Bromide, Aluminum Phosphate Gel, Anileridine Hydrochloride, Methacholine Chloride, Potassium Bromide, Sodium Bromide, Tridihexethyl Chloride, NF and First Supplement. Ammonium Chloride, Dimenhydrinate, Potassium Bromide, and Potassium Chloride are some of the products found in the BP which make use of the Volhard method. Potassium Chlorate, BPC, is reduced to chloride by nitrite:

$$ClO_3^- + 3NO_2^- \rightarrow 3NO_3^- + Cl^-$$

Excess nitrite is removed by addition of potassium permanganate. Excess of the latter is removed by addition of ferrous sulfate which would also reduce any unreacted chlorate. Since nitrites would interfere with the titration, urea is added to decompose any residual traces to nitrogen and water.

Among the products listed in Table 4.2, theophylline and its various forms and thiourea require additional comment.

Theophylline will form a silver theophyllinate completely only in an ammonia solution after a period of heating. The product does not form in acid media (dilute nitric acid), because it would be solubilized. Precipitation in water alone is incomplete and of a different physical form, more colloidal than flocculent. The acidic hydrogen at C-7 is substituted.

The argentimetric method[21,22] used for the assay of thiourea is based on the following reactions:

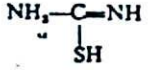
$$2AgNO_3 + NH_4OH \rightarrow Ag_2O + 2NH_4NO_3 + H_2O$$

$$Ag_2O + (NH_2)_2CS \rightarrow Ag_2S + (NH_2)_2CO$$

The latter may be represented by

$$Ag_2O + (NH_2)_2CS \rightarrow NH_4CN + Ag_2S + H_2O$$

Black silver sulfide is formed on addition of silver nitrate to a solution of thiourea in the presence of ammonia. (A white precipitate is formed by these in acid solution. The sulfhydryl form



would be the reactive form and precipitate as the silver salt.) Silver sulfide is removed by filtration following boiling, cooling, and acidifying with nitric acid solution. The excess silver nitrate is titrated with thiocyanate, using ferric ammonium sulfate indicator. Since 2 moles of $AgNO_3$ react with each thiourea, the milliequivalent weight of the latter is 76.12/2000.

Methimazole, Methimazole Tablets, Propylthiouracil, Propylthiouracil Tablets, USP; Ethchlorvynol, Ethchlorvynol Capsules, Ethinamate, Ethinamate Tablets, NF. An equivalent amount of acid (HNO_3) is formed when sulfur compounds capable of forming a sulfhydryl (mercapto) group react with silver nitrate. In each, the acid formed (HNO_3) may be titrated with standard alkali. In the case of thiouracil derivatives, the carbonyl group is also capable of enolizing and reacting with $AgNO_3$ to form HNO_3. The sample is usually dissolved in a known volume of standard alkali, and following the addition of excess silver nitrate solution, the alkali required to complete the titration is added. Acetylenic hydrogens from certain ethynyl substituted compounds react with silver nitrate to form a silver acetylide derivative and HNO_3.

Ammoniated Mercury, Sodium Mercaptomerin, Sterile Sodium Mercaptomerin, Mercuric Acetate (R), Mercuric Nitrate (R), Mercuric Sulfate (R), USP; Yellow Mercuric Oxide, Yellow Mercuric Oxide Ointment, Mercurophylline (for Mercuri Compound), Mercurophylline Injection, Mercurophylline Tablets, Nitromersol, Nitromersol Solution, Nitromersol Tincture, Phenylmercuric Nitrate, Thimerosal, NF; Mersalyl Acid, Phenylmercuric Nitrate, Thimerosal, BP. Inorganic mercuric compounds and certain organic mercurials of the type RHgX can be assayed in a dilute nitric acid solution by direct titration with a standard thiocyanate, using ferric ammonium sulfate or ferric nitrate as the indicator. The equation $Hg^{2+} + 2SCN^- \rightarrow Hg(SCN)_2\downarrow$ does not give a true picture of the titration. Although mercuric thiocyanate is only very slightly soluble in water, precipitation may not occur until near the equivalence point because of the formation of various complexes such as $Hg(SCN)^+$ and $Hg(SCN)_3^-$. The colored ferric indicator end point is usually not so distinct as in the silver-thiocyanate titration. The rust-red color is usually more distinct at lower temperatures and the reaction mixture may be cooled to about 15 to 20°.

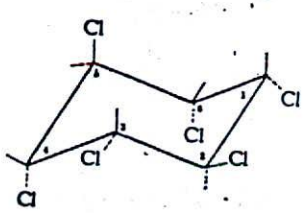
For certain mercurials special preliminary procedures are necessary before titration with thiocyanate.[23] The mercury in ammoniated mercury is separated as zinc amalgam from a hot acetic acid solution. The amalgam is dissolved in a nitric acid solution and oxidized with permanganate to ensure that all the mercury is in the Hg(II) state. The mercury in phenylmercuric nitrate and thimerosal also is separated as the zinc amalgam. Sodium mercaptomerin is digested in hot sulfuric-nitric acids and diluted previous to oxidation with permanganate.

Nitromersol is digested in hot sulfuric acid and then with 30% hydrogen peroxide until the mixture is decolorized. This is followed by a slight excess of permanganate. In all instances excess permanganate is removed with peroxide or oxalic acid previous to acidifying and titrating with thiocyanate.

Halogens in Organic Combination. Some organic halogen derivatives are assayed by the Volhard method following the conversion to inorganic halide. This changeover is made with varying degrees of difficulty.[24–26]

Procedures range from shaking an alcoholic solution of the sample with silver nitrate and allowing it to stand overnight, to treatment with aqueous or alcoholic NaOH or KOH, or refluxing with alcohol and sodium. With the exception of gamma benzene hexachloride, the milliequivalent weight of the sample will be the mol. wt./*n* (1000), where *n* equals the number of halogens present.

Gamma Benzene Hexachloride. Structure (I) is the 1e2e3e4a5a6a isomer of γ-benzene hexachloride. Most likely, by *trans*-elimination the 4a,6a-chlorines are removed followed by the 2e-chlorine, which becomes sterically unstable. Consequently, only three of the six chlorines are made available for reaction with the 0.1 *N* $AgNO_3$. The milliequivalent weight is 290.83/3000.



(I)

Table 4.3 summarizes the methods for converting halogen in organic combination to inorganic halide.

TABLE 4.3: Preparatory Methods for Organohalogen Compounds*

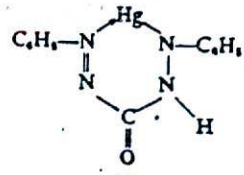
Compound	Compendium	Method
Chlorobutanol	USP	Alcoholic KOH (3 in 10); reflux
Chlorophenothane	USP	Benzene-isopropyl alcohol and sodium; reflux
Gamma-benzene hexachloride	USP	Alcoholic KOH; room temperature
Methyl iodide (R)	USP	Alcohol; add $AgNO_3$; shake 2 hr; let stand overnight
Chlorotrianisene	NF	Alcohol and sodium; reflux
Iodoform	NF	Alcohol; add silver nitrate; let stand overnight
Tribromoethanol	NF	Aqueous NaOH; reflux
Carbromal	BP	2 *N* NaOH; heat
Chlorambucil	BP	Alcoholic KOH; reflux
Dibromopropamidine isethionate	BP	Amyl alcohol and sodium
Dicophane (DDT)	BP	Alcoholic KOH; reflux
Gamma-benzene hexachloride	BP	Alcoholic KOH
Chlorotrianisene	BPC	Alcohol and sodium; reflux
Dyflos (isofluorophate)	BPC	Alcohol and sodium; reflux
Eosin	BPC	Heat; fuse with KNO_3, K_2CO_3, and Na_2CO_3

* Sodium sulfobromophthalein USP and BP: Bromine content or assay, respectively, by Volhard method following oxygen flask combustion method. (See general discussion of oxygen combustion methods, Section 4.26.)

D. MISCELLANEOUS PRECIPITATION METHODS

Mercuric Nitrate

Chlorides[27,28] may be determined by titration with a standard mercuric nitrate solution using diphenylcarbazide or diphenylcarbazone as the indicator. The solution being titrated must be maintained at about pH 1.5 to 2, and 3.0 to 3.5 for the respective indicators. At the end point a blue-violet diphenylcarbazone complex (II) is formed with excess Hg^{2+}.



(II)

The determination of bromides and iodides is usually less satisfactory. A mercurimetric method[29] is used in the assays of Methylthiouracil and Propylthiouracil, BP. Mercuric acetate (0.05 *M*) is the titrant, and diphenylcarbazone is the indicator with the end point the appearance of a rose-violet color. Two moles of the sulfhydryl form of the uracil compound react with 1 mole of the mercuric titrant.

Potassium Ferrocyanide

Diphenylthiocarbazone (dithizone) is used to determine zinc by titration with a standard potassium ferrocyanide solution[30]:

$$2K_4Fe(CN)_6 + 3Zn^{2+} \rightarrow K_2Zn_3[Fe(CN)_6]_2 + 6K^+$$

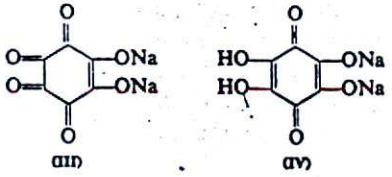
A pink zinc diphenylthiocarbazone complex is visible until all the zinc has been precipitated. Ferrocyanide may also be titrated with a zinc solution with the same indicator (7 mg dithizone in 100 ml CCl_4, use 1 drop). The color change is from green to pink. Zinc ferrocyanide precipitation titrations may also be carried out using an indicator such as diphenylamine or diphenylbenzidine, which undergo color change with a change in potential at the end point.[31]

Acetylenic Compounds

Compounds which contain an acetylenic hydrogen react with silver nitrate (ammoniacal) to precipitate a silver acetylide, AgC≡CR (explosive if dry).[32] The excess silver nitrate is determined by the usual Volhard method.

Barium-Sulfate Precipitation Titrations

Limit tests for sulfate are based on a comparison of turbidity obtained on adding barium chloride solution to a solution of the test sample and to a solution containing a known amount of sulfate. Various analytical methods[23–34] for measuring barium or sulfate are based on this reaction. Although the procedures are usually rapid, the error is seldom less than 1 to 3%. Sodium rhodizonate (III), which forms a barium salt in various shades of red, depending on the ions present, and disodium tetrahydroxyquinone (IV), which also forms a red precipitate with barium, are the best known indicators.



(III) (IV)

Residual Volhard Method with Adsorption Indicator

A procedure for the residual titration of excess $AgNO_3$ with standard thiocyanate solution using tartrazine indicator has been reported by Berry.[27] The end point is rather difficult to identify because the precipitate is colored yellow, and the titration is continued until the supernatant liquid turns yellow. This change can only be noted if the supernatant liquid is observed at eye level.

4.2 OXIDATION-REDUCTION METHODS IN PHARMACEUTICAL ANALYSIS

A. POTASSIUM BROMATE; POTASSIUM BROMATE–BROMIDE (0.1 *N* BROMINE)

W. F. Koppeschaar (1876) is usually given credit for being the first to use a combination of bromate and bromide in acid solution as a substitute for free bromine:

$$BrO_3^- + 5Br^- + 6H^+ \rightarrow 3Br_2 + 3H_2O$$

Bromination methods involve either substitution or addition. Whenever bromate-bromide solutions are used in substitution reactions, 1 mole of HBr is formed for each bromine substituted. Such reactions are usually not sufficiently rapid for direct titration, and residual titration procedures must be employed. Excess $KBrO_3$-KBr solution is added and, following a suitable

waiting period, the residual bromine is determined by adding excess iodide. The iodine liberated is titrated with a standard thiosulfate solution:

$$Br_2 + 2I^- \rightarrow 2Br^- + I_2$$

$$I_2 + 2S_2O_3^{2-} \rightarrow 2I^- + S_4O_6^{2-}$$

The waiting period may vary from a few minutes to an hour, depending upon the directional groups present. Ruderman[38] studied the effect of acid concentration, bromination time, and temperature on the extent of bromination for a representative group of phenols and phenol alcohols. Callan and Henderson[39] added bromide and hydrochloric acid to the solution of the sample, and $KBrO_3$ solution as the titrant to the end point of free bromine as detected by starch-iodide paper. Day and Taggart[40] compared this method with the residual titration method for a group of phenols and amines and report the residual method preferable. Kolthoff and Belcher[41] have reviewed the principles and the extensive applications of the bromate and bromate-bromide methods of analysis.

Bromine forms Br_3^- in aqueous bromide solutions. It is less convenient, less stable, and requires greater precautions to prevent loss of bromine during handling. Bromine in glacial acetic acid or bromine dissolved in an inert organic solvent such as carbon tetrachloride is used for some bromination methods.

Factors such as the reaction period and the amount of excess bromine must be regulated to avoid both low and high results. Too large an excess of bromine and too lengthy an exposure may cause substitution on a side-chain in addition to the desired substitution on an aromatic ring. The possibility of addition and substitution occurring together must be avoided.

Catalysts may be used to speed addition reactions if addition is slowed by the presence of electron-withdrawing groups (COOH, NO_2, etc.) conjugated with the unsaturated group. According to Lucas and Pressman,[42] Hg^{2+} when present as a catalyst will form $[Hg\text{-}Br\text{-}Br]^{+}$, which reacts with an unsaturated group

to form $BrCH_2\dot{C}H_2$ more rapidly than Br_2 alone.

All residual titrations, substitution or addition, are carried out in glass-stoppered flasks at room temperature (25°C) or lower, whenever necessary, to minimize the loss of bromine because of its volatility. Since light may affect the results, proper precautions should be taken in long bromination procedures.

When potassium bromate alone is used as a titrant, it acts as an oxidizing agent in acid media (about 1.5 to 2 *N* HCl) and is converted to a bromide by reducing agents:

$$BrO_3^- + 6H^+ + 6e \rightarrow Br^- + 3H_2O$$

When all the reducing agent has been oxidized, an excess of bromate will react with bromide and liberate bromine. So-called reversible and irreversible indicators are available.[41,43,44] Certain indicators such as Methyl Orange, Methyl Red, Naphthol Blue-Black, Brilliant Ponceau 5R, and others, are irreversibly oxidized and their color destroyed in the presence of excess oxidant. Local excesses of bromate resulting from too rapid titration and insufficient mixing may cause decolorization prematurely. The indicator is therefore added as the end point is approached, or an additional drop of indicator may be added at the end point if necessary. An indicator blank will determine the volume of titrant needed to decolorize the volume of indicator solution used in the titration. Several less well-known reversible indicators, *p*-Ethoxychrysoidine Hydrochloride and Quinoline Yellow, which permit back titration, have also been used.

Standardization. Potassium bromate is commonly available in at least 99.8% purity and, after drying at 120°, it can be weighed directly and diluted to volume to give a solution of known normality. It is usually advisable to standardize it against a standard arsenite solution, against arsenic trioxide, or by adding a known volume of bromate solution to an excess of iodide and titrating the liberated iodine from an acid solution with a standard thiosulfate solution:

$$BrO_3^- + 3H_3AsO_3 \rightarrow Br^- + 3H_3AsO_4$$
$$2BrO_3^- + 3As_2O_3 \rightarrow 2Br^- + 3As_2O_5$$
$$BrO_3^- + 6I^- + 6H^+ \rightarrow 3I_2 + Br^- + 3H_2O$$

Details of the latter procedure are to be found in the USP.[1]

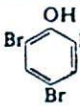
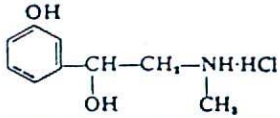
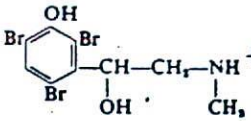
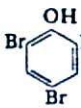
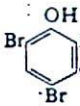
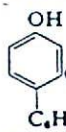
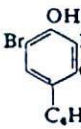
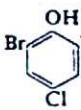
Bromine solution (0.1 *N*) (bromate-bromide solution) is not standardized ahead of time, but a blank determination is run using the same volume of solution and under the same conditions as indicated in the assay. The volume of thiosulfate used for the blank less that used for the back titration in the assay will give the volume of thiosulfate equivalent to the bromine solution required for the addition or substitution.

Pharmaceutical Applications

Phenol, Liquefied Phenol, Phenylephrine Hydrochloride, Phenylephrine Hydrochloride Injection, Phenylephrine Hydrochoride Solution, Resorcinol, Salicylic Acid Plaster, USP; Hexylresorcinol, Hexylresorcinol Pills, Parachlorophenol, NF. Table 4.4 summarizes the assay procedures for a group of phenols which substitute bromine on addition of $KBrO_3$-KBr (0.1 *N* bromine) in acid media.

Calcium Cyclobarbital, Calcium Cyclobarbital Tablets, Sodium Hexobarbital, Sterile Sodium Hexobarbital, NF. The above barbiturates, under the conditions of the assay, add bromine to the unsaturated cyclohexenyl group at C-5. To avoid possible substitution which might occur at vulnerable

Table 4.4: Bromine Substitution Methods

Compound	Bromine substitution products	Milli-equivalent weight	Reaction time, interval before KI addition, min
 Phenol	 + 3HBr*	$\frac{94.11}{6000}$	45
 Phenylephrine hydrochloride	 + 3HBr	$\frac{203.56}{6000}$	15
 Salicylic acid	 + 3HBr†	$\frac{138.12}{6000}$	45
 Resorcinol	 + 3HBr	$\frac{110.11}{6000}$	3
 Hexylresorcinol	 + 2HBr	$\frac{194.28}{4000}$	10
 *p*-Chlorophenol	 + 2HBr	$\frac{128.56}{4000}$	45 (4°)

* The tetrabromo derivative $C_6H_3Br_3OBr$ is an intermediate.
† Formed after decarboxylation of dibromosalicylic acid.

positions, such as for the acidic N-hydrogens, the reaction with bromine is carried out at low temperature, and large excesses of bromine are to be avoided. The milliequivalent weights are $(C_{12}H_{13}N_2O_3)_2$ Ca/4000 and $(C_{12}H_{13}N_2O_3)$ Na/2000 since four bromines and two bromines will add per mole of calcium cyclobarbital and sodium hexobarbital, respectively.

Oxophenarsine Hydrochloride, Oxophenarsine HCl for Injection (for total arsenic), USP. The organic arsenical is treated with 30% hydrogen

peroxide and concentrated sulfuric acid to decompose the organic matter. Any pentavalent arsenic formed during the reaction is reduced to the trivalent state by reaction with hydrazine sulfate. Excess hydrazine which would affect the titration is destroyed by boiling the acid mixture; nitrogen is given off. The mixture is cooled and titrated with standard $KBrO_3$ solution with Methyl Orange being used as the irreversible indicator. If the amount of indicator is kept at a minimum, the indicator blank is negligible and may be omitted. However, it is best to determine the amount of titrant required to decolorize the amount of indicator that has been added. Thorough mixing during titration and slow dropwise titration near the end point should prevent any premature decolorization of the indicator.

$$2BrO_3^- + 3N_2H_4 \xrightarrow{\text{acid}} 2Br^- + 3N_2 + 6H_2O$$

The oxidation of As(III) to As(V) may be indicated as follows:

$$3H_3AsO_3 + BrO_3^- \xrightarrow{\text{acid}} Br^- + 3H_3AsO_4$$

Each milliliter of 0.1 *N* $KBrO_3$ is equivalent to

$$0.1 \times KBrO_3/6000 \times 3As/KBrO_3 \times 1000\ mg = 3.746\ mg\ As$$

or

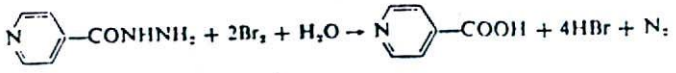
$$11.77\ mg\ oxophenarsine{\cdot}HCl$$

The BPC uses an iodimetric method for the assay; the digestion is with sulfuric and fuming nitric acids.

Carbarsone Suppositories, NF. The sample is digested in a Kjeldahl flask with sulfuric acid to which potassium sulfate has been added to raise the boiling point. The solution turns colorless when the organic matter has been destroyed. After dilution, the resultant As(III) is oxidized to As(V) by titration with bromate as indicated above. Even though carbarsone is a pentavalent arsenical, the sulfuric acid digestion reduces the arsenic to the trivalent state. Since no hydrogen peroxide is used which might oxidize the arsenic to the pentavalent state, the reduction with hydrazine as in the case of oxophenarsine is unnecessary.

In addition to some of the compounds already discussed, bromination methods are used in the assay of following substances:

Isoniazid, Methyl Hydroxybenzoate, Phenindione, BP; Calcium Hypophosphite, Potassium Hydroxyquinoline Sulfate, BPC. Bromimetric and iodimetric methods are used by the USP and BP, respectively, for the assay of isoniazid. The two methods are compared by Haugas and Mitchell.[43] The bromimetric reaction is based on the reaction



Methyl hydroxybenzoate (Methylparaben) is also assayed by different methods in the BP and USP. A bromimetric method is used in the former, whereas the latter uses a residual titration of excess 1 *N* NaOH to pH 6.5.

The bromine used in the assay of Phenindione BP, is in the form of a 10% v/v solution of bromine in alcohol.

Bromine substitution reactions are sometimes effectively coupled with a precipitation reaction. 8-Hydroxyquinoline (oxine) forms insoluble oxinates with certain metals, for example, $Mg(C_9H_6ON)_2$ and $Al(C_9H_6ON)_3$, which can be dried and weighed. 8-Hydroxyquinoline is also capable of substituting two bromines to form 5,7-dibromo-8-hydroxyquinoline and 2HBr:

If, for example, $Mg(C_9H_6ON)_2$ is precipitated and separated by filtration and then dissolved in 2 to 4 *N* HCl solution, the amount of bromine substituted on the liberated oxine may be determined by direct or residual titration. The appearance of excess bromine can be determined by various standard methods for the direct titration; the addition of KI and titration of liberated iodine is used for the residual titration.

The oxine liberated from a divalent metal oxinate substitutes 4 Br and 4 HBr are formed. If magnesium is being assayed, each mole of magnesium is equivalent to 8 Br. A trivalent metal such as aluminum accounts for 12 Br and the milliequivalent weights are Mg/8000 and Al/12,000, respectively.

$KBrO_3$ has been used to determine Fe(II) in the presence of $CuCl_2$ as a catalyst.[46]

Antimony potassium tartrate (tartar emetic), a water-soluble antimonial, can be determined in an acid medium by direct titration with $KBrO_3$ solution, using a reversible or an irreversible indicator.[47] Sb(III) is oxidized to Sb(V), but a lesser acid concentration is used for a reversible indicator, and tartaric acid or potassium sodium tartrate is added to prevent hydrolysis of the antimony. An iodimetric method is used for the assay by the USP and BP.

B. POTASSIUM IODATE

Potassium iodate is an oxidizing agent available in a high degree of purity, and standard solutions of known molarity may be prepared by diluting a known weight of the previously dried material to an exact volume. Solutions, if properly stored, are stable for long periods. They can be checked against As_2O_3, standard iron [Fe(II)] solution, or sodium thiosulfate. The significant applications of this oxidizing agent in analysis date back to the work of Andrews,[48] in 1903. The most widely used reaction is that which occurs in concentrations of hydrochloric acid ranging from 3 to 4 *N* to 9 *N*.

$$IO_3^- + 6H^+ + 4e \rightarrow I^- + 3H_2O$$

The iodine, at the end point, is converted to ICl, which is considered to exist as a stable complex ICl_2^- in the presence of a sufficient concentration of hydrochloric acid. The intermediate reaction, which is most noticeable during the titration, involves the liberation of free iodine:

$$IO_3^- + 5I^- + 6H^+ \rightarrow 3I_2 + 3H_2O$$
$$IO_3^- + 2I_2 + 6H^+ \rightarrow 5I^+ + 3H_2O$$

Morgan et al.[49] should be referred to for details of the mechanism of the iodate-iodide reactions. Briefly, IO_2^+, a dissociation product of HIO_3, reacts with I^- to form a complex, $IO_2^+—I^-$, which either decomposes to IO^+ and IO^- or reacts with additional iodide to form $IO_2^+—2I^-$. The latter decomposes to I^+ and $2IO^-$, and I^+ and I^- form I_2.

The reaction between iodine and iodate is dependent on the concentration of hydrochloric acid.[50] Usually, the acid concentration will be varied according to the reducing agent being titrated; for example, potassium or sodium iodide is titrated in a higher concentration of acid than mercuric iodide. The disappearance of free iodine cannot be based on the color of the starch-iodine complex, since the latter is usually not formed in the high acid concentrations used in iodate titrations. A small volume of an immiscible solvent, usually carbon tetrachloride or chloroform, is added to the reaction mixture as an aid in identifying the end point.[51] The liberated iodine is partitioned between the aqueous and immiscible solvent layers, and thorough mixing it required in order to prevent overtitration. In the immediate vicinity of the end point, the addition of each drop of titrant must be followed by thorough shaking. As a result of the greater solubility of iodine in the organic solvent, the aqueous layer is always first to become free of iodine, and at the end point, the immiscible solvent layer loses its purple color. Because of the great difference in the solubility of iodine in the two solvents, the volume of the immiscible solvent should be quite small in relation to the aqueous layer. It may be advantageous to withhold the addition of the $CHCl_3$ or CCl_4 until the end point is neared.

Dyes which are destroyed or change color in the presence of an oxidizing titrant may be used as indicators. Amaranth, for example, is decolorized, as soon as the KIO_3 is in excess, but it must be added only in the immediate vicinity of the endpoint, otherwise the temporary excesses of titrant which usually occur in the area of addition will gradually decolorize the indicator before the end point is reached. The volume of indicator solution should be quite small to avoid any significant indicator blank.[52]

The I^- formed during reduction of iodate need not always be as ICl. In the presence of acetone, iodoacetone is formed and with cyanide, iodocyanide:

$$IO_3^- + 5H^+ + CH_3\overset{O}{\overset{\|}{C}}CH_3 + 4e^- \rightarrow ICH_2\overset{O}{\overset{\|}{C}}CH_3 + 3H_2O$$
$$IO_3^- + 6H^+ + CN^- + 4e^- \rightarrow ICN + 3H_2O$$

With the latter, due consideration must be given to the toxicity of cyanides, but these I^+ forms do not require the high acid concentration of ICl. Iodide can be titrated with iodate at a much lower acid concentration in the presence of acetone[53]:

$$IO_3^- + 2I^- + 3CH_3\overset{O}{\overset{\|}{C}}CH_3 + 3H^+ \rightarrow 3ICH_2\overset{O}{\overset{\|}{C}}CH_3 + 3H_2O$$

An oxidation-reduction reaction involving KIO_3 as the standard, and KI and $Na_2S_2O_3$ in excess, can be titrated with an acid solution. The iodine released is immediately decolorized by the thiosulfate. At the end point, an excess of acid will produce a color change with an indicator such as Methyl Red.

A number of significant studies[53–56] compare iodate methods with other oxidizing titrants; potassium permanganate, ceric sulfate, and potassium bromate for the analysis of iodide, thiocyanate, and hydrazine.

Pharmaceutical Applications

Benzalkonium Chloride, Benzalkonium Chloride Solution, USP; Methylbenzethonium Chloride, NF First Supplement.

$$\left[C_6H_4CH_2\overset{CH_3}{\overset{|}{\overset{+}{N}}}(CH_3)R\right]Cl^- + KI \rightarrow \left[C_6H_4CH_2\overset{CH_3}{\overset{|}{\overset{+}{N}}}(CH_3)R\right]I^- + KCl$$

The iodide formed in the above reaction is chloroform-soluble with excess KI remaining in the aqueous layer of a $CHCl_3$-H_2O system. The water layer is separated and titrated with 0.05 *M* KIO_3 following strong acidification with hydrochloric acid. In the blank, water replaces the sample, and the same volume of KI is carried through the identical procedure. The difference in the volumes of 0.05 *M* KIO_3 used for the two titrations is equivalent to the benzalkonium chloride present.

$$IO_3^- + 2I^- + 6H^+ + 3Cl^- \rightarrow 3ICl + 3H_2O$$

Each milliliter of 0.05 *M* KIO_3 is equivalent to

$$0.05 \times \frac{2 \times 360 \text{ (avg. mol. wt.)}}{1000} \times 1000 \text{ mg of benzalkonium chloride}$$

Echothiophate Iodide, Echothiophate Iodide for Ophthalmic Solution, USP.

$$\left[(CH_3)_3\overset{+}{N}CH_2CH_2S\overset{O}{\overset{\uparrow}{P}}(OC_2H_5)_2\right]I^-$$

The alkaline hydrolysis of the thiophosphate ester results in the formation of a mercaptan, $[(CH_3)_3\overset{+}{N}CH_2CH_2SNa]OH^-$. The reaction mixture is

slightly acidified and titrated with KIO_3 solution. The free iodine formed from the IO_3^-/I^- reaction oxidizes the mercaptan to a disulfide (RS—SR), and the appearance of excess iodine is indicated by a pale yellow color:

$$2RSH + I_2 \rightarrow RSSR + 2HI$$

The determination is repeated but without subjecting the sample to alkaline hydrolysis. The volume of KIO_3 solution used is subtracted from that recorded in the first titration to correct for the presence of any free thiol present as a contaminant.

The milligrams of echothiophate iodide equivalent to 1 ml of 0.01 *M* potassium iodate may be calculated as follows:

$$0.01 \times \frac{KIO_3}{1000} \times \frac{3I_2}{KIO_3} \times \frac{2RSH}{I_2} \times \frac{\text{echothiophate iodide}}{RSH} \times 1000$$

$$= 0.01 \times 6(383.23)\text{ mg}$$

Iodine Tincture (for NaI), Strong Iodine Solution (for KI), Potassium Iodide, Sodium Iodide, Red Mercuric Iodide (R), USP; Iodine Ampuls, Iodine Solution, Potassium Iodide Solution, NF. Samples containing iodide alone are titrated from a strongly acidified solution with a standard KIO_3 solution using an immiscible solvent (5 ml $CHCl_3$) to aid in the detection of the end point. The end point for samples containing both free iodine and iodide, when titrated with iodate solution, will come when both the free iodine and iodide have been converted to ICl. The free iodine is determined by titration with potassium arsenite solution. The milliliters of 0.05 *M* KIO_3 less the milliliters of 0.1 *M* potassium arsenite ($KAsO_2$) equals the milliliters of iodate used by the iodide alone:

$$AsO_2^- + I_2 + H_2O \rightarrow AsO_3^- + 2H^+ + 2I^-$$

or

$$H_2AsO_3^- + I_2 + H_2O \rightarrow H_2AsO_4^- + 2H^+ + 2I^-$$

The equations for the titration of iodine and iodide with iodate have been previously presented.

Hydralazine Hydrochloride, Hydralazine Hydrochloride Injection, Hydralazine Hydrochloride Tablets, NF. Hydrazines may be titrated directly with a standard iodate solution in about a 4 to 6 *N* concentration of HCl. Free iodine is liberated first and at the end point, ICl[34,37]:

$$N_2H_4 + IO_3^- + 2H^+ + Cl^- \rightarrow ICl + N_2 + 3H_2O$$

The milligrams of hydralazine hydrochloride equivalent to 1 ml of 0.02 *M* KIO_3 is calculated as follows:

$$0.02 \times \frac{196.64}{1000} \times 1000$$

Indicators such as amaranth solution (0.2%) have been used, and are preferably added near the end point.

Although hydrazine can be titrated directly with iodate, isoniazid cannot, owing to the slow release of iodine and the absence of a detectable end point. If an irreversible indicator (*p*-ethoxychrysoidine) is used, the titration with iodate to the discharge of color gives good results.[58]

Stannous Fluoride (for Stannous Ion), NF. Stannous salts are oxidized to Sn(IV) by iodine. The assay of stannous fluoride may be accomplished either by titration with a standard iodine solution or by the use of a standard iodate solution in the presence of iodide in an acid medium, to liberate iodine for the oxidation:

$$I_2 + Sn^{2+} \rightarrow Sn^{4+} + 2I^-$$

As soon as all the tin is in the Sn(IV) form, an excess of iodine will react with the starch.

Upon omitting the iodide from the reaction, the oxidation of Sn(II) may be represented as:

$$IO_3^- + 2Sn^{2+} + Cl^- + 6H^+ \rightarrow ICl + 2Sn^{4+} + 3H_2O$$

Stannous fluoride in solution undergoes various types of decomposition: namely, oxidation, hydrolysis, and reaction with glass in acid media. The titration is performed in an inert atmosphere to protect against oxidation; any stannous hydroxide formed during hydrolysis would be solubilized in the acid medium of the titration. Solutions, freshly prepared and titrated immediately, will show a minimum of etching and hydrolysis.

The milligrams of Sn per milliliter of 0.1 *N* KIO_3 may be calculated as follows:

$$0.1^* \times \frac{KIO_3}{4000} \times \frac{2Sn}{KIO_3} \times 1000$$

Hefferren[59] presents a study of the analysis of stannous fluoride and the standardization procedure for potassium iodate solution against pure tin or sodium thiosulfate.

The determination of fluorides with $AlCl_3$ depends on the formation of a stable complex, AlF_6^{3-}, in a nearly neutral solution using Eriochromcyanine R as an internal indicator.[60,61]

Other examples of pharmaceutical products assayed by iodate methods are the following.

Ascorbic Acid, Ferrous Sulfate Tablets, Hydralazine Hydrochloride, Aqueous Iodine Solution (for KI), Weak Iodine Solution, Iodized Oil Viscous Injection, Potassium Iodide, Sodium Acetrizoate Injection, Sodium Iodide, BP; Caffeine and Sodium Iodide (Content of NaI), BPC. In this list, an immiscible solvent method (chloroform) is used to determine the end point for ascorbic acid and ferrous sulfate tablets; the disappearance of the blue color with starch is the end point with iodized oil viscous injection and sodium acetrizoate injection; the remainder use an irreversible indicator, amaranth. The iodine in organic combination is reduced to inorganic iodide by refluxing for 1 hour with

powdered zinc and glacial acetic acid. The titration is carried out in the presence of cyanide but with a lesser acid concentration.

Ascorbic acid may be titrated in a low acid concentration to the appearance of iodine in the immiscible solvent layer or in a high concentration to the loss of color, the usual ICl end point.

C. IODIMETRIC, IODOMETRIC DETERMINATIONS

Included here are direct or residual titrations involving oxidation with iodine as the triiodide and thiosulfate titrations of iodine resulting from the oxidation of iodide. Only substances with oxidation potentials less than that of $I_2 + 2e \rightleftharpoons 2I^-$ can undergo oxidation with iodine.

Iodine

Iodine solutions of desired normality may be prepared by dissolving and then diluting to volume a known weight of iodine which has been resublimed from lime and potassium iodide and dried in a desiccator to eliminate other halogens and water. The purified iodine is weighed into a glass-stoppered weighing bottle previously tared with the required KI and a convenient quantity of water, the entire contents of the bottle transferred to a volumetric flask and diluted to volume. The triiodide formed is water-soluble, acts as an oxidizer in acid and alkaline solutions, and has the advantage of greater stability because of lesser volatility. The solution should be stored in a light-protected container not above room temperature.

In most instances it is more expedient to use resublimed iodine that is commercially available, weigh approximately the amount needed, solubilize with KI in water, and then standardize. Iodine solutions are stabilized by the addition of several drops of hydrochloric acid.

Standardization. Various methods of standardization are available to the analyst. One of the most common, uses primary standard arsenic trioxide previously dried at 105°. The standard is solubilized by sodium hydroxide and the excess alkali is neutralized with diluted hydrochloric acid as indicated by Methyl Orange. After addition of $NaHCO_3$ and dilution with water, the solution is titrated with the iodine solution, using starch as the indicator. For details of the method refer to the USP.[1]

$$H_3AsO_3 + I_3^- + H_2O \rightarrow H_3AsO_4 + 3I^- + 2H^+$$

The reaction goes to completion if $NaHCO_3$ is present to react with the HI formed during the titration. Strong alkali such as NaOH or Na_2CO_3 cannot be used for this purpose, because they react with iodine to form various oxyhalogen compounds. Borate or phosphate buffers are sometimes used. McAlpine[42] has studied the effect of pH on the iodimetric titration of alkali solubilized arsenic trioxide.

normality of iodine = (weight in grams of As_2O_3)/(milliliters of I × As_2O_3/4000)

Iodine solutions can also be standardized by titration with a standard sodium thiosulfate solution, adding starch as the indicator near the end point:

$$I_2 + 2S_2O_3^{2-} \rightarrow S_4O_6^{2-} + 2I^-$$

Stouts et al.[63] give the details for a method of standardization using pure silver as the primary standard.

Standardization against arsenite may be represented:

$$I_2 + HAsO_2 + 4OH^- \rightarrow HAsO_4^{2-} + 2I^- + 2H_2O$$

Sodium Thiosulfate

Standard solutions of sodium thiosulfate are prepared from the pentahydrate or the equivalent amount of the anhydrous product. To obtain the greatest stability against sulfur bacteria which may attack the solution, the water used for the preparation should have been recently boiled for at least several minutes and then cooled. An 0.02% concentration of sodium carbonate or just sufficient to produce a slight alkaline reaction, but below pH 10, tends to check bacterial activity. An acid solution is avoided to prevent the formation of free sulfur and bisulfites which would react with iodine in a different ratio than thiosulfate. Chloroform has been recommended as a preservative.[64] Solutions should be stored in light-resistant containers. The titer of thiosulfate solution may increase or decrease, depending on the type of decomposition that occurs.

Quantitative reduction with thiosulfate involves the reaction

$$2S_2O_3^{2-} \rightarrow S_4O_6^{2-} + 2e$$

Oxidation of thiosulfate to sulfate with strong oxidizers is usually not encountered.

Standardization. The various methods of standardization usually depend upon the release of iodine from KI by an oxidizing agent. The potassium dichromate method is used in the USP[1] to standardize a 0.1 *N* solution:

$$K_2Cr_2O_7 + 6KI + 14HCl \rightarrow 3I_2 + 8KCl + 2CrCl_3 + 7H_2O$$

Primary standard, potassium dichromate is previously dried at 120° for 4 hr. The HI formed by the excess KI and HCl may cause erroneous results, owing to its ease of air oxidation unless sodium bicarbonate is present. Starch should not be added until the end point is neared.

Thiosulfate solution may be standardized against a standard iodine solution previously standardized against arsenic trioxide or a standard arsenite solution.

The iodine released from excess KI by oxidation with standard potassium permanganate solution is the basis for another method of standardization:

$$2KMnO_4 + 10KI + 16HCl \rightarrow 5I_2 + 12KCl + 2MnCl_2 + 8H_2O$$

Pure copper in the form of turnings is used if the thiosulfate will be used for the assay of copper salts. Cupric nitrate, which will release iodine from KI,[65–69] is formed.

$$2Cu(NO_3)_2 + 4KI \rightarrow I_2 + 4KNO_3 + 2CuI$$

A neutral medium is best for the titration. The chief difficulties of the method are: (1) Any nitrites formed during nitration must be removed. Urea is added for this purpose:

$$2HNO_2 + CO(NH_2)_2 \rightarrow 2N_2 + CO_2 + 3H_2O$$

(2) The precipitated cuprous iodide tends to adsorb iodine. If some ammonium or potassium thiocyanate is added near the end point, the adsorbed iodine will be released as a result of the formation of CuSCN. The thiocyanate should not be added prematurely. A sufficient excess of KI will decrease adsorption by increasing the solubility of the CuI.

A reverse procedure is sometimes used for standardization of thiosulfate solutions. If excess KI is added to an accurately measured volume of thiosulfate solution, the solution can be titrated with standard solutions of oxidants. The end point is reached when excess oxidant liberates free iodine as noted with starch indicator.

In most instances the color of iodine alone cannot be relied upon for the detection of the end point.

Starch

The source of the starch is not specified in most discussions. Arrowroot starch is specified in the USP for the preparation of its indicator solution, Starch Test Solution. Potato starch is used in some preparations, but corn starch is not used because of its high α-amylose content. Soluble starch, which has the advantage that it disperses in hot water and produces almost clear indicator solutions, is not as sensitive as regular starch, which must be boiled in water for a longer period but just to the production of translucent liquid. Overboiling decreases the sensitivity. Starch indicator should be capable of detecting approximately a 10^{-5} N concentration of iodine as the triiodide. The hydrolysis products, β-amylose and amylopectin, form a blue-purple and a red-purple color with starch, respectively. The disadvantages of starch indicator are well-known. Its ease of decomposition makes the preparation of fresh samples desirable. The addition of preservatives such as chloroform, thymol, or mercuric bromide are less desirable, especially when considering the small cost involved in preparing fresh solutions. Solutions of starch in glycerine (glycerite) and formamide have been used for greater stability. Polyvinyl alcohol has been recommended as a substitute for starch.[70–74]

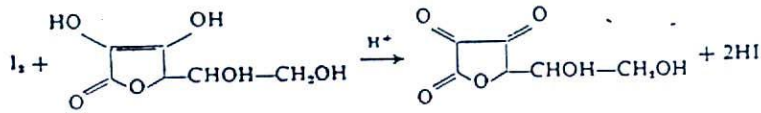
When titrating iodine, starch should be added only when the end point is approached. This decreases the errors resulting from the formation of slightly soluble complexed or adsorbed iodine. Starch may be added at the beginning of a titration when the end point is to be noted by the appearance of free iodine. Starch cannot be used in highly acid media. The sensitivity of starch to iodine decreases with an increase in temperature. This is of very

little concern since the increased volatility and loss of iodine precludes the use of warm solutions.

Small volumes of immiscible solvents such as chloroform or carbon tetrachloride are frequently added to aid in the detection of iodine end points.

Pharmaceutical Applications

Ascorbic Acid, Sodium Ascorbate, USP. Samples are dissolved in recently boiled water and titrated in acid media with 0.1 *N* iodine, using starch as the indicator. Since 2 moles of I react with each ascorbic acid, its milliequivalent weight is 176.13/2000:



Sodium Thiosulfate, Sodium Thiosulfate Injection, USP. Thiosulfate is titrated from a neutral solution with 0.1 *N* iodine. The injection, whose pH is between 8 and 9.5, must be previously neutralized, if necessary:

$$I_2 + 2Na_2S_2O_3 \rightarrow Na_2S_4O_6 + 2NaI$$

Carbarsone, Carbarsone Tablets, Tryparsamide, Sterile Tryparsamide, USP; Carbarsone Capsules, NF. The arsenic in these pentavalent organic arsenicals is converted to trivalent inorganic As before titration with iodine. Carbarsone is digested in a Kjeldahl flask with concentrated sulfuric acid and potassium sulfate to remove organic matter and reduce the arsenic to inorganic As(III). The excess acid is neutralized and then made slightly acid. $NaHCO_3$ is added in slight excess, and the solution is titrated with 0.1 *N* iodine.

Tryparsamide is digested in a nitric-sulfuric acid mixture which converts the As to inorganic As(V), owing to the oxidizing acid. Potassium iodide reduces this to As(III), liberating iodine which is titrated with 0.1 *N* thiosulfate. Excess acid is neutralized with alkali, made slightly acid, and the As(III) is titrated with 0.1 *N* iodine:

$$H_3AsO_3 + I_3^- + H_2O = H_3AsO_4 + 2H^+ + 3I^-$$

This reversible reaction goes to the right if the HI is removed by the $NaHCO_3$ added to the reaction mixture just before titration with iodine. Since each As reacts with 2I, the milliequivalent weight of each arsenical will be the mol. wt./2000.

Arsanilic Acid (R), USP. The sample is digested in sulfuric and nitric acids and then 30% H_2O_2 is added to ensure complete oxidation of the arsenic to inorganic As(V). Excess peroxide is removed by heat, and the iodine liberated following the addition of KI is titrated in a strongly acid medium (at least 4 *N*) with thiosulfate.

Drocarbil (for Acetarsone), NF. The sulfuric acid-permanganate digestion is followed by additional oxidation with 30% H_2O_2. Following the addition of KI, the reaction mixture must be allowed to stand in a cool, dark place for 1 hr before the slowly liberated iodine is titrated with thiosulfate solution.

Antimony Potassium Tartrate, USP. Trivalent antimony is oxidized to the pentavalent state by iodine. The reaction involving antimony potassium tartrate may be represented as either of the following:

$$\begin{array}{l} KOOC{-}CHOH{-}CHOH{-}COO(SbO) + I_2 + H_2O \rightarrow \\ \qquad\qquad\qquad\qquad\qquad\qquad\quad OH \end{array}$$

$$\begin{array}{l} KOOC{-}CHOH{-}CHOH{-}COO(SbO) + 2HI \quad \text{(removed with } NaHCO_3\text{)} \\ \qquad\qquad\qquad\qquad\qquad\quad OH \end{array}$$

$$\begin{array}{l} KOOC{-}CHOH{-}CHOH{-}COO(SbO) + I_2 + 6NaHCO_3 \rightarrow Na_3SbO_4 + \\ \quad KNaC_4H_4O_6 + 2NaI + 6CO_2 + 3H_2O \end{array}$$

Antimony precipitates as a hydroxide quite easily; excess tartrate, $KNaC_4H_4O_6$, aids in preventing this. The reaction mixture must be close to neutral; if alkaline, hypoiodite formation is possible as is precipitation of antimony hydroxide. In acid, the reaction undergoes reversal.

Stibophen, Stibophen Injection, USP. Oxidation of Sb(III) to Sb(V) occurs with the 0.1 *N* iodine titration being carried out in a weakly acid solution at 50° which tends to decrease the sensitivity of the starch to excess iodine at the end point. The formaldehyde that is added to the sample of the injection evidently is needed to form addition products with any sulfonate hydrolysis products which may form in the aqueous solution during storage. The analysis of organic Sb is reported by Wilkinson.[75]

Dimercaprol, Dimercaprol Injection, Potassium Xanthogenate (R), Sodium Thioglycollate (R), USP; Methionine, Methionine Capsules, Methionine Tablets, NF. Thiols (mercaptans) are oxidized quantitatively to disulfides by iodine:[76]

$$2RSH + I_2 \rightarrow RSSR + 2HI$$

Hydrogen sulfide, if present, will react with iodine and give high results:

$$H_2S + I_2 \rightarrow S + 2HI$$

Dimercaprol may be contaminated with H_2S and a trimercapto derivative. The former is removed by bubbling an inert oxygen-free gas through the sample. The latter is separated by column chromatography and titrated with 0.1 *N* iodine. When the H_2S-free sample is titrated with iodine both the dithiol and any tri-thiol present are determined. The calculation of the per cent dimercaprol is

$$\left[\frac{\text{ml.} \times 0.1 \times \frac{1}{1000} \times \frac{2(124.22)}{41}}{\text{wt. of sample in g}} \times 100\right] - \left[\frac{C_3H_4OS_2/2}{C_3H_5S_3/3} \times \%\ \text{1,2,3-trimercaptopropane}\right]$$

Since dimercaprol will decompose in the presence of water, only organic solvents are used until the final titration with the iodine solution.

Potassium xanthogenate is oxidized to dixanthogenate by iodine. The excess iodine is titrated after 5 min from a slightly acid medium:

$$2C_2H_5OCSSK + I_2 \rightarrow 2KI + \begin{matrix} C_2H_5OCSS \\ | \\ C_2H_5OCSS \end{matrix}$$

Sodium thioglycollate is oxidized to a disulfide by direct titration with iodine. The water used in the assay as well as the solution to be titrated must be free of oxygen to avoid low results.

Methionine ($CH_3—S—CH_2—CH_2—\underset{\displaystyle NH_2}{\underset{|}{CH}}—COOH$) is a thioether and will be represented as R_2S. According to Lavine,[77] methionine was considered to form a periodide in a slight excess of iodine and 1 *M* KI at pH 7:

$$R_2S + I_2 \rightarrow R_2S{\cdot}I_2$$

Under these conditions the hydrolysis of the periodide to the sulfoxide was retarded. Dibasic potassium phosphate and monobasic potassium phosphate were added in the proper ratio for pH 7. Later Lavine[78] showed that the products formed were dehydromethionine ($CH_3—\overset{+}{S}—C—C—C(COO^-)NH$) and 2HI. Since 2I react with each methionine, the milliequivalent weight for calculations is 149.21/2000.

Sorbitol, Sorbitol Solution, USP; Mannitol, Mannitol Injection, NF. Oxidation of polyalcohols by periodic acid (Malaprade reaction) has been studied by many investigators.[79–83] The hydroxyl groups in polyhydroxy compounds undergo oxidation by periodates in acid solutions. Each CHOH group will form HCOOH and each CH_2OH group will become HCHO.

$$HOCH_2—\underset{\displaystyle OH}{\underset{|}{CH}}—\underset{\displaystyle OH}{\underset{|}{CH}}—\underset{\displaystyle OH}{\underset{|}{CH}}—\underset{\displaystyle OH}{\underset{|}{CH}}—CH_2OH + 5IO_4^- \rightarrow$$

$$2HCHO + 4HCOOH + 5IO_3^- + H_2O$$

$$IO_4^- + 7I^- + 8H^+ \rightarrow 4I_2 + 4H_2O$$

$$IO_3^- + 5I^- + 6H^+ \rightarrow 3I_2 + 3H_2O$$

The difference in the volume of thiosulfate solution required to titrate the iodine formed in the latter two reactions is equivalent to the iodine released by the periodate used for the oxidation of the mannitol. Sorbitol and sorbitol solution are assayed for their content of D-sorbitol which must be separated from other polyhydric alcohols by column chromatography previous to oxidation with potassium periodate. Since mannitol and sorbitol are

isomers, the calculation of the titer is identical for both. The following may assist in showing the relationship of the reagents involved.

$$0.02 \times \frac{Na_2S_2O_3}{1000} \times \frac{I_2}{2Na_2S_2O_3} \times \frac{KIO_3}{I_2} \times \frac{C_6H_{11}O_6}{5KIO_3} \times 1000 = .02 \times \frac{C_6H_{11}O_6}{10}$$

= mg mannitol or sorbitol equivalent to 1 ml 0.02 N $Na_2S_2O_3$

Cupric Sulfate, NF. Cupric sulfate is reduced by iodide in a weakly acidic solution to cuprous iodide and the iodine released is titrated:

$$2Cu^{2+} + 4I^- \rightarrow 2CuI_\downarrow + I_2$$

Acetic acid is added to obtain a weakly acidic solution, between pH 4 and 5.5. Although the assay procedure is quite simple, the adsorption of iodine by the precipitated CuI may give low results. The addition of potassium or ammonium thiocyanate very near the end point forms CuSCN and tends to free adsorbed iodine.[66–69]

Ferric Citrate (R), Iodine Pentoxide (R), Potassium Bromate (R), Potassium Periodate (R), Sodium Chromate (R), USP. All the above reagents are capable of oxidizing iodide in acid media, and the iodine released is titrated with thiosulfate solution. Only the equation for the chromate-iodide reaction is illustrated:

$$2CrO_4^{2-} + I^- + 10H^+ \rightarrow IO_3^- + 2Cr^{3+} + 5H_2O$$
$$IO_3^- + 5I^- + 6H^+ \rightarrow 3I_2 + 3H_2O$$

Sodium Bisulfite, Sulfur Dioxide, Sodium Metabisulfite (R), Sodium Sulfite (R), Sulfurous Acid (R), USP. Sulfur dioxide is absorbed in 0.1 N NaOH solution and titrated with 0.1 N iodine solution. The remainder of the above are assayed using a residual titration of excess iodine with thiosulfate solution. The sample should be added to excess iodine solution rather than the reverse. Solid samples are added directly but may react slowly unless previously powdered. Liquids are added close to the surface of the iodine solution, and the reaction flask is quickly stoppered. Products containing sulfites or bisulfites as contaminants may be treated with formaldehyde to form an addition product which prevents their reaction with iodine.

$$SO_2 + NaOH \rightarrow NaHSO_3$$
$$HSO_3^- + I_2 + H_2O \rightarrow SO_4^{2-} + 3H^+ + 2I^-$$
$$SO_3^{2-} + I_2 + H_2O \rightarrow SO_4^{2-} + 2H^+ + 2I^-$$
$$S_2O_5^{2-} + H_2O \rightarrow 2HSO_3^-$$

Methenamine Mandelate, Methenamine Mandelate Tablets, USP. The methenamine is decomposed by refluxing in acid solution. An aliquot of the sample containing the liberated aldehyde is pipetted into a special modified Nessler type reagent which has a ratio of I to Hg(II) slightly higher than the 4:1

ratio of potassium mercuric iodide. This reagent is made alkaline just before use and chilled. If the ratio is varied, an iodide of mercury rather than free mercury is precipitated:

$$HCHO + K_2HgI_4 + 3NaOH \rightarrow HCOONa + Hg + 2KI + 2H_2O + 2NaI$$

Acacia or agar is added and acts as a protective colloid for the mercury. Acetic acid solution is added and then excess iodine which reacts with the Hg:

$$Hg + I_2 \rightarrow HgI_2$$

Excess iodine is back-titrated. Since methenamine contains six methylene groups which are converted to formaldehyde, and each aldehyde group is equivalent to Hg and to 2I, the equivalent weight of methenamine mandelate is 292.34/12,000.

Ruck and Johnson[84] referred to the reagent as mercural reagent and found that a ratio less than 4:1 caused HgI_2 precipitation and a ratio over 5:1 produced mercury precipitation that was less reactive and caused low results.

Halazone, Halazone Tablets, Sodium Hypochlorite Solution, Diluted Sodium Hypochlorite Solution, NF. N-chloramines in water form hypochlorites, and the assay for all of above may be considered based on the reaction

$$ClO^- + 2I^- + 2H^+ \rightarrow Cl^- + I_2 + H_2O$$

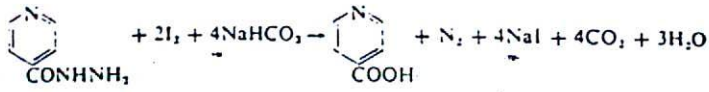
Halazone with two chlorines has a milliequivalent weight of $C_7H_5Cl_2NO_4S$/4000; sodium hypochlorite = NaOCl/2000.

Povidone-Iodine, Povidone-Iodine Solution, NF. The iodine in the above is complexed with PVP. It is not related to the chlorine in chloramines. In both instances, however, the chlorine and the iodine released from their acid solutions are referred to as available chlorine or available iodine. The iodine in povidone-iodine is titrated in an acid medium with thiosulfate solution.[85]

Antipyrine, First Supplement, NF XII (Phenazone, BPC). Antipyrine in aqueous solution, buffered with sodium acetate to maintain a neutral or weakly alkaline medium, forms 4-iodoantipyrine and HI on addition of excess 0.1 *N* iodine solution. After 20 min the crystalline precipitate which forms is solubilized with alcohol or chloroform, NF and BPC, respectively, and the excess iodine is titrated with 0.1 *N* sodium thiosulfate solution. Excess iodine is strongly adsorbed or complexed with the crystalline 4-iodoantipyrine. The residual titration is therefore inaccurate without the previous solubilization of the precipitate.

Isoniazid, Isoniazid Injection, Isoniazid Tablets, USP. After the introduction of isoniazid as an antitubercular agent, numerous methods of analysis

were proposed. Canbäch,[86] in 1952, proposed an assay for isoniazid based on a reaction analogous for hydrazine:



The reaction is slow and 90 min are needed for the completion of the reaction before back-titration of excess iodine. Appropriate measures should be taken to protect against loss of iodine by volatilization. Sodium bicarbonate is added to remove the HI formed during the reaction. The syrup also may be assayed by this procedure, but sugars such as lactose, as well as certain flavoring vehicles, interfere. A bromimetric method

$$RCONHNH_2 + 2Br_2 + H_2O \rightarrow RCOOH + N_2 + 4HBr$$

is recommended when interfering agents such as lactose are present in the formulation.[38,87]

Selenium Sulfide, Selenium Sulfide Detergent Suspension, USP. Selenium sulfide is solubilized in fuming nitric acid and diluted; the aliquot which is removed is heated to boiling with urea, cooled, an excess of KI is added, and the iodine released is titrated with sodium thiosulfate. Any nitrite present must be removed with urea:

$$2HNO_2 + (NH_2)_2CO \rightarrow 2N_2 + CO_2 + 3H_2O$$
$$H_2SeO_3 + 4HI \rightarrow Se + 2I_2 + 3H_2O$$

The selenium content is calculated as follows:

$$\frac{\text{ml} \times N \times (\text{Se}/4000)}{\text{g of sample}} \times 100 = \%\text{Se}$$

Sodium Levothyroxine Tablets, Sodium Liothyronine Tablets, Thyroid, Thyroid Tablets, USP. The sample, mixed with potassium carbonate and covered with a layer of potassium carbonate, is ignited at 675 to 700° to convert the organically combined iodine into potassium iodide. Bromine water, chlorine water, or hypochlorite solution will oxidize iodide to iodate. The former is used in the official procedure for the oxidation. Sadusk and Ball[38] use bromine vapor rather than bromine water for the oxidation:

$$Br_2 + 2OH^- \rightarrow BrO^- + Br^- + H_2O$$
$$3BrO^- + I^- \rightarrow IO_3^- + 3Br^-$$

The over-all reaction may be represented

$$I^- + 3Br_2 + 3H_2O \rightarrow 6Br^- + IO_3^- + 6H^+$$

Excess bromine is removed by boiling and finally by the addition of phenol. Any hypobromite present will react with phenol slowly unless phosphoric acid is present to free the bromine for the elimination of the bromine as tribromphenol. Phosphoric acid is also preferred for acidification of the

final iodide-iodate reaction, since it will serve as a complexing agent for some interfering ions. Once the iodate has been formed and excess bromine removed, the iodine released on addition of excess KI is titrated with standard thiosulfate solution.

Since each iodine in organic combination is converted to iodide and then to iodate and each iodate forms 6I, the milliequivalent weights of sodium levothyroxine and sodium liothyronine are 798.86/24,000 and 672.96/18,000, respectively. Thyroid and thyroid tablets are assayed for their iodine content, and 1/6000 is the milliequivalent weight used for the calculation.

Antimony Potassium Tartrate, Antimony Sodium Tartrate, Chlorinated Lime, Dimercaprol, Iodine, Aqueous Iodine Solution, Weak Iodine Solution, Mannitol, Phenelzine Sulfate, Sodium Metabisulfite, Tryparsamide, BP; and Arsenic Trioxide, Bismuth Glycollyl Arsanilate, Carbarsone, Chloramine, Chromium Trioxide, Copper Sulfate, Leptazol, Oxophenarsine Hydrochloride, Phenazone, BPC. Iodi-iodometric titrations of these compounds are included in the BP and BPC.

Acriflavine (BPC) and Proflavine Hemisulfate (BP). Assays of these substances involve titration of iodine from the oxidation of KI by the excess potassium ferricyanide solution used to precipitate the flavines. Zinc sulfate is present to precipitate zinc ferrocyanide and permit the quantitative oxidation of the iodide.

Methylbenzethonium Chloride (NF). A quarternary ammonium compound is assayed by a similar procedure. The quaternary ammonium-ferricyanide is precipitated and filtered, the excess ferricyanide is determined as above[89]:

$$2[Fe(CN)_6]^{3-} + 2I^- \rightarrow 2[Fe(CN)_6]^{4-} + I_2$$

Other titrations which may be of special interest in the possible assay of pharmaceuticals are the estimation of organic peroxides by iodide reduction[90] and the estimation of vinyl ethers[91] based on the reaction:

$$ROCH{=}CH_2 + I_2 + CH_3OH \rightarrow ROCH(OCH_3)CH_2I + HI$$

Excess iodine is titrated with sodium thiosulfate.

D. CERIC SULFATE

The relatively impure ceric sulfate and ceric ammonium sulfate originally used for the preparation of standard solutions have generally been replaced by ceric ammonium nitrate (ammonium hexanitrato cerate), $Ce(NO_3)_4{\cdot}2NH_4NO_3$, which usually assays at least 99%. A reference standard grade also is available or a lesser grade can be purified by recrystallization.[92] The product should not be dried at temperatures above 85°. Thus, some of the problems resulting from the use of the impure forms are avoided, such as excessive contamination

with other rare earths, lack of solubility, the need for a substantial but unpredictable excess of reagent, and slow precipitation before a relatively stable titer can be achieved. Ceric oxide or hydroxide also have been used for the preparation of standard solutions.[93,94]

Preparation of Solutions

Sulfuric, nitric, perchloric, and hydrochloric acids are used to solubilize the ceric salt and the anion complexes $Ce(SO_4)_3^{2-}$, $Ce(NO_3)_6^{2-}$, $Ce(ClO_4)_6^{2-}$, and $(CeCl_6)^{2-}$ are considered to form.[95] King and Pandow[96] do not consider a complex possible with perchloric acid. At the 1 to 2 *N* acid concentration usually used, the oxidation potential for ceric solutions decreases in the order: $HClO_4 > HNO_3 > H_2SO_4 > HCl$. Regardless of the acid used, only one reversible reaction can occur:

$$Ce^{4+} + e \rightleftharpoons Ce^{3+}$$

For the preparation of 1 liter of 0.1 *N* ceric sulfate solution, the weight of ceric ammonium nitrate equivalent to 33.2 g of $Ce(SO_4)_2$ is mixed with sufficient concentrated sulfuric acid to give a 1 *N* concentration of acid on dilution. Generally, acid concentrations are about 1 to 2 *N*. Water is added in 15 to 20 ml portions, slowly and with constant stirring, until the ceric salt is dissolved. This concentrated solution should be permitted to stand overnight in a closed container and then filtered (sintered-glass) and diluted with distilled water to volume. If precipitation occurs in a few days, it is usually due to improper solution, which results in the formation of less soluble salts. Ceric ammonium nitrate and ceric ammonium sulfate are used by the USP[1] and BP,[3] respectively, for the preparation of standard solutions.

Properties

Generally speaking, ceric sulfate solutions are stable to light, heat, and acids in a wide range.[97] They maintain their titer for long periods if they are aged for several days or longer before standardization. As a result of their higher oxidation potential, ceric solutions made with perchloric or nitric acid are less stable than ceric sulfate solutions. If the acid concentration is less than about 0.5 *N* the precipitation of insoluble ceric hydroxide will occur. Ceric solutions which are about 0.1 *N* usually maintain their titer for longer periods than at 0.01 *N* or less. Light may possibly affect the stability of very dilute solutions. There is some danger of chloride oxidation occurring when hot solutions (over 80°C) are titrated.

$$2Ce^{4+} + 2Cl^- \rightarrow 2Ce^{3+} + Cl_2$$

According to Smith and Getz,[98] lower normality solutions made from nitric or perchloric acid undergo a greater per cent change with time, but the stability of all solutions increases with time. A stability test of cerate solutions prepared with nitric or perchloric acids, in a range of normalities,

showed that 0.1 *N* hexanitrato ammonium cerate in 2 *N* perchloric acid underwent the least average change per day. Storage in the dark improved the life of solutions. Ceric sulfate solutions have a large coefficient of cubical expansion,[99] 0.39 ml per °C per 1000 ml in the range 15 to 30°.

Standardization

Although it is possible to prepare solutions of known normality from an accurately weighed quantity of previously dried reference standard grade ceric ammonium nitrate, changes which may occur during solution and storage make standardization desirable.

(1) Arsenic Trioxide. A known weight of previously dried primary standard As_2O_3 is solubilized in sodium hydroxide solution with the aid of heat. The solution is cooled to room temperature, acidified with dilute sulfuric acid (1 in 3), and titrated with the ceric sulfate solution using orthophenanthroline test solution (ferroin) (phenanthroline-ferrous complex solution) as the indicator and osmic acid as the catalyst. Variations of this procedure with regard to acidifier, catalyst, and indicator have been widely reported.[95,97,100–104] The arsenic trioxide method of standardization has been adopted for USP XVII[1] and should be consulted for exact details. The titration may be represented by the following:

$$2Ce^{4+} + H_3AsO_3 + H_2O \rightarrow 2Ce^{3+} + H_3AsO_4 + 2H^+$$

The presence of ICl also serves as a means of detecting the end point. Carbon tetrachloride or chloroform is added, and titration is carried out to the disappearance of the iodine color in the immiscible solvent. Free iodine is released just before the end point according to the reaction

$$H_3AsO_3 + 2ICl + H_2O \rightarrow H_3AsO_4 + I_2 + 2HCl$$

ICl is reformed on the addition of excess oxidant.

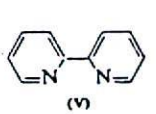
(2) Sodium Oxalate. Acidified solutions of sodium oxalate are usually titrated between 40 and 50°, using ferroin indicator and a catalyst such as ICl, or at 70° without a catalyst.[95,97,100,101] Based on the action of manganese salts as catalysts in permanganate titrations, Watson[105] has recommended manganous sulfate as a catalyst for the ceric titration as well.

(3) Iron. Iron, in the form of clean, dry iron wire, and a number of ferrous salts can be used for standardization of ceric solutions. The method using iron wire was official in USP XVI. In this method, the iron wire is solubilized in diluted sulfuric acid with heat while protected from air by the use of a Bunsen valve. To ensure that the iron is in the ferrous state, the solution is passed through a Jones reductor tube (see Fig. 4.2) containing zinc amalgam and is titrated with ceric sulfate solution using orthophenanthroline test solution (ferroin) indicator. Iron wire may be solubilized with hydrochloric

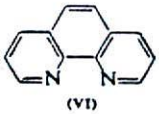
acid and reduced to the ferrous state with stannous chloride. Excess Sn(II) must be removed with mercuric chloride solution before titration with the ceric sulfate solution. Since Fe(II) is readily oxidized, an inert atmosphere of CO_2 and the use of recently boiled water for dilutions is recommended. The Sn(II) method is used for the assay of several ferrous salts.

Indicators

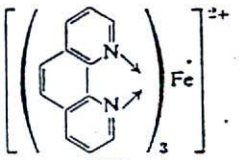
Substances such as 2,2′-dipyridyl (2,2′-bipyridine) (V), 1,10-phenanthroline (VI) and its derivatives, such as the 5-nitro, the 5,6-dimethyl, the 4,7-dimethyl,



(V)



(VI)



(VII)

and the 3,4,7,8-tetramethyl, form complexes with Fe, Cu, Zn, and other metals unless sterically hindered by substitution in the 2,9-positions.[106–112] According to Cagle and Smith,[112] the configuration

accounts for their similarity in forming an iron complex (VII) which undergoes color change at the end point. The formal potentials of these vary, and each is selected to match the potential of the oxidant used. With ferroin the Fe^{2+} complex is red and titration is to a faint blue color:

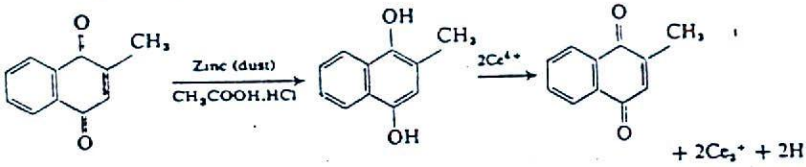
$$(C_{12}H_8N_2)_3Fe^{2+} \rightleftharpoons (C_{12}H_8N_2)_3Fe^{3+} + e$$

It is fairly stable to air oxidation but unstable over 50°. The temporary excesses of Ce^{4+} introduced during titration tend to oxidize the indicator and cause slow fading especially in the vicinity of the end point. Nitroferroin, which has less of this tendency, has a higher oxidation potential. Thus, nitroferroin is chiefly used for the so-called nitrato and perchlorato cerate solutions, and ferroin for the ceric sulfate solutions.

Dephenylamine and diphenylamine sulfonates (see Titanous Solutions) are the best indicators for some ceric titrations which require a lower oxidation potential (approximately 0.8 compared to 1.0 for ferroin). Orthophenanthroline test solution or diphenylamine test solution are the indicators called for in the USP and NF assays which use ceric sulfate solution and a noninstrumental titration.

Pharmaceutical Applications

Menadione, Menadione Tablets, USP; Menadione Sodium Bisulfite, Menadione Sodium Bisulfite Injection, NF. Menadione is reduced in the absence of air (Bunsen valve) and is titrated with standard ceric sulfate solution using orthophenanthroline test solution (USP) as the indicator:



The bisulfite addition product is hydrolyzed first in alkali and the precipitated menadione is extracted, purified, and freed of organic solvent. A similar solvent extraction and purification is used to liberate menadione from tablets before reduction and titration. A blank is necessary.

Menadione is sensitive to light and due consideration to this fact should be given during the manipulations of the assay procedure:

$$\% \text{ menadione} = \frac{\text{ml (ceric)} \times N \times C_{11}H_8O_2/2000 \times 100}{\text{sample wt in g}}$$

Hydroquinone (R) (USP) is titrated in weakly acid media with 0.1 *N* ceric sulfate solution using diphenylamine as the indicator. The reaction involved is similar to the titration of menadione following reduction to the dihydroxy derivative.[113]

Ferrous Fumarate, Ferrous Fumarate Tablets, Ferrous Sulfate Tablets, Iron Wire (for Reducing Power as Fe) (R), USP; Ferrous Gluconate, Ferrous Gluconate Tablets, NF. Ferrous fumarate and ferrous gluconate may contain not more than 2% of ferric iron as determined by the volume of thiosulfate solution required to titrate the iodine liberated from an acid solution of the sample. In the assay of these ferrous salts and their dosage forms, any iron in the ferric state is first reduced. Stannous chloride in dilute hydrochloric acid (3 in 10) is added dropwise to an acid solution of ferrous fumarate until the yellow color is discharged plus 2 drops in excess. The excess stannous compound is removed with a solution of mercuric chloride. A large excess of stannous compound is avoided to prevent reduction of the mercuric compound to free mercury. The precipitate formed here should not be excessive but definitely should be white indicating Hg_2Cl_2:

$$Sn^{2+} + 2HgCl_2 + 4Cl^- \rightarrow SnCl_6^{2-} + Hg_2Cl_2$$

The reduction step in the ferrous gluconate assay is accomplished with the use of zinc dust in diluted sulfuric acid and with air being excluded from the reaction flask by means of a Bunsen valve.

The Fe(II) formed, from either reduction method, is titrated with 0.1 *N* ceric sulfate using orthophenanthroline test solution as the indicator. To prevent any oxidation of the chloride present in the assay involving stannous chloride and hydrochloric acid, phosphoric acid is added to form a complex with the Fe^{3+} formed. The danger of chloride oxidation is slight, and probably the addition of phosphoric acid is precautionary and acts primarily to speed the reaction. Any ferric iron present will be included in the per cent of the ferrous compound; however, the ferric iron may not be more than 2%.

Ferrous sulfate in tablets is assayed by dissolving the sample portion in dilute sulfuric acid (1 in 5), filtering, washing with an acid solution of the same strength, and titrating with 0.1 *N* ceric sulfate using orthophenanthroline as the indicator. Since there is no previous reduction of any ferric compound present, only Fe(II) is determined. Certain precautions are taken to avoid oxidation of the ferrous compound, such as the use of previously boiled water and rapid filtration.

Iron wire is tested for reducing power. The iron is converted to ferrous sulfate in an oxygen-free atmosphere, the air having been replaced from the reaction flask with oxygen-free carbon dioxide. A known weight of potassium dichromate, 150 mg ± 1 mg less than weight of sample and dissolved in oxygen free water, is slowly added with stirring. The remaining ferrous iron is titrated with 0.01 *N* ceric sulfate.

Ringer's Injection (for KCl), USP; Ringer's Solution (for KCl), NF. The potassium is precipitated as the dipotassium sodium cobaltinitrite from a hydroalcoholic solution by the addition of sodium cobaltinitrite test solution. The mixture is allowed to stand for 1 hr, and then it is centrifuged. The precipitate is washed with 70% alcohol, centrifuged, drained, and dried at 80°. The dry precipitate is dissolved with the aid of heat in an excess of 0.02 *N* ceric sulfate and additional sulfuric acid. The solution is cooled and the excess Ce^{4-} titrated with 0.02 *N* ferrous ammonium sulfate, using orthophenanthroline test solution as the indicator.

On the surface it would appear that the basis for the assay is the ability of nitrites to be oxidized by ceric sulfate:

$$2Ce^{4-} + NO_2^- + H_2O \rightarrow 2Ce^{3+} + NO_3^- + 2H^-$$

The potassium sodium cobaltinitrite which precipitates is believed to have a formula somewhat intermediate between $K_2Na[Co(NO_2)_6]$ and $KNa_2[Co(NO_2)_6]$. The formula $K_{1.84}Na_{1.16}[Co(NO_2)_6]$ is considered to represent the composition of the precipitate. Slight variations in procedure may alter its composition, and therefore strict adherence to the recommended procedure is necessary. On addition of ceric sulfate solution, 1/11 of the NO_2 is oxidized by the Co(III) and 11/12 by the Ce(IV). The factor relating potassium or potassium chloride to ceric sulfate is therefore empirical.[114,115]

If $KMnO_4$ is replaced by ceric sulfate, 11 moles of $KMnO_4$ react with

5 moles of $K_2Na[Co(NO_2)_6]$; however, 25 NO_2 are oxidized by the MnO_4 and 5 by the Co.[116–118]

Sodium cobaltinitrite is tested for its suitability for determination of potassium[1] by a procedure similar to the above; however, the precipitated dipotassium sodium cobaltinitrite is dried at 105° and weighed:[114,115]

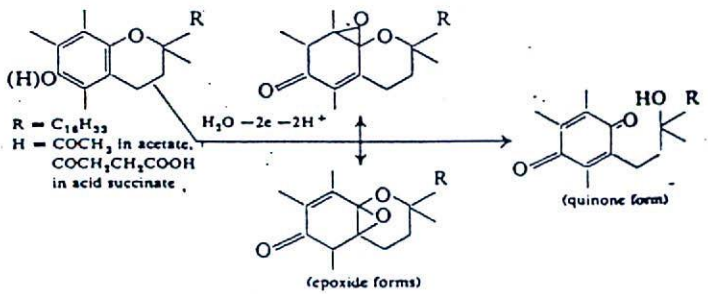
$$5K_2Na[Co(NO_2)_6] + 11KMnO_4 + 14H_2SO_4 \rightarrow$$
$$5CoSO_4 + 9MnSO_4 + 2Mn(NO_3)_2 + 5NaNO_3 + 21KNO_3 + 14H_2O$$

$$2K_2Na[Co(NO_2)_6] + 24Ce(SO_4)_2 + 12H_2O \rightarrow$$
$$Co_2(SO_4)_3 + 11Ce_2(SO_4)_3 + 2NaNO_3 + 4KNO_3 + 2Ce(NO_3)_3 + 12H_2SO_4$$

Based on this equation, the titer value for 1 ml of 0.02 *N* ceric sulfate in terms of mg of KCl would be:

$$0.02 \times \frac{KCl}{6000} \times 1000 = 0.2485 \text{ mg}$$

dl-Alpha Tocopherol, *d*-Alpha Tocopheryl Acetate, *dl*-Alpha Tocopheryl Acetate, *d*-Alpha Tocopheryl Acid Succinate, NF. *dl*-Alpha tocopherol is titrated directly with 0.01 *N* ceric sulfate from a hydroalcoholic sulfuric acid solution using diphenylamine as the indicator. For the acetate or the acid succinate derivatives, acid and alkaline hydrolysis are used, respectively, to free the tocopherol. Ether extraction of the tocopherol is necessary in the latter process. The ceric sulfate oxidation probably occurs in two stages[119]:



The milliequivalent weight of tocopherol or either of its derivatives is the molecular weight of the substance/2000. Lehman[120] has reviewed the assay of vitamin E in pharmaceutical products.

Acetomenaphthone, Acetomenaphthone Tablets, Ascorbic Acid Tablets, Ferrous Gluconate, Ferrous Gluconate Tablets, and Phanquone. The BP ceric ammonium sulfate titrations include these above compounds.

Miscellaneous Applications. The assay of polyhydroxy compounds by cerate oxidimetry using perchloratocerate is discussed by Smith and Duke.[121]

The mechanism of the reaction is explained, and the final total reaction using citric acid is

$$C_6H_8O_7 + 14Ce(ClO_4)_4^{2-} + 5H_2O \rightarrow 4CO_2 + 2HCOOH + 8ClO_4^- + 14H^+ + 14Ce^{3+}$$

A variety of polyhydroxy alcohols, and hydroxyacids of pharmaceutical importance may be assayed, including glycerol, glucose, sucrose, tartaric acid, citric acid, and malic acid.

A volumetric determination of iodide[122,123] by titration with ceric sulfate in the presence of sulfuric acid and acetone (Berg method) is based on the reaction

$$KI + 2Ce(SO_4)_2 + C_3H_6O \rightarrow KHSO_4 + Ce_2(SO_4)_3 + ICH_2\overset{O}{\overset{\|}{C}}CH_3$$

E. POTASSIUM PERMANGANATE

$KMnO_4$ has been used for over a century as an inexpensive titrant in oxidation-reduction analyses. The most common reactions attributed to the oxidizing effect of permanganate as a titrant are:

$$MnO_4^- + 8H^+ + 5e^- \rightarrow Mn^{2+} + 4H_2O \qquad E^\circ = 1.51 \text{ volts}$$
$$MnO_4^- + 2H_2O + 3e^- \rightarrow MnO_2 + 4OH^- \qquad E^\circ = 0.59 \text{ volt}$$

In alkaline or near neutral media, permanganate is a weaker oxidizing agent than in acid media, not considering the effect of the media on the reactivity of the reducing agent. The equivalent weights are $KMnO_4/5$ and $KMnO_4/3$, respectively, in acids and alkali.

Properties

Potassium permanganate is relatively stable unless contaminated with Mn(II) or MnO_2. The latter is usually present in very small quantities in commercially available potassium permanganate and must be removed in preparing solutions of the latter to prevent autodecomposition. Any Mn(II) present will react with Mn(VII) to form MnO_2, which catalyzes further decomposition. For detailed explanations of the mechanism of decomposition see Polissar[124] and Waterbury et al.[125]

Potassium permanganate solutions are unstable in the presence of direct sunlight and upon contamination with organic matter. Tests found in official compendia such as "substances reducing permanganate" and "readily oxidizable substances" make use of this property to test for or to limit the presence of certain contaminants capable of decolorizing a dilute solution of potassium permanganate. The test is usually performed on substances not affected by $KMnO_4$ (for example, acetic acid, methanol, acetone, strong ammonia solution).

In view of the above, special precautions are taken to prepare standard

solutions of potassium permanganate which will be free of reducing contaminants. Usually a slight excess of $KMnO_4$ is dissolved in distilled water and boiled for at least 15 min or kept hot on a steam bath for at least 1 h. The solution, protected from light and dust, is allowed to stand for at least 2 days before it is filtered through an inert filter such as asbestos or through a clean sintered-glass filter. The resulting solution is stored in a glass-stoppered light-resistant container which has been cleaned with a great deal of care. Any dilution of the solution should be made with water distilled from a solution of potassium permanganate. A 0.1 *N* solution, if properly prepared and stored, will maintain its titer for at least several months; however, it is best to make a periodic check. More dilute solutions require frequent standardization. It is sometimes best not to store dilute solutions but to prepare them as needed from 0.1 or 0.2 *N* solutions. No indicator is required unless the solution is 0.01 *N* or less. Ferroin or a diphenylamine derivative may be used, but for obvious reasons they should not be added until the end point has almost been reached.

Standardization

Sodium oxalate, arsenic trioxide, and, less frequently, pure iron are used for standardization of potassium permanganate solutions. The sodium oxalate method is described in the USP. The mechanism of the oxalate-permanganate reaction is reported by Adler and Noyes.[126] The official procedure does not follow the recommendations of Fowler and Bright,[127] who found that best results are obtained by adding the main portion (about 90%) of the permanganate at room temperature and with moderate mixing to the production of a pink color which slowly becomes decolorized during mixing. The solution is then heated to the recommended temperature (about 60°) and the titration is completed slowly to the production of a pink color. The older method by McBride,[128] also based on standardization against sodium oxalate, is usually considered to give slightly low results. The acidified oxalate solution is heated to about 80° and then titrated slowly with vigorous stirring so that no pink color is formed until the end point is reached. The rate of titration, the temperature of the solution (not below 60° at end point), the acidity, and other factors affect the results, and sometimes it is a good practice to standardize under the same conditions at which an unknown sample of oxalate is to be analyzed.

The first few drops of permanganate are slow to be decolorized. This is especially noted with the slow titration procedure. The explanation for the mechanism of decomposition also explains the tendency for slow decolorization until sufficient manganous catalyst is formed.

When arsenic trioxide is used as the primary standard, ICl, iodate, or iodine is selected as the catalyst.[129] After solution of the As_2O_3 in alkali, neutralization and slight acidification, the titration of the arsenite proceeds

slowly unless a catalyst is present. Mn(III), an intermediate in the reduction of Mn(VII) to Mn(II), is considered complexed by arsenate. The following equations depict the action of ICl as the catalyst and its ability to be reformed:

$$2ICl + H_3AsO_3 + H_2O \rightarrow I_2 + H_3AsO_4 + 2H^+ + 2Cl^-$$
$$5I_2 + 2MnO_4^- + 10Cl^- + 16H^+ \rightarrow 10ICl + 2Mn^{2+} + 8H_2O$$

When pure iron (iron wire or electrolytic iron) is used as the standard, the ferrous salt formed ($FeSO_4$) must be protected from air oxidation which would decrease the volume of permanganate needed for the titration. Phosphoric acid is usually added and forms a complex $[Fe(HPO_4)]^+$ with the ferric iron which is less colored than ferric chloride. With hydrochloric acid as the acidifier, the well-known Zimmermann-Reinhardt solution is added to prevent oxidation of chlorides. The $MnSO_4$ present in this solution lessens the oxidizing strength of the permanganate but the oxidation of the Fe(II) to Fe(III) proceeds more rapidly. It is less effective at elevated temperatures. The phosphoric acid in the preventive solution also speeds the reaction by forming a complex with the Fe(III) and aids in making the visual end point more easily detected. Laitinen[130] should be consulted for a review of Zimmermann-Reinhardt solution and alternative protective solutions. The lowering of the oxidation potential of the MnO_4^--Mn(II) couple so that chloride cannot be oxidized is considered inadequate, and an explanation based on the formation of higher oxidation states of Fe, Fe(IV)-Fe(V)-Fe(VI), and intermediate oxidation states of Mn is given.

Potassium iodide may be used to standardize permanganate solutions. A potentiometric method[131] was reproducible to within ±0.01%. The visual titration gave results which were on an average of 0.04% lower at a final acidity of 3.7 to 5.5 *N*. The potassium iodide was synthesized from purified $KHCO_3$ and pure HI. A visual method[132] uses ferroin as the indicator and the 0.1 *M* permanganate is titrated into an acid aqueous-acetone solution of potassium iodide. The method gave low results with more dilute solutions of permanganate,[133] owing to the reduction of the oxidizing agent by acetone and iodoacetone near the end point.

Pharmaceutical Applications

Dibasic Calcium Phosphate, USP. The sample, previously ignited, is solubilized in a hydrochloric acid solution, precipitated as calcium oxalate and filtered; the precipitate is washed with a rather dilute ammonium oxalate solution and then with water. The wash solutions should be below 20°. The washed precipitate is solubilized by acid liberating oxalic acid which is titrated with 0.1 *N* $KMnO_4$, about 80% of the titrant being added at room temperature, and the titration completed following heating to about 70°. Based on the equation

$$2MnO_4^- + 5H_2C_2O_4 + 6H^+ \rightarrow 2Mn^{2+} + 10CO_2 + 8H_2O$$

the titer value may be calculated as follows:

$$0.1\ N \times \frac{KMnO_4}{5000} \times \frac{5H_2C_2O_4}{2KMnO_4} \times \frac{CaHPO_4}{H_2C_2O_4} \times 1000 = 6.803 \text{ mg } CaHPO_4$$

or

$$6.352 \text{ mg } Ca_2P_2O_7 \quad \text{(calcium pyrophosphate)}$$

However, because the sample of dibasic calcium phosphate is previously ignited and weighed as the pryophosphate, 1 ml of 0.1 N $KMnO_4$ is made equivalent to 6.352 mg of $CaHPO_4$.

Cherry Juice (for Malic Acid), USP. The sample of juice is heated on a steam bath with an excess of calcium carbonate. Filtration separates the excess calcium carbonate from the water-soluble calcium malate, which is then precipitated as calcium oxalate. From this, the oxalic acid is liberated and titrated by the older method of heating the mixture to 80° previous to titration with permanganate. Since calcium reacts with malic acid or oxalic acid in the same ratio, 1 ml of 0.1 N $KMnO_4$ equals 6.705 mg of malic acid.

Hydrogen Peroxide Solution, Hydrogen Peroxide, 30% (R), Sodium Peroxide (R), USP; Sodium Perborate, NF. The peroxide or perborate, properly acidified and diluted, is titrated with permanganate at room temperature. It is important that the titration not be performed too rapidly in order to prevent formation of MnO_2 which catalyzes peroxide decomposition:

$$2MnO_4^- + 5H_2O_2 + 6H^+ \rightarrow 2Mn^{++} + 10O + 8H_2O$$

Hydrogen peroxide may also be assayed by titration with ceric sulfate solution. A comparison of the ceric and permanganate methods are reported by Hurdis and Romeyn.[134] Although both methods gave excellent results, the cerate method was preferred as a result of the lesser number of precautions required and the greater convenience. Cahill and Taube[135] have studied the titration of H_2O_2 with permanganate and other oxidant titrants using heavy oxygen. They report that the oxygen liberated is derived completely (>99.8%) from the H_2O_2.

Potassium Permanganate, Potassium Permanganate Tablets, USP. The sample is dissolved and treated with an excess of 0.1 N oxalic acid in an acidified media (sulfuric acid), heated to 80°, and back titrated with 0.1 N potassium permanganate.

Ferrous Sulfate, Dried Ferrous Sulfate, USP. These are titrated in acid media directly with potassium permanganate solution as represented by the equation

$$5Fe^{++} + MnO_4^- + 8H^+ \rightarrow Mn^{++} + 5Fe^{+++} + 4H_2O$$

Since iron present in the ferrous state other than total iron is assayed, no previous reduction of Fe(III) is indicated.

Sodium Nitrite, Potassium Nitrite (R), Sodium Nitrite (R), USP. Two slightly different procedures are used for determining nitrite, but the basic

reaction between the nitrite and permanganate is the same:

$$2MnO_4^- + 5NO_2^- + 6H^+ \rightarrow 2Mn^{2+} + 5NO_3^- + 3H_2O$$

Nitrites cannot be titrated directly, owing to the volatility of HNO_2 and because the titration of permanganate with nitrite proceeds too slowly, especially as the end point is approached. Cool and Yoe[136] used a direct titration of permanganate with a solution of the nitrite sample, the error was $\pm 0.1\%$ provided the titration was conducted very slowly near the end point. The usual procedure is to add the nitrite from a pipette to a known volume of standard permanganate solution present in excess, with the tip of the pipette held beneath the surface of the permanganate solution. In the assay of sodium nitrite, the mixture is warmed to 40° and permitted to stand for 5 min to allow for the sluggish reaction; then a known volume of standard oxalic acid is added in excess, the solution is warmed to 80°, and the excess oxalic acid is titrated with standard permanganate solution.

For the two reagents, following the addition of the nitrite in the usual manner, a known volume of excess 0.1 *N* ferrous ammonium sulfate solution is added and after 5 min the excess ferrous solution is titrated with 0.1 *N* potassium permanganate. The titer, mg of $NaNO_2$ equivalent to 1 ml 0.1 *N* $KMnO_4$, may be calculated as follows:

$$0.1\,N \times \frac{KMnO_4}{5000} \times \frac{5KNO_2}{2KMnO_4} \times 1000$$

Titanium Dioxide, USP. In summary, the titanium dioxide is solubilized as the sulfate by heating in sulfuric acid containing ammonium sulfate, diluted with water and filtered to remove insoluble silicates. The filtrate and washings are diluted and NH_4OH is added to reduce the acid concentration to about 5%. A Jones reductor tube (Fig. *A*.2) is prepared; directions for its preparation are given in the USP monograph. The Ti(IV) is reduced to Ti(III) in passage through the reductor and is collected in a suction flask containing a large excess of 8% ferric ammonium sulfate solution previously titrated to a pink color with 0.1 *N* $KMnO_4$. The Fe(II) formed by the reducing action of Ti(III) is titrated with 0.1 *N* $KMnO_4$.

Unlike zinc, amalgamated zinc is a rapid acting reducing agent, whose action is enhanced by its large surface area. It is essential that the reductor in the column always be covered with a layer of water, sample solution, or diluted acid to prevent its exposure to air, which causes the slow decomposition illustrated by the equations

$$Zn + O_2 + 2H_2O \rightarrow Zn(OH)_2 + H_2O_2$$
$$Zn + O_2 + H_2SO_4 \rightarrow ZnSO_4 + H_2O_2$$

The acid that is used to wash the column must not be affected by the reductor, such as nitric acid which would be reduced to hydroxylamine. Amalgamated zinc is less readily attacked by acids than zinc. Hydrochloric acid is also avoided because it attacks zinc too readily. Sulfuric acid is usually best

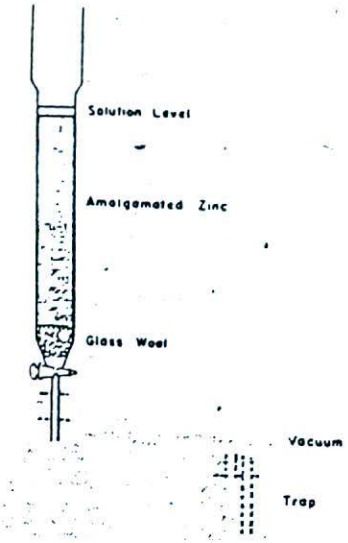


FIGURE 4.2: Jones reductor.

with the zinc reductor and hydrochloric acid with the silver reductor. The latter combination is the weaker reducing agent of the two. Each ml of 0.1 *N* $KMnO_4$ is equivalent to

$$0.1 \times \frac{TiO_2}{1000} \times 1000 \text{ mg } TiO_2$$

Ammonium Persulfate (R), USP. The persulfate sample is treated with excess standardized acid ferrous sulfate test solution and after 1 hr the excess Fe(II) is titrated with 0.1 *N* $KMnO_4$:

$$S_2O_8^{2-} + 2Fe^{2+} + 2H^+ \rightarrow 2Fe^{3+} + 2HSO_4^-$$

Various modifications of this procedure are available which do not require the 1 hr standing.[47] The reaction is completed in a few minutes if about 5 ml of phosphoric acid is added. The formation of the complex $Fe(HPO_4)^+$ speeds the reaction.

Persulfates may also be reduced with oxalic acid using silver sulfate as the catalyst; the excess oxalic acid is titrated with standard $KMnO_4$ solution.

Hydroxylamine Hydrochloride (R), USP. This assay is based on the reaction between ferric salts and hydroxylamine:

$$2NH_2OH + 4Fe^{3+} \rightarrow N_2O + 4Fe^{2+} + 4H^+ + H_2O$$

Each ml 0.1 *N* $KMnO_4$

$$= 0.1 \times \frac{KMnO_4}{5000} \times \frac{5Fe(II)}{KMnO_4} \times \frac{Fe(III)}{Fe(II)} \times \frac{2NH_2OH \cdot HCl}{4Fe(III)} \times 1000$$

$$= 0.1 \times \frac{NH_2OH \cdot HCl}{2000} \times 1000 \text{ mg } NH_2OH \cdot HCl$$

Manganese Dioxide (R), USP. The assay is related to the ability of manganese dioxide to decompose peroxide solutions.[137] A volume of peroxide in excess is added to the manganese dioxide sample in an acid medium. When the reaction is complete (the black specks of MnO_2 not visible), the excess peroxide is titrated with standard $KMnO_4$. An equal volume of peroxide is titrated directly. The difference in the two volumes of titrant represents the milligrams of MnO_2 in the sample.

$$MnO_2 + H_2O_2 + 2H^+ \rightarrow Mn^{2+} + 2H_2O + 2O$$
$$5H_2O_2 + 2MnO_4^- + 6H^+ \rightarrow 2Mn^{2+} + 10O + 8H_2O$$

$$\text{Each ml } 0.1\ N\ KMnO_4 = 0.1 \times \frac{KMnO_4}{5000} \times \frac{5H_2O_2}{2KMnO_4} \times \frac{MnO_2}{H_2O_2} \times 1000 \text{ mg } MnO_2$$

Vanadyl Sulfate (R), USP. VO^{2+} is oxidized to VO^{3+} by permanganate[138] as follows:

$$10VOSO_4 + 8H_2SO_4 + 2KMnO_4 \rightarrow 5(VO)_2(SO_4)_3 + K_2SO_4 + 2MnSO_4 + 8H_2O$$

Based on the above equation, the titer for $0.1\ N$ $KMnO_4$ in terms of milligrams of vanadyl sulfate is calculated as follows:

$$0.1 \times \frac{KMnO_4}{5000} \times \frac{10VOSO_4}{2KMnO_4} \times 1000 = 16.30 \text{ mg } VOSO_4$$

Ferrous Sulfate, Strong Hydrogen Peroxide Solution, and Hydrogen Peroxide Solution, BP; Aluminum Powder, Iron, and Iron Phosphate, BPC.

BP and BPC assays of the above involve a permanganate titration.

F. POTASSIUM DICHROMATE

Solutions of known normality may be prepared by diluting to volume an accurately weighed sample of primary standard grade potassium dichromate which has been previously powdered, if necessary, and dried between 120 and 200°. Samples of lesser purity should be recrystallized several times from an aqueous solution before drying, or solutions may be prepared directly and standardized against iron wire, ferrous ammonium sulfate, or potassium iodide.[139] The reactions usually involve the following:

$$Cr_2O_7^{2-} + 6I^- + 14H^+ \rightarrow 2Cr^{3+} + 3I_2 + 7H_2O$$
$$I_2 + 2Na_2S_2O_3 \rightarrow 2NaI + Na_2S_4O_6$$
$$Cr_2O_7^{2-} + 6Fe^{2+} + 14H^+ \rightarrow 2Cr^{3+} + 6Fe^{3+} + 7H_2O$$

Even though dichromate standard solutions are readily prepared and remain stable for long periods[140] if protected against evaporation and contamination, they are less widely used than the stronger oxidizing titrants such as ceric sulfate or potassium permanganate.

The titration of ferrous iron is one of the oldest uses for dichromate solution. If the determination is for ferric iron, the sample is first reduced with stannous chloride. Substances which are capable of reducing ferric iron quantitatively may be determined by titrating the ferrous iron formed with dichromate. Substances capable of being reduced with ferrous iron are determined by adding a known excess of ferrous solution and the residual ferrous iron is titrated.

Diphenylamine and chemically related substances, and 5,6-dimethylferroin may be used as internal indicators. These have replaced the external indicator, 0.1 % potassium ferricyanide solution.[131] When diphenylamine indicators are used for iron determinations, phosphoric acid is usually added to form a complex with the ferric ions and to lower the oxidation potential of the titrating system to the needs of the indicator and permit sharper end points.

Pharmaceutical Applications

Quinacrine Hydrochloride, Quinacrine Hydrochloride Tablets, Lead Monoxide (R), USP; Ferrous Fumarate, BPC. The use of potassium dichromate in the assay of quinacrine hydrochloride is based on the precipitation of quinacrine dichromate in a buffered medium. After filtration, an aliquot of the filtrate is acidified and the excess dichromate is determined iodometrically. Similar precipitates are formed with solutions of quinine or quinidine salts, chloroquine phosphate, dioxylene phosphate, and other related substances. To correct for loss due to the solubility of quinacrine dichromate, a correction is made by adding 0.38 ml to the ml of titrant used in the blank. The difference in the volume of 0.1 *N* sodium thiosulfate used to titrate the liberated iodine in the blank less that used for the sample is equivalent to the dichromate used for the precipitation. The per cent quinacrine hydrochloride is calculated as follows:

$$[\text{ml } 0.1\ N\ Na_2S_2O_3\ (\text{blank}) + 0.38\ \text{ml}] - [\text{ml } 0.1\ N\ Na_2S_2O_3\ (\text{sample})]$$

$$= \text{ml } 0.1\ N\ K_2Cr_2O_7 \text{ used for precipitation}$$

$$\left\{\left[\text{ml } (K_2Cr_2O_7 \text{ used}) \times 0.1 \times \frac{K_2Cr_2O_7}{6000} \times \frac{C_{23}H_{30}ClN_3O\cdot 2HCl\cdot 2H_2O}{K_2Cr_2O_7}\right] \Big/ \text{g sample in aliquot}\right\} \times 100 = \text{per cent quinacrine hydrochloride}$$

Lead monoxide is ignited at 600° and dissolved in a solution acidified with glacial acetic acid. Lead chromate is precipitated by addition of a known volume of 0.1 *N* dichromate solution and the mixture is boiled. On cooling and diluting to volume, an aliquot of the supernatant liquid is removed and the excess potassium dichromate is determined iodometrically. After

filtration, the precipitate may be dissolved in diluted hydrochloric or sulfuric acid and the dichromate titrated iodometrically:

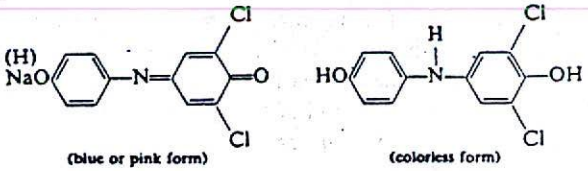
$$2Pb^{2+} + Cr_2O_7^{2-} + H_2O \rightarrow 2PbCrO_4 + 2H^+$$

It is important that sufficient time be given for the digestion of the precipitate before filtration in order to prevent fine particles of the precipitate from contaminating the filtrate.

In the assay of ferrous fumarate, Fe(II) is oxidized to Fe(III) by titration with potassium dichromate using barium diphenylamine sulfonate as the indicator and phosphoric acid to complex the Fe(III).

G. MISCELLANEOUS OXIDIZING AND REDUCING TITRANTS

Sodium-2,6-dichlorophenol-indophenol is an indicator dye chiefly used as a titrant for the determination of ascorbic acid in pharmaceutical products, in foods, and in other instances where the titration of ascorbic acid with iodine is not feasible. It is a dark green powder, usually occurs as the dihydrate, is soluble in water and in alcohol, and its aqueous solutions are blue if neutral or alkaline, pink if acid, and colorless if reduced to a hydroxy compound.



(blue or pink form) (colorless form)

It can be assayed by titrating the iodine liberated from an acid solution of the dye on the addition of KI.

The dye used for the preparation of standard solutions should be stored over soda lime to protect it from carbon dioxide. Solutions usually contain about a 0.02% concentration of sodium bicarbonate, and they are filtered to remove traces of less soluble contaminants. Solutions are standardized against ascorbic acid reference standard and the concentration of the solution is expressed in terms of milli- or micrograms of ascorbic acid equivalent to 1 ml of solution. The ascorbic acid must be protected against the action of ascorbic acid oxidase and trace metals which catalyze its autooxidation. Metaphosphoric and acetic acids (see metaphosphoric-acetic acids test solution, USP) protect against these, and the titration with dichlorophenol-indophenol solution should be carried out rapidly (BP specifies not more than 2 min).

The procedures for the standardization with ascorbic acid and the assay for the per cent of ascorbic acid are very similar. The titration with dichlorophenol-indophenol is to the appearance of a pink color which persist for 5 or 10 sec (USP and BP, respectively). The dye color is discharged until the end point is reached.

Pharmaceutical Applications

Ascorbic Acid Injection, Ascorbic Acid Tablets, Decavitamin Capsules (for Ascorbic Acid), USP; Hexavitamin Capsules (for Ascorbic Acid), NF; Titrations with Titanous Solutions (Titanium Trichloride or Titanous Sulfate)

Titanium trichloride (titanous chloride, $TiCl_3$) is an unstable substance which exists as dark-violet, almost black, deliquescent crystals or powder. It is water soluble and is usually available commmercially as a 20% solution from which standard solutions are prepared by dilution. It is one of the most powerful reducing titrants used in analysis, being readily oxidized by contact with air. Solutions of titanous salts must be preserved in an inert atmosphere usually under hydrogen or nitrogen, and carbon dioxide or nitrogen is used to displace the air in the titration vessel. For utmost accuracy, the gases are passed through wash bottles containing dilute solutions of $TiCl_3$ (about 2%).

Titanous sulfate (titanium sesquisulfate, $Ti_2(SO_4)_3$ occurs as a green crystalline water-insoluble powder. It is solubilized by dilute hydrochloric or sulfuric acids. Although solutions of the sulfate are claimed to be slightly more stable than the chloride, it is customary to store them as indicated above. The preparation of titanous sulfate solutions from titanium hydride (TiH_2) is generally recommended for its lower cost and lower iron content.[142,143] The iron content may or may not be of significance, depending on the use of the titanous solution. The determination of the iron content, correction for the iron content, and the calculation of the effective normality of the titanous solution based on standardization against potassium dichromate have been studied.[143]

Titrations involving the use of solutions of titanous salts are usually carried out using a Machlett Auto-Burette or a burette fitted with a side tube and a three-way stopcock (Fig. 4.3). The burette and the reservoir which holds the titanous solution are linked in a closed system with the titration vessel, an Erlenmeyer flask equipped with a magnetic stirrer and fitted with a three-hole rubber stopper to hold the burette tip, an inlet tube for introducing inert gas, and an exit tube. With so many manipulations involved (for example, the need for heating, refluxing, automatic stirring, replacement of air with inert gases, and titration), the use of a two- or a three-neck flask should be given some consideration for a titration vessel.

The use of titanous solutions as reducing titrants is best illustrated by the equation

$$Fe^{3+} + Ti^{3+} \rightarrow Fe^{2+} + Ti^{4+}$$

a method for the determination of iron and the method for standardizing titanous solutions against ferric ammonium sulfate.

Preparation of 0.1 *N* Solution. Add 75 ml of 20% titanium trichloride solution to sufficient 1 *N* hydrochloric acid to make 1 liter. Store in a well-filled tightly stoppered container, and when ready for use store in the closed-system

apparatus in an inert atmosphere. For increased stability of the solution, the titanous solution is mixed with concentrated hydrochloric and heated to boiling for about 1 min (only if H_2S and other volatile impurities are suspected), cooled, and diluted with recently boiled water.

Standardization. Titanous solutions are standardized against ferric ammonium sulfate solutions of known normality.[1,2] This method is less affected by iron impurities usually present in titanous solutions. An accurately measured volume of 0.1 *N* ferric ammonium sulfate is introduced into the titration flask which is then arranged for a closed-system titration. The air

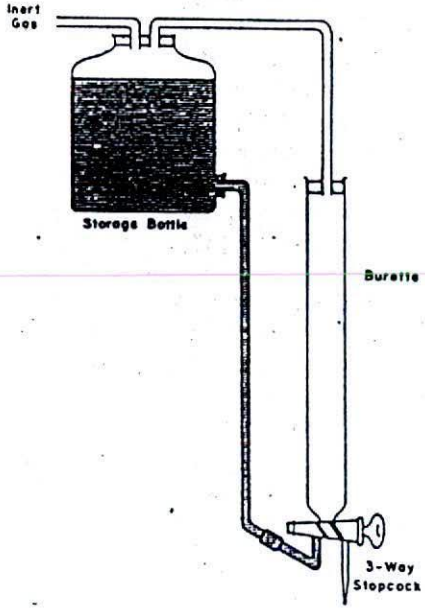


FIGURE 4.3: Burette arrangement for use of an inert gas.

in the vessel is displaced by a stream of carbon dioxide. A volume of titanous solution is introduced which is a few milliliters short of the end point (ascertained by calculation or color of the reaction mixture) and 5 ml of 10% ammonium thiocyanate is added (generally through the outlet opening to avoid introduction of air). The titration with the titanous solution is continued to the disappearance of the red color due to the presence of a red ferrithiocyanate.

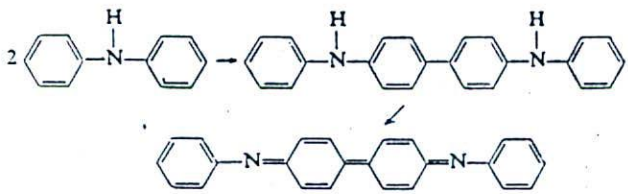
Since the reaction at the end point is slow, sufficient time should be allowed for the reaction to take place after each addition of titrant. In some instances it may be necessary to heat the reaction mixture and then cool to about 35° before continuing the titration.

The standardization may also be performed using a number of variations:

(1) Methylene blue solution may be substituted for ammonium thiocyanate as the indicator. The indicator is decolorized at the end point. Since some decolorization of the indicator may occur previous to the end point, owing to excess Ti^{3+} at the point of addition, the indicator is best added as the end point is approached. Here again the use of heat may be needed to hasten the reaction. An indicator blank is essential and is carried out using the same solvents and under the same conditions as the titration. A trace of sodium salicylate (1 drop of 10% soln) speeds the reaction at the end point.[47]

(2) Pipette standardized potassium dichromate solution into the reaction flask, acidify with dilute sulfuric acid (2 in 5), displace the air in the flask with nitrogen or carbon dioxide and add an exact volume of titanium trichloride solution which will provide about 10 ml in excess. Mix and titrate with 0.1 *N* ferric ammonium sulfate and, when approaching the end point (purple color becomes faint), add 5 ml of 10% ammonium thiocyanate solution. Titrate to the appearance of a pink color which persists for 1 min and indicates the formation of the ferrithiocyanate complex.[47]

(3) Potassium dichromate solution (acidified) can be titrated directly with titanium trichloride solution using 1% diphenylamine, 0.25% sodium diphenylamine sulfonate, or 0.5% sodium diphenylbenzidine sulfonate. In the presence of oxidizing agents they are converted into highly colored (violet or red-violet) resonating forms similar to those obtained from their parent forms diphenylamine and diphenylbenzidine, which are considered to form diphenylbenzidine violet.[144–146] These colored forms are slowly destroyed and in part this accounts for their addition to titration mixtures only near the end point.



Usually 3 to 6 drops of indicator is added as the end point is approached and an indicator blank is run. The end point is the disappearance of purple and the appearance of a blue-green color due to Cr^{3+} ions.

(4) Add a known volume of potassium dichromate to a ferrous salt present in excess in an acid media. The ferric compound formed is titrated with titanium trichloride solution, with addition of ammonium thiocyanate solution as the end point is approached. In the blank the $K_2Cr_2O_7$ is omitted.[63,147]

The necessity for standardization just before use, and special precautions required during storage and use, have limited the use of titanous solutions in assays. In organic analyses they are best known for their ability to convert nitro, azo, hydrazo, and related compounds to amines or for the conversion of

colored resonating forms into colorless nonresonating forms.[32] In some instances the latter serve as useful indicators.

$$R{-}N{=}N{-}R + 4H^+ \rightarrow 2RNH_2$$

$$RNO_2 + 6H^+ \rightarrow RNH_2 + 2H_2O$$

The kinetics of the reduction by titanous chloride of nitrobenzene and dinitrotoluene has been studied.[148,149]

By using semimicro- or micromethods and, with potassium citrate as an alkaline buffer, the time needed for the reduction has been decreased to about 2 to 3 min.[150]

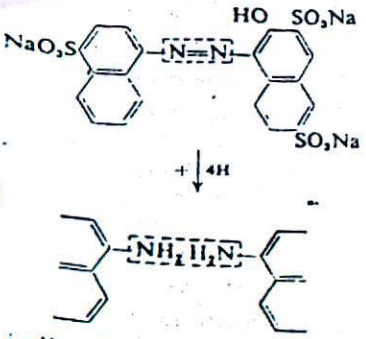
The determination of certain organic peroxides has been accomplished by titration of the Fe(III) formed with a titanous solution.[151]

The presence of peroxides in Ether (R), USP is determined by means of titanium tetrachloride. The limit is 0.0001% and is based on an orange or yellow complex formed by Ti^{4+} in the presence of peroxides. Ti^{4+} ions are complexed by other substances as well and the absorption maximum of the peroxide complex may be measured.

Pharmaceutical Applications

Amaranth, Amaranth Solution, Gentian Violet, Gentian Violet Solution, Sodium Indigotindisulfonate, Sodium Indigotindisulfonate Injection, USP; Crystal Violet, Indigo Carmine, Menaphthone, Menaphthone Sodium Bisulfite, Methylene Blue, BP; Congo Red, Magenta, BPC. The sample of amaranth is dissolved in water and buffered with sodium citrate. The latter speeds the reduction by increasing the reduction potential of Ti^{3+}. A strong acid reaction would inhibit the reduction by Ti^{3+}. The sample is heated to boiling and titrated with 0.1 *N* titanium trichloride until colorless in an inert atmosphere. Two amino compounds are formed which require 4H.

$$1 \text{ ml of } 0.1\ N\ TiCl_3 = 0.1 \times \frac{604.48}{4000} \times 1000 \text{ mg amaranth}$$



A solution of sodium indigotindisulfonate buffered with sodium bitartrate is heated to boiling and titrated with 0.1 *N* $TiCl_3$ to the disappearance of the

blue color. The end point may be colorless or slightly reddish brown. The addition of 2H results in the formation of lesser resonating forms and possibly accounts for the variations in the color of the end point:

$$1 \text{ ml } 0.1\ N \text{ TiCl}_3 = 0.1 \times \frac{466.36}{2000} \times 1000 \text{ mg sodium indigotindisulfonate}$$

An acidified solution of gentian violet is treated with an accurately measured volume of 0.1 *N* titanium trichloride in excess. The mixture is boiled for 10 min since the reaction is slow. Excess Ti(III) is titrated with 0.1 *N* ferric ammonium sulfate using ammonium thiocyanate indicator. The appearance of the faint red color of ferrithiocyanate is the end point. A blank is required. Since two hydrogens are involved in the change to the nonresonating form,

$$1 \text{ ml of } 0.1\ N \text{ TiCl}_3 = 0.1 \times \frac{407.99}{2000} \times 1000 \text{ mg gentian violet}$$

Ascorbic acid is one of the newer reducing agents introduced for volumetric analysis. Its standard solutions are used for the determination of various oxidants based on the ascorbic acid-dehydroascorbic acid system:

$$C_6H_8O_6 \rightleftharpoons C_6H_6O_6 + 2H^+ + 2e$$

Several recent reviews by Erdey[152,153] should be consulted.

Although more stable than many reducing agents used as titrants, its solutions, even if refrigerated, require standardization at the time of use. Light, heat, and the presence of metal ions as contaminants, hasten decomposition.

Of pharmaceutical interest may be the use of ascorbic acid solutions for the determination of Hg(II), Fe(III), XO_3^-, and $Fe(CN)_6^{3-}$ based on the following:

$$2Hg^{2+} + C_6H_8O_6 \rightarrow Hg_2^{2+} + C_6H_6O_6 + 2H^+$$
$$2Fe^{3+} + C_6H_8O_6 \rightarrow 2Fe^{2+} + C_6H_6O_6 + 2H^+$$
$$XO_3^- + 3C_6H_8O_6 \rightarrow X^- + 3C_6H_6O_6 + 3H_2O$$
$$2Fe(CN)_6^{3-} + C_6H_8O_6 \rightarrow 2Fe(CN)_6^{4-} + C_6H_6O_6 + 2H^+$$

In addition to the reaction with iodine, which is also a method for standardization, potassium ferricyanide may be used as a standard. The latter, in a bicarbonate-buffered medium, is titrated with ascorbic acid solution using dichlorophenol-indophenol as the indicator.

Numerous applications involving direct or indirect analysis with ascorbic acid have been found suitable for the determination of silver, Cu(II), peroxides, zinc, hypochlorite, hydroxylamine, permanganate, glycerol, formaldehyde, iodine number, and dissolved oxygen.

Variamine Blue B, N-(*p*-methoxyphenyl)-*p*-phenylene amine hydrochloride, is the redox indicator recommended for many of the above analyses. The indicator is blue in the presence of the oxidant and turns colorless when the first excess ascorbic acid appears at the end point.

Other reducing agents which are receiving attention as titrants are hydrazine sulfate and hydroquinone. *Hydrazine sulfate* is available in a highly purified form and its solutions are stable even if heated. Although many of its applications involve potentiometric or deadstop end points, the visual titration of iodine or bromine is possible using starch or starch-KI as indicators.

$$NH_2NH_2{\cdot}H_2SO_4 + 2I_2 + 6NaHCO_3 \rightarrow 4NaI + Na_2SO_4 + 6CO_2 + 6H_2O + N_2$$

Hydroquinone solutions are usually prepared in about a 2% concentration of H_2SO_4. If stored in light-resistant containers and protected from light, the solutions are stable for a month or longer. Slight discoloration usually has no effect on the titer. Diphenylamine and ferroin are suitable as visual indicators and the standardization is against potassium dichromate either as a direct or a residual titration.

For years, *hydrogen peroxide* has been used to oxidize organic matter especially following nitric or sulfuric acid digestion of the sample for analysis. It has also served to remove excess permanganate from an oxidation digestion. Volumetric solutions, such as 0.1 *N* H_2O_2, have been found to be stable for long periods if the solutions are prepared from a highly purified H_2O_2 which has been freed of ions which catalyze its decomposition and are stored in polyethylene containers.

Several other products which have been used as pharmaceuticals for many years have also found application as titrants in a variety of analyses. *Sodium hypochlorite* and *chloramine-T* (the latter more stable) solutions may be standardized by titrating the iodine liberated on the addition of excess KI. Sodium bicarbonate is used to maintain a slight alkalinity for the former. Reactions which proceed too slowly with hypochlorite may be improved by substituting hypobromite by supplying bromide for the reaction

$$OCl^- + Br^- \rightarrow OBr^- + Cl^-$$

Slow reactions may also be handled by adding excess hypochlorite and after a suitable waiting period, a measured excess of standard arsenite solution is added and the titration is completed with hypochlorite, using an irreversible indicator such as amaranth.

Some possible pharmaceutical applications may be noted from the following:

$$CO(NH_2)_2 + 3OBr^- \rightarrow N_2 + CO_2 + 3Br^- + 2H_2O$$
$$NO_2^- + OCl^- \rightarrow NO_3^- + Cl^-$$
$$2NH_3 + 3OBr^- \rightarrow N_2 + 3Br^- + 3H_2O$$
$$H_2O_2 + OCl^- \rightarrow O_2 + Cl^- + H_2O$$

A review by Zyka and Berka[134] of the new oxidizing and reducing titrants should be consulted. Although none of these are used at present in the analysis of official products, the possibility of adapting them to the analysis

H. OXYGEN COMBUSTION

The analysis of compounds containing chlorine, bromine, iodine, or sulfur in organic combination usually involves some preliminary steps to remove the organic material and convert the halogen or sulfur into an analyzable form. The oxygen-flask combustion method is such a preparatory procedure. The recent applications of this method are usually based on the work of Schöniger[155] and others.[156–158]

The sample for analysis is enclosed in halide-free paper or a methyl cellulose or cellulose acetate capsule and attached to a platinum sample

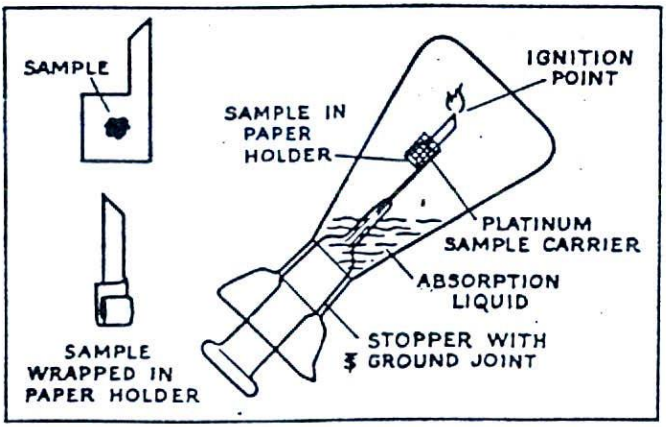


FIGURE 4.4: Oxygen combustion apparatus.

holder—essentially platinum wires fused to the stopper of a thick-walled iodine flask. The proper absorbing solution is placed in the flask, the air in the flask is replaced with oxygen, and the sample is ignited by an electric current or a paper or cotton fuse with the flask inverted so that the absorbing liquid serves as a seal (Fig. 4.4). The apparatus should be scrupulously clean and the operator should take proper safety precautions during the ignition.

The oxygen-flask method has been adopted for a number of official determinations. These are listed according to the titrant used for the final estimation. Both the USP and BP provide a general procedure with the variations and additional steps to complete the assay given in the specific monographs.

Sodium Thiosulfate Methods

Diiodohydroxyquin, Diiodohydroxyquin Tablets, Iophendylate Injection, Propyliodone, Sterile Propyliodone Suspension, Sterile Propyliodone Oil Suspension, Sodium Levothyroxine, Sodium Liothyronine, USP; Iodized Oil, NF; Acetrizoic Acid, Diiodohydroxyquinoline, Diodone Injection,

Iopanic Acid, Iophendylate Injection, Propyliodone, Sodium Diatrizoate, BP; Chiniofon Sodium, BPC

$AgNO_3$-NH_4SCN *Methods.* Sodium Sulfobromophthalein (for bromine content), USP; Assay, BP; Iodochlorhydroxyquin, USP; Butyl Chloride, First Supplement, NF; Sulfur in Sodium Sulfobromophthalein (USP) is estimated gravimetrically following oxygen combustion.

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