THE
ANALYSIS
OF
DRUGS
AND
CHEMICALS

N. EVERS
&
W. SMITH

THE ANALYSIS OF DRUGS AND CHEMICALS

NORMAN EVERS, B.Sc., PH.D., F.R.I.C.
Formerly Chief Chemist, Messrs. Allen & Hanburys Ltd.
Editor, "Analytical Abstracts"

AND

WILFRED SMITH, B.Sc., F.R.I.C.
Chief Analyst, Messrs. Allen & Hanburys Ltd.

Published in 1955

Printed in Great Britain
by Butler & Tanner Limited, Frome

PREFACE

The Analysis of Drugs and Chemicals was originally written by Norman Evers and G. D. Elsdon, being first published in 1929. Owing to Mr. Elsdon's untimely death and the intervention of World War II the book was not brought up to date. After this lapse of time, we have found it necessary to rewrite the book entirely, while retaining the general form and arrangement.

Since 1929 three editions of the *British Pharmacopœia* have been published, namely in 1932, 1948 and 1953, and two editions of the *British Pharmaceutical Codex* in 1934 and 1949. In all of these, analytical tests and methods have been developed in much greater detail with each new edition. Because of the authority of these works and the general excellence of the analytical methods contained in them it has been necessary to repeat many of these methods in this book, and for permission to include them we are indebted to the General Medical Council and to the Pharmaceutical Society of Great Britain.

Nevertheless, analysts should not regard this book as a substitute for those works but rather as supplementary to them, since it includes many substances not contained in them and in some instances gives alternative methods which are preferred by the authors. Generally speaking, official standards and limits have not been included.

It is impossible to make a cut-and-dried classification of drugs and chemicals, and often it is a matter of opinion whether a substance should be included in one or other of the chapters. We must hope that if the reader does not find a substance under the expected heading, the index will not fail him.

A description of micro methods of analysis has not been included, in spite of the undoubted importance of these methods and their probable development in the future. We feel that the techniques have been adequately described elsewhere and can usually be adapted to the methods described in this book, if it is desired to do so.

We wish to express our thanks to Professor E. J. Conway for allowing us to quote from his valuable book, *Micro-diffusion Analysis and Volumetric Error* (Crosby, Lockwood and Son, Ltd). We are also grateful to Miss V. G. Barnett for drawing some of the figures, and to her and Mrs. G. Short for typing the manuscript.

N. E.
W. S.

LIST OF ABBREVIATIONS

cm.	centimetre
mm.	millimetre
μ	micron = 10^{-3} millimetre
mμ	millimicron = 10^{-6} millimetre
A.	angström = 10^{-7} millimetre
g.	gramme
mg.	milligram
μg.	microgram = 10^{-3} milligram
gr.	grain
oz.	ounce (Avoirdupois)
l.	litre
ml.	millilitre
fl. oz.	fluid ounce
fl. dr.	fluid drachm
w/w	per cent weight in weight
w/v	per cent weight in volume
v/v	per cent volume in volume
v/w	per cent volume in weight
p.p.m.	parts per million
u.p.g.	units per gram
$x°$	degrees centigrade
$x°$ F	degrees Fahrenheit
N.T.P.	normal temperature and pressure
wt. per ml. ($t°$)	weight per millilitre at $t°$
S.G.t_1/t_2	specific gravity at $t_1°$ compared with water at $t_2°$
m.p.	melting-point, corrected
m.p. (uncorr.)	melting-point, uncorrected
m.p. (dec.)	melting-point with decomposition
f.p.	freezing-point
b.p.	boiling-point
Ref. index ($t°$)	refractive index at $t°$
O.R.	optical rotation
$[\alpha]_D^{t°}$	specific rotation (sodium light)
$[\alpha]_{5461}^{t°}$	,, ,, (mercury light)
$[\alpha]_D^{t°}$ (c = 1 in water)	,, ,, of a 1 w/v solution in water at $t°$ (sodium light)
pH	hydrogen ion exponent

U.V.	ultra-violet
E 1%/1 cm.	extinction of a 1 w/v solution in a 1 cm. cell
0·1N	decinormal
0·1M	decimolar
D-	dextro-
L-	laevo-
A.R.	analytical reagent grade
B.P.	*British Pharmacopœia 1953*
B.P.C.	*British Pharmaceutical Codex 1949*
U.S.P.	*United States Pharmacopœia XIV*
B.S.	British Standard
S.P.A.	Society of Public Analysts and Other Analytical Chemists (now the Society for Analytical Chemistry)
A.O.A.C.	Association of Official Agricultural Chemists (U.S.A.)
A.O.C.S.	American Oil Chemists' Society
sp. spp.	species
p. pp.	page, pages

JOURNALS

Journal	*Abbreviation*
Analytical Chemistry	Anal. Chem.
Archiv der Pharmazie	Arch. Pharm.
Biochemical Journal	Biochem. J.
Helvetica Chimica Acta	Helv. Chim. Acta
Industrial and Engineering Chemistry	Ind. Eng. Chem.
Industrial and Engineering Chemistry (Analytical Edition)	Ind. Eng. Chem. (Anal. edn)
Journal of the American Chemical Society	J. Amer. Chem. Soc.
Journal of the American Pharmaceutical Association (Scientific Edition)	J. Amer. Pharm. Assn.
Journal of the Association of Official Agricultural Chemists	J. Ass. Off. Agric. Chem.
Journal of Biological Chemistry	J. biol. Chem.
Journal de Pharmacie de Belgique	J. Pharm. Belg.
Journal de Pharmacie et Chimie	J. Pharm. Chim.
Journal of Pharmacology and Experimental Therapeutics	J. Pharmacol.
Journal of Pharmacy and Pharmacology	J. Pharm. Pharmacol.
Journal of the Society of Chemical Industry	J. Soc. Chem. Ind.
Manufacturing Chemist	Mfg. Chem.
Pharmaceutical Journal	Pharm. J.
Pharmaceutica Acta Helvetica	Pharm. Act. Helv.
Quarterly Journal of Pharmacy and Pharmacology	Quart. J. Pharm.
Schweizerische Apotheker-Zeitung	Schweiz. Apoth.-Ztg.
Verhandlungen der Schweizerischen Naturforschenden Gesellschaft	Verh. Schweiz. naturf. Ges.
Zeitschrift für analytische Chemie	Z. anal. Chem.
Hoppe-Seyler's Zeitschrift für physiologische Chemie	Z. physiol. Chem.
Zeitschrift für angewandte Chemie und Zentralblatt für technische Chemie	Z. angew. Chem.

CONTENTS

APPENDIXES

CHAPTER 1

Introduction

The analysis of drugs and chemicals covers the examination of a large variety of materials. A laboratory which has to deal exclusively with these products must therefore be fitted up in such a way that many types of analysis can be carried out. Almost every drug or chemical will have its own analytical methods and it will not be possible to effect as much in the way of standardization as can be done in a laboratory dealing with a few products only. Nevertheless, much economy in time and space can be effected by fitting up benches for specific purposes such as titrations, alkaloid determinations, and the determination of impurities, such as lead, arsenic, chlorides, sulphates, etc.

Micro methods

Although no micro methods are normally described in the book, their use is recommended where it is shown that they are not less accurate than the corresponding macro methods. It is possible to carry out operations, such as the assay of crude drugs, by taking one-tenth of the prescribed quantity and using micro methods for the final titration or weighing. Apart from the normal methods of qualitative and quantitative micro-analysis, much useful information may be obtained on general principles and the special types of balances and glass apparatus required from *Methods of Quantitative Micro-Analysis*, by R. F. Milton and W. A. Walters (Edward Arnold & Co., London).

Apparatus required

It should not be necessary to emphasize the importance of accurate and reliable balances and of accurately calibrated measuring apparatus. Balances should be checked at regular intervals, and much time can be saved by the use of the aperiodic prismatic reflecting types. Volumetric apparatus should be checked when received from the makers or purchased unmarked and calibrated in the laboratory, preferably by the actual user. Apart from the apparatus usual in any chemical laboratory a good supply of the following is essential: (a) accurately graduated specific-gravity bottles or pyknometers; (b) separating funnels with accurately fitting stoppers and taps; (c) reflux apparatus for saponification, acetylation, etc.; (d) apparatus for solvent extraction of drugs; (e) stills for the recovery of volatile solvents; (f) stills for the determination of boiling-points; (g) stills for alcohol determination—these are preferably arranged in "batteries" of four or more on suitable

stands ; (h) a centrifuge ; (i) grinding machines ; (j) sieves ; (k) a shaking machine, and (l) electrical stirring gear.

Physical apparatus

It is best to segregate physical apparatus in a room separate from the laboratory. This prevents corrosion of metal parts and deposition of fumes on lenses and mirrors. Needless to say, physical apparatus should be carefully treated and kept in a spotless condition. Indispensable items include a good microscope, a polarimeter, a refractometer, pH meter, colorimeter, absorptiometer, fluorimeter, spectrophotometer and viscometers. Apparatus for potentiometric titrations and a polarograph will be found useful.

Moisture determination

Electrically heated constant-temperature ovens are most generally used, but it is also necessary to have apparatus for drying *in vacuo* at a controlled temperature. Apparatus for moisture determination by the Dean & Stark and Karl Fischer methods should also be available.

THE BALANCE

It is essential in analytical work to have good and reliable balances and to see that these are properly adjusted, very carefully used and correctly housed. It is desirable to keep the balances in a separate room which is as free as possible from vibration, draughts, acid fumes and damp, and which is not subject to violent changes in temperature. The interior of each balance case should also be kept dry by means of a small jar containing anhydrous calcium chloride, soda lime or similar desiccant.

For general work an aperiodic balance weighing to 0·1 mg. will be found most generally useful and more rapid than the ordinary type. For microanalysis a special type of balance is required.

Most balances exhibit a decreasing sensitivity with increasing load and this change of sensitivity provides the criterion of the maximum load of the balance. Usually the maximum load is established by the makers and the information is supplied with the balance, but if this is not done, it may be assumed that no greater load should ever be placed on a balance than the load at which the sensitivity becomes 40 per cent of its maximum value.

Determination of sensitivity of the balance

The sensitivity of a balance is usually defined as the number of scale divisions that the rest or equilibrium point is displaced by an excess of weight of 1 mg. The sensitivity may be measured as follows. The beam is released and allowed to oscillate, the position of the pointer on the scale at the end of each swing being noted. The scale is usually divided into 20 divisions and it will be found convenient to number them from 0 to 20, when the centre point will become number 10. The mean of six swings, three to the left and three to the right, is found and the result is the true zero of the balance.

A 1 mg. weight is now placed on one pan, the operations repeated, and the second zero point found. The difference between the two means is a measure of the sensitivity of the balance.

Calibration of weights

All weights should be calibrated at regular intervals and the simplest and best method of doing this is to check each weight against a corresponding standard weight which has been calibrated by the National Physical Laboratory. If this is impracticable, it is desirable to have at least two N.P.L. calibrated standard weights, say a 1 g. and a 10 g. weight, and to calibrate the corresponding weights from these. Then, since in most cases only *weight ratios* and not *absolute weights* are required in analytical chemistry, it is necessary to see that the pieces in the set to be calibrated agree among themselves.

The method of calibration is one of substitution and for this a subsidiary set of weights is required to act as tares during the operation. The weight to be calibrated is placed on the right-hand pan of the balance and is balanced by the corresponding weight from the subsidiary set and the point of the balance ascertained by the method of swings. The weight from the right-hand pan is then removed and the corresponding standard weight is placed on the pan and the rest point of the balance again determined with the same counterpoise on the left-hand pan. From the difference between these two rest points and the known sensitivity of the balance for that load it is possible to compute the error in weight.

Having obtained the error in weight of one of the weights, it is necessary to adjust this weight either by gentle rubbing of the bottom of the screw head or by variation of the ballast until the weights agree exactly with the corresponding weights when tested by the method outlined. Having once adjusted the 1 g. and 10 g. weights, these can then be used to calibrate the other similar weights and the sum of these weights used to calibrate the larger weights. In the case of fractional weights, it is usual to obtain a third decigram weight, so that the fractional weights, whose sum is 1 gramme, may be standardized against that weight and subsequently made to correspond one with another. If a fractional weight is too heavy the edge may be rubbed down; if it is too light it should be replaced by another.

Precautions in weighing

When the weights have been accurately calibrated and the uncorrected weight (i.e. against brass weights in air) only is required, thereby disregarding the effects of buoyancy, there are only two chief sources of error to be considered. The first is the change of condition of the object between successive weighings and the second is due to inequality of the lengths of the balance arms. In order to minimize the change of condition of the object, the following precautions should be taken. No object should be weighed until it is at the same temperature as the air in the balance case. The normal procedure

after heating is to allow the object to remain in a desiccator for a minimum of 20 minutes and then in the balance case for a further 10 minutes before weighing. Some authorities state that in weighing objects which, owing to adsorption or to loss of moisture, change in weight by several milligrams in successive weighings, it is inadvisable to have a desiccating agent in the balance case. Powders should be weighed in a glass-stoppered weighing bottle, part of the powder removed for analysis, the bottle and remaining powder being weighed again and the weight of powder removed thus being obtained by difference. Liquids are best weighed in bottles or in one of the many types of weighing-pipettes. Salts containing water of crystallization should not, of course, be placed in a desiccator before weighing but should be weighed after air-drying.

It is stated that appreciable errors may be caused by electrification of the vessel being weighed by rubbing and also electrification of glass and quartz weights by friction with the velvet lining of the weight box. Such changes may be dispelled by the presence of a radioactive substance in the balance case in order to ionize the air.

The second source of error, the possible inequality of the lengths of the balance arms, may be overcome by the method of double weighing (Gauss's method), but this is unnecessary when only the relative weight of an object is required. Similarly Borda's method of weighing is independent of any inequality of the balance arms. In this method the object is counterbalanced by lead shot or similar substance and the rest point of the balance is determined. The object is then removed and replaced by weights to give the same rest point.

When the weights are always placed on the right-hand balance pan, and the uncorrected weight only is required, the error due to inequality of the arms is insignificant and may be disregarded.

Reduction of weight in air to weight *in vacuo*

In every case where absolute weights are required, a correction for buoyancy must be made. A correction for buoyancy is also required when it is necessary to determine the specific gravity of a liquid of density less than 0·9 or greater than 1·2 with an accuracy of $\pm$ 0·0001.

The true weight, that is the weight *in vacuo*, is given by the following formula to a very close approximation :—

$$W_v = W_a + W_a \times D_a\left(\frac{1}{D_s} - \frac{1}{D_w}\right)$$

where W_v is the true weight
W_a ,, ,, weight in the air
D_s ,, ,, density of the substance being weighed
D_w ,, ,, ,, ,, weights used
D_a ,, ,, ,, ,, air.

Under normal conditions of temperature, pressure and humidity D_a is about 0·0012, and for brass weights D_w is 8·4, so that the equation becomes

$$W_x = W_a + W_a \times 0{\cdot}0012\left(\frac{1}{D_s} - \frac{1}{8{\cdot}4}\right)$$

or

$$W_x = W_a + \frac{k\,W_a}{1000} \quad \text{where} \quad k = 1{\cdot}20\left(\frac{1}{D_s} - \frac{1}{8{\cdot}4}\right)$$

The values of k for $D_a = 0{\cdot}0012$ and $D_w = 8{\cdot}4$ have been calculated and collected in Table 1. If the substance has a density lower than that of brass the correction is positive, and if the density is greater than that of brass the correction is negative.

Table 1

D_s	k	D_s	k	D_s	k	D_s	k
0·5	+ 2·26	1·4	+ 0·72	3·5	+ 0·20	10·0	− 0·02
0·6	+ 1·86	1·5	+ 0·66	4·0	+ 0·16	11·0	− 0·03
0·7	+ 1·57	1·6	+ 0·61	4·5	+ 0·13	12·0	− 0·04
0·8	+ 1·36	1·7	+ 0·56	5·0	+ 0·10	13·0	− 0·05
0·9	+ 1·19	1·8	+ 0·52	5·5	+ 0·08	14·0	− 0·06
1·0	+ 1·06	1·9	+ 0·49	6·0	+ 0·06	15·0	− 0·06
1·1	+ 0·95	2·0	+ 0·46	7·0	+ 0·03	16·0	− 0·07
1·2	+ 0·86	2·5	+ 0·34	8·0	+ 0·01	17·0	− 0·07
1·3	+ 0·78	3·0	+ 0·26	9·0	− 0·01		

CALIBRATION OF VOLUMETRIC APPARATUS

Before any attempt is made to calibrate glass apparatus it must be thoroughly clean. If, on filling a vessel with distilled water and then pouring it out, an unbroken film of water remains, there is no necessity to clean, other than rinsing with distilled water. If the water collects in drops, the vessel is dirty and must be cleaned either with hot soapy water or with a hot solution of one of the proprietary detergents commonly used for glass cleaning. If this is not effective, the vessel should be filled with hot concentrated sulphuric acid in which powdered potassium dichromate has been dissolved, and allowed to stand overnight. Whichever method is used, the vessel should be finally rinsed with several lots of distilled water. If it is necessary to weigh the vessel, it must be thoroughly dried either in a steam-oven or by washing out first with alcohol and then with ether. In either case, any water or ether vapour should be blown out with a current of air and the vessel allowed to assume laboratory temperature without the stopper in position before weighing.

Apparatus is calibrated on the basis of the litre which is defined as the volume occupied by one kilogram of water at the temperature of its maximum

density (4°) at 760 mm. pressure. One thousandth part of a litre is the millilitre (ml.).

The standard temperature for the calibration of apparatus up to 1934 was 15·5°, but since the issue of the *Report on the Standard Temperature of Volumetric Glassware* (No. 554) in that year by the British Standards Institution, 20° has been generally adopted. Calibration of apparatus is, however, best carried out at the temperature of the laboratory and corrections made from Table 2 on page 8.[1] The temperature should preferably be between 17° and 23° and extremes should be avoided.

The marking of volumetric apparatus

Although this may be done with a file, the method is not very satisfactory, especially in the case of pipettes (on account of the risk of fracture), and the method given below is much better. A strip of gummed paper having a straight edge is fitted round the stem of the instrument to be marked, and placed accurately in position so that the edge of the paper occupies the line where the mark is to be placed ; the stem is then coated with a thin layer of hard paraffin. When the paraffin has hardened, a ring is cut with a knife along the edge of the gummed paper and the exposed glass is then etched with hydrofluoric acid rubbed in with a pad of cotton-wool attached to a piece of copper wire.

Reading a meniscus

The important points in reading a meniscus are (a) to avoid errors of parallax by reading with the eye at the same level as the meniscus, and (b) to read against a uniform background. Errors of parallax are best avoided by carrying the graduation mark right round the tube. A uniform background may be obtained by placing a piece of white paper behind the tube. The lowest point of the dark part of the meniscus should be taken as the reading.

Standards

The Class A Standards of the National Physical Laboratory as laid down in the pamphlet issued in April 1934 have been adopted in this book.

Errors of measurement

Errors of measurement of volume by means of glass apparatus may be divided into two groups—*constant errors* and *variable errors*. When a volume of liquid is measured, say from a pipette, the mean of a large number of measurements will vary from the stated volume by an amount which depends on the position of the graduation mark on the stem. This difference is the *constant error* of the pipette and may be allowed for in making a measurement. At the same time, the measurements from which the mean was calculated vary among themselves in a manner which may be regarded as following the normal distribution curve (see p. 485). These variations are due to differences

[1] For comprehensive tables for use in the calibration of volumetric glassware see B.S. 1797 : 1952.

in draining, temperature, etc., and make up the *variable error* which may be expressed as a standard deviation in the usual way (see p. 484). No allowance can be made for variable error, but a knowledge of its value enables us to assess the maximum volume by which any single measurement is likely to differ from the mean.

Errors of pipettes

Provided that the adjustment of the meniscus to the mark is carefully made and the diameter of the tube is not too great, the variable error from this cause is small and may be neglected in comparison with errors due to other causes such as the amount of fluid left on the walls and on the tip of the pipette after draining. Errors due to changes of temperature are, of course, important, but can be corrected as will be explained later. The variable error is increased by reducing the delivery time, and it is important that pipettes should conform to the standard times of delivery given below. The variable error is, in fact, directly proportional to the volume of liquid remaining on the walls of the pipette and inversely proportional to the square root of the time of delivery.[1] It is therefore important that in filling a pipette the liquid should not be drawn up more than is necessary above the mark.

The most accurate type of pipette for a small volume is the straight tube of the blow-out type. Those which measure between two marks are less accurate, since they are liable to the errors which will be discussed under burettes. The glass at the end of a pipette should be as thin as possible, the taper should be gradual and the end should be ground off. If these points are observed, it is easy to obtain 1 ml. pipettes having the same percentage variation as a 25 ml. standard pipette merely by prolonging the delivery time.

Errors of burettes

The delivery error of burettes, as with pipettes, is proportional to the liquid remaining on the walls and varies according to the delivery time. Since in practice the delivery time may vary from one titration to another, the variable error from this cause is greater than with pipettes. In macro-titrations the end-point is often taken to the nearest drop, which from a good burette should not exceed 0·04 ml. This source of error may be reduced by taking fractions of a drop by touching the side of the vessel with the tip of the burette. Errors of reading are also not negligible as is the case with pipettes, owing to the larger diameter of the tube and the fact that two readings are necessary. The last two errors are independent of the volume used for titration, but the delivery error is proportional to the volume delivered. Using a 50 ml. burette a delivery volume of about 35 ml. should be aimed at as being subject to the least variable error due to the burette alone. Burettes with graduation marks extending round the tube are less liable to parallax error in reading than those with the usual type of scale or the Schellbach type.

[1] See *Micro-diffusion Analysis and Volumetric Error* by E. J. Conway (Crosby Lockwood & Son, Ltd) for a very valuable discussion of this subject.