ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

CHEMISTRY

ALKALOIDS

Protoveratrine A and B and Germitetrine B from *Veratrum album*. H. A. Nash and R. M. Brooker. (*J. Amer. chem. Soc.*, 1953, **75**, 1942.) Protoveratrine from *Veratrum album* was found to be a mixture of two alkaloids, protoveratrine A and B. They were separated by a Craig countercurrent distribution procedure using a chloroform–water–acetic acid system. Protoveratrines A and B are remarkably alike in many of their properties. Protoveratrine A was found to conform to the accepted structure of protoveratrine except that it yielded two instead of one mole of acetic acid on hydrolysis. Protoveratrine B yielded protoverine, 2-methylbutyric acid, 2:3-dihydroxy-2-methylbutyric acid and two moles of acetic acid on hydrolysis, corresponding to an empirical formula of $C_{41}H_{63}O_{15}N$. Analytical data and equivalent weight determinations support this formula. After removal of "protoveratrine" from the total alkaloids, considerable hypotensive activity remained in the residual "amorphous alkaloid" fraction. Application of paper chromatographic methods to the examination of the fraction showed the presence of at least 15 alkaloids. By means of countercurrent distribution and fractional crystallisation, an alkaloid named germitetrine B was isolated. It yielded germine, 2-methylbutyric acid, 2:3-dihydroxy-2-methylbutyric acid and two moles of acetic acid on alkaline hydrolysis. A. H. B.

***Veratrum eschscholtzii* Gray, Alkaloids of.** M. W. Klohs, M. D. Draper, F. Keller, M. Malesh and F. J. Petracek. (*J. Amer. chem. Soc.*, 1953, **75**, 2133.) *iso*Rubijervosine, a new glucosidic alkaloid, was isolated from the hitherto uninvestigated species, *Veratrum eschscholtzii* Gray, as well as the known glycosides, pseudojervine and veratrosine. *iso*Rubijervosine was obtained from the hypotensively active amorphous bases by the use of chromatography and fractional crystallisation as fine needles, m.pt. 279° to 280° C., $[\alpha]_D^{24^\circ C.} - 20 \pm 2$ (*C* 1·45 in pyridine). The formulation $C_{33}H_{53}O_7N$ for the alkaloid was derived by analysis of the base and its pentacetyl derivative. On acid hydrolysis, *iso*rubijervosine yielded the aglycone *iso*rubijervine and D-glucose, and from this and other evidence its structure was established as 3-(β)-D-glucosyl-$\triangle^5$-solanidene-18-ol. A. H. B.

ANALYTICAL

Barbiturates, Argentimetric Determination of. P. Chavanne and H. Marie. (*Ann. pharm. franç.*, 1953, **11**, 91.) The method is based on that of Danielsson (*Svensk farm. Tidskr.*, 1951, **55**, 125) but avoids the use of potassium metaborate which is difficult to obtain: 0·6 g. of the barbiturate is dissolved in 7 ml. of N ethanolic potash, and the solution is diluted with 33 ml. of water and treated with 0·45 g. of boric acid. After warming to dissolve the boric acid, and cooling, the mixture is titrated with 0·1 N silver nitrate in presence of 1·5 ml. of 10 per cent. potassium chromate solution as indicator. The end-point is indicated when the solution assumes permanently a colour different from that of a comparison solution containing 2 g. of precipitated calcium carbonate,

1·5 ml. of potassium chromate solution and 55 ml. of water. One molecule of silver nitrate corresponds to 2 molecules of barbiturate. The method has been applied to barbitone, allobarbitone and butobarbitone. It may also be used for hexobarbitone, but in this case there is no precipitate and the comparison solution does not contain calcium carbonate. For phenobarbitone, a different *p*H value is necessary and the amount of boric acid is therefore increased to 1·20 g. G. M.

Bismuth, Direct Titration of with Complexone. K. E. Grönkvist. (*Farm. Revy*, 1953, **52**, 305.) A method is proposed for the determination of bismuth in compounds and preparations occurring in pharmacy. The procedure is based on the titration at a *p*H of 2·5 to 4·0 with 0·05M complexone III (the disodium salt of ethylenediamine tetra-acetic acid) using the yellow-coloured complex between bismuth and thiourea as indicator. The general method consists of neutralising the colourless solution of bismuth (corresponding to 0·10 to 0·20 g. Bi) in nitric or hydrochloric acid with 5M sodium hydroxide added drop by drop until the first permanent turbidity is obtained and then adjusting the volume to about 30 ml. with water. Thiourea (6 g.) is added and solution effected by gentle heat on a water bath. After the addition of 30 ml. of water, 3 g. of potassium hydrogen phthalate and one drop of a gentian violet solution, the bismuth is titrated by means of complexone III 0·05M to a violet colour. The solutions to be determined must be colourless, and the pre-treatments of the pharmaceutical coloured substances to obtain the bismuth in colourless solution are described. The results by the proposed method are compared with results obtained by official methods of assay. Most metal ions in large amounts interfere with the determination but NH_4, K, Na, Ca, Ba, Sr and Mg in 5 times the amount of bismuth did not interfere. A. H. B.

Calomel, Assay of, in Preparations Containing Vegetable Drugs. L. Domange and L. Seguin. (*Ann. pharm. franç.*, 1953, **11**, 193.) Calomel may be determined by reaction with an excess of standard iodine solution, followed by back-titration with thiosulphate, but the method gives high results in the presence of cascara, tansy, santonica, etc., which react with iodine. It is preferable to destroy the organic matter before carrying out the determination, care being taken to avoid loss of calomel by volatilisation. The following method is recommended. Place the sample, containing 0·06 to 0·12 g. of calomel and preferably not more than 0·25 g. of vegetable matter in a conical flask, add 10 ml. of sodium persulphate, 100 ml. of water and 20 ml. of sodium hydroxide solution, shake frequently during 1 hour, cover with a small funnel and heat for 2 hours on a boiling water bath. This destroys organic matter and produces a yellow precipitate. Cool, add 10 ml. of sodium hydroxide solution and 10 ml. of formaldehyde solution and allow to stand for 15 hours to reduce the precipitate to mercury. Decant the supernatant liquid through a filter, wash with a dilute solution of sodium hydroxide and place the filter in the flask. Add 2 ml. of acetic acid (50 per cent.) and 20 ml. of 0·1N iodine, and, when all the mercury has dissolved, titrate the excess of iodine with sodium thiosulphate in the presence of starch mucilage. Preparations containing a large proportion of sucrose or lactose should be washed with water to remove the sugars before assay. Suppositories should be prepared for assay by removal of the oil of theobroma with ether, but pills may be assayed without prior treatment. G. B.

Glycerol, Periodate Determination of. L. Hartman. (*J. appl. Chem.*, 1953, **3**, 308.) A study of the determination of glycerol by the potassium periodate method has been carried out with the object of developing a rapid and exact analytical procedure. It was found that the time of oxidation recommended in the existing methods could be substantially reduced from 1 hour or more to 5 minutes without affecting the accuracy of the determination. Results and conditions are given for the oxidation procedures based on the titration of the formic acid produced and on the estimation of the residual periodate. The use of potassium dimesoperiodate (dipotassium periodate) instead of the much less soluble metaperiodate (monopotassium periodate) made it possible to increase the size of the glycerol sample and thus to achieve a greater reproducibility of results; the salt in solution can be prepared by dissolving potassium periodate in an equivalent amount of aqueous potassium hydroxide. R. E. S.

Hexoses, Determination of. B. Klein and M. Weissman. (*Analyt. Chem.*, 1953, **25**, 771.) A new colour reaction is described for the identification and determination of hexoses in the presence of pentoses, based upon the action of chromotropic acid in 15M sulphuric acid. The reaction depends on the conversion of hexoses to 5-hydroxymethylfurfural followed by the splitting of the methylol group to form formaldehyde which reacts with chromotropic acid. Under these circumstances, pentoses which form furfural, incapable of splitting off formaldehyde, do not react, but the common hexoses—glucose, galactose, mannose and fructose—react and give a characteristic violet colour, the intensity varying linearly with the concentration; the common disaccharides —lactose, sucrose and maltose—also react. The pentoses arabinose, xylose, and rhamnose failed to react under the conditions of the experiment. No quantitative stoichiometric relationship was apparent between the amount of hexose used and the formaldehyde produced. Pentoses did not interfere with either the quantitative or the qualitative reaction; 1 ml. of a solution containing 0·1 mg. of glucose with increasing amounts of arabinose produced the same optical density in the reaction and a quantitative glucose determination was unaffected. R. E. S.

***iso*Nicotinyl Hydrazide, Analysis of.** A. Anastasi, E. Macarelli and L. Novacic. (*Mikrochemie*, 1952, **40**, 113.) Alkaline solutions of *iso*nicotinyl hydrazide may be assayed polarographically. A solution, of 0·1 to 1 millimols/l., is prepared by dilution with Britton-Robinson buffer (*p*H 9) containing 0·01 per cent. of gelatin, and examined in the range $-0{\cdot}8$ to $-1{\cdot}8$ volts. A corresponding sodium acetate-hydroxide buffer may also be used, in which case the half-wave potential is $-1{\cdot}33$ in place of $-1{\cdot}2$ (saturated calomel electrode). The presence of *iso*nicotinic acid, in the case of acetate buffer, is indicated by a reduced potential step proportional to the amount of hydrolysis of the hydrazide. Free hydrazine cannot be detected polarographically. The amount of hydrazine produced by decomposition of a solution can be determined by the increase in the titre of the solution with nitrous acid, in accordance with the following equations:

$$RCO{\cdot}NH{\cdot}NH_2 + HNO_2 = RCON_3 + 2H_2O$$

$$H_2N{\cdot}NH_2 + 2HNO_2 = 3\,N + 3H_2O + NO$$ G. M.

***iso*Nicotinyl Hydrazide, Electrometric Determination of.** A. Anastasi, E. Mecarelli and L. Novacic. (*Mikrochemie*, 1952, **40**, 53.) The following methods are given. 1. Addition of excess of cerium sulphate, heating for 1 hour on the water bath, and back titration with ferrous sulphate. The results show a variation of about 2 per cent. 2. Titration with nitrous acid, with formation of $RCON_3$. The end-point may be determined with starch iodide paper, but better electrometrically. The method may be applied directly to tablets, injections and syrups. The end-point may be obtained from the potential curve of a platinum electrode against a saturated calomel electrode, by observation of the polarisation current between two platinum electrodes (applied voltage about 15 mV) or by observation of the current between a platinum and a tungsten electrode. 3. Titration of the hydrazide group in glacial acetic acid with perchloric acid, using crystal violet or, better, an electrometric method. 4. Titration of the *iso*nicotinic acid group with sodium methylate in diethylamine solution. The end-point is determined with thymol blue, or electrometrically. For the latter an antimony-glass electrode pair is used. G. M.

***iso*Nicotinyl Hydrazide, Volumetric Determination of.** H. Harting. (*J. Amer. pharm. Ass. Ass. Sci. Ed.*, 1953, **42**, 323.) A simple and rapid method for the determination of isoniazid depends upon the evolution of the hydrazine nitrogen in the presence of an excess of oxidising agent such as a mixture of potassium ferricyanide and potassium hydroxide. The nitrogen evolved is measured by displacement in a burette and its weight calculated. Each g. of nitrogen is equivalent to 4·893 g. of isoniazid. 10 ml. of a 20 per cent. solution of potassium ferricyanide and 10 ml. of a 20 per cent. solution of potassium hydroxide is sufficient to oxidise 50 mg. of isoniazid dissolved in 20 ml. of water. The method is applicable to tablets, mixtures and biological fluids, but cannot be used in the presence of thiacetazone which yields nitrogen under the same conditions.

G. B.

Phenyl Thiohydantoins, Paper Strip Identification of. J. Sjöquist. (*Acta chem. scand.*, 1953, **7**, 447.) In Edman's method (*Acta chem. scand.*, 1950, **4**, 283) for determining the amino-acid sequence in peptides, phenyl thiohydantoins are formed. A paper chromatographic procedure for the direct identification of these latter compounds is described. The descending technique was used, and solvent of heptane–pyridine and heptane–butanol with formic acid applied. The R_F values of a number of phenyl thiohydantoins prepared from naturally occurring amino-acids are recorded. A. H. B.

Platyphylline, New Reaction for. V. F. Kramarenko. (*Aptechnoe Delo*, 1953, **2**, No. 2, 52.) The present identity test for platyphylline tartrate in the Soviet Pharmacopœia VIII is based on the tartrate radical. The following reaction which is based on the presence of a carboxylic ester group in the molecule of platyphylline base is proposed. Place a small quantity of the substance in a small test-tube; add 1 drop of saturated ethanolic solution of hydroxylamine hydrochloride and 1 drop of saturated ethanolic potassium hydroxide. Heat to boiling and, after cooling, add two drops of ethanolic hydrogen chloride. Add a few pieces of marble to remove excess of hydrochloric acid and when the evolution of carbon dioxide has ceased add one drop of hydrogen peroxide and, after a minute, 1 drop of 5 per cent. ferric chloride solution. A transient blue-violet colour is observed. E. H.

Potassium, Colorimetric Determination of, with Dipicrylamine. R. Faber and T. P. Dirkse. (*Analyt. Chem.*, 1953, **25**, 808.) The method of Amdur (*Industr. Engng Chem. (Anal.)*, 1940, **12**, 731) was examined in an attempt to determine the extent to which zinc and other ions interfered in the estimation. It was evident that the presence of even small amounts of ammonium ion caused considerable errors in the potassium determination. The effect of *p*H was studied and it was found that a *p*H of 3 or 3·5 was the lower limit for accurate results, the upper *p*H limit being about 11. In the presence of zinc it was found that only when the weight of zinc in a sample was greater than the weight of potassium need the zinc be removed or reduced in amount before proceeding with the determination of potassium. R. E. S.

Riboflavine, Polarographic Determination of. W. J. Seagers. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 317.) Purified and partially purified riboflavine were assayed by dissolving 20 to 30 mg. in 5 ml. of 0·4M sodium hydroxide and adding 20 ml. of buffer solution (0·2M acetic, boric and phosphoric acids and 0·1M potassium chloride) and sufficient water to produce 50 ml. The solution (*p*H 2·8) was placed in a polarographic cell with a dropping mercury electrode, and nitrogen passed through the solution to remove dissolved oxygen. The polarograph was taken over the range of potential 0 to –1 volt. The experiment was repeated using dried riboflavine U.S.P. reference standard and the result calculated from the ratio of the diffusion currents. Solubilised riboflavine compounds were assayed by dissolving 0·5 to 0·6 g. in 100 ml. of water and adding to a 10-ml. aliquot, 10 ml. of a mixture of 1 part of 0·4N sodium hydroxide and 4 parts of buffer solution. Another 10-ml. aliquot was similarly treated, but 20 mg. of dried reference standard was added. The solutions were diluted to 50 ml. and polarographed as before. Vitamin mixtures containing riboflavine were mixed with a solution of equal quantities of 0·1M hydrochloric acid and acetone, 10 ml. of the supernatant liquid, after standing, being placed in each of 2 flasks. To one was added 5 ml. of a mixture of 4 parts of the acetone —0·1M hydrochloric acid and 1 part of 0·4M sodium hydroxide. To the other was added 5 ml of riboflavine standard solution. The solutions were deoxygenated and polarographed as before. The results agreed satisfactorily with the fluorimetric assay, taking into account the precision of the methods, ± 2 per cent. for the fluorimetric and ± 3 per cent. for the polarographic assays. G. B.

Sugars, Colorimetric Estimation of, with Benzidine. J. K. N. Jones and J. B. Pridham. (*Nature, Lond.*, 1953, **172**, 161.) A quantitative technique, using a solution of benzidine in acetic acid, has been perfected for the majority of common sugars and their methylated derivatives, with the exception of ketoses. 1 ml. of sugar solution and 5 ml. of a 0·2 per cent. (w/v) benzidine solution in glacial acetic acid were placed in a boiling-tube and heated in a boiling water bath for 15 to 60 minutes according to the sugar under test. After cooling to room temperature the orange-yellow coloration was measured photometrically (Ilford No. 601 filter), the relationship between concentration and absorption being linear. Polysaccharides could be analysed quantitatively by the new method using paper chromatography; sugars were extracted from the paper with hot methanol (90 per cent.), the methanolic sugar solution was evaporated to a syrup *in vacuo* and distilled water added, a 1-ml. aliquot of the solution being taken for analysis. An error of 3 to 5 per cent. was found in the estimation of standard sugar mixtures. R. E. S.

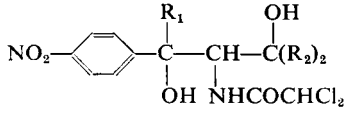
Sulphate and Organic Sulphur, Microdetermination of. L. Andersen. (*Acta chem. scand.*, 1953, **7**, 689.) A spectrophotometric method is presented for the determination of sulphate and organic sulphur on the micro- and ultramicro-scale depending on the fact that benzidine and its salts absorb strongly in the ultra-violet, $\lambda_{max.}$ at 250 mμ. The sulphate is precipitated as benzidine sulphate, is isolated, dissolved in hydrochloric acid and diluted to a certain volume, and its absorption measured in a spectrophotometer. The solubility of benzidine sulphate in water is 98 mg./l. at room temperature and the precipitation cannot, therefore, be made from a highly diluted solution. Strong acidity raises the solubility further, but it is most advantageous to work with slightly acid solutions. For ultramicro quantities the solubility can be reduced by using ethanolic solutions. For the determination of sulphur in organic material, the destruction is generally carried out by the method of Carius in a small sealed tube of about 0·5 cm. diameter; the nitric acid is evaporated, and the remaining traces of free acid carefully neutralised by means of ammonia. R. E. S.

Sulphonamide Separations based on Ion Exchange Chromatography. H. H. Hutchins and J. E. Christian. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 310). The distribution coefficients of sulphanilamide and sulphadiazine were determined for 5 cation exchange resins, by placing 10 or 25 ml. of a standardised solution of the S-35 labelled sulphonamide in hydrochloric acid solution in bottles containing various weighed amounts of the cation exchangers in the free acid form and shaking for 72 hours to bring to equilibrium. 1-ml. quantities of the solutions were analysed by Geiger-Muller counter technique. The ratio of average distribution coefficients for sulphanilamide and sulphadiazine with any of the resins was greater than 1·2, indicating the possibility of separation by chromatographic elution. The effects of flow rate, type and depth of resin, mesh size and elutriant were investigated for 6 sulphonamide derivatives. Sulphanilamide was separated from its acetyl derivative by placing 25 ml. of aqueous acidic solution on a column of Amberlite IR-120 (H) A.G., adding distilled water and collecting a 500-ml. fraction which contained the acetyl derivative, sulphanilamide being retained on the column until eluted with 1000 ml. of 4N hydrochloric acid. In a method of analysis of mixtures of sulphonamides in tablets, etc., solutions of S-35 labelled sulphadiazine and sulphanilamide of known specific activity were added to a solution prepared from tablets containing sulphanilamide and sulphadiazine. After separation on a column of Amberlite IR-120(H)A.G. the specific activity of suitable eluate fractions was determined and the content of sulphonamide calculated. This method is applicable to many types of sulphonamide mixture and to the determination of trace amounts in biological fluids. G. B.

Thioureas, Paper Chromatography of. A. Kjaer and K. Rubinstein. (*Acta chem. scand.*, 1953, **7**, 528.) A detailed description of the technique used, and the results obtained by the application of paper chromatography to the separation of a large number of thioureas is given: *N*-substituted and *NN'* and *NN*-disubstituted thioureas were investigated. The ascending technique was used and the most suitable solvent mixture proved to be chloroform/water. Exploratory experiments indicated that the application of the method to quantitative studies gave results with an accuracy of ± 5 per cent. Some related compounds, viz. thioamides, thiohydrazides, thiosemicarbazides and thiobarbituric acids were also briefly studied by the same technique. A. H. B.

ORGANIC CHEMISTRY

Chloramphenicol, Related Compounds of: Some Tertiary Alcohols. M. C. Rebstock and A. C. Moore. (*J. Amer. chem. Soc.*, 1953, **75**, 1685.) A series of compounds related to chloramphenicol were prepared in which the "1" hydrogen or the two "3" hydrogens were replaced by *p*-nitrophenyl or methyl groups. The compounds were of general formula.



and the following substances were prepared: (*a*) R_1 = *p*-nitrophenyl-, R_2 = H, (*b*) R_1 = H, R_2 = CH_3, (*c*) R_1 = H, R_2 = *p*-nitrophenyl-, (*d*) R_1 = CH_3, R_2 = H. None of the compounds was found to possess significant antibacterial, antirickettsial or antivirus properties. A. H. B.

Gentisic Acid, Infra-red Spectra of Esters of. J. F. Nash, F. W. Bope and B. V. Christensen. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 250.) Infra-red spectra were obtained by examination of solutions in chloroform, ether and nujol gel, and evaluated by comparison with the spectra of known compounds of gentisic acid. Decreased absorption in the region 3·0 to 4·2 μ occurred when the 5-hydroxyl group was modified or replaced by ether or methoxyl. Changes were also observed in the region 5·6 to 6·0 μ. Examination of the spectra of 2-diethylaminoethyl gentisate hydrochloride, 2-diethylaminoethyl 5-methoxysalicylate hydrochloride and the corresponding 3-diethylaminopropyl compounds showed the presence of 1 carbonyl group, indicating the esterification of the carboxyl group of gentisic acid. 2-Diethylaminoethyl 5-acetylgentisate hydrochloride, 2-diethylaminoethyl 5-benzoylgentisate hydrochloride and the 3-diethylaminopropyl analogues appeared to contain 2 carbonyl groups, suggesting the esterification of the carboxyl and 5-hydroxyl groups. Reaction between gentisic acid and the acid chlorides of phenylacetic, anisic, *p*-nitrobenzoic and succinic acids in the presence of alkali produced compounds with 2 carbonyl groups, esterification having occurred at the 5-hydroxyl group of gentisic acid. An exception was observed in the product of the reaction between gentisic acid and acetylsalicylic acid chloride, which appeared to have only 1 hydroxyl group. The constitution of this compound is doubtful. G. B.

Gentisic Acid, Preparation and Analyses of New Esters of. J. F. Nash, F. W. Bope and B. V. Christensen. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 207.) 8 esters of gentisic acid were prepared by reaction with the chlorides of 2-diethylaminoethanol and 3-diethylamino-1-propanol. The best yield was generally obtained by dissolving the acid in *iso*propanol, adding the chloralkamine and heating under a reflux condenser for several hours. A dry method consisted of mixing the chloralkamine with an ethanolic solution of the carboxylic acid, removing the ethanol under reduced pressure and heating the residue at 90° to 100° C. for 10 hours. 5 esters of gentisic acid were prepared by the Schotten-Baumann reaction involving the 5-hydroxyl group of gentisic acid with phenylacetic, *p*-nitrobenzoic and *o*- and *p*-methoxybenzoic acid chlorides or the mono-gentisic ester of succinic acid chloride in 10 per cent. sodium hydroxide solution. Melting points, molecular weights and elementary analyses are given for these compounds. G. B.

BIOCHEMISTRY

GENERAL BIOCHEMISTRY

Bacitracin A, Nature of. G. G. F. Newton and E. P. Abraham. (*Biochem. J.*, 1953, **53**, 604.) Bacitracin A has been purified by counter-current distribution between solvents and shown to be a polypeptide, the minimum stoichiometric unit of which has a molecular weight of about 1500. The amino-acids cysteine 1, ornithine 1, lysine 1, histidine 1, aspartic acid 2, glutamic acid 1, phenylalanine 1, *iso*leucine 2, and leucine 1, have been identified. The minimum molecular weight of 1500 for each stoichiometric unit of bacitracin A hydrochloride follows from the weights of the amino-acid residues, if allowance is made for 3 molecules of hydrochloric acid; the figure is confirmed by the amide nitrogen liberated on acid hydrolysis and the spans of the titration curve. The latter indicate that the peptide contains groups which ionise in 3 distinct ranges of hydrogen ion concentration attributable to the presence of two carboxyl groups, the glyoxaline ring of histidine and a free α-amino group, and either the δ-amino group of ornithine or the ϵ-amino group of lysine. Reaction of bacitracin A with 1-fluoro-2:4-dinitrobenzene and hydrolysis of the dinitrophenyl bacitracin gave products which indicated that the peptide contains a free α-amino group belonging to a leucine or an *iso*leucine residue, a free NH in the glyoxaline ring of a histidine residue and a free δ-amino group belonging to ornithine. The ϵ-amino group of lysine is not free, and it is suggested that bacitracin A may be a cyclic polypeptide in which a carboxyl group is condensed with the ϵ-amino group of lysine. Bacitracin A is rapidly inactivated by 0·5N hydrochloric acid at 100° C., a reaction which is paralleled by the liberation of a free thiol group. This reaction is also accompanied by hydrolysis of an amide group and the disappearance of an absorption maximum at 252 mμ. Bacitracin A is similarly inactivated by treatment with 0·1N sodium hydroxide at 37° C. with liberation of 1 equivalent of ammonia. The potential thiol group is unaffected by this treatment, and there is little change in the ultra-violet absorption spectrum. Hydrogenolysis with Raney nickel converts the cysteine residue to an alanine residue with a free α-amino group. Hydrolysis of the product of hydrogenolysis yields two new substances which give a blue colour with ninhydrin; one of these substances appears to be an amino-alcohol. The possible presence of a thiazoline ring system in bacitracin A is discussed in the light of the experimental evidence; the latter is not conclusive. J. B. S.

Corticotrophin and Corticotrophin-B, Preparation of Potent Concentrates of. A. W. Bazemore, J. W. Richter, D. E. Ayer, J. Finnerty, N. G. Brink and K. Folkers. (*J. Amer. chem. Soc.*, 1953, **75**, 1949.) The investigations were carried out on whole frozen hog pituitary glands or a commercial acid–acetone extract. Procedures are described for the preparation in good yields of corticotrophin and corticotrophin-B concentrates at activity levels of approximately 80 units/mg. The process applied to whole frozen hog pituitary glands consisted of acetone defatting, extraction with a methanol–acetic acid mixture, purification with oxycellulose and pepsin digestion. Commercial acid–acetone powder was processed by oxycellulose treatment and pepsin digestion to yield similarly purified concentrates of corticotrophin-B.

A. H. B.

Corticotrophin-B, Nature of. N. G. Brink, G. E. Boxer, V. C. Jelinek, F. A. Kuehl, Jr., J. W. Richter and K. Folkers. (*J. Amer. chem. Soc.*, 1953, **75**, 1960.) The final steps in the isolation of corticotrophin-B involved the use of ion exchange resins and then countercurrent distribution to yield material of 250 to 300 units/mg. of activity. This material was believed to be corticotrophin-B in essentially pure form. It appears to be a peptide of molecular weight in the range of 5000 to 7000. It contains 14 common amino-acids in amounts corresponding to a chain of some 60 amino-acid units. The amino-acids and ammonia isolated from the hydrolysis experiments accounted for 99·5 per cent. of the total nitrogen. The preponderance of basic amino-acids reflects the known basic nature of corticotrophin-B. If a prosthetic group occurs in corticotrophin-B it must be of low molecular weight and contain so little nitrogen and exhibit so little characteristic ultra-violet or infra-red absorption that these properties would be obscured by those of the gross peptide. The substance is highly active clinically in rheumatoid arthritis. It possesses adrenal weight-increasing activity and causes melanophore expansion in frog skins. A. H. B.

Corticotrophin-B, Purification of, by Ion Exchange Techniques. J. W. Richter, D. E. Ayer, A. W. Bazemore, N. G. Brink and K. Folkers. (*J. Amer. chem. Soc.*, 1953, **75**, 1952.) Procedures are described which gave a threefold enhancement of the activity of corticotrophin-B concentrates. The active material of these concentrates was allowed to undergo exchange with a sodium-buffered column of Amberlite IRC–50. A substantial amount of inactive proteinaceous material was removed from the column by washing with aqueous pyridine, and other proteins, pyridine and sodium ions then eluted with aqueous acetic acid. The active principle was then removed from the resin with dilute hydrochloric acid, and recovered as a solid hydrochloride free from inorganic salts. It possessed an activity of 250 to 300 units/mg. It was also shown that fractionation on columns of oxycellulose led to considerable purification. The presence of a reducing agent such as sulphite or hydrogen sulphide during the above processes inhibited inactivation and made possible better separations and more highly active products. A. H. B.

Esterified Fatty Acids and Total Fatty Acids in Blood, Determination of. I. Stern and B. Shapiro. (*J. clin. Path.*, 1953, **6**, 158.) Higher fatty acid esters react at room temperature with hydroxylamine in alkaline solutions in aqueous ethanol. This reaction was examined to establish optimal conditions for an analytical procedure. The procedure adopted consisted of adding serum (0·1 to 0·3 ml.) containing 2 to 5 mEq. of fatty acid esters to 8 ml. of an ethanol–ether mixture. The mixture is brought to the boil, cooled, made up to 10 ml., filtered and 3 ml. of the filtrate measured into a 16 mm. test tube. A blank containing 3 ml. of the ethanol–ether mixture is included in every run. Then 0·5 ml. of a 2M hydroxylamine solution and 0·5 ml. of a 3·5N sodium hydroxide solution are added and mixed, and the tubes stoppered and allowed to stand for 20 minutes at room temperature. After this period, 0·6 ml. of hydrochloric acid solution is added and, after mixing, 0·5 ml. of a 0·37M ferric chloride solution dissolved in 0·1N hydrochloric acid solution is introduced. The tubes are mixed again and the colour developed is measured in a Fisher electrophotometer with an F.525^{B} filter using micro test tubes. This hydroxamic method gives results comparable with those of a gravimetric macro-method,

in which the fatty acids are extracted from 3 ml. of serum and are weighed after saponification and acidification. The method gives a high reproducibility with a standard deviation of ±4 per cent. A. H. B.

BIOCHEMICAL ANALYSIS

Barbiturates in Urine; Paper Chromatographic Identification of. L.-G. Allgén. (*Svensk. farm. Tidskr.*, 1953, **57**, 188.) Extract 50 ml. of urine with 100 ml. of redistilled chloroform; filter, and evaporate. Dissolve the residue in 0·1 to 1·0 ml. of chloroform and apply 5 μl. diluted once, twice and 4 times on a large filter paper on a line 5 cm. from 1 side as 3 spots at a distance of 3 or 4 cm. Standards consisting of a number of barbiturates are also applied along the line, one of the standards being applied alone and also mixed with urine extract (5 μl. diluted twice). The paper is shaped to a cylinder by connecting the two sides with glass needles, and development carried out as ascending chromatography in large glass cylinders with ground tops covered with glass plates, the joints being made airtight with grease. 2 or 3 chromatograms are developed, using different solvents, the following being the most suitable: *iso*amyl alcohol or *n*-hexyl alcohol saturated with concentrated ammonium hydroxide solution in saturated ammonia atmosphere, or butanol saturated with water. The solvents are poured on the bottom of the glass cylinders and the saturated ammonia atmosphere produced by placing a small beaker with concentrated ammonium hydroxide solution in the centre of the bottom, this being renewed 1 hour before each new chromatogram is started. In the first two solvents mentioned separation is obtained of the various barbiturates tested, giving R_F values between 0·2 and 0·8, whereas in butanol-water the barbiturates give R_F values of the order of 0·9 at room temperature. Butanol-water is used to distinguish barbiturates from other ultra-violet absorbing substances and from metabolic breakdown products of barbiturates. The barbiturate spots are identified on the paper by illumination with an ultra-violet lamp of maximum intensity (250 mμ). After the paper has been treated with a saturated ammonia atmosphere for a few minutes the dark barbiturate spots are easily recognised when illuminated with ultra-violet light, and may be marked with a pencil. S. L. W.

7-Dehydrocholesterol, 7-Hydroxycholesterol and Bile Acids in Serum, Estimation of. A. E. Sobel, M. Goldberg and S. R. Slater. (*Analyt. Chem.*, 1953, **25**, 629.) Methods were developed for the determination of the two steroids related to cholesterol, 7-dehydrocholesterol and 7-hydroxycholesterol, and of bile acids in serum. Preliminary experiments indicated that the 3 sterols were not present in equal amounts, cholesterol concentrations being 1000 times greater than concentrations of either the provitamin or hydroxysterol. Digitonin precipitation experiments showed that cholesterol and 7-dehydrocholesterol were readily precipitated with small amounts of water, but the hydroxysterol was quantitatively recovered as the digitonide with 54 per cent. of water and with a large excess of digitonin. The digitonides were split with pyridine, specific colour reactions being applied to the petroleum ether-extracted sterol mixture for the evaluation of individual concentrations. 7-Dehydrocholesterol was determined by ultra-violet absorption at 281·5 mμ and by the Rosenheim-Callow reaction; 7-hydroxycholesterol was determined by reaction with activated glycerol dichlorohydrin while total sterols were measured by the Liebermann-Burchard reaction. A modified fluorimetric serum bile acid procedure is given which requires 0·2 ml. of serum; results are quoted for sterol determinations in six human subjects. R. E. S.

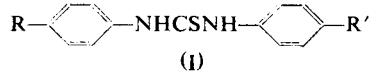
Œstrogens in Plasma, Determination of. A. H. Veldhuis. (*J. biol. Chem.*, 1953, **202**, 107.) A fluorimetric method for the determination of œstrogens in plasma is described. The method is based on the development of fluorescence when œstrogens are heated in the presence of sulphuric acid. It was found necessary to keep non-specific fluorescent materials in the reagents and solvent at a minimal concentration. The optimal time of heating the œstrone or œstradiol–sulphuric acid mixture was 4 minutes at 100° C. The œstrogens are determined in the unconjugated forms. Data on the analytical recoveries of œstrogen added to plasma are presented and the utility of the method for the determination of œstrogens in plasma demonstrated. A. H. B.

Steroid Hormones, Solubilised, Spectrophotometric Determination of. P. Ekwall, L. Sjöblom and J. Olsen. (*Acta chem. scand.*, 1953, 7, 347.) The spectrophotometric determination of α-œstradiol, desoxycorticosterone, testosterone, testosterone propionate and cortisone-21-hemisuccinate in aqueous solutions of sodium lauryl sulphate was investigated. The absorption curves of the hormones were similar to those recorded in ethanol, but a bathochromic shift occurred except in the case of œstradiol. The absorption at the maxima increased linearly with the hormone concentration. In the concentration range investigated, the solubilities of the hormones in sodium lauryl sulphate increased linearly with the colloid concentration. A direct spectrophotometric determination of the steroid hormones on these solutions is possible. A. H. B.

CHEMOTHERAPY

Acid Hydrazides, their Derivatives and Related Compounds, Synthesis of. H. L. Yale, K. Losee, J. Martins, M. Holsing, F. M. Perry and J. Bernstein. (*J. Amer. chem. Soc.*, 1953, **75**, 1933.) The preparation of a large number of aliphatic, aromatic and heterocyclic carboxylic acid hydrazides and their derivatives and related compounds is described. Because no outstanding antituberculous activity was found among the aliphatic, alicylic and aromatic carboxylic acid hydrazides the emphasis was placed on hydrazides of heterocyclic carboxylic acids. The majority of the compounds prepared were derivatives of *iso*nicotinyl hydrazide. A. H. B.

Thiocarbanilides as Antitubercular Compounds. C. F. Huebner, J. L. Marsh, R. H. Mizzoni, R. P. Mull, D. C. Schroeder, H. A. Troxell and C. R. Scholz. (*J. Amer. chem. Soc.*, 1953, **75**, 2274.) The high antituberculous activity of 4:4′-diethoxythiocarbanilide (I; $R=R'=OC_2H_5$) in mice infected with the H37RV strain prompted the synthesis and testing of over 300 thiocarbanilides and related substances. Rather specific structural features



(I)

necessary for activity were revealed. Shortening the 4-substituent to methoxy destroys activity, while lengthening the chain results in a fourfold increase to a maximum of activity in the neighbourhood of three to four carbon atoms. Increase beyond this causes activity to decline and then disappear. Replacement of alkoxy by an alkyl of equivalent length results in similar activity. Branching of the alkyl chain at the carbon adjoining the ring causes complete loss of activity. One of the 4-alkoxy groups may be replaced by halogen or dialkylamino and still retain some activity. Replacement of both of them

causes total loss of activity, while removal of one of the 4-alkoxyl groups also results in loss of activity. The 2- and 3-position isomers are inactive. A second substituent in the ring destroys activity as does substitution of methyl on the ureido nitrogen. The thiocarbanilide moiety is shown to be essential by the inactivity of the corresponding carbanilide, guanidine, guanylthiourea, dithiobiuret and the *cyclo*hexyl substituted thiourea. Some of the more active compounds gave favourable results in delayed and limited therapeutic trials in both mice and guinea-pigs. Resistant strains did not develop. The compounds have only a low toxicity. A. H. B.

Thioureas, Substituted, Antituberculous Activity of. R. L. Mayer, P. C. Eisman and E. A. Konopka. (*Proc. Soc. exp. Biol., N.Y.*, 1953, **82**, 769.) The authors have investigated the antimycobacterial and antifungal activities of more than 350 thiourea derivatives and related compounds. Many of the disubstituted thioureas, especially the thiocarbanilides, possessed *in vitro* activity and also had excellent chemotherapeutic effects in mice and guinea-pigs. The results obtained with 11 compounds of the general formula $R{\cdot}C_6H_4{\cdot}NHCSNH{\cdot}C_6H_4{\cdot}R'$ are reported. Mice were infected intravenously with 0·5 ml of a 1:10 dilution of a 7-day culture of H37Rv. Immediately after infection, groups of 10 mice were fed for 21 days on a diet containing from 0·01 to 0·1 per cent. of the test compound. After a further 15 days on a normal diet the surviving animals were killed. Guinea-pigs were infected subcutaneously with 1 ml. of a 1:200 dilution of the culture. After 21 days, the tuberculin-positive animals were given the medicated diet and this was continued for 115 days, when the surviving animals were killed. With some of the compounds examined there was a parallelism between antifungal and antimycobacterial activity suggesting a biochemical relation between the two forms of activity which could be useful in the search for new antituberculosis agents. There was no correlation between *in vitro* and *in vivo* tuberculostatic activity. *In vitro* resistance developed only slowly; no resistance developed *in vivo*. Streptomycin-resistant strains of H37Rv were sensitive to the thiocarbanilides, and isoniazid-resistant strains were sensitive to the thiourea derivatives. Certain of the compounds are suggested for clinical trial. H. T. B.

PHARMACY

GALENICAL PHARMACY

Vitamin B_{12}-Folic Acid Parenteral Solutions, Stability of. A. Taub and H. Lieberman. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 183.) Cyanocobalamin in aqueous solution has optimum stability at *p*H 4 to 6·5, but under these conditions folic acid is not sufficiently soluble to provide a suitable combined injection solution. A number of solubilisers were examined and citrate buffer, gluconic acid, glutamic acid, sodium benzoate, sodium gentisate, ethylenediamine, triethanolamine, urea, lysine, polyoxyethylene 20, sorbitan monooleate and propylene glycol were not satisfactory, but aminoacetic acid, nicotinamide, methylglucamine and glucose could be used. It was observed that folic acid could be brought into solution in a concentration of 5 mg./ml. by dissolving in water containing sodium hydroxide (*p*H 8·5) and adjusting to *p*H 6·0 with dilute hydrochloric acid, clouding of the solution being prevented by the addition of 10 per cent. of nicotinamide or aminoacetic acid. The latter caused discoloration after several weeks' storage. Solutions containing 30 μg./ml. of cyanocobalamin and 5 mg./ml. of folic acid were prepared with

glucose or methylglucamine at *p*H 6·5, but only nicotinamide prevented cloudiness developing at *p*H 6·0. These solutions showed no material loss of folic acid on storage for 3 weeks at 45° C. or 6 months at room temperature. It is concluded that solutions containing up to 30 μg./ml. of cyanocobalamin and 5 mg./ml. of folic acid with 10 per cent. of nicotinamide, at *p*H 6·0 to 6·5 remain clear and stable sufficiently long for practical purposes. G. B.

NOTES AND FORMULÆ

Chloramphenicol, Diffusion of, from Ointments. C. Trolle-Lassen. (*Arch. Pharm. Chemi*, 1953, **59**, 243.) The rate of diffusion of chloramphenicol from a number of ointment bases was tested by two methods. In the first the preparation was drawn up into a glass tube, the end of which was then closed by a film of cellophane. This tube was suspended in a test tube so that the cellophane-covered end was just below the surface of water in the test tube. After a certain period the concentration of chloramphenicol in the water was determined from the optical density of the aqueous liquid at 278 mμ. Plotting the amount of chloramphenicol in the solution against the square root of the time gave a straight line, the slope of which (α) was a measure of the diffusion. In the second method subcutaneous depots of the ointment were formed in the shoulder region of mice and, after 2 hours, the mice were killed and the concentration of chloramphenicol in the blood determined microbiologically. The results obtained were as follows:

Base	Results of tests	
	in vitro α	*in vivo* chloramphenicol μg./ml. of blood
Tragacanth gel	573	346
Oil-in-water emulsion	157	227
Water-in-oil emulsion	24·4	(14)
Soft paraffin base	0·313	88
Hydrophilic base	26·8	162

For the 3 preparations with an external aqueous phase the results of both methods give the same order of efficiency, and it would appear that the *in vitro* method represents sufficiently closely the conditions *in vivo*. For the water-in-oil emulsion and the soft paraffin base the order of efficiency as determined by the two methods is different. G. M.

Chlormerodrin (Neohydrin). (*New and Nonofficial Remedies; J. Amer. med. Ass.*, 1953, **152**, 331.) Chlormerodrin is [3-(chloromercuri)-2-methoxypropyl]urea, $ClHg{\cdot}CH_2{\cdot}CH(OCH_3){\cdot}CH_2{\cdot}NH{\cdot}CO{\cdot}NH_2$, and occurs as a stable, white, odourless powder, with a bitter, metallic taste, m.pt. 145·0° to 155·0° C., very soluble in sodium hydroxide solution, very slightly soluble in chloroform, and soluble in about 180 parts of ethanol, about 90 parts of methanol, and about 90 parts of water (a 0·5 per cent. solution has *p*H 4·3 to 5·0). It yields no immediate precipitate or colour when dissolved in alkali and treated with sodium sulphide (absence of ionisable mercury and other heavy metals). It also yields not more than 0·3 per cent. of sulphated ash and loses not more than 1·0 per cent. in weight when dried at 105° C. for 5 hours. The content of mercury, determined gravimetrically by precipitation with hydrogen sulphide in acid solution, is 53·5 to 55·7 per cent., equivalent to 98·0 to 102·0 per cent. of chlormerodrin. Chlormerodrin is administered by mouth as a mercurial diuretic. G. R. K.

PHARMACOGNOSY

Betulinic Acid in *Menyanthes trifoliata* L. A. Stabursvik. (*Acta chem. scand.*, 1953, **7**, 446.) The details of the isolation of betulinic acid from fresh rhizomes of *Menyanthes trifoliata* are recorded. A table is given showing that the acid has rather a scattered distribution throughout the plant kingdom and does not seem to be characteristic of certain families. A. H. B.

***Digitalis purpurea*, Paper Chromatography of.** K. B. Jensen. (*Acta pharm. tox. kbh.*, 1953, **9**, 99.) The paper describes the increase of separation effected by combining the previously used liquid system (chloroform–methanol–water) and the system chloroform–benzene–formamide. Details of the apparatus and technique for the one dimensional paper chromatography are given. Spot tests for the detection of the glycosides and aglycones are described which utilise the production of fluorescent and coloured spots by heating with trichloroacetic acid, trichloroacetic acid–chloramine and antimony trichloride. The Baljet reaction with alkaline picric acid, the Legal reaction with alkaline sodium nitroprusside and the Raymond reaction with alkaline *m*-dinitrobenzene were modified for chromatographic use on paper. The acid reagents gave both colour and fluorescence so that the A series and B series of substances (purpureaglycoside A, digitoxin, digitoxigenin—and purpureaglycoside B, gitoxin, gitoxigenin) and glycosides and aglycones could be distinguished. Diagrams of the chromatograms are given together with a description of the colours produced by the reagents and their sensitivity. R. E. S.

PHARMACOLOGY AND THERAPEUTICS

Aminosalicylic Acid, Effect of, on Thyroid and Adrenal. T. Wong, J. R. Hogness and R. H. Williams. (*Proc. Soc. exp. Biol., N.Y.*, 1953, **82**, 598.) The authors have investigated the effect of prolonged administration of aminosalicylic acid on thyroid and adrenal function in rats. 4 groups each of 10 rats were treated respectively with 1 per cent. of aminosalicylic acid in the diet for 2 weeks, stock diet alone, stock diet with 0·2 per cent. of propylthiouracil, and stock diet plus 1 mg. daily of corticotrophin gel (Armour) subcutaneously. After the 2-week period, a tracer dose of ^{131}I was injected intraperitoneally into each animal and food was removed. After 4 hours, blood was removed from the aorta, the thyroid glands were weighed and their radioactive iodine content determined. For the adrenal function investigation the following were determined—weight of gland, its ascorbic acid content and cholesterol content, thymic weight, liver glycogen, and circulating eosinophils. Aminosalicylic acid exerted a marked goitrogenic effect though not as great as the propylthiouracil in the quantities taken. The thyroid glands of the animals receiving aminosalicylic acid were about twice the weight of the controls, while those of the propylthiouracil treated group were about 3 times the weight of the controls. Corticotrophin had no significant effect on the gland weight. The total radioactive iodine content of the glands of the aminosalicylic acid group was only 40 per cent. of that of the controls, and there was no significant difference in this respect in the groups treated with aminosalicylic acid and with propylthiouracil. No differences in the adrenals of the 4 groups were noted, but the group treated with aminosalicylic acid showed a raised liver glycogen concentration, possibly due to a direct action of the compound on the liver. It is

suggested that aminosalicylic acid, like propylthiouracil, inhibits iodine concentration by the gland, thus lowering the production of thyroxine and stimulating the secretion of thyrotropic hormone by the anterior pituitary. This in turn results in thyroid stimulation and enlargement. H. T. B.

Ascorbic Acid, Glucose-1-phosphate and Lysergic Acid Diethylamide in Rheumatoid Arthritis. R. R. H. Lovell, J. A. Osborne, H. C. Goodman and B. Hudson. (*Lancet*, 1953, **264**, 970.) Various factors are known to influence experimental tuberculin hypersensitivity and on the basis of a possible analogy between tuberculin hypersensitivity in man and the tissue reaction in rheumatoid arthritis, the effects of some of these factors were investigated in the latter disease. On the basic analogy, ascorbic acid deficiency would enhance the inflammation and diminish the responsiveness of the disease to hormones. Sensitivity to tuberculosis in guinea-pigs is diminished by α-D-glucose-1-phosphate and lysergic acid diethylamide and the effect of these substances in man was investigated in regard to both rheumatoid arthritis and tuberculin sensitivity. Lysergic acid diethylamide has been reported to cause transient mental changes and disturbances in the autonomic nervous system in doses as small as 0·02 mg. The investigation was carried out on 10 patients with rheumatoid arthritis and 1 with polyarteritis nodosa. All were treated by rest, controlled exercises, splinting if necessary and salicylates or analgesics. Ascorbic acid was given orally. Patients treated with glucose-1-phosphate and lysergic acid diethylamide received inert preparations for up to 14 days prior to treatment. Cortisone, 200 mg. daily, was given orally in 1 case and intramuscularly in 1 case. Adrenocorticotrophic hormone, 100 mg. daily, was given by intramuscular injection. Glucose-1-phosphate was given intravenously as a solution of the dipotassium salt in normal saline in doses of 4 to 800 mg. daily. Lysergic acid diethylamide was given orally in doses of from 0·005 to 0·1 mg. daily in about 30 ml. of water before breakfast. The disease was not influenced by ascorbic acid depletion or saturation, nor did these factors influence the effect of cortisone or adrenocorticotrophic hormone. The other substances also had no effect on rheumatoid arthritis nor on the tuberculin reaction in man. It is suggested that the action of cortisone on the tuberculin reaction in man reflects, at least in part, a non-specific diminution of an inflammatory response rather than an interruption of the mechanism which initiates a hypersensitivity reaction. H. T. B.

Butylscopolammonium, Curarising Action of. E. Philippot and M. J. Dallemagne. (*Arch. int. Pharmacodyn.*, 1953, **93**, 337.) Butylscopolammonium bromide (buscopan) is a curarising agent similar in action to tubocurarine. Injected in doses between 5 and 10 mg./kg. it inhibits neuro-muscular transmission at the level of the limbs in the cat. The action is as marked for the tibial as for the soleus, and is preceeded by a transitory potentiation especially in the latter muscle. The injection of adrenaline or neostigmine frees the neuromuscular junction. The action of butylscopolammonium bromide is antagonised by decamethonium iodide, which removes the muscle block. If, subsequently the block is restored with decamethonium bromide, it can be relieved by a further injection of butylscopolammonium bromide. G. B.

Cortisone and Desoxycorticosterone (Deoxycortone), Effects of, on the Toxicity of Barbiturates. C. K. Gorby, C. A. Leonard, J. L. Ambrus and W. E. Harrisson. (*J. Amer. pharm. Ass. Sci. Ed.*, 1953, **42**, 213.) Groups of mice were treated with 100 mg./kg./day of cortisone acetate or deoxycortone

acetate administered subcutaneously for 6 days. Pentobarbitone sodium or phenobarbitone sodium was then given intraperitoneally. Cortisone exerted a slight protective effect when doses of pentobarbitone sodium below the LD50 were given, and deoxycortone slightly increased the effect of doses above the LD50. Both substances increased the toxic effect of phenobarbitone sodium, and this was also observed when the hormones were injected immediately after the phenobarbitone sodium. G. B.

***NN'*-Dibenzylethylenediamine Penicillin: Oral Use in Children.** I. A. B. Cathie and J. C. W. MacFarlane. (*Brit. med. J.*, 1953, **1**, 805.) A total of 101 children of all ages were given 300,000 units of *NN'*-dibenzylethylenediamine penicillin (penidural) by mouth and their blood levels estimated 3 to 4 hours after administration for reliability of absorption. In addition a series of 17 adult volunteers were investigated with the same preparation. Reliable absorption was shown in all of the 118 cases. In several cases hourly penicillin levels were estimated and a cumulative effect was seen following a second oral dose. In the treatment of acute infections, blood levels during the first 6 hours with this oral penicillin alone are inadequate and treatment should be augmented by a single intramuscular injection of a large amount of crystalline penicillin. A rational scheme of treatment in acute infection would appear to be an initial intramuscular injection of 600,000 units of penidural and 100,000 units of crystalline penicillin, with 300,000 units of penidural orally, the treatment continuing thereafter with oral penidural alone. S. L. W.

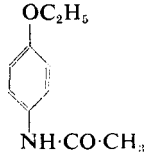
Hydralazine in the Treatment of Hypertension. J. H. Moyer. (*Arch. intern. Med.*, 1953, **91**, 419.) The results are recorded of treating hypertension for periods of 1 to 2 years with hydrallazine (1-hydrazinophthalazine, apresoline) alone and with hydrallazine either before or after ganglionic blockade with hexamathonium. 54 patients received hydrallazine alone in progressively increasing doses. Initially 25 mg. was given orally with each meal and at bedtime. After a few days the amount of drug being taken was increased until a significant reduction in blood pressure occurred or side reactions prohibited further increase in the dose. Patients treated with hydrallazine and hexamethonium included 20 who had previously received hexamethonium alone without adequate regulation of blood pressure and 32 who had previously received hydrallazine alone, but had failed to obtain significant reduction in blood pressure without prohibitive side reactions. Although 35 per cent. of the patients on hydrallazine alone obtained a significant reduction in blood pressure after 3 months' treatment, only 9 per cent. continued to obtain adequate control after treatment for 1 to 2 years. Of 20 patients in whom treatment with hexamethonium alone was a failure and 32 in whom treatment with hydrallazine alone was a failure, treatment with the two drugs combined gave satisfactory results in 75 per cent. Patients with malignant hypertension greatly improved on combined therapy. Hexamethonium is by far the more potent hypertensive agent, but hydrallazine minimises wide fluctuations in blood pressure. The side effects which most often necessitated discontinuing hydrallazine were tachycardia and palpitations or gastrointestinal disturbances. Headache was common but was usually relieved spontaneously after several weeks. Except for paræsthesias and hyperæsthesias the side effects decreased in intensity with time and some degree of tolerance with loss of therapeutic response also occurred. H. T. B.

Levorphan, Dextrorphan, Racemorphan (Dromoran), Absorption and Excretion of. A. L. Fisher and J. P. Long. (*J. Pharmacol.*, 1953, **107**, 241.) Although these drugs have a close similarity of structure to morphine and have similar analgesic properties, the excretion patterns are shown to be only qualitatively similar to those of morphine. A comparison of excretion by non-tolerant and tolerant dogs indicates similar excretions. The highest percentage of excreted conjugate by normal dogs was found to be about the average excretion of conjugate in tolerant dogs. With morphine, tolerant dogs excreted less conjugate than normal dogs, while excretion of unconjugated morphine remained about the same. The excretions of levorphan and dextrorphan, the *l*- and *d*-isomers, and of a mixture of the two were found to be similar to the excretion of racemorphan hydrobromide, the racemic compound. Following intravenous infusion, the drug was found to disappear very rapidly from the plasma, only small amounts being found even 1 minute following the injection. The rate of absorption from the gastrointestinal tract of fasting cats was found to be very rapid when the concentration was high, absorption being about 75 per cent. complete after 2 hours. It is partially excreted as the glucuronide and possibly in some other conjugated form. A method for the determination in plasma, urine and gastrointestinal contents, using a modification of the methyl orange technique for basic amines, is described. S. L. W.

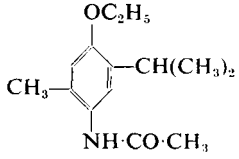
Levorphan Tartrate for Relief of Postoperative Pain. R. D. Hunt and F. F. Foldes. (*New Engl. J. Med.*, 1953, **248**, 803.) Animal experiments and observations on patients addicted to drugs have shown that dextrorphan (*d*-dromoran, 3-hydroxy-*N*-methylmorphinan) has no morphine-like effects, and that the analgesic potency of the racemic form resides wholly in the *l*-isomer. An investigation was therefore undertaken to determine whether levorphan could be substituted for the racemic form of the drug (racemorphan) in clinical medicine. In a series of 311 surgical patients a dose of 3 mg. of levorphan tartrate was administered for the relief of postoperative pain, and complete relief was obtained in 77·2 per cent. of cases. The average duration of analgesia was 5½ hours. The results are compared with those obtained from a similar group of surgical patients who received the theoretical equivalent of racemorphan hydrobromide (5 mg.). A higher percentage of patients derived complete analgesia from the first dose of 3 mg. of levorphan than from the first dose of 5 mg. of racemorphan, but results from subsequent doses were approximately equal. The incidence of side effects was no greater than with the racemic compound, and clinically the two drugs would appear to be interchangeable. 3 mg. of levorphan tartrate compared favourably with 10 mg. of morphine sulphate in analgesic potency. H. T. B.

Narcotics and Sedatives, Influence of on Blood Coagulation. D. I. Macht. (*Arch. int. Pharmacodyn.*, 1953, **93**, 325.) Drugs were administered to rabbits and the change in clotting time determined at intervals. Morphine, ethylmorphine and diamorphine produced a marked acceleration of clotting, whereas codeine, papaverine, dihydromorphinone and dihydrocodeinone did not. Freshly prepared solutions of cocaine had no effect on the clotting time, but solutions which had been allowed to stand for several days were found to shorten it. Ether, chloroform and a number of barbiturates did not affect the clotting time. Procaine, even in small doses markedly accelerated clotting. When diamorphine or procaine were given repeatedly or in large quantities, an entire change in the blood clotting mechanism took place and the blood remained fluid for several hours. *N*-allylnormorphine and pethidine showed a thromboplastic effect, but methadone did not. G. B.

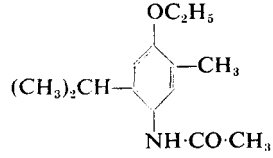
Phenacetin, Thymacetin and an Isomer, 4-Ethoxy-2-*iso*propyl-5-methylacetanilide, Comparative Study of. J. R. Lewis. (*Arch. int. Pharmacodyn.*, 1953, **93**, 450.) These chemically related compounds were examined for analgesic potency by intraperitoneal injection of 10 per cent. suspensions in gelatin solution into rats (radiant heat stimulus test) for sedative and antipyretic action in the rat, for the production of methæmoglobinæmia by oral administration in the cat, and for toxicity when administered to mice by stomach tube.



Phenacetin



Thymacetin



4-Ethoxy-2-*iso*propyl-5-methylacetanilide

Thymacetin was shown to have greater analgesic, antipyretic and sedative activity than phenacetin. It was less toxic and did not produce methæmoglobinæmia. 4-Ethoxy-2-*iso*propyl-5-methylacetanilide was considerably less active. In a limited clinical trial, thymacetin produced good analgesic effects, but there was a high incidence of side effects, mainly nausea. G. B.

Primidone—a New Anticonvulsant Drug. J. Yule Bogue and H. C. Carrington. (*Brit. J. Pharmacol.*, 1953, **8**, 230.) Primidone (mysoline, 5-ethyl-5-phenylhexahydropyrimidine-4:6-dione), a new anticonvulsant drug, chemically related to phenobarbitone, has been tested for anticonvulsant activity against electroshock and leptazol seizures in rats. Against electrically induced seizures an oral dose of 5 mg./kg. was more effective than the same dose of phenobarbitone, and its activity compared favourably with other convulsant drugs. Against leptazol-induced convulsions 20 mg./kg. protected 60 per cent. of the rats, compared with 10 mg./kg. of phenobarbitone. The acute and chronic toxicities in the rat, mouse and monkey were remarkably low; and it was much less toxic than phenobarbitone. G. F. S.

Propyliodone (*n*-Propyl-3:5-diiodo-4-pyridone-*N*-acetate). E. G. Tomich, B. Basil and B. Davis. (*Brit. J. Pharmacol.*, 1953, **8**, 166.) The effects of aqueous and oily suspensions of this compound, a bronchographic agent with a rapid lung clearance, have been studied in the lungs of rabbits. Intratracheal injections of aqueous propyliodone, its aqueous vehicle (consisting of a mixture of sodium carboxymethylcellulose), and an oily suspension, caused congestion of the lungs maximum at 3 days and absent after 2 weeks. Bronchographic studies showed the absence of alveolar filling and the disappearance of the contrast media within 3 days, making serial bronchography a possibility. With iodised poppy-seed oil marked congestion developed after 16 hours and persisted for 1 week when opaque oil was still present. Metabolic studies with propyliodone labelled with ^{131}I in man have shown the compound to be completely hydrolysed and eliminated as an iodine metabolite 3: 5-diiodo-4-pyridone-*N*-acetate ion. 50 per cent. of the iodine was eliminated after bronchography in 72 hours. G. F. S.

Pyrimethamine for Prophylaxis Against *Plasmodium falciparum*. G. Covell, P. G. Shute and M. Maryon. (*Brit. med. J.*, 1953, **1**, 1081.) An investigation was carried out to discover whether one weekly dose of 25 mg. of pyrimethamine is sufficient to protect non-immune subjects from an attack of falciparum malaria when repeatedly exposed to mosquito infection. Blood from an African child suffering from the disease was inoculated intramuscularly into a patient suffering from neurosyphilis, and sub-inoculations were made into two other neurosyphilitic patients. All developed overt malarial attacks but only one produced gametocytes in the peripheral blood in sufficient numbers to infect a batch of *Anopheles stephensi*. About 700 insects were allowed to feed on this gametocyte carrier. 14 patients, arranged in pairs, were each given a weekly dose of 25 mg. of pyrimethamine on different days, and subjected to infection by 10 to 12 of the mosquitoes at intervals of 1 to 7 days after the first dose of pyrimethamine had been given. Two of the patients suffered a rise in temperature, both due to irrelevant causes, otherwise there were no side effect and protection from malarial attack was complete. The authors consider that a weekly dose of 25 mg. of pyrimethamine is a practical and economical method of preventing malarial attack by the particular strain of *P. falciparum* used. H. T. B.

Sodium Salicylate in Rheumatic Fever. L. L. Henderson. (*Amer. J. med. Sci.*, 1953, **225**, 480.) A clinical investigation was conducted on rheumatic fever patients to ascertain the effect of adjuvant medication on the blood salicylate level. The patients were given 1·6 g. of enteric-coated sodium salicylate every 4 hours (a total of 10 g. a day) together with the adjuvant drug being studied. The adjuvant drugs employed, and the doses given with each dose of salicylate, were as follows:—sodium bicarbonate 1·6 g., sodium bicarbonate 0·65 g., magnesium trisilicate 1 g., aluminium hydroxide gel 8 to 16 ml. The respective average plasma salicylate levels (mg./100 ml.) obtained were 29·8, 35·9, 43·0 and 49·8; using sodium salicylate without any adjuvant the average figure was 42·2. While sodium bicarbonate depresses the plasma salicylate level it tends to return the carbon dioxide combining power, which is lowered by massive doses of salicylate, to normal. Magnesium trisilicate and magnesium hydroxide gel do not lower the plasma salicylate level, but neither do they raise the carbon dioxide combining power. Massive doses of salicylate seldom result in any harmful effect, but cause considerable discomfort. Most patients will tolerate the somewhat smaller doses of sodium salicylate sufficient to relieve rheumatic discomfort without any adjuvant medication. For those complaining of gastric distress magnesium trisilicate, aluminium hydroxide in tablet form or sodium bicarbonate in 0·65 g. doses will give satisfactory relief. S. L. W.

Succinic Acid and Choline, Response in Dogs to Relaxants derived from. L. W. Hall, H. Lehmann and E. Silk. (*Brit. med. J.*, 1953, **1**, 134.) Experiments in anæsthetised dogs have shown them to be more sensitive than man to the short-acting neuromuscular relaxants suxethonium and suxamethonium. Doses equivalent to one fifth of those used in man produced apnœa of about 5 times the duration. These findings could not be correlated with a low activity of pseudo-cholinesterase in dog plasma, which is the cause of an occasional prolonged effect in man. True cholinesterase in dog red cells was only one seventh of that in man, which suggests a correlation between higher sensitivity and lower true cholinesterase. Raising the pseudo-cholinesterase, by injection of a concentrated preparation (cholose), shortened the apnœa after both relaxants equivalent to that seen in man. G. F. S.