ABSTRACTS OF PAPERS PUBLISHED IN OTHER JOURNALS

CHEMISTRY

ALKALOIDS

Morphine, Isolation and Purification of. L. B. Achor and E. M. K. Geiling. (*Analyt. Chem.*, 1954, **26**, 1061.) A method is given for the isolation of morphine (and particularly ^{14}C labelled morphine) from the opium poppy which is satisfactory for the recovery of quantities of the order of 1 to 10 mg. The opium is extracted with aqueous sodium carbonate solution from which, after adjustment to *p*H 8·6, the morphine is removed by shaking with a 1-butanol-benzene mixture. After purification dilute sulphuric acid is used to remove the morphine from the organic phase, the aqueous solution being treated with barium hydroxide in potassium hydroxide before being passed down a column of Nalcite SAR. Elution of morphine from the column is accomplished with 0·1N hydrochloric acid. The Nalcite effluent containing morphine is adjusted to *p*H 7·0 with 10 per cent. sodium hydroxide and passed through a column of Amberlite IRC–50 buffered at *p*H 7·0 with 0·5M dihydrogen potassium phosphate-disodium monohydrogen phosphate buffer. The final effluent is evaporated to dryness over phosphorus pentoxide; the solid material remaining is morphine suitable for crystallisation. R. E. S.

ANALYTICAL

***Digitalis purpurea*, Chromatography of Glycosides and Aglycones from.** K. B. Jensen. (*Acta pharm. tox. Kbh.*, 1954, **10**, 69.) Details are given for the paper chromatographic separation and fluorimetric determination of the cardioactive glycosides and aglycones purpurea glycoside A, digitoxin, digitoxigenin, purpurea glycoside B, gitoxin, and gitoxigenin. Separation was effected by descending, one-dimensional chromatography on formamide-impregnated filter paper with chloroform or benzene-chloroform as the mobile phase. For the purpurea glycosides A and B the chromatograms were developed for 3 to 4 days with chloroform, for gitoxin and gitoxigenin for 3 to 5 hours with chloroform, and for digitoxin and digitoxigenin for 3 to 4 hours with benzene-chloroform (6:4). Localisation of the substances on the chromatogram was obtained by means of parallel chromatograms on known substances. Quantitative determinations were made by the simultaneous chromatography of known amounts of standard substances at different levels. The substances of the A series were located with trichloroacetic acid-chloramine-R and those of the B series with the same reagent or trichloroacetic acid-R (*Acta pharm. tox. Kbh.*, 1953, **9**, 99). The cut out paper strips with test substance were eluted directly with the fluorescence producing test solution. The fluorescence curves were linear for quantities ranging from 2 to 15 μg. of digitoxin and 1 to 10 μg. of gitoxin, or equimolecular amounts of the corresponding glycosides and aglycones, per 15 ml. of test solution. For the estimation of both glycosides the standard deviations varied from about 6 per cent. at the lowest concentrations to about 2 per cent. at the highest concentrations employed. R. E. S.

Diphenan and Urethane, Assay of. P. Ekeblad. (*Svensk. farm. Tidskr.*, 1954, **23**, 557.) Heating a carbamic ester with perchloric acid in acetic acid solution gives the corresponding ester of acetic acid, and ammonia. The method

may be applied to the assay of diphenan and urethane, and is simpler than the usual one of heating with sulphuric acid. For diphenan, one millimole (0·2273 g.) is dissolved in 10 ml. of anhydrous acetic acid neutralised with 0·1N perchloric acid in acetic acid, using Blue BZL as indicator, treated with a further 25·0 ml. of the standard acid, and heated on the water bath for 30 minutes. A silica gel tube is used to keep steam out of the mixture. After cooling and the addition of indicator, the solution is titrated with a standard 0·1N solution of trimethylamine in acetic acid. One ml. of acid corresponds to 0·02273 g. of diphenan. For urethane 60 minutes' heating with 3 equivalents of the acid is required. The addition of acetic anhydride to remove traces of water is not permissible before heating, but may be employed afterwards. G. M.

Penicillin, Determination of, by Hydroxylamine. P. Mørch. (*Dansk. Tidsskr. Farm.*, 1954, **28**, 157.) A detailed study was made of the Boxer and Ewerett (*Analyt. chem.*, 1949, **21**, 670) method of determination of penicillin, in which the penicillin is converted to a hydroxamic acid which is determined colorimetrically after the addition of ferric iron. The colour is due to a mono-complex having an absorption maximum at 490 mμ. No higher complex is formed with penicillin. Fading may result from reduction of the ferric salt by excess of hydroxylamine. In applying the method of Boxer and Ewerett to penicillin cultures, a preliminary extraction is necessary. About 10 ml. of the filtered liquid (10,000 to 30,000 units) is mixed with 20 ml. of water and 40 ml. of amyl acetate. After cooling to 0° to 5° C., 20 ml. of ice-cold glycine buffer (*p*H 1·1) is added and the mixture is shaken for 1 minute. The aqueous layer is removed, the amyl acetate solution is dried with sodium sulphate and filtered: 30 ml. of the filtrate is shaken with 5 ml. of 2 per cent. sodium bicarbonate solution and the aqueous phase is used for the colorimetric determination. The determination is repeated with a corresponding amount of culture to which has been added 15,000 to 25,000 units of sodium penicillin. In the case of penicillin ointments, the material is dissolved in chloroform and shaken out into 2 per cent. sodium bicarbonate solution. Another portion of the ointment is treated similarly, but shaken with bicarbonate solution containing a known amount of penicillin standard. Combination preparations may be treated as follows. Total penicillin is determined after hydrolysis of the penicillin esters by allowing to stand for 2 hours in 2 per cent. bicarbonate solution. Sodium penicillin + procaine penicillin are determined after removal of penicillin esters by shaking with amyl acetate at *p*H 2, the esters remaining in the aqueous phase. Finally, procaine penicillin is determined from the absorption of the procaine at 290 mμ. G. M.

Pyrethrum Extracts, Determination of Pyrethrins in. J. A. Cornelius. (*Analyst*, 1954, **79**, 458.) A quantitative chromatographic method is given for the separation of pyrethrum extracts in *n*-hexane. Alumina is used as the adsorbent and it is modified by exposure to a humid atmosphere or by drying until standardisation with Sudan yellow/Sudan red solution shows it to be of the required activity. The pyrethrins are eluted from the alumina with *n*-hexane containing 10 per cent. and 20 per cent. diethyl ether. The eluates are evaporated to dryness, dissolved in ethanol and the ultra-violet absorption determined at 224 mμ for the "pyrethrin-I" fraction and at 229 mμ for the "pyrethrin-II" fraction; empirical conversion factors are used based on parallel analyses of a pyrethrum extract by the current A.O.A.C. mercury reduction method and the present chromatographic method. For pyrethrum extracts in mineral oil, the solvent is first removed by distillation at a low pressure (less than 0·001 mm. of

mercury) not exceeding 40° C., the residue being dissolved in *n*-hexane. A table is given showing a comparison of results obtained by chromatography with those obtained by the A.O.A.C. mercury reduction method. R. E. S.

Vitamin B_1, Fluorimetric Determination of, by the Thiochrome Method. G. Pruner. (*Rendiconti Ist. Sup. Sanit.*, 1954, **17**, 129.) The method of Jansen is modified by reducing the amount of potassium ferricyanide and carrying out the oxidation of the vitamin B_1 to thiochrome at a higher temperature. To 1 ml. of solution containing 1 to 10 μg. of vitamin B_1 1 ml. of 0·01N hydrochloric acid is added, followed by 1 ml. of 3 per cent. potassium ferricyanide and 2 ml. of buffer solution [prepared by mixing: (1) anhydrous monopotassium phosphate 0·283 g., disodium phosphate dodecahydrate 1·665 g., distilled water to 100 ml., and (2) anhydrous sodium carbonate 0·857 g., water to 100 ml.]. The tube is covered with a Kjeldahl bulb and placed in a boiling water bath for 5 minutes; a standard solution containing a known amount of vitamin is treated in the same way. The tubes are removed and cooled for 10 minutes, and 8 ml. of *iso*butanol is added to each. The tubes are shaken for 2 minutes and the *iso*butanol removed by means of a pipette and dried over sodium sulphate. The fluorescence is then compared in a suitable instrument. The fluorescence is proportional to the vitamin content. For quantities of vitamin B_1 of 1 μg. or less, 1 mg. of potassium ferricyanide is used; 3 mg. of ferricyanide oxidises 5 μg. of vitamin B_1 even in the presence of 200 μg. of ascorbic acid, but larger quantities of ascorbic acid cause complete decolorisation at the heating stage and a reduction in the fluorescence intensity. E. H.

ESSENTIAL OILS

Essential Oils, Chromatography of. R. H. Reitsema. (*Analyt. Chem.*, 1954 **26**, 960.) Chromatoplates prepared by coating glass plates with silicic acid using starch as a binder were used for the rapid analysis of the constituents of essential oils. Following the application of 1 to 2 μg. of material, development was accomplished in a covered jar using 10 to 15 per cent. ethyl acetate in hexane as developing agent. The plates were first inspected under ultra-violet light and were then sprayed with an acidic solution of 2 : 4-dinitrophenylhydrazine and inspected under visible and ultra-violet light to detect ketones. After heating, the plate was again inspected under visible and ultra-violet light to detect heat- and acid-sensitive materials. Results are given of a chromatographic comparison of spearmint-type oils, and various compounds including common constituents of essential oils were run on chromato-plates. The R_F values of carvone varied widely but the relative positions of any two materials on a path were nearly constant, and a fairly consistent ratio of R_F values could be calculated.
R. E. S.

GLYCOSIDES, FERMENTS AND CARBOHYDRATES

Digitalis, Relation between Digitoxigenin and Gitoxigenin Glycosides in. F. H. L. van Os, C. H. Galenkamp and A. R. Kliphuis. (*Pharm. Weekbl.*, 1954, **89**, 429.) The method used for the assay of the relative proportions of the two groups of glycosides was that of Tattje (*J. Pharm. Pharmacol.*, 1954, **6**, 476.) In commercial Dutch samples of digitalis, 40 to 50 per cent. of the glycosides were of the gitoxigenin group. This explains the low yields of digitoxin obtained commercially, especially in view of the fact that of the "digitoxin" a large proportion is present as purpurea glycoside A. In French samples the glycosides contained about 20 per cent. of the B series. The difference in these results

appears to be due to hereditary factors, since wild plants from Belgium had the same composition as the Dutch ones but other plants, known to have been descended from specimens from the Botanical Gardens at Cambridge, resembled the French specimens. With regard to the method of assay, it is known that gitoxin is practically insoluble in both water and chloroform. The authors' experiments show, however, that even with high gitoxin content, the extraction with chloroform is practically complete. It is known that the solubility of gitoxin in water and in chloroform is influenced by the presence of digitoxin. Objections to the shaking out of gitoxin into chloroform must therefore be regarded as merely theoretical. G. M.

BIOCHEMISTRY

GENERAL BIOCHEMISTRY

Cholinesterase, Selective Inhibitors of. M. P. Fulton and G. A. Mogey. (*Brit. J. Pharmacol.*, 1954, **9**, 138.) A series of bis-quaternary ammonium salts is described which have in common a bis-[*p*-(trialkyl ammonium)-phenyl] structure joined by a four carbon-atom chain. Alterations of the molecule were confined to the second carbon atom of the chain and the quaternary nitrogens. The series had a highly selective reversible action on true cholinesterase, activity being measured by the Warburg method using acetylcholine as substrate for true cholinesterase activity and benzoylcholine for pseudocholinesterase activity. The compounds increased the twitch height of the isolated rat diaphragm indirectly stimulated and had an anticurare action, and in high doses a neuromuscular blocking action, on the same preparation. Adrenaline increased this blocking action. One of the compounds contracted the isolated frog rectus; the others had no effect other than as anticholinesterases. The effect on the blood pressure of the cat varied from compound to compound. Death with toxic doses in mice appeared to be due to respiratory failure. *d*-Tubocurarine, dibenamine, hexamethonium, nicotine tartrate or a combination of nicotine and atropine had no effect on the toxicity. G. P.

Cholinesterases of the Central Nervous System, Inhibition of. L. Austin and D. R. Davies. (*Brit. J. Pharmacol.*, 1954, **9**, 145.) To establish whether cholinesterase inhibition was responsible for chronic paralysis in chickens caused by some phosphorus-containing anticholinesterases, dyflos, sarin, tabun, soman and ethyl-sarin were administered, in doses within the lethal range, to chickens treated with atropine. Only dyflos produced any signs of chronic paralysis, although all five were powerful anticholinesterases. Determination, in the birds treated with dyflos and sarin, of both true- and pseudo-cholinesterase levels in the blood and central nervous system showed that with both drugs these levels fell soon after administration, but most recovered their original ranges by the time chronic paralysis set in. The exception was the pseudocholinesterases of the spinal cord after dyflos. Their level only returned to 50 to 75 per cent of the original and remained there. To repeat the state of cholinesterase inhibition found with dyflos, sarin was given in small repeated doses. Under these conditions no paralysis was obtained with sarin. Plasma pseudocholinesterase levels were markedly lowered by each of the inhibitors, but recovery was rapid and in all cases rose higher than normal. With dyflos this rise was particularly marked. Paralysis in chickens with dyflos therefore did not appear to be due to inactivation of the cholinesterases of the central nervous system or of the blood. G. P.

BIOCHEMICAL ANALYSIS

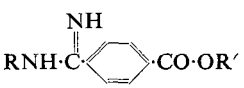
Adrenaline and Noradrenaline on Paper Chromatograms, Detection of. M. E. Pitkänen. (*Scand. J. clin. Lab. Invest.*, 1954, **6**, 78.) The sensitivity of the usual paper chromatographic detection of adrenaline and noradrenaline using either potassium ferricyanide or iodine solution as an oxidative agent can be considerably increased by subsequently spraying the paper with a solution of *p*-dimethylaminobenzaldehyde. This reduces the limit of sensitivity from 1 to 2 μg. of adrenaline or noradrenaline to 0·2 μg. of either. M. M.

Bilirubin, A New Tablet Test for. J. A. Tallack and S. Sherlock. (*Brit. med. J.*, 1954, **2**, 212.) A simple method is described for the detection of bilirubin in urine. Five drops of the urine is placed on a test mat composed of a mixture of asbestos and cellulose fibres. A tablet, containing a stable diazo dye (*p*-nitrobenzene diazonium *p*-toluene sulphonate), sulphosalicylic acid, sodium bicarbonate and boric acid, is placed on the centre of the mat. Two drops of water are allowed to flow on the tablet and the colour developing on the mat is recorded within 30 seconds. If bilirubin is present the mat around the tablet turns purple, and the amount of bilirubin is roughly proportional to the speed of development and intensity of the colour. Any colour developing after 30 seconds is ignored. The method is specific, as sensitive as the Fouchet test, and superior to the iodine test. No false positives were obtained with 100 urines from normal, non-jaundiced patients, receiving a variety of drug treatments. The presence of urobilinogen also gave no false positives. Between 0·1 and 0·15 mg. of bilirubin per 100 ml. can be detected. G. F. S.

Chloramphenicol Esters, Assay of. C. Trolle-Lassen. (*Arch. Pharm. Chemi.*, 1954, **61**, 435.) For the assay of chloramphenicol esters it is first necessary to hydrolyse them, and as chemical hydrolysis leads to destruction of the chloramphenicol, the author uses a commercial bacterial lipase (Lipase A. Rohm and Haas Co.). A study of the effect of different conditions led to the following method. A suitable quantity of the ester is dissolved in ethanol and diluted with buffer solution (*p*H 6) to give a suspension containing about 0·1 mg. of ester per ml. To this is added 1 mg. of the lipase per ml., and after vigorous shaking, the mixture is kept at 37° C. for 4 hours. The chloramphenicol is then assayed in this solution microbiologically in the usual way. G. M.

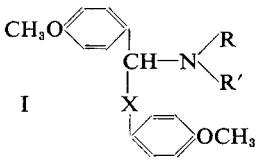
CHEMOTHERAPY

Amidines, Anaesthetic Activity of. O. Gisvold. (*J. Amer. pharm. Ass., Sci. Ed.*, 1954, **43**, 372.) The duration of anaesthesia caused by application of a 1 per cent. solution of several *p*-carboxybenzamidines to the eyes and skin of guinea-pigs was determined.



The compounds tested included R=H, methyl, ethyl, propyl and butyl,R′= 2-chloroethyl, benzyl, *cyclo*hexyl and *cyclo*pentyl. Some of the compounds showed a high anæsthetic activity, but in some cases irritation of the skin was reported. Most of the substances were bactericidal at a concentration of 0·1 per cent. against *Staphylococcus aureus* and *Salmonella typhosa*. G. B.

Antispasmodics, New Synthetic. J. Cymerman-Craig, K. V. Martin, P. C. Wailes, R. H. Thorp, R. Ladd and G. Thorburn. (*Nature, Lond.*, 1954, **174**, 231.) A series of *N*-alkyl-1:2-di-(*p*-methoxyphenyl)-ethylamines (I; X = CH_2), N-alkyl-α-aminodeoxyanisoins (I; X = CO), and *N*-alkyl-2-amino-1:2-di-(*p*-methoxyphenyl)-ethanols (I; X = CHOH) were prepared and, in view of the promising activity of certain 1-phenyl*iso*quinoline compounds, a

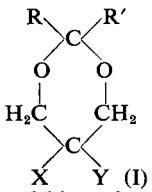


I

number of *N*-alkyl-di-(*p*-methoxy-phenyl)-methylamines (I; X = —) were also synthesised. All compounds exhibited spasmolytic activity when tested on isolated guinea-pig ileum against spasms produced by barium chloride and carbachol, using papaverine and atropine as standards. Some of the compounds were more active than papaverine. Relative neurotropic activity was between 0·005 and 0·2 of atropine, and acute toxicities (intravenous in mice) were uniformly between 60 and 80 mg./kg. for the compounds of the series. A. H. B.

Benzotriazines, Compounds having Antimalarial Activity. F. J. Wolf, K. Pfister, 3rd, R. M. Wilson, Jr. and C. A. Robinson. (*J. Amer. chem. Soc.*, 1954, **76**, 3551.) In an attempt to obtain compounds having the desirable therapeutic properties of sulphaquinoxaline, without the undesirable effect of forming a highly insoluble 3-hydroxy compound which may cause the formation of kidney stones when high levels of the drug are administered, the preparation of a nitrogen isostere, 3-sulphanilamido-1:2:4-benzotriazine was undertaken. The substitution of nitrogen for carbon eliminates the possibility of hydroxylation. The method of synthesis is described. A series of chloro substituted compounds was prepared because of the antimalarial properties exhibited in the intermediate bases. 7-Chloro-3-amino-1:2:4-benzotriazine-1-oxide and 7-chloro-3-amino-1:2:4-benzotriazine had excellent activity as suppressive agents in avian malaria. On a weight basis, these compounds are about 4 times as potent as quinine and 5 times as potent as sulphadiazine, or about equivalent to sulphaquinoxaline. Effectiveness was limited to the series having a halogen in the 7-position and maximum activity was obtained with an amino group in position 3. Activity could not be detected when the halogen was in other positions, or when other groups were substituted in the 7-position. Replacement of the amino group by hydroxyl, or acylation of the amino group, gives a great reduction in the activity. A. H. B.

1:3-Dioxanes, Basic, as Antispasmodics. F. F. Blicke and E. L. Schumann. (*J. Amer. chem. Soc.*, 1954, **76**, 3153.) A series of substituted 1 : 3-dioxanes of type I: where R = H and C_6H_5–; R′ = $-CH_2N(CH_3)_2$, $-CH_2N(CH_3)_3I$, and C_6H_5 ; X = CH_3, C_2H_5, C_6H_5, H, and CH_2OH ; Y = $CH_2N(CH_3)_2$, $-CH_2N(CH_3)_3I$, CH_2OH, C_6H_5, NH_2, $N(CH_3)_2$, $-CH_2N(C_2H_5)_2$, $-CH_2CH_2NH_2$ were prepared. The pharmacological activity is recorded against acetylcholine and barium chloride induced spasm of isolated rabbit jejunum and histamine induced spasm of isolated guinea-pig intestine. A. H. B.



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3-Indolecarboxaldehyde Thiosemicarbazone, a New Antitubercular Compound. L. E. Weller, H. M. Sell and R. Y. Gottshall. (*J. Amer. chem. Soc.*, 1954, **76**, 1959.) The synthesis of 3-indolecarboxaldehyde thiosemicarbazone is described. It has been shown to have high bacteriostatic activity *in vitro* and to suppress tuberculosis in mice after injection of virulent tubercle bacilli. A. H. B.

Local Anæsthetics Derived from *p*-Cymene. B. Samdahl, G. Gjerstad and E. Rydström. (*Ann. pharm. franç.*, 1954, **12**, 125.) Diethylaminoacetylcarvacrylamine, the *p*-cymene derivative analogous to lidocaine (lignocaine) was prepared by reaction of carvacrylamine with chloracetyl chloride and treatment of the product with diethylamine. The substance was anæsthetic to the tongue and rabbit cornea, but the hydrochloride, sulphate and nitrate were too acid for injection. Certain organic acid salts are being investigated, and the possibility of making a long-acting anæsthetic solution of the base in a mixture of propylene and polyethylene glycols and water is being examined. Carvacryloxyethanol and chlorcarvacryloxyethanol were obtained by reaction of glycol monohydrin with the corresponding phenolate. The substances, analogous to phenoxyethanol, were not sufficiently soluble in water to show antibacterial properties. A feeble local anæsthetic activity was observed in phenoxyethanol, carvacryloxyethanol and chlorcarvacryloxyethanol. The *p*-aminobenzoic esters were prepared with the object of intensifying this property, and the products may be useful as local anæsthetics for use in propylene/polyethylene glycol solution. G. B.

PHARMACY

NOTES AND FORMULÆ

Cetrimide and Acriflavine in Ointments, Bactericidal Action of. R. Frank and G. Stark. (*Pharm. Acta Helvet.*, 1954, **29**, 81.) The action of cetrimide and acriflavine was tested in a number of ointment bases. In water in oil emulsions the action of both of these compounds was poor. Oil in water emulsions (e.g., emulsifying ointment B.P.) gave much better results. The diffusion of acriflavine from pure soft paraffin was good, at least in fairly high concentrations. The best results were obtained with a glycerine ointment and with sodium alginate jelly. It was observed that there was no incompatibility between cetrimide and sodium alginate, in spite of opposite electrical charges. There is a marked incompatibility between acriflavine and cetrimide on the one hand and hydrous emulsifying ointment B.P. on the other hand. The two antiseptics in question have no synergistic action together. Optimum concentration of the antiseptics is between 1 and 1.5 per cent. G. M.

Steroids, Preparation of, in Microcrystalline Form by Freeze-Sublimation. A. P. Lemberger, T. Higuchi, L. W. Busse, J. V. Swintosky and D. E. Wurster. (*J. Amer. pharm. Ass., Sci. Ed.*, 1954, **43**, 338.) A solution of the steroid in chloroform or carbon tetrachloride was allowed to fall from a funnel into liquid air or liquid nitrogen, so that droplets were formed before they came into contact with the frozen liquid. The frozen solution was transferred to a chilled sample tube, which was then connected to a condensing vessel and vacuum pump. The sample tube and condenser were surrounded with liquid air and the apparatus evacuated. The cooling material around the sample tube was replaced by acetone/solid carbon dioxide mixture and sublimation allowed to proceed, about 24 hours being required to dry 50 ml. of the frozen solution,

The particle size of the resulting powder was determined by making measurements of the specific surface area, using the low temperature nitrogen absorption method. The process appeared to be well suited to the production of submicroscopical particles, a diameter as low as 0·25 μ being achieved under optimum conditions. Differences were observed with the various steroids and solvents employed, but generally an optimum concentration of solution could be found which would yield the smallest particles. Larger particles were produced when the temperature of the subliming sample approached the melting point of the solvent. The addition of ethanol, methanol or tween mixture increased the particle size. When caffeine or polyvinylpyrrolidone were introduced as protective agents, the powders were readily wetted and the steroid particles tended to aggregate. There was however no tendency towards aggregation with powders prepared with tween mixture and polyvinylpyrrolidone, and dispersions of almost colloidal characteristics could be obtained. G. B.

Water, Demineralisation of, by a Single Column. J. Büchi and M. Soliva. (*Pharm. Acta Helvet.*, 1954, **29**, 221.) A column of 60 cm. height and 5 cm. diameter was layered first with 250 ml. of cation exchange resin (Amberlite IR 120) upon which rested the widened end (2 cm. diameter) of a 50 cm. length of glass tubing of 1 cm. diameter. 500 ml. of anion exchange resin (Amberlite IRA 410) was then added around the tubing giving a column of resin 40 cm. in height. 300 ml. of 15 per cent. sodium hydroxide solution was run through the column which was afterwards rinsed with tap water to nearly neutral reaction. The cation exchange resin was then treated with 300 ml. of 10 per cent. hydrochloric acid run in through the glass tubing. After rinsing, the two beds of resin were mixed by bubbling air through the column. Water was run at the rate of about 12 to 15 l. per hour until the column was exhausted, as shown by the presence of chloride in the issuing fluid. For bacteriological reasons it is best to run the apparatus so that all the water required is produced in one operation. The quality of the product corresponds in every way to Swiss Pharmacopœial requirements and the authors consider that demineralised water should be permitted for all preparations except eye-drops and solutions for injection. G. M.

PHARMACOGNOSY

Alkaloid Production in *Datura*, Influence of Shoot and Root on. E. Steinegger. (*Pharm. Acta Helvet.*, 1954, **29**, 141.) The influence of root and shoot on alkaloidal production was followed by experiments in grafting the diploid *Datura tatula* with the tetraploid *Datura tatula* var. *inermis*. It was found that the 4n root produces a somewhat lower proportion of alkaloids as compared with the 2n root. On the other hand, a 4n shoot graft gives a considerably higher yield than a 2n graft. The highest alkaloidal production was obtained with a 4n shoot and 2n root, but calculated on a basis of alkaloid per sq. m. of soil area the highest yield is given by the wholly 4n plant. The increase in the size of the plant in the latter case is however undesirable in practice. The composition of the alkaloids was not influenced by the chromosome number. The results appear to indicate a balance between two opposing factors: a slightly increased production with the 2n root, and a decisive increase in the 4n shoot. Although it has been observed that the alkaloids are produced in the root of *Datura*, it is now shown that the shoot can influence the production, e.g., by influencing the root growth. G. M.

***Cinchona succirubra*, Biosynthesis of Alkaloids in.** P. de Moerloose. (*Pharm. Weekbl.*, 1954, **89**, 541.) Plants of *Cinchona succirubra* were grown under laboratory conditions in an atmosphere containing radio-active carbon dioxide, which after a varying time was replaced by ordinary carbon dioxide. The results show that the view that the alkaloids are formed in the leaf and transported to other parts of the plant is incorrect. Study of the time factor showed that synthesis occurs both in the leaf and in the bark. The leaf produces mostly quinine and quinidine, while cinchonine and cinchonidine are formed mainly in the bark. It does not appear probable that one of the alkaloids is a precursor of the others. Apparently all of the alkaloids are derived from a common precursor, the syntheses being independent of one another. Biosynthesis in the leaves is inhibited by illumination, but not in the bark. G. M.

***Digitalis purpurea* L. and *Digitalis lutea* L., Carbohydrate and Chlorophyll Content of Leaves of, after Freeze-drying and Oven-drying.** F. P. Cosgrove and E. P. Guth. (*J. Amer. pharm. Ass., Sci. Ed.*, 1954, **43**, 268.) A comparison was made between leaves dried at 50° C. and freeze-dried. An ethanolic extract was made, diluted with water and the ethanol removed on a water bath. Phenols, tannins, etc., were removed with lead acetate, the excess of which was precipitated with sodium carbonate. After hydrolysis with hydrochloric acid, followed by neutralisation, the product was analysed for sugars (as glucose) and for chlorophylls A and B by the A.O.A.C. method. No significant difference was found in sugar content between the freeze-dried and oven-dried leaves of *D. purpurea*, whereas oven-dried *D. lutea* contained appreciably more sugars than freeze-dried. No correlation was established between sugar content and total glycosides, as estimated by a colorimetric method; therefore it is unlikely that there is a greater yield of total glycosides from oven-dried leaves. A more likely explanation is that the heat renders the glycosides more soluble in ethanol. Freeze-drying appeared to preserve more chlorophyll A, but there was little difference in the yield of chlorophyll B after freeze- or oven-drying. G. B.

PHARMACOLOGY AND THERAPEUTICS

Adrenaline and Noradrenaline in Urine after Adrenalectomy. U. S. von Euler, C. Franksson and J. Hellström. (*Acta physiol. scand.*, 1954, **31**, 1.) The adrenaline and noradrenaline content of the urine of patients, suffering either from hypertension or from cancer of the prostate or mammary gland, was estimated both before and after either unilateral or bilateral adrenalectomy. Unilateral adrenalectomy did not significantly alter the output of adrenaline or noradrenaline but after bilateral removal the adrenaline output fell considerably while the noradrenaline output was either maintained or increased. These results indicate that most of the adrenaline excreted in the urine is derived from the adrenals while the noradrenaline comes from other sources, probably the adrenergic nerves. M. M.

***N*-Allylnormorphine, Effect of, on the Antidiuretic Action of Morphine.** C. Winter, C. Gaffney and L. Flataker. (*J. Pharmacol.*, 1954, **111**, 360.) *N*-Allylnormorphine (nalorphine) effectively blocks the antidiuretic effect of morphine in rats. The drug itself has neither a diuretic nor an antidiuretic action nor does it antagonise the antidiuretic effect of vasopressin. It is suggested that morphine stimulates the release of antidiuretic hormone from the

posterior lobe of the pituitary gland and that nalorphine blocks this action, the site of action being the hypothalamus. M. M.

Autonomic Ganglia, Relation of Chemical Structure to Action on. R. Wien. (*Arch. int. Pharmacodyn.*, 1954, **97**, 395.) Spatial configuration of the molecule appears to play a greater part in determining the ganglionic blocking activity of the bisquaternary ammonium series than does the interquaternary chain length. In the methonium series the maximum potency (blockade of the superior cervical ganglion) for chain lengths of 5 and 6 carbon atoms was obtained where the "onium" radical was dimethylethyl and for a chain length of 4, where the radical was diethylmethyl. In hexamethonium, replacement of the nitrogen by sulphur, or a primary amino group for one of the quaternary nitrogens, or an amino group for one of the methyl groups on each nitrogen, or tertiary nitrogens for the quaternary nitrogens all reduced the activity on the superior cervical ganglion. Phenylethane *p*-ω-bis(trimethyl ammonium) iodide was 3 times more active than hexamethonium iodide. The fact that the interquaternary distance was intermediate between penta- and hexa-methonium might be significant. Also in this compound saturation of the phenyl radical gave two geometric isomers, one of which was as potent as the unsaturated compound while the other had only one-tenth the activity. With the two isomers the interquaternary distance differed only slightly. Other polymethylene series, where the "onium" radicals were the nitrogens of quaternary heterocyclic nuclei, confirmed these results. The peak activity in a polymethylene bis(1-methylpyrrolidinium) series was very sharp with the pentane member, while in similar morpholinium and piperidinium series the peaks were comparatively flat. To extend these results to parasympathetic ganglia in the cat several procedures were used: (i) mydriasis; in the presence of light the sympathetically denervated eye is reduced to a slit. The compounds when injected dilated the pupil quantitatively; (ii) salivary flow on stimulation of the chorda-lingual nerves; (iii) vagal ganglionic-blocking activity—the fall in blood pressure being the response measured; (iv) the bladder-pelvic nerve preparation. Further results were obtained with the peristaltic reflex of the isolated guinea-pig ileum and in the mouse with the standard mydriasis test for atropine-like agents. There was good agreement of activity between the superior cervical ganglion and the mydriasis and salivary flow tests in cat for hexamethonium, penta- and hexamethylene pyrrolidinium, and phenylethane (bis-trialkyl) ammonium derivatives. For various reasons discrepant results were obtained on the guinea-pig ileum, cat blood pressure, cat bladder-pelvic nerve preparation and mouse mydriasis test. G. P.

Cortisone Acetate *v.* Cortisol in Rheumatoid Disease. H. F. West and G. R. Newns. (*Lancet*, 1954, **267**, 168.) Cortisol (hydrocortisone "free alcohol"), the natural adrenocortical hormone, has been administered to 22 patients who had received cortisone acetate daily for from 1 to 3 years and had not improved. The patients were observed over three months. The physical ability of 6 patients was considerably improved, 15 showed no change and one was worse. The erythrocyte sedimentation rate fell on an average from 27 to 21·5 mm. Increased effectiveness was however paralleled by an increase in side effects—increased deposition of fat, increased leucocytosis and a very definite rise in blood pressure. Cortisol is a more potent antirheumatic than cortisone acetate but because of the increased side effects the authors do not advocate its use in place of cortisone acetate. G. F. S.

Cortisone, a Chloro-derivative of, with Enhanced Activity. R. K. Callow, J. Lloyd, and D. A. Long. (*Lancet*, 1954, **267**, 20.) The biological activity of a new chloro derivative of cortisone (9-α-chloro-17-α-hydroxy-corticosterone) has been studied in mice, rats and guinea-pigs. A dose of 50 mg./kg. injected intramuscularly daily for ten days killed 7 out of 10 mice, compared with 3 out of 10 with cortisone. There was a greater fall in body weight and an increase in kidney size. In nestling rats 9α-chlorohydrocortisone (2·5 and 1·25 mg./kg.), and cortisone at the same doses, inhibited growth to a comparable degree; but 9α-chlorohydrocortisone was 3·4 times as active as cortisone in causing thymus involution, and 4·76 times as active in producing liver hypertrophy. In the tuberculin sensitivity test in guinea-pigs, 2 mg./kg. of 9α-chlorohydrocortisone had approximately the same desensitising activity as 10 mg./kg. of cortisone. In all tests therefore 9α-chlorohydrocortisone was more active than cortisone, and it is suggested that a high therapeutic activity in man may be expected.

G. F. S.

Curarising Agents, Central Action of Some. R. Hazard, J. Cheymol, P. Chabrier, Y. Gay and P. Muller. (*Thérapie*, 1954, **9**, 314.) Considerable sedative action, in addition to curariform action, was found in some members of a series of bisquaternary ammonium compounds with a piperazine nucleus in the interquaternary chain. Activity was measured directly, and by the prolongation of the sedative action of hexobarbitone, in mice. There was no correlation with curarising and central depressant activity. The quaternary nitrogens could be replaced by corresponding tertiary groups, but with loss of central depressant potency. On the "onium" radical, ethyl was more effective than either methyl or benzyl substitution. The piperazine nucleus in the interquaternary chain appeared essential for sedative action since a similar series where this nucleus was replaced by an ethylene oxide ($-O-CH_2-CH_2-O-$) grouping was without activity.

G. P.

β-Diethylaminoethyldiphenylpropylacetate Hydrochloride, Pharmacological Effect of. L. Cook, J. T. Toner and E. J. Fellows. (*J. Pharmacol.*, 1954, **3**, 131.) This paper describes the effects of the compound on the duration of hypnosis (loss of righting reflex) induced by hexobarbitone sodium in mice and rats. Premedication with 25 mg./kg. i.p. enhanced a subhypnotic dose of hexobarbitone (50 mg./kg. i.p.) in rats, which slept for an average time of 35·2 minutes. In mice oral or i.p. doses of 50 mg./kg. administered simultaneously with 100 mg./kg. of hexobarbitone i.p., significantly prolonged the duration of hypnosis, indicating quick absorption and onset of action. The optimal premedication time was 40 to 60 minutes, but the duration of action was long, the compound being still effective after 15 to 20 hours. There was a linear relationship between the log. sleeping time and the log. dose. The compound had a wide margin of safety, 1/500th of the oral LD50 (= 538 mg./kg.) in mice enhancing the duration of hexobarbital hypnosis. Normally the duration of hypnosis with hexobarbitone is limited because of toxic effects. Premedication with the compound prolonged hypnosis with very little effect on the toxicity of hexobarbitone. Neither kidneys nor adrenals were essential for the compound to produce its effects. Mice did not become tolerant but some rats did. When it was administered to rats immediately after recovery from hexobarbitone hypnosis, further hypnosis did not occur. Diphenylpropylacetic acid, which has been supposed to be a metabolite of the compound, showed only a weak activity. The paper also describes other pharmacological actions of this compound.

G. F. S.

Diethyl-*p*-nitrophenyl Phosphate (Paraoxon), Acetylcholine Production in Animals Poisoned by. J. M. Barnes and J. I. Duff. (*Brit. J. Pharmacol.*, 1954, **9**, 153.) Anæsthetised fully atropinised rabbits, cats and dogs recovered from a lethal dose of paraoxon (E600) if artificial respiration were administered. The animals were then insensitive to further injections of the same dose of the anticholinesterase. Concomitant determinations of the venous blood level of acetylcholine showed that in the cat and the rabbit this rose with each dose of paraoxon, but in the dog the rise was not as great and doses subsequent to the first had no further effect. Evisceration of the cat resulted in a slower acetylcholine blood level rise after the anticholinesterase. Acetylcholine production by the isolated rat diaphragm stimulated at a rate of 50 per second through the phrenic nerve for twenty minutes, was assayed on the cat blood pressure and found to be constant over a long period, in presence of either eserine or paraoxon. An explanation is advanced for this tachyphylaxis to paraoxon. G. P.

Isoniazid and *p*-Aminosalicylic Acid, Study of a Combination of. R. Kourilsky, S. Kourilsky and S. Micoulaud. (*Thérapie*, 1954, **9**, 273.) The compound formed by the interaction between *p*-aminosalicylic acid and isoniazid was well tolerated by guinea-pigs when given in doses 8 times those used in isoniazid therapy. Tolerance in man was good at an oral dose level of 10 mg./kg. Increasing the dose to 15 mg./kg. also increased the incidence of side effects, mainly of an intestinal nature. In experimental tuberculosis in the guinea-pig and in pulmonary tuberculosis in man the activity of the compound was comparable with those of isoniazid, *p*-aminosalicylic acid and dihydrostreptomycin. Experimetally its activity was similar to that of isonizid, but was more rapid and more regular with the same dose. *In vitro* studies on H37Rv sensitive, H37Rv resistant and "D" strains of *Myco. tuberculosis* showed the drug to be more active and more constant in activity over period of time than was isoniazid. Also, isoniazid-resistant strain "D" organisms were not resistant to the compound. G. P.

Isoniazid and Weight Gain. I. S. Mudie, N. W. Horne and J. W. Crofton. (*Brit. med. J.*, 1954, **1**, 1304.) In groups of patients with pulmonary tuberculosis treated with isoniazid a remarkable gain in weight has been one of the principal effects reported. 8 healthy males, aged between 25 and 40, none of whom had had tuberculosis, were divided into a treatment group and a control group. The control group received capsules containing lactose and the treatment group capsules containing 100 mg. isoniazid twice daily. The total period of trial was 14 weeks, including an initial observation period of 2 weeks before administration of any drugs and a final 2 weeks period after stopping administration. The control group received lactose for the whole 10 weeks. During the 10 weeks of administration the treatment group received isoniazid for 8 weeks and lactose for 2 weeks. The average weekly weight gain in the 4 subjects receiving isoniazid for 8 weeks was similar to the gain in the same group treated for 2 weeks with capsules containing lactose and to the gain during 4 weeks on observation alone. The average weekly gain was less than that of the group treated for 10 weeks with lactose alone. The authors conclude that there is no evidence from this trial to suggest that isoniazid in tuberculosis patients has any effect on weight gain other than that due to its effect on the disease. S. L. W.

Mephenesin and Gallamine Triethiodide in Tetanus. C. M. Parkes. (*Brit. med. J.*, 1954, **2**, 445.) The case under review deals with the treatment of tetanus in a young man of 22. In spite of the administration of 100,000 units of tetanus antitoxin intravenously, soluble penicillin 500,000 units 4-hourly, and 14 ml. of paraldehyde in 100 ml. of saline rectally, painful muscle spasms began, increasing in frequency and severity. An intravenous infusion of normal saline was set up and run in at the rate of 1 pint in 8 hours (5000 units of heparin to each pint of saline); this was kept going for 9 days. To control the spasms a 10 per cent. solution of mephenesin was injected into the drip tubing in doses of 5 to 10 ml.; this was followed by immediate relief of the spasm. To reduce the general muscular rigidity between spasms the patient was also given mephenesin elixir by mouth, 1 fl. oz. (1 g.) 5 times daily, and 30 ml. of the 10 per cent. solution was added to each bottle of normal saline. The dosage of mephenesin needed rose daily and on the 5th day hæmoglobinuria occurred and the mephenesin was replaced by gallamine triethiodide, given in a maximum single intravenous dose of 0·5 ml. (10 mg.), when immediate loss of spasm and pain followed. With the diminution in the frequency and intensity of spasms and the reduction in the amount of gallamine required to control them the respiratory condition improved and the patient made an uneventful recovery. The author concludes that the absence of respiratory depression makes mephenesin a highly useful drug in the treatment of tetanus, but it has the disadvantage that increasing dosage is required which may result in hæmoglobinuria. Gallamine triethodide should be reserved for those cases which fail or cease to respond to mephenesin since it carries a risk of respiratory depression. S. L. W.

Mercurial Diuresis, Observations on the Character of. R. A. Dale and P. H. Sanderson. (*Brit. J. Pharmacol.*, 1954, **9**, 210.) 0·2 g. mersalyl injected intravenously into normal male human subjects caused a fall in potassium excretion where normal diet was being taken; on salt-poor diets the potassium excretion rate rose. With normal diet the mersalyl increased urinary *p*H, but had no constant effect on inorganic phosphate excretion. There was a rise in ammonia excretion and a fall in bicarbonate excretion where the subjects were in normal acid-base balance. These results were reversed where the mersalyl was given after acidosis was induced by ingestion of ammonium chloride, although in both normal and acidotic subjects a fall in urinary *p*H occurred. In subjects made alkalotic with sodium bicarbonate, mersalyl diuresis was inhibited. The creatinine excretion fell transiently between 30 and 90 minutes of the injection, but the fall was not marked. Uric acid excretion rose within 30 minutes of injection whereas changes in the rates of water, sodium and chloride excretion generally occurred only after 60 minutes. The significance of the results in relation to the sites of excretion of the urinary constituents and to the site of action of mersalyl is discussed. Four points are put forward as worthy of further attention: the changes in potassium excretion; the delay in onset of diuresis; the reversal of the usual changes in ammonia and bicarbonate excretion in acidosis; and the abolition of diuresis in alkalosis. G. P.

Nitrogen Mustard in Treatment of Systemic Lupus Erythematosus. E. L. Dubois. (*Arch. intern. Med.*, 1954, **93**, 667.) 20 patients with active systemic lupus erythematosus on maintenance cortisone dosage were treated with mustine hydrochloride or triethylene melamine. The mustine hydrochloride was administered in a single dose of 20 mg. injected into the tubing of an intravenous infusion

of 5 per cent. dextrose in water. The drug was given during the evening after pre-medication with 0·1 g. of phenobarbitone and 0·1 g. of pentobarbitone sodium. The triethylene melamine was given in 5 mg. tablets, taken with water on an empty stomach an hour before breakfast. Initial courses consisted of a total dose of 10 to 15 mg. over a 2- to 3-day period and the interval between courses was not less than 2 weeks. 4 of 5 very œdematous patients with the nephrotic syndrome of lupus nephropathy had good results, diuresis appearing in 3 of the patients within 3 to 14 days after administration of the drug. 5 of 6 relatively dry nephrotic patients had improvement in their renal and in their general condition. 2 patients with hypertension and nephropathy and no œdema were not benefited. None of 7 patients without evidence of renal damage were benefited. Triethylene melamine was administered by mouth to 5 patients in 11 courses. Agranulocytosis occurred in 1 case and fatal aplastic anæmia in another despite the usual precautions. No serious toxicity was noted from the 34 courses of intravenously administered nitrogen mustard. S. L. W.

Octylamine and Compound 48/80—Comparison of Histamine Release by. W. Feldberg and J. L. Mongar. (*Brit. J. Pharmacol.*, 1954, **9**, 197.) Octylamine is a more potent liberator of histamine from minced guinea-pig lung than is 48/80. The situation reverses when the two are compared by the triple response of human skin *in vivo*. To investigate the comparison further the activities of the two agents on isolated perfused tissues were determined. Throughout the range of preparations used 48/80 was in most cases much more active than octylamine. The ratios of potency (48/80 : octylamine) on the preparations were:—cat skin flaps and gastrocnemius muscle, 200 : 1; perfused hind-quarters—rat 1000 : 1,—guinea-pig 60 : 1; perfused lung, 1 : 1 to 20 : 1 depending on species. With the lung preparations large doses of the drugs had to be used to liberate the histamine. G. P.

Pilocarpine, as an Antagonist to the Undesired Effects of Ganglion-blocking Agents. J. A. Gunn and A. M. Cooke. (*Brit. med. J.*, 1954, **1**, 1473.) A hypertensive patient, under treatment first with hexamethonium and later with pentolinium tartrate, was treated successfully with pilocarpine to abolish the dryness of the mouth and eyes, constipation, difficulty with micturition and loss of accommodation consequent to the parasympathetic ganglionic blockade caused by the blocking agents. With 6·6 mg. of pilocarpine nitrate given orally, salivation was excessive and the dose was reduced to 5 mg. which appeared adequate. The side effects of the ganglionic blockade disappeared within half an hour of taking the pilocarpine and returned in 6 to 10 hours, depending on the dose. G. P.

Serotonin, (5-Hydroxytryptamine), Species Difference in the Respiratory and Cardiovascular Response to. J. A. Schneider and F. F. Yonkman. (*J. Pharmacol.*, 1954, **111**, 84.) The authors have, by pharmacological and surgical means, attempted to localise species differences in respiratory and cardiovascular responses to 5-hydroxytryptamine. The study was conducted on dogs, cats and rabbits under barbiturate anæsthesia. Blood pressure was recorded by a glass membrane manometer, respiration by body plethysmography, afferent nervous activity was recorded from the intact vagus and heart rate by an electronic frequency recorder. In all 3 species a period of apnœa and bradycardia was observed after 5-hydroxytryptamine, but in the dog this apnœa was preceded by a short period of hyperpnœa. The blood pressure

response in the dog was mainly pressor, preceded in some instances by a brief fall. In cats and rabbits only a sustained vasodepressor response was obtained. On the isolated perfused Langendorff heart preparations of the 3 species 5-hydroxytryptamine had the same action, any difference being quantitative. Heart rate, rate of coronary flow and amplitude of contraction were all increased. Activation of pulmonary stretch receptors as evidenced by afferent nerve activity in the vagal fibres subserving this function was demonstrable after 5-hydroxytryptamine intravenous injection in the cat and dog, but not in rabbit. The bradycardia and initial blood pressure fall were considered to be due to such a reflex mechanism, since in all 3 species these effects could be abolished by procaine (in a dose which had been found to abolish similar effects by the veratrum alkaloids), by cutting the vagi, by ganglionic blockade with pendiomide, and by atropine. This reflex effect of 5-hydroxytryptamine was also responsible for the respiratory arrest in expiratory position in the cat; both bilateral vagotomy and intravenous procaine abolished the response. In the rabbit respiratory stimulation could be abolished only by combining spinal cord section at C6 and bilateral vagotomy. Neither of these measures alone completely eliminated the stimulation, nor did procainisation. The initial respiratory stimulation in the dog, however, was easily blocked by either cord section at C6 or bilateral vagotomy. In the dog the rise in blood pressure was due to a direct vasoconstriction and probably also to a direct effect on the heart. Regitine effectively blocks this action, but ganglionic blockade by Pendiomide potentiates the rise. The prolonged fall in blood pressure in rabbits produced by 5-hydroxytryptamine was reversed by cord section at C6, but not by ganglionic blockade. It would thus appear to be reflex in origin, but of a different nature from that seen in the cat. It was concluded that reflexes originating from the heart and lungs were responsible for the differences in cardiovascular and respiratory responses to 5-hydroxytryptamine in the species studied. A direct stimulation of the carotid body or of the brain is unlikely to be the cause of these effects. G. P.

Stanolone (Dihydrotestosterone) in the Treatment of Mammary Cancer. A. Gellhorn, J. Holland, J. B. Herrmann, J. Moss and A. Smelin (*J. Amer. med. Ass.*, 1954, **154**, 1274.) This is a report on the evaluation of stanolone in the treatment of 26 patients with carcinoma of the breast, in all of whom the extent of metastatic disease was such that radiotherapy was not feasible. The disease had been present for 1 to 5 years in 18 of the patients, for more than 5 years in 4, and for less than a year in the remaining 4. The stanolone was given as a suspension in isotonic sodium chloride solution in a dose of 100 mg. intramuscularly either daily or 6 times weekly. 3 patients received less than 3 g. of the hormone, 3 were given more than 10 g., and the remainder received between 3 and 10 g. Treatment was continued for as long as improvement was maintained. 11 of the patients experienced subjective relief for periods of 14 to 183 days, and 4 of these showed objective evidence of temporary tumour regression; 19 of the patients died within 1 to 11 months. There was no indication that the therapy achieved any significant prolongation of life. Of the 26 patients, 24 had side-reactions which were predominantly those of virilisation. From the results obtained in this study the authors conclude that stanolone has no qualitative or quantitative advantages over testosterone propionate in the therapy of metastatic cancer of the female breast. A much larger group of similarly studied cases will be required before it can be determined whether its therapeutic efficacy equals that of testosterone propionate.

S. L. W.