CCXCI.—*Physiologically Active Constituents of the Yew,* Taxus baccata. *Part I. Taxine.*

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Taxine, the poisonous alkaloid of the yew, *Taxus baccata*, was isolated by Lucas in 1856 (*Arch. Pharm.*, **85**, 145), but the first real advances in our knowledge of its chemistry are due to Winterstein and his collaborators (Winterstein and Iatrides, *Z. physiol. Chem.*, 1921, **117**, 240; Winterstein and Guyer, *ibid.*, 1923, **128**, 175; Danuser, Diss., Zurich, 1925; Schaller, Diss., Zurich, 1928), some of whose results have been confirmed and extended by Kondo and Amano (*J. Pharm. Soc. Japan*, 1922, 1074) and by Kondo and Takahashi (*ibid.*, 1925, No. 524, 2).

The alkaloid is poorly characterised, for it is amorphous and yields amorphous salts. Moreover, processes of degradation generally give amorphous products; only occasionally are small yields of crystalline material obtained. Some doubt as to its complete homogeneity is therefore unavoidable, in spite of the marked increase in purity recorded in this paper.

The early statements that taxine is crystalline (Marmé, *Bull. Soc. chim.*, 1876, **26**, 417; Amato and Capparelli, *Gazzetta*, 1880, **10**, 349) have not been substantiated by later workers, and even the physical constants are divergent. Marmé (*loc. cit.*), Hilger and Brande (*Ber.*, 1890, **23**, 462), and Thorpe and Stubbs (J., 1902, **81**, 874) give the melting point as 82°, Kondo and Amano as 105—111° after sintering at 97°, and Winterstein and Guyer as 111° after sintering at 100°. A value of $[\alpha]_D + 35°$ in alcohol is quoted by Kondo and Amano, whereas Winterstein and Iatrides found $+ 51{\cdot}5°$ in the same solvent. We also have failed to crystallise the alkaloid, but have effected purification involving material alteration of the constants. As an alternative to the conversion into salts, distillation, etc., employed by others, we have investigated a lengthy process combining solution in acids and reprecipitation, treatment of solutions with charcoal, and precipitation from different solvents. The purest taxine obtained formed colourless flakes or granules, which showed no crystalline structure under the polarising microscope, had $[\alpha]_D + 95{\cdot}7°$ in alcohol, and melted at 121—124° after precipitation from an acid solution by ammonia, at 110—115° when precipitated from ether by light petroleum, and at 85° when obtained by evaporating a benzene solution at a pressure of 0·1 mm. of mercury. The last specimen retained benzene which could not be removed, and the melting point is clearly dependent to some extent on the nature of the solvent from which the alkaloid has separated. The possibility has been considered that the relatively high rotation of our product was due to alteration of the taxine during manipulation, but as the processes were gentle and were only repetitions of those employed in extraction, we regard it as the result of the removal of lævorotatory material.

Further evidence of the increased purity of this taxine as compared with previous specimens is afforded by the following points. First, it fails to develop the striking colour reactions with concentrated mineral acids described by Winterstein and his collaborators; a comparison of their results and ours suggests that their material was contaminated by substances which develop a violet colour with mineral acids. Secondly, pure taxine does not reduce Fehling's solution, although the crude alkaloid does so; the reducing power of crude specimens is due to a small amount of impurity containing a carbonyl group. Thirdly, Mrs. T. Bryan Brown of the Pharmacology Department, Oxford University, has determined the minimum lethal dose to be about 40% lower than that quoted by other workers (*e.g.*, Winterstein and Iatrides; Pfenninger, *Z. ges. Exp. Med.*, 1922, **29**, 310) in the case of intravenous injection into rabbits. Her results in this connexion are in agreement with those of Winterstein and Guyer, and will be published elsewhere.

Contrary to the statement of Winterstein and Iatrides that taxine does not deteriorate, it has now been observed that even purified specimens decompose slowly into an ill-defined substance when kept in corked tubes in a dry atmosphere. This product is not basic, but contains half the percentage of nitrogen of the original taxine.

Hilger and Brande, Thorpe and Stubbs, Winterstein and Iatrides, and Kondo and Amano all concur in assigning to taxine the molecular formula $C_{37}H_{51}O_{10}N$, with which our analytical data are in fair agreement, and Winterstein and Guyer have proposed the partial formula $C_6H_5{\cdot}CH(NMe_2){\cdot}CH_2{\cdot}CO{\cdot}O{\cdot}C_{24}H_{34}O_6{\cdot}O{\cdot}CO{\cdot}CH_3$. This is based principally on the quantitative estimation of one acetyl group after alkaline hydrolysis, and on the isolation after acid hydrolysis of cinnamic acid and of optically inactive β-dimethylamino-β-phenylpropionic acid hydrochloride, which is reported to be identical with a synthetic *dl*-specimen prepared by Danuser by interaction of dimethylamine with β-iodo-β-phenylpropionic acid and by Schaller by the reduction of ethyl β-dimethylaminocinnamate. Presumably, racemisation of the β-carbon atom has occurred during the hydrolysis or subsequent manipulation. Following the method of Winterstein and Guyer, we also have isolated a hydrochloride having the same m. p. as their product, but at the request of Professor Winterstein we have refrained from intervention in this side of the problem and have restricted our attention to the non-nitrogenous products.

During the hydrolysis of crude taxine by boiling dilute sulphuric acid, the solution deposits a resin. From this Winterstein and Guyer were unable to isolate any homogeneous material capable of being characterised, but by specially devised methods (see p. 2145) it has now been possible to obtain a nitrogen-free substance which contained small quantities of an aldehyde or ketone, which yielded a semicarbazone. This carbonyl compound was not formed as a result of the hydrolysis, since the same semicarbazone has been isolated by treating crude taxine with semicarbazide. Our experiments show that it is responsible for the reduction of Fehling's solution by crude taxine.

After the removal of this impurity, the hydrolysis product melted at 155—165° and the analytical figures agreed with a formula $C_{24}H_{32}O_7$ (or slightly less closely with $C_{24}H_{34}O_7$). It is proposed to name this substance *anhydroxatine*. Assuming the correctness of Winterstein's partial formula for taxine as a basis for discussion, the name " xatine " is proposed for the group $C_{24}H_{34}O_6(OH)_2$, taxine itself being the acetyl and β-dimethylamino-β-phenylpropionyl derivative. Anhydroxatine is to be regarded as being formed from taxine essentially by the hydrolysis of these acyl groups and elimination of the elements of water, changes which might be expected through the agency of hot dilute sulphuric acid. The first-mentioned formula suggests that two hydrogen atoms may also be removed, possibly in the oxidation of a dihydroaromatic structure to an aromatic one, but certainty on this point is at present impossible owing to the amorphous character of anhydroxatine.

Anhydroxatine contains no methoxyl groups. By implication taxine also contains no methoxyl, and this finding confirms the statement by Schaller that the estimation of half a methoxyl in taxine in Zeisel determinations by himself, by Winterstein and Iatrides, and by Kondo and Takahashi is in reality due to the instability of the dimethylamino-group of the β-dimethylamino-β-phenylpropionyl residue.

Anhydroxatine contains no carbonyl group and is not oxidised by boiling Fehling's solution; nevertheless, it reduces Tollens's reagent (ammoniacal silver oxide containing sodium hydroxide). Further, it is unsaturated towards permanganate and behaves as a lactone with alkalis. These facts suggest that it is an unsaturated lactone of the type of angelicolactone, and since it has no reaction with sodium nitroprusside it resembles bufagin rather than digitalin and strophanthin (Jacobs, Hoffmann, and Gustus, *J. Biol. Chem.*, 1926, **70**, 1).

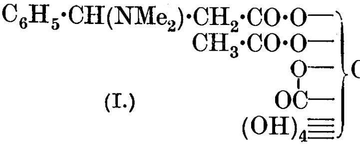
Taxine, though not attacked by carbonyl reagents, reduces Tollens's reagent, and by inference from anhydroxatine is also an unsaturated lactone. All the oxygen atoms of taxine (I) may now be accounted for. Two form a lactone group, and four take part in the ester linkages comprised by the acetyl and dimethylamino-β-phenylpropionyl groups. The remaining four are now shown by micro-Zerewitinoff estimations to be present as hydroxyl. We take this opportunity of expressing our indebtedness to Dr. H. Roth of the Kaiser Wilhelm Institut für Medizinische Forschung, Heidelberg, who has carried out these determinations.

It is, unfortunately, impossible to deduce the number of carbocyclic rings in taxine from the foregoing results, since no exact knowledge is available of the hydrogen content and the number of unsaturated linkages. It would be unwise to base an estimate of the former solely on the analytical data for amorphous substances, and the quantitative reduction experiments of Winterstein and Iatrides and of Danuser are inconclusive.

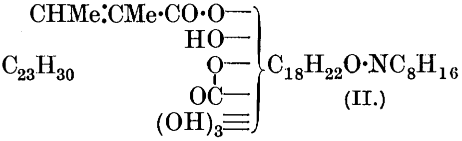
During an investigation of the possibility that ephedrine may occur in crude taxine (see following paper) or might be formed during its hydrolysis, it was observed on one occasion only that a smooth decomposition into a neutral non-nitrogenous *substance* was effected by a few minutes' shaking with sodium hydroxide solution and *p*-nitrobenzoyl chloride in ether at room temperature. This material may have been acetylxatine, $C_{26}H_{38}O_9$, but the analytical figures are unsatisfactory, and it has not been possible to repeat this degradation in spite of many attempts.

It is interesting to compare taxine with other chemically related substances. On the one hand, there is a close general resemblance between the partial formulæ for taxine (I) and cevadine (veratrine) (II; Freund and Schwarz, *J. pr. Chem.*, 1917, **96**, 236; Macbeth and Robinson, J., 1922, **121**, 1571), the most completely investigated of the alkaloids of the Gymnosperms, to which class the yew also belongs.

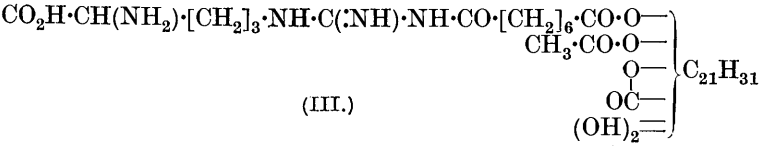
On the other hand, the lethal action of taxine is essentially on the heart, and this, together with its character as an unsaturated lactone, might relate it to the cardiac poisons of the digitalis, strophanthus, and toad-poison groups (bufotoxin, III; Wieland and Alles, *Ber.*, 1922, **55**, 1789), since Jacobs and Hoffmann (*J. Biol. Chem.*, 1926, 67, 333) suggest that the unsaturated lactone group may be essential for the " pharmacodynamic action " of these substances. There is,


(I.)

$$\left.\begin{array}{r} C_6H_5\cdot CH(NMe_2)\cdot CH_2\cdot CO\cdot O- \\ CH_3\cdot CO\cdot O- \\ O- \\ OC- \\ (OH)_4\equiv \end{array}\right\}C_{23}H_{30}$$


(II.)

$$\left.\begin{array}{r} CHMe{:}CMe\cdot CO\cdot O- \\ HO- \\ O- \\ OC- \\ (OH)_3\equiv \end{array}\right\}C_{18}H_{22}O\cdot NC_8H_{16}$$


(III.)

$$\left.\begin{array}{r} CO_2H\cdot CH(NH_2)\cdot[CH_2]_3\cdot NH\cdot C({:}NH)\cdot NH\cdot CO\cdot[CH_2]_6\cdot CO\cdot O- \\ CH_3\cdot CO\cdot O- \\ O- \\ OC- \\ (OH)_2= \end{array}\right\}C_{21}H_{31}$$

however, a significant difference in pharmacological behaviour, since taxine stops the frog's heart in diastole (Winterstein and Iatrides; Pfenninger; Bryan Brown, unpublished), whereas the digitalis effect is characteristically systolic.

The difficulty of obtaining supplies of leaves of high and constant alkaloid content and of handling large quantities has unfortunately limited this investigation, but the results are published now since it is improbable that the present authors will be able to continue the research.

EXPERIMENTAL.

Extraction of Taxine.—The yield was slightly improved by drying and mincing the leaves before extraction.

Extracts were made by two methods, *viz.*, (i) by soaking in 1% sulphuric acid, and (ii) by digesting with slaked lime and benzene. In preliminary experiments, method (i) was decidedly the more advantageous, and a large quantity of leaves was extracted by this method. Twigs from Kew Gardens were dried at 37°, stripped, and the leaves (2·15 kg.) minced and extracted with dilute sulphuric acid (8 l. of 1%) for 4 days. The mixture was filtered, mixed with " norite " charcoal, and stirred mechanically for 12 hours at a low temperature. This procedure yielded a clear yellow solution, which darkened when made alkaline, but neither deposited amorphous material nor gave rise to emulsions during ether extraction (below), as is the case when treatment with norite is omitted. The filtrate was made alkaline with ammonia, extracted thoroughly with ether, and the ethereal solution shaken with 1% sulphuric acid. The taxine, thus freed from non-basic impurities, was reprecipitated by ammonia and again taken up in ether, and the extract was dried and evaporated first at about 45° and then in a vacuum desiccator over sulphuric acid. The taxine (20 g.) thus obtained was a colourless powder, m. p. 90—98° after softening at 85°, and had $[\alpha]_D^{17^\circ}$ + 82·7°

in dry alcohol (α 8·24°, $l = 1$, $c = 10{\cdot}040$). It dissolved readily in dilute acids and in the usual organic solvents except light petroleum.

Purification of Taxine.—The taxine obtained above was submitted to a prolonged process of purification in an unsuccessful attempt to crystallise it. During the following stages the alkaloid was not allowed to remain in acid solution longer than necessary, and such solutions were kept ice-cold. The rotations were observed in absolute-alcoholic solution.

The taxine was dissolved in ether, shaken out with normal sulphuric acid, liberated with ammonia, and again taken up in ether, and the solution was dried, concentrated to small volume, and slowly mixed with light petroleum. The viscous precipitate which separated at first was discarded, and more light petroleum was gradually added to the solution, cooled to 0°. The powdery precipitate thus obtained was dissolved in 5% sulphuric acid, precipitated with ammonia, taken up in ether, and dried. Removal of the solvent yielded a colourless specimen of taxine having $[\alpha]_D^{17^\circ} + 90{\cdot}3^\circ$, ($\alpha = 3{\cdot}69^\circ$, $l = 1$, $c = 3{\cdot}178$). The acid–ammonia process was repeated, and the product subjected to four successive precipitations from ether by means of light petroleum. After being dried in a vacuum desiccator over sulphuric acid, it melted at 108—115° after softening at 87°, and had $[\alpha]_D^{17^\circ} + 95{\cdot}4^\circ$ ($\alpha = 5{\cdot}12^\circ$, $l = 1$, $c = 4{\cdot}590$). This material was dissolved in 1% sulphuric acid, precipitated with ice-cold ammonia, collected, washed with water, and dried in a vacuum desiccator. It formed a colourless granular powder, m. p. 121—124° after softening at 115°, which reduced Tollens's reagent but not boiling Fehling's solution [Found: C, 67·4, 65·9, 65·8; H, 7·8, 7·9, 7·9; N, 2·1. Calc. for $C_{37}H_{51}O_{10}N$: C, 66·4; H, 7·6; N, 2·1. Found by micro-Zerewitinoff: at 26°, OH, 10·2, 10·1; at 90°, OH, 12·7, 12·5. Calc. for $C_{37}H_{47}O_6N(OH)_4$: OH, 10·2; and for $C_{37}H_{46}O_5N(OH)_5$: OH, 12·7%]. The apparent existence of 5 active hydrogen atoms at 90° is presumably due to elimination of dimethylamine from the β-dimethylamino-β-phenylpropionyl group, a change which can be shown to occur on heating taxine in a boiling water-bath.

Since the m. p. of this specimen was at least 10° higher than that of the same material when precipitated from ether by light petroleum, it was decided to obtain it from benzene solution in order to ascertain if the m. p. varied with the solvent used. This supposition proved correct. The specimen of taxine, m. p. 121—124°, was dissolved in pure benzene, and the solvent removed at room temperature under reduced pressure in presence of phosphoric oxide. The product melted at 80°, frothing up the tube, and the m. p. was raised only to 85° by prolonged exposure at 0·1 mm. (Hyvac pump).

The rotation, however, remained unchanged at $[\alpha]_D^{17°} + 95{\cdot}7°$ ($\alpha = 6{\cdot}03°$, $l = 1$, $c = 5{\cdot}450$). In order to confirm this interdependence of m. p. and solvent, the taxine obtained thus from benzene was dissolved in 1% sulphuric acid, liberated with ammonia, and extracted with ether. The dried extract was freed from solvent first by drawing a stream of dry hydrogen through it and then by exposure to a Hyvac pump. The resulting taxine melted at 110—115°, the same temperature as observed previously after precipitation from ether (above) (Found : C, 65·2; H, 7·7; N, 2·0. Calc. for $C_{37}H_{51}O_{10}N$: C, 66·4; H, 7·6; N, 2·1%).

Decomposition of Taxine by 5% *Sulphuric Acid.*

This experiment has been repeatedly successful as regards the isolation of anhydroxatine, but as indicated below, the other products have not always been obtained.

A solution of crude taxine (8 g., from Kew Gardens leaves) in sulphuric acid (65 c.c. of 5%) was heated under reflux on the water-bath for 17 hours. The liquid, which smelt strongly of crushed yew leaves, was decanted from a considerable layer of dark resin which had separated on the bottom of the flask.

A. *Examination of the Acid Solution.*—(i) *Cinnamic acid.* The solution was extracted with ether, which yielded a small amount of cinnamic acid, identified by mixed m. p.

(ii) *Nitrogenous product.* The extracted liquid was mixed with an excess of phosphotungstic acid solution (20% in 5% sulphuric acid), and the precipitated phosphotungstate was decomposed by dissolving it in acetone (30 c.c.) and water (40 c.c.) and adding baryta solution until a filtered test portion gave no further precipitate. The solid was removed, the excess of barium precipitated by a stream of carbon dioxide, and the filtrate acidified with hydrochloric acid and evaporated to dryness at 40°/12 mm. The residue, freed from a little barium chloride by solution in absolute alcohol, could not be crystallised in spite of many attempts. In a similar experiment, however, in which the taxine was *freshly prepared* from leaves from the Forest of Dean and used *at once*, no difficulty was experienced in crystallising this product, and after being ground with acetone, in which it is sparingly soluble, and crystallised by adding ether to an alcoholic solution, it formed colourless flat needles, m. p. 186—187°, the m. p. recorded by Danuser (*loc. cit.*) for β-dimethylamino-β-phenylpropionic acid hydrochloride. Nevertheless, in a subsequent experiment carried out with the same specimen of taxine some weeks later, only an uncrystallisable gum was obtained. This may be related to our frequent observations that taxine, even after purification, decomposes slowly into non-basic nitrogenous products.

(iii) *Non-nitrogenous water-soluble products.* An excess of baryta was added to the filtrate from the phosphotungstate precipitate, the solid removed, and the solution freed from excess of baryta with carbon dioxide and evaporated to small volume in a vacuum desiccator. The addition of alcohol threw out a caseous, deliquescent precipitate, which was collected. This did not reduce Fehling's solution and was neither an acetate nor a cinnamate; the amount was too small for further examination. The filtrate contained barium acetate, which was converted into sodium acetate, m. p. 320° (Found in material dried at 100°: C, 29·1; H, 3·6. Calc. for $C_2H_3O_2Na$: C, 29·3; H, 3·7%).

B. *Non-basic Resin. Anhydroxatine.*—The resin was extracted with boiling benzene (100 c.c.) and the addition of light petroleum to the filtered solution yielded a tarry precipitate which was repeatedly extracted with boiling light petroleum (b. p. 100—120°). On cooling, these extracts deposited a flocculent, pale yellow precipitate of very crude anhydroxatine, m. p. 75—90° (1·8 g.). An alternative procedure was to grind the resin with cold $2N$-sodium hydroxide to remove dark-coloured acidic material, to extract the residue with benzene, and to precipitate the crude anhydroxatine with light petroleum. Further purification was effected by solution in cold ether, decantation from any residue, and precipitation with light petroleum, and after four repetitions of this treatment, the product was of a pale buff colour, m. p. 115—120° after sintering at 105° (Found: C, 67·8; H, 7·3%). It contained neither nitrogen nor methoxyl, and reduced boiling aqueous-alcoholic Fehling's solution. In view of the possibility that this substance might contain a carbonyl group, an attempt was made to prepare a semicarbazone. The substance (0·45 g.), semicarbazide hydrochloride (0·7 g.), and sodium acetate (0·6 g.) in 80% alcohol (15 c.c.) were heated under reflux on the water-bath for 5 hours. A trace of crystalline material, m. p. 259° (? semicarbazone), separated, but the amount did not permit further study. The liquid was poured into water, the precipitate collected, dried, dissolved in dry alcohol, and mixed with benzene. The small precipitate which separated was discarded, and the solution evaporated to dryness. The residual solid, m. p. 155—165°, contained no nitrogen and did not reduce Fehling's solution. When dissolved in benzene and precipitated with light petroleum, the *anhydroxatine* thus obtained formed a colourless powder, m. p. 155—165° (Found: C, 67·0; H, 7·2; M, 440. $C_{24}H_{32}O_7$ requires C, 66·7; H, 7·4%; M, 432). It gave no colour with alcoholic ferric chloride. It rapidly reduced permanganate in aqueous-alcoholic solution, but in chloroform it did not decolorise bromine at room temperature; on warming, a little hydrogen bromide was evolved. It readily reduced Tollens's reagent. It was insoluble in cold sodium hydroxide solution, but when its solution in aqueous-alcoholic sodium hydroxide was warmed for a few moments and acidified with dilute hydrochloric acid, an acid was precipitated which redissolved in excess of aqueous sodium hydroxide.

Action of p-*Nitrobenzoyl Chloride and Alkali on Taxine.*

Taxine (0·5 g., from leaves gathered in Oxford in July) in ether (7 c.c.) was mixed with *p*-nitrobenzoyl chloride (0·35 g.); a colourless solid, presumably a quaternary chloride, separated. 2*N*-Sodium hydroxide (1·5 c.c.; 4 mols.) was added, and the mixture shaken at room temperature. After a few minutes, sodium hydroxide (2 c.c.) and *p*-nitrobenzoyl chloride (0·35 g.) were added, the shaking was continued, and two minutes later a further 2 c.c. of sodium hydroxide were introduced.

The colourless, crystalline precipitate of *p*-nitrobenzoic anhydride which had separated was collected and crystallised from acetone, from which it separated in needles, m. p. 179—180° alone or mixed with an authentic specimen (Found: C, 53·2; H, 2·5; N, 8·9. Calc. for $C_{14}H_8O_7N_2$: C, 53·2; H, 2·5; N, 8·9%).

The ethereal layer was washed with water, dried, and evaporated. The residual gum solidified when stirred with light petroleum, and was purified by repeated precipitation by this solvent from benzene and ethereal solutions alternately. The resulting *acetylxatine* (?) formed a cream-coloured amorphous powder, m. p. 125—130° (decomp.), which did not contain nitrogen (Found: C, 63·5; H, 6·7. $C_{26}H_{38}O_9$ requires C, 63·2; H, 7·7. $C_{24}H_{36}O_8$ requires C, 63·7; H, 8·0. $C_{23}H_{30}O_8$ requires C, 63·6; H, 6·9%). Repeated fruitless attempts were made to obtain this substance from other specimens of taxine prepared from Kew Gardens leaves gathered in November; 75—90% of the alkaloid was recovered even after prolonging the reaction for periods up to 4 days.

Colour Reactions.—

Reagent.	Purified taxine.	Anhydroxatine.
Conc. H_2SO_4	Deep red.	Deep red.
Salkowski	Acid, reddish-orange. Chloroform, pink.	Acid, reddish-orange. Chloroform, colourless.
Lifschütz	Red, changing rapidly to dark blue, and slowly to dull green with green fluorescence.	Orange-red.
Conc. H_2SO_4 and $K_2Cr_2O_7$	Yellow, becoming dull olive brown.	—
Conc. HCl	No colour.	No colour.
Conc. HNO_3	No colour.	No colour.

Other sterol reactions were negative in both cases.

Deterioration of Pure Taxine.—A specimen of purified taxine, m. p. 121—124°, which had been preserved for about 3 months in a corked specimen tube in a desiccator (calcium chloride) was shaken with a large excess of ice-cold 2*N*-hydrochloric acid. The clear solution was filtered from undissolved solid and made alkaline with ice-cold ammonia. The colourless granular precipitate of taxine melted at 122° after sintering at 110° (Found: C, 66·0; H, 7·6. Calc. for $C_{37}H_{51}O_{10}N$: C, 66·4; N, 7·6%). It reduced Tollens's reagent, but not boiling Fehling's solution.

The acid-insoluble solid (above) was dissolved in ether, and the solution shaken first with dilute hydrochloric acid, then with water, dried, and concentrated. The addition of light petroleum yielded a colourless amorphous precipitate, m. p. 122—124° after softening at 115° (Found : N, 1·1%). It was not oxidised by boiling Fehling's solution but reduced Tollens's reagent, and readily decolorised cold alcoholic permanganate solution. It formed a colourless solution in concentrated sulphuric acid, which slowly became light brown. On being warmed for a few moments with dilute sodium hydroxide solution containing a little alcohol, it dissolved, and did not separate on cooling. When this solution was acidified with hydrochloric acid and concentrated in a vacuum desiccator, cinnamic acid, m. p. 133°, separated in leaflets and was identified by mixed m. p. determination.

The Carbonyl Impurity of Crude Taxine.—This experiment was carried out with a month-old specimen of crude taxine, m. p. about 90°, which reduced boiling Fehling's solution. It was shaken with a mixture of dilute hydrochloric acid and ether, and the two layers were separated.

(i) The acid solution was made alkaline with ammonia and extracted with ether, and the extract was dried and evaporated. The resulting gum still reduced Fehling's solution to a slight extent, but the remainder of the carbonyl compound was removed as follows. A solution of the gum in dilute acetic acid was filtered from undissolved solid, mixed with semicarbazide hydrochloride and sodium acetate, and kept at room temperature for 12 hours. The solution was again filtered, ammonia was added, and the resulting precipitate of still crude taxine melted at 100—115° after sintering at 90° (Found : N, 2·3. Calc. for $C_{37}H_{51}O_{10}N$: N, 2·1%). It did not reduce Fehling's solution, and the analysis shows that not more than traces of semicarbazide can have been present.

(ii) The ethereal solution was dried and evaporated. The resulting gum, which readily reduced Fehling's solution, was dissolved in 60% alcohol and mixed with semicarbazide hydrochloride and sodium acetate. After standing over-night, the solution was boiled under reflux for 1 hour, and part of the solvent was evaporated.

On cooling, a small amount of a semicarbazone separated in colourless needles, m. p. 259°, which were probably identical with those isolated after the hydrolysis of crude taxine by sulphuric acid. The mother-liquors were diluted with water, and the non-basic precipitate melted at 100—115° after sintering at 90°. This was evidently the normal product of the deterioration of taxine.

We wish to express our gratitude to Professor Wright Smith of Edinburgh University; to the Curator of the Royal Botanic Gardens, Kew; and to Mr. R. Grant Broadwood of the Forestry Commission, Parkend, who have supplied us with yew clippings; and to Messrs. T. and H. Smith, who have extracted large quantities of material for us. The work recorded in the following paper was also made possible by the good offices of these gentlemen.

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