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DIFFERENCES IN SPECIES OF TAXUS.*

BY IVOR JONES AND E. V. LYNN.

During the course of an extended investigation into the conifers of Washington, attention was ultimately called to the western yew, *Taxus brevifolia*. This tree is little known except to the woodsmen who call it simply "yew." It inhabits the forests on the coast from central British Columbia to central California, being usually found on the shaded banks of mountain streams, deep gorges or damp ravines. Most of the thriving and finest trees occur directly under larger conifers. Whenever they are accidentally exposed to direct sunlight, the leaves acquire a yellowish brown, apparently unhealthy color. The largest specimens are encountered in Oregon and Washington, particularly on the Olympic Peninsula.

The tree attains a height of 20 to 75 feet and a diameter of 6 to 30 inches, but the growth is very slow. One specimen cut down in Washington was found to be over 100 years old, although only 4.4 inches in diameter. The trunk is straight and conical with long, slightly dropping branches which give the tree a weeping appearance. The bark is conspicuously thin, rarely over one-fourth inch thick, and covered with a layer of thin, papery, purple, easily detached scales, beneath which the newer bark is a clear rose color. There are no pitchy pockets as is characteristic of most western conifers.

The leaves, which usually remain on the branches for three or four years, are dark yellowish green on top and much paler on the under surface and are very soft to the touch. They are usually about one-half inch long and one-sixteenth inch wide. The midribs are proportionately large and prominent and the petioles are slender and yellow.

The fruit is a bright red berry which differs from that of other conifers in consisting of red pulp enclosing a single seed which is almost as large, about the size of a small pea. It becomes ripe in September and begins to fall in October.

The wood, like that of other yews, is heavy, hard, strong, brittle, closely grained and slightly resinous. It is of a light red color with a thin white sap-wood and obscure medullary rays. By the natives and pioneers it was very highly prized for bows, paddles, spears, fence posts and many other articles.

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Of the several known species of *Taxus*, only three have been examined chemically. The bark of *Taxus canadensis* has been analyzed for moisture, ash and tannin (1). The only one which has been investigated extensively, however, is the European species, *Taxus, baccata,* together with the Asiatic variety, *Taxus cuspidata*. For centuries these have had a reputation as being toxic, although it is certain that the poisonous character is not always existent (2). At various times the seeds, fruit, leaves and young shoots have been reported as causing harmful effects in animals and man. On the other hand, the seeds are used to fatten poultry, the fruit has frequently been eaten with impunity, and cattle are known to have browsed on the shoots many times without being poisoned. The explanation of these contradictory facts is not yet certain.

A specific alkaloid was isolated by Lucas (3) in 1856 and twenty years later by Marme (4), who named it taxine. This was confirmed by Amato and Capparelli (5) in 1881, and the alkaloid was studied later by several others (6). A similar or identical base was extracted from the Asiatic yew by Ueda and Haseda (7) in 1914 and later by Kondo (8).

A glucoside, taxicatin, was extracted by Bourquelot (9) and later by Lefebre (10). The amount seemed to vary according to time of the year. A small amount of volatile oil was obtained from the needles by Amato and Capparelli (5).

The material used in the investigation of *Taxus brevifolia* was obtained from trees near the shore of Paradise Lake, twenty miles northeast of Seattle. Some of the leaves were dried in the air and some of them were used immediately. Collections were made at varying times from October 1930, to the spring of 1932.

Histological examination of the leaves and of the bark brought to light nothing unusual in cell structure. There was a complete absence of resin cells in the wood and in the tangential section was observed a spiral thickening of the vertical tracheids, far more pronounced than in other conifers with the possible exception of *Torreya*.

Volatile Oil.—Approximately 1000 Gm. of washed leaves were placed in a large flask and steam was passed through until three liters of distillate had been collected. No oil could be detected even after cohobation or after collection by means of ether, and repetitions with larger quantities of leaves were likewise fruitless. Samples of the wood and of the bark were distilled in the same way, with equally negative results. In each case the residual liquid was made acid and again distilled, but no oil could be detected at any time.

Alkaloid.—In a preliminary examination some of the leaves were extracted with ether and the ethereal solution was then exhausted with acidified water. The aqueous solution so obtained gave some precipitate with Wagner's and Mayer's reagents and with phosphomolybdic or tannic acid, but none with phosphotungstic or picric acid.

Since this left doubt as to presence of alkaloids, extracts were made from larger quantities, using four different methods. In the first the samples were macerated with ether and ammonia water (2:1). In the second the solvent consisted of very dilute sulphuric acid, while the third was similar except that fresh leaves were used in place of the dried. In each of these the extracted material was purified largely by repeated transference from aqueous to ethereal solution in the usual way. In the fourth method the solvent consisted of boiling alcohol acidified with tartaric acid. The alcohol was ultimately evaporated under reduced pressure, the resins were removed by pouring into very dilute acid and the resulting liquid was submitted to a regular Dragendorff extraction, using petroleum ether, benzene and chloroform. After evaporating the final solvents in each method, the residues were submitted to a sodium-fusion test for nitrogen. In no instance was a positive result obtained.

In order to test efficiency of the processes, controls were made using samples of the leaves from *Taxus baccata*. All of the extracts made from ammoniacal-aqueous solutions gave decidedly positive reactions. It is safe to conclude that the leaves of *Taxus brevifolia* contain no alkaloid, or at least not more than a trace.

The experiments were repeated with other parts of the tree, especially the bark, the wood and the roots, but as before the tests were entirely negative.

Glucoside.—Approximately 500 Gm. of the leaves were digested on the steam-bath with an excess of water containing 50 Gm. of finely powdered calcium carbonate. After straining through muslin, the volume was reduced by evaporation under reduced pressure and the syrupy mass was then extracted several times with ethyl acetate, as was done by Bourquelot (9) to isolate taxicatin from *Taxus baccata*. The grayish powder so obtained amounted to 0.2 Gm. It gave none of the reactions of taxicatin and, from reduction tests before and after hydrolysis, apparently contained no glucoside.

Other extractions were made with alcohol and with water in the usual manner for isolating glucosides, but none of them gave any indications of such a substance. One cannot say positively that the leaves do not contain a glucoside, but the evidence certainly indicates that they do not.

Anthocyanins.—About 200 Gm. of the dried bark were completely extracted with hot water to give a deep red solution. Upon acidifying with hydrochloric acid, the color brightened considerably, while sodium hydroxide gave an intense greenish blue. Amyl alcohol did not extract any of the red color from an acidified solution. Nascent hydrogen from zinc dust removed the color rapidly and, as soon as the excess of zinc was filtered off, the color reappeared. All of these reactions are typical of conjugated anthocyanins.

Pectins.—Solutions of the root-bark often became very viscous and tended to gelatinize. A determination of pectic acid by the A. O. A. C. method showed 0.26 per cent in the bark.

The reasons for the striking differences in composition in the species of *Taxus* constitute a very interesting problem in plant physiology. Practically every other conifer contains a leaf oil and exudes an oleoresin, but not *Taxus brevifolia*. More peculiar yet is the absence of alkaloid and probably glucoside, both of which are characteristic of other species of *Taxus*. Significant are the facts that the western yew grows very slowly and best in the absence of direct sunlight.

SUMMARY.

Taxus brevifolia differs from other species of the genus in containing no oil, no alkaloid and probably no glucoside. The bark yields anthocyanin pigments and that of the root gives a small amount of pectic acid.

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STUDIES ON THE PREPARATION, TOXICITY AND ABSORPTION OF BISMUTH COMPOUNDS. V. BISMUTH COMPOUNDS OF CATECHOL, PYROGALLOL AND GALLIC ACID.*

BY W. M. LAUTER, A. E. JURIST AND W. G. CHRISTIANSEN.

The bismuth compounds of catechol, pyrogallol and gallic acid have received only a limited amount of study as anti-syphilitics. Substantially no information is available comparing them to other bismuth compounds to determine their relative toxicity and absorption. The preparation of bismuth subgallate was discussed by Schamelhout (1) and others, and other bismuth gallates by Bianquis (2) and by Sazerac and Levaditi (3), and the biological characteristics of these substances has been described by Didry (4) and by Sazerac and Levaditi (5). The preparation of bismuth compounds of catechol has been described by Weinland and Sperl (6) and by Rosenheim and Baruttschisky (7). The use of bismuth compounds of pyrogallol has been discussed by Sazerac and Levaditi (5) who studied their action as anti-syphilitics.

In this paper the preparation of several bismuth compounds of gallic acid is described and they are compared as regards their toxicity and absorption. Also the preparation of bismuth derivatives of catechol and pyrogallol is described. The latter, owing to their insolubility, were not studied to determine their toxicity and absorption because it was most unlikely that they would be readily absorbed.

TABLE I.

Compound Injected.	Medium for Injection.	Concentration Mg. Bi/Cc.	Approximate Maximum Tolerated Dose—Mg. / Bi Kg. Body Weight.	Per cent Absorption.
Sodium bismuth digallate	Water	50	Less than 400	60
Sodium bismuth digallate	Olive oil	40	Less than 400	60
Sodium bismuth methyl gallate	Olive oil	50	Less than 400	60
Sodium bismuth gallate	Water	40	Less than 40	68*

* The rats injected died in less than 72 hours.

* Scientific Section, A. PH. A., Toronto meeting, 1932.