A PHYTOCHEMICAL SURVEY OF SOME PLANTS OF NORTH BORNEO

By H. R. Arthur

From the University of Hong Kong

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The aim of the survey was to sort out plants worthy of detailed research. In addition, the results reveal some interesting correlations between the chemical constituents of plants and their botanical classifications. Many plants used therapeutically by natives show strong tests with alkaloid reagents.

The plant species were collected in North Borneo from an area of the West Coast bounded by Jesselton, Ranau, and Keningau, during June and July 1952, and were tested there. After collection, specimens were pressed between double thickness bamboo paper and the papers were changed daily until the specimens were dry. From over 200 species recorded in this paper, all but 27 have been completely identified and these 27 have been given generic classification. In some cases, e.g., *Sonerila spp.* (Melastomaceæ) collected at high altitudes from Mt. Kinabalu, the failure in identification is because the species have not been fully worked out.

With the aid of interpreters, native names (Bajau, Dusun, and Malay) and in many cases, native uses for the plants collected were recorded. These data are recorded here only for the more interesting plants. The Dusun language varies considerably with the locality and the names recorded are those spoken by Dusuns living in the Dalas area. The names which were checked at two kampongs, correspond favourably with those recorded by Keith[1].

Many of the plants used therapeutically by natives are clouded with superstition. However, the following species used therapeutically by natives of North Borneo, have been selected as an illustration of the value to phytochemical and pharmacological investigation, which can be derived from a knowledge of native uses.

The leaves of *Brucea javanica* (Dusun: *towarr*) are used against malaria and the fruit is eaten to alleviate stomach complaints. The leaves are very bitter and show strong positive alkaloid tests. *Fagræa fragrans* (Bajau: *ombiatong*, Dusun: *dundorok*, Malay: *temasuk*) is well known by the natives. The leaves and fruit which give highly positive alkaloid tests are very bitter to the taste and are used against fever. This member of the Loganiaceæ also shows strong positive tests for essential oil, and triterpenoids, and should repay investigation. *Adenostemma lavenia* (Bajau: *tomali mali;* Dusun: *nonokot;* Malay: *tomali mali*) a member of the Compositæ has a very bitter leaf which gives strong alkaloid reactions. The leaf extract is applied for healing purposes after childbirth. Another member of the Compositæ, *Blumea balsamifera* (Bajau: *sambong;* Dusun: *towarr; Malay*: *sambong*) is used for cuts and abrasions. Tests of the

leaf show probable presence of alkaloids. The leaves of *Vernonia cinerea*, a member of the same family, are used therapeutically for chest complaints and are given to children who have "vacant stares". The leaves show highly positive alkaloid tests and a positive saponin test. It is not surprising that *Datura metel* (Bajau and Malay: *kosubang*) has a variety of therapeutic uses. A leaf extract of a *Polygonum* species (Dusun: *tawawa perai*) is used by the Dusuns against fever. The extract is very bitter and the leaves show highly positive alkaloid tests.

Chemical investigation of plants has already been of assistance to taxonomy. Jones[2], on the basis of an investigation of the constitutents of essential oils, showed that a species of *Melaleuca* existed in two varieties.

Members of certain botanical families, e.g., Solanaceæ, Loganiaceæ, are noted for their alkaloid content, while other botanical families are noted for other chemical constituents. Thus, Apocynaceæ and Scrophulariaceæ are noted for cardiac glycosides and Pittosporaceæ for saponins.

In a survey of 753 species from 110 families of Queensland plants, Webb[3] has shown that 145 species gave heavy precipitates with alkaloid reagents. 41 families apparently contain appreciable quantities of alkaloids, while 42 families appear entirely to lack alkaloids. In a preliminary phytochemical investigation of flora of New Guinea, Webb (personal communication) found similar results regarding families containing alkaloids and saponins.

According to Hilditch[4], in the seed fat of species of certain plant families—e.g., Coniferæ, Urticaceæ—linoleic acid and linolenic acid are the major component acids of the fat, whilst in species of some other familes—e.g., Oleaceæ, Myrtaceæ—linoleic acid and oleic acid predominate. In a third large group of plant families including the Menispermaceæ, Rutaceæ, and Rubiaceæ, palmitic acid and oleic acid are the major component acids of the seed fat.

All saponins have strong frothing powers. A negative froth test accordingly indicates absence of plant saponin. In this survey only very strongly positive tests for frothing power have been taken as evidence for the presence of saponin. It is considered that plants giving weakly positive or positive tests are without saponin or else contain only small amounts.

It is becoming clearer that a knowledge of the systematic positions of plants broadly allows a decision on whether detailed investigation of any particular plant for certain classes of chemicals is worth while.

Experimental

The methods used by Webb[3] were mainly employed in testing plant material, but some modifications and extensions, particularly in the tests used to detect triterpenoid and steroid compounds, have been made.

When testing for any particular class of constituent, as far as was possible, tests were applied to equivalent amounts of plant material.

The results of tests for all classes of constituents were classified as negative, weakly positive, strongly positive or very strongly positive on a comparative basis. Results of the tests are recorded as positive only if they were consistently strongly positive or consistently very strongly positive.

In this work ethereal extracts of plants were tested first with concentrated sulphuric acid and then by the Liebermann-Burchard test. A negative result to both these tests indicates that steroid and triterpenoid compounds are absent. If no colour reaction is obtained with sulphuric acid alone, a positive Liebermann-Burchard test is more diagnostic for steroid and triterpenoid compounds. However, a positive Liebermann-Burchard reaction, usually a good guide for steroid and triterpenoid compounds, is then still not specific, for some other plant products, e.g., abietic acid, are known to give colour-reactions in this test. Many triterpene acids form insoluble sodium salts and are commonly isolated by extraction of an ethereal solution with sodium hydroxide solution. The triterpene acid is precipitated at the interface. Other triterpene acids are partly precipitated and partly extracted. The concentration of sodium hydroxide solution influences this ratio. In this survey, plant extracts giving positive Liebermann-Burchard tests were extracted with sodium hydroxide solution and the presence of a precipitate at the interface was noted. The alkaline extract was acidified, extracted with ether, and tested again with sulphuric acid and by the Liebermann-Burchard test. Positive tests were taken as strong indications of the presence of triterpene acids. As a control experiment, the tests for triterpene acids were applied to *Psidium guaijava*, *Rhodomyrtus tomentosa*, to *Centrella asiatica* and to various *Melaleucas*, all of which are known to contain triterpene acids, and satisfactory results were obtained.

Test for Alkaloids. The plant material was finely chopped with a razor, introduced to half fill a small stoppered specimen tube (5 cm. $\times$ 1 cm.), covered with Prollius fluid and left for 2 days. The extract was evaporated on a small watch-glass at room temperature. The residue was thoroughly stirred with 4 drops of hydrochloric acid (2 per cent.), then left for half an hour. Drops were transferred to a microscope slide and tested by mixing with a drop of each of the following alkaloid reagents: picric acid solution, Mayer's reagent, Wagner's reagent, Sonnenschein's reagent and phosphotungstic acid. When precipitates were obtained the strength of precipitation was judged by eye and classified according to the aforementioned scheme.

Tests for Triterpenoids and Steroids. Chopped plant material (2·0 g.) was covered with ether (15 ml.) and left, with occasional shaking, for 2 days. 2 drops of the extract were evaporated on a watch glass at room temperature. A drop of concentrated sulphuric acid was added and the mixture stirred. Any colouration was noted.

To a further portion of 2 drops of the etheral extract dried on a watch-glass was added 2 drops of acetic anhydride and the mixture was stirred. A drop of concentrated sulphuric acid was added and the colouration was noted. If either or both of the foregoing tests produced a colouration, the ether extract was poured into a separating funnel and 3 per cent. sodium hydroxide solution (15 ml.) was added: the liquids were then shaken and allowed to settle. The strength of precipitation at the interface of the liquids was judged by eye and classified on a comparative basis. The lower alkaline layer was separated, acidified with dilute hydrochloric

acid, and extracted with ether (15 ml.). The ethereal extract was washed with water (15 ml.), dried over anhydrous sodium sulphate, and divided into two portions, each of which was evaporated on a watch-glass. As before, one portion was tested with concentrated sulphuric acid and the other with acetic anhydride, followed by concentrated sulphuric acid. Colourations were noted.

Test for Hydrogen Cyanide. As stated by van Der Walt[5]. No enzyme was used.

Test for Essential Oil. The fresh plant material was chopped with a razor and introduced to half fill a test-tube, then covered with water and the mixture boiled. The strength of any characteristic odour was noted. Fresh leaves were also crushed and smelt in the hand.

Test for Glycoside, Alkaloid etc. The plant material was chewed in the mouth. More particularly bitterness was noted and classified.

Test for Saponin. The fresh plant material was finely chopped with a razor and a small amount (0·1 g.) introduced into a test-tube (6 in. × ½ in.). Hot water (5 ml.) was added and the mixture boiled for 1 minute, then allowed to cool to room temperature. After vigorous shaking, the mixture was left for 3 minutes and the amount of frothing was classified as follows: No froth—negative; froth less than 1 cm. high—weakly positive; froth greater than 1 cm. high but less than 2 cm. high—positive; froth greater than 2 cm. high—strongly positive.

TABLE I

Tests were carried out on leaves, or in the case of herbs, on the whole plant. Positive tests are indicated in parenthesis after the species name by use of the following symbols: A for alkaloid; B for bitterness; E for essential oil; H for hydrogen cyanide; S for saponin; T for triterpenoid. Each plant was tested for each class of chemical.

ACANTHACEÆ—*Asystasia coromandeliana, Hemigraphis sp.* (E), *Pseuderanthemum sp.* (T, A), *Strobilanthes involucratus, Thunbergia alata* (A).
AMARANTHACEÆ—*Amaranthus frumentoceus, Celosia argentea* (T, A), *Gomphrena globosa* (T).
AMPELIDACEÆ—*Leea angulata* (E).
ANACARDIACEÆ—*Anacardium occidentale* (E), *Buchanania lucida* (B), *Melanochyla beccariana* (T).
ANNONACEÆ—*Desmos chinensis* (T, E).
APOCYNACEÆ—*Allamanda cathartica* (T), *Alstonia macrophylla* (T, A), *Ichnocarpus volubilis* (T), *Lochnera rosea* (T, A), *Plumeria rubra, Tabernæmontana sphærocarpa* (T).
ARACEÆ—*Amorphophallus sp.* (B, H. S).
ASCLEPIADACEÆ—*Asclepias curassavica* (T, A) *Hoya sp.* (B), *Toxocarpus sp.* E).
BALSAMINACEÆ—*Impatiens griffithii*
BOMBACEÆ—*Ceiba petandra*
BORAGINACEÆ—*Cynoglossum micranthum*
BURMANNIACEÆ—*Burmannia disticha*
CAMPANULACEÆ—*Pratia borneensis* (T).
CANNACEÆ—*Canna indica.*
CAPPARIDACEÆ—*Cleome sp.* (A).
CARICACEÆ—*Carica papaya* (B, T, A).
COMMELINACEÆ—*Commelina elegans.*
COMPOSITÆ—*Adenostemma lavenia* (B, A, E) *Ageratum conyzoides* (A, E), *Bidens pilosa, Blumea balsamifera* (A, E), *Dicrocephalla latifolia* (E), *Erichtites valerianifolia* (B, A, E), *Pluchea indica* (A), *Synedrella nodiflora* (A), *Vernonia cinerea* (T, A), *Vernonia patula* (T, A), *Wedelia biflora* (A).
CONVOLVULACEÆ—*Hewittia sublobata, Ipomœa cairica, Ipomœa crassicaulis, Ipomœa gracilis* (T), *Ipomœa pes-caprœ* (T), *Ipomœa tuba* (H), *Jacquemontia tomentella* (B, T, A), *Merremia tridentata.*
CUCURBITACEÆ—*Melothria affinis.*
CYPERACEÆ—*Scleria sp.*
DILLENIACEÆ—*Wormia excelsa* (T).
ELÆOCARPACEÆ—*Elæocarpus brevipes* (T, A), *Elæocarpus griffithii.*
EPACRIDACEÆ—*Leucopogon suaveolens* (B, T).
ERICACEÆ—*Rhododendron acuminatum* (T), *Rhododendron durionifolium* (T), *Rhododendron ericoides* (T), *Rhododendron Lowii* (T), *Rhododendron rugosum* (T, E), *Rhododendron stenophyllum* (A), *Vaccinium cordifolium* (T).
EUPHORBIACEÆ—*Antidesma ghœsembila, Croton tiglium, Euphorbia hirta. Glochidion littorale, Glochidion glaucum, Hevea brasiliensis* (H, T), *Macaranga maanoides* (E), *Macaranga triloba* (A), *Manihot esculenta* (H, T), *Phyllanthus niuri.*
FLACOURTIACEÆ—*Pangium edule.*
GESNERIACEÆ—*Didissandra sp.* (T).

GLEICHENIACEÆ—*Gleichenia linearis.*
GRAMINEÆ—*Axonopus compressus, Imperata cylindrica* (T, A), *Ischæmum muticum* (T).
GUTTIFERÆ—*Cratoxylon ligustrinum* (E), *Garcinia mangostana* (T).
HYPERICACEÆ—*Hypericum mutilum.*
LABIATÆ—*Dysophylla auricularia* (A, E), *Hyptis brevipes* (T, A), *Hyptis suaveolens* (E), *Leucas zeylanica* (T).
LECYTHIDIACEÆ—*Barringtonia racemosa* (T, S).
LEGUMINOSÆ—*Adenanthera pavonina, Albizzia littoralis, Bauhinia kochiana* (E), *Bauhinia purpurea, Cæsalpinia pulcherrima, Cassia fistula, Cassia occidentalis, Cassia siamea* (A), *Clitoria ternatea, Crotalaria anagyroides* (T, A), *Crotalaria striata* (A), *Crotalaria verrucosa, Delonix regia, Desmodium heterocarpum, Desmodium sp. Dioclea lasiocarpa, Mimosa pudica, Pithecolobium dulce* (S, A), *Tamarindus indicus.*
LOGANIACEÆ—*Fagræa fragrans* (B, E, T, A), *Fagræa pendula* (E).
LYTHRACEÆ—*Lagerstræmia speciosa* (H?).
MALVACEÆ—*Hibiscus esculentus, Hibiscus mutabilis* (A), *Hibiscus rosa-sinensis, Hibiscus surattensis Hibiscus tiliaceus, Sida rhombifolia, Urena lobata.*
MARANTACEÆ—*Donax canneformis.*
MELASTOMACEÆ—*Clidemia hirta, Medinilla speciosa, Melastoma asperum* (T), *Melastoma malabathricum, Melastoma sp., Sonerila spp.* (six different) (E), *Sonerila tenuifolia.*
MENISPERMACEÆ—*Hypserpa cuspidata* (A).
MORACEÆ—*Artocarpus spp.* (two different), *Conocephalus cordifolia* (E).
MYRISTICACEÆ—*Knema conferta* (E).
MYRSINACEÆ—*Ardisia elliptica* (T), *Ardisia spp.* (two different).
MYRTACEÆ—*Eugenia tormeda* (E), *Eugenia zeylanica* (T, E), *Leptospermum recurvum* (T, E), *Psidium guaijava* (T, E), *Rhodomyrtus tomentosa* (T, E).
NEPENTHACEÆ—*Nepenthes gracilis, Nepenthes rafflesiana.*
NYCTAGINACEÆ—*Bougainvillia spectabilis*
OLEACEÆ—*Jasminum bifarium* (A).
ONAGRACEÆ—*Jussiæa linifolia* (B), *Jussiæa suffruticosa* (B).
ORCHIDACEÆ—*Chelistonelle sp., Coelogyne sp., Cystorchis sp., Eria grandis* (T), *Vanda teres.*
OXALIDACEÆ—*Averrhoa bilimbi* (T, E).
PALMÆ—*Arenga sp., Carota mitis.*
PASSIFLORACEÆ—*Passiflora fœtida* (A, H), *Passiflora laurifolia* (A, H).
PITTOSPORACEÆ—*Pittosporum ferrugineum* (B, S, T).
POLYGONACEÆ—*Antigonon leptopus, Polygonum chinense* (T), *Polygonum sp.* (A, E).
PONTEDERIACEÆ—*Eichornia crassipes* (H, T, A), *Monochoria vaginalis.*
RHAMNACEÆ—*Alphitonia zizyphoides* (S).
ROSACEÆ—*Angelesia splendens, Rubus angulosus.*
RUBIACEÆ—*Argostemma sp., Argostemma brachyantherum, Argostemma hameliæfolium* (S), *Hedyotis macrostegia* (T), *Ixora stenophylla, Mussænda hirsuta* (H, T), *Mussænda villosa, Randia anisophylla* (T), *Randia longifolia.*
SAPINDACEÆ—*unidentified* (T).
SAXIFRAGACEÆ—*Polyosma hookeri* (T).
SCROPHULARIACEÆ—*Euphrasia borneensis, Ilysanthes ciliata, Scoparia dulcis* (B), *Vandellia crustacea.*
SIMARUBACEÆ—*Brucea javanica* (B, A).
SOLANACEÆ—*Capsicum frutescens* (A), *Datura metel* (B, A), *Physalis minima* (B), *Solanum mammosum, Solanum nigrum* (A), *Solanum torvum.*
STERCULIACEÆ—*Kleinhovia hospita* (T, E), *Melochia umbellata.*
TERNSTRŒMIACEÆ—*Ploiarium alternifolium* (A).
THEACEÆ—*Schima brevifolia.*
UMBELLIFERÆ—*Didiscus saniculæfolius.*
URTICACEÆ—*Antiaris toxicaria* (T), *Elatostemma sp., Leucosyke capitellata.*
VERBENACEÆ—*Clerodendron myrmecophilum, Gmelina elliptica* (B), *Lantana camara* (T, E), *Stachytarpheta indica* (A), *Vitex pubescens* (B, A).
ZINGIBERACEÆ—*Catimbium assimile* (E), *Costus speciosus, Hedychium sp.* (T, E).

CONCLUSIONS

Table I shows that in the species tested, about 1 in every 6 gives strong precipitates with alkaloid reagents. Webb found about the same rate from a random selection of Queensland flora. Bornean members of the following plant families contain alkaloids: Acanthaceæ, Apocynaceæ, Capparidaceæ, Compositæ, Convolvulaceæ, Elæocarpaceæ, Ericaceæ, Euphorbiaceæ, Gramineæ, Labiatæ, Leguminosæ, Loganiaceæ, Menispermaceæ, Oleaceæ, Passifloraceæ, Polygonaceæ, Simarubaceæ, Solanaceæ, Ternstrœmiaceæ, Verbenaceæ. Webb has found similar results in the Queensland flora. From the data obtained to date, Bornean members of the following families apparently lack alkaloids: Melastomaceæ, Myrtaceæ, Orchidacæ, Rubiaceæ (?), Scrophulariaceæ. Webb finds that in members of the Queensland Compositæ, less than 1 in 10 contain alkaloids, while in the Bornean species of the same family which were tested about 7 in 10 give strong precipitates with alkaloid reagents. It is obvious, however, that insufficient members have been tested for a true comparison to be made.

The families of Bornean species with interesting triterpenoids are: Apocynaceæ, Ericaceæ, Myrtaceæ, Urticaceæ, Rubiaceæ, while saponins occur in Araceæ, Pittosporaceæ, and Rhamnaceæ.

Hydrogen cyanide was detected in the following plant families: Araceæ, Convolvulaceæ, Euphorbiaceæ, Lythraceæ (?), Passifloraceæ, Pontederiaceæ, Rubiaceæ and the positive species possibly contain cyanogenetic glycosides.

The following very bitter species gave negative tests to alkaloid reagents: *Buchanania lucida* (Anacardiaceæ), *Leucopogon suaveolens*, (Epacridaceæ *Jussiæa linifolia, Jussiæa suffruticosa* (Onagraceæ) *Scoparia dulcis* (Scrophulariaceæ) and are considered, therefore, possibly to contain glycosides.

The tables of results bring out an interesting feature among the Ericaceæ. 5 of the 6 species of *Rhododendron* which were tested, show practically identical results in the triterpenoid and steroid tests and all are negative to alkaloid reagents. The other species, *Rhododendron stenophyllum*, is strongly positive to alkaloid reagents and gives a reaction in the Liebermann-Burchard test quite different from the other species.

The presence of saponin in *Pittosporum ferrugineum* is not surprising. In addition the tables of results show that investigations of the Melastomaceæ would probably be time wasted.

Significance of the Results

With few exceptions Webb[3] found that when Prollius fluid was used for extraction, later detailed laboratory examination confirmed the presence of alkaloid in species giving strongly positive and very strongly positive tests. The results from hydrochloric acid extraction compared favourably, but discrepancies between the two methods are known to occur. In this survey, good agreement was obtained by the two methods for the few species where both were applied. In general, Prollius fluid was used for extraction. 8 species which were tested in this survey have already been tested by Webb in his Queensland survey. The results agree very well for 7 of the species. *Vernonia cinerea* (Compositæ) from Queensland, however, gave negative alkaloid tests whereas a Bornean specimen gave highly positive tests. There are many factors which can account for discrepancies of this sort. The age of the plant, the soil and climatic conditions, and the season when the tests are applied undoubtedly influence the result. Hilditch (*loc. cit.*, p. 151) records a geographical variation of the fatty acid constituents of palm oils.

Negative alkaloidal tests are generally certain. Tests taken as positive in this survey are those which were classified strongly positive and very strongly positive to all reagents used.

Summary

1. 205 species of plants of North Borneo have been examined for alkaloids, triterpenoids and steroid compounds, saponins, essential oils, glycosides and hydrogen cyanide.

2. Native names and uses for the plants were collected and some of these are recorded.

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