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# Host specificity and phytochemical constituents of mistletoe and twigs of parasitized plants: Implications for blanket application of mistletoe as cure-all medicine

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## ABSTRACT

A study on host specificity and phytochemical constituents of leaves of mistletoe and parasitized twigs of seven host trees/shrubs was conducted in University of Benin, Benin City, Nigeria. Plant height and location were measured with Nikon Laser Rangefinder (Forestry 550) and GPS respectively while occurrence and count of mistletoe on host plants was largely anecdotal/visual. Phytochemical analyses were conducted at Faculty of Agriculture Science/Soil Laboratory, University of Benin, Benin City, Nigeria while data were subjected to One-way Analysis of Variance and means separated using Duncan Multiple Range Test ( $p=0.05$ ). Results revealed mistletoe were largely generalists, and contained more phytochemicals than host plants. *Tapinanthus ogowensis* from *Moringa oleifera* was richest in alkaloid; *Phragmanthera nigriflora* on *Citrus sinensis*, *Tapinanthus ogowensis* on *Moringa oleifera* and *Phragmanthera capitata* on *Calliandra portoricensis* recorded highest phenol content and differences among them were not significant ( $p<0.05$ ). *Phragmanthera capitata* on *Spondias mombin* had significantly higher tannins than other mistletoe and host plants ( $p<0.05$ ). *P. capitata* on *S. mombin* recorded best for saponin and flavonoid while *P. capitata* on *Psidium guajava*, *Loranthus micranthus* on *Persia americana* and *P. capitata* on *C. portoricensis* recorded more anthraquinone and were not significantly different from each other  $p<0.05$ . While the marked variation in phytochemicals in leaves of mistletoe and twigs of host plants is noteworthy; the study has sufficiently established that ecological idiosyncrasies, time/season and varied environmental phenomena strongly influence incidence and/or buildup of secondary metabolites. The foregoing should serve as benchmark in selection and/or use of mistletoe for resolving infirmities.

**KEYWORDS:** Mistletoe, ethnomedicine, generalists, host preference, parasitized plants, phytochemicals

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## INTRODUCTION

Host specificity is the restricted use of available potential host species at a local scale, while host preference refers to the hierarchical ranking of host use [1]. Several factors such as seed dispersal vectors, host availability, host abundance, host compatibility and suitable niche for the parasite determine host specificity in mistletoes [2]. Mistletoe are hemiparasitic plants which access water and nutrients from other living plants [3]. They physically attach at least one point either to the above ground (aerial/stem/shoot parasites) or below ground (root parasites) parts of the host plants [4]. As hemi-parasites that parasitize and obtain nourishment from the branches of trees/shrubs with the aid "haustoria", mistletoe possess chlorophyll

and photosynthesize but the quantum of nutrients and water they obtain from their host plants tend to vary in relation to host use preference and/or specificity [5,6,7]. The term 'host specificity' is used to connote the restricted use of available potential host species at a local scale, host preference refers to the hierarchical ranking of host use [1].

In Africa, 70% of mistletoe are generalist species, that is, species that parasitize hosts from several families; 12% are specific on hosts from one family but occasionally parasitize a few genera of other families and 18% are specific to one or a few host species of a single genus [8]. A number of mistletoe species are specialized in living on different hosts due to frequent encounters between mistletoe seeds and commonest plants [9]. Tizhe *et al* [10]

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reported that mistletoe could parasitize a variety of tree species with few of them having special preference to certain type of host plants. Some studies have indicated that non-random perch preferences of seed dispersers are important for determining host specificity [11]. This may result in concentration of mistletoe seeds on either the most abundant trees or the less abundant ones [9].

As semi-parasitic plants, mistletoe are considered as unwanted plant but are also known for their medicinal value and use in the treatment of various ailments including cough, diabetes, cancer, ulcer, etc. [12,13]. Mistletoe are now known as “cure all” and have been used as a drug/remedy for more than twenty health problems [14]; this is dependent on the phytochemicals they contain and the efficacy of drug produced which are linked to the host plant(s) and that mistletoe harvested there from, are suited for the treatment or cure of ailments [15]. Dibong *et al* [16] reported that 82% of the healers in a region in Cameroon recommended *Phragmanthera capitata* for the treatment of some 22 different diseases, including hypertension, hypotension, irregular menstruations, menopause, convulsions, diabetes, rheumatism related pains, epilepsy, kidney and chronic muscular pains. This may not have a scientific backing as constituents of *Korthalsella japonica* did not change or disappear irrespective of its host trees [17].

Host specificity is an elusive concept and the mechanism behind it remains one of the most challenging areas of mistletoe biology [18]. These aerial parasites have received little research attention compared to their counterparts – the root parasites [19,20]. There is lot of confusion regarding the taxonomy of Loranthaceae, particularly in West Africa; for example, in Nigeria, many authors have referred to most *Tapinanthus spp.* as *V. album*, a foreign plant that does not occur in the country [21]. Some people often identify mistletoe through the host plant it is found on; this can be very misleading as different mistletoe species could grow on a particular tree host or vice versa [22].

The phytochemical constituents of mistletoe largely depend on plant characteristics, season of the year, mode of extraction, etc. Using ‘methanolic extract of the leaves harvested from *Persea americana*, Moghadamtousi *et al.* [23] recorded higher concentrations of alkaloids during April compared to July which is the peak of raining season; but saponins, glycosides, etc. were not found in July samples. But Osadebe *et al.* [24] recorded higher amounts of flavonoids and oils in July compared to April samples. Phytochemical studies on *Loranthus micranthus* leaves harvested from six different host trees, namely, *Persia americana*, *Baphia nitida*, *Kola acuminata*, *Pentaclethra macrophylla*, *Azadirachta indica*, and *Irvingia gabonensis*, revealed that alkaloids were in significant quantities in the extracts of *K. acuminata*, *P. americana*, and *I. gabonensis*; moreover, the phytochemical constituent host dependency was also shown [13, 25]. A petroleum ether extract of *L. micranthus* leaves parasitic on *P. americana* harvested at different seasons (January, April, July, and November) showed only presence of alkaloids in April and July and proved harvesting period dependency in phytochemicals of *L. micranthus* [26].

In the light of the forgoing this paper evaluates host specificity / dependency and use preference alongside important secondary metabolites of medicinal relevance that mistletoe share with selected parasitized host trees/shrubs. The intertwining roles played by intrinsic (e.g. biological characteristics and idiosyncrasies of host and hemi-parasitic plants) and ecological/environmental factors, particularly season, dispersal agents and phytochemical screening methods in shaping the presence and/or availability of secondary metabolites in mistletoe and parasitized plants are highlighted.

## METHODOLOGY

### Study Area

The study was carried out in University of Benin, Benin City, Edo State, Nigeria (Latitudes 6° 11' and 6° 29' N and Longitudes 5° 33' and 5° 47' E; 109m *asl*). Benin City, the capital of Edo State, is located in the rainforest region of Nigeria. The Mean annual rainfall ranges from 1800 to 2000mm; mean relative humidity is about 75% at noon and 95% at the early hours of the day while the maximum and minimum temperatures averaged 33°C and 23°C respectively [27]. The general topography of Benin City typically low, sloping gently from about 105m above sea level in the north-east to about 55m above sea level in the south-west area [28].

### Materials and Methods

Leaf and twig samples were collected from healthy plants with the aid of a sickle (*go-to-hell*) at various locations in University of Benin, Benin City, Nigeria in April, 2018. The height and positions of each sampled host tree/shrub were measured using Nikon Laser Rangefinder (Forestry 550) and geographical positioning system (GPS). The study on host specificity was field based and largely visual/anecdotal. Two sets of specimens were collected from the lower, middle and upper crown regions during the pre-mature to mature growth stages. One was primed for identification / authentication in the Forestry Research Institute of Nigeria (FRIN) herbarium at Ibadan (FHI) while the other was prepared (air dried, ground into uniform powder using a Thomas Willey milling machine before extraction, dehydration) for phytochemical screening at the Faculty of Agriculture crop and soil laboratory, University of Benin, Benin City, Nigeria.

### Tests for Secondary Metabolites

The aqueous extract of each sample was prepared by soaking 100g of dried powdered sample in 200ml of distilled water for 12 h; and the extracts were filtered using Whatman filter paper No. 42 (125mm) [29].

- (a) Test for Alkaloid was carried out using methods described by Boham and Kocipai-Abyazan [30]: 20% ethanol was added to 1g of each extracted sample; allowed to stand for 2-4 hours and then filtered. Concentrated ammonium hydroxide was added to the extract to precipitate the alkaloid and the precipitate filtered, oven dried for 30mins at 60°C and reweighed.

- (b) Total Phenols were determined by spectrophotometric method [29]. The fat free sample was boiled with 50 ml of ether for the extraction of the phenolic component for 15 minutes. 5 ml of the extract was pipetted into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of concentrated amylocholol were added. The samples were made up to mark and left to react for 30 min for colour development and measurement was taken at 505 nm.
- (c) Test for Tannins was carried out using methods described by Van-Burden and Robinson [31]. 1g of each of the sample was weighed and macerated with 50ml of distilled water. The mixture was filtered and 1ml of filtrate pipetted into test tubes. 2ml of the saturated picric acid was then added. Absorbance was measured at 530nm and tannin acid 1.3% in 100ml H<sub>2</sub>SO<sub>4</sub>.
- (d) Saponin test followed methods described by Obadoni and Ochuko [32]. 20 g of ground sample was weighed into a conical flask and 100 cm<sup>3</sup> of 20% aqueous ethanol added. It was heated over a hot water bath for 4 h while stirring continuously at about 55°C and the mixture was filtered and residue re-extracted with 100ml of 20 % ethanol. Extracts were reduced to 40ml over water bath at about 90°C and the concentrate transferred into a 250ml separating funnel and 20ml of diethyl ether added and shaken vigorously. The aqueous layer was recovered and the ether layer discarded. The purification process was repeated; 30ml of n-butanol was added and the combined n-butanol extracts were washed twice with 10ml of 5% aqueous Sodium chloride. The solution was heated in a water bath; after evaporation, samples were dried in the oven to a constant weight at 60°C for 2hrs and the saponin content calculated as percentage.
- (e) Test for Flavonoid followed Boham and Kocipai-Abyazan [30]. 1g of samples were extracted repeatedly with 100ml of 80% aqueous methanol at room temperature for 2 to 6hours while soaking and filtering. The whole solution was filtered through Whatman filter paper no 42 (125mm). The filtrate was later transferred into a crucible and evaporated to dryness over water and weighed to a constant weight.
- (f) Anthraquinone was determined by methods described by Soladoye and Chukwuma [33]. 1g of extracted sample was soaked in 50ml distilled water for 16 hours. The suspension was heated in a water bath at 70°C for 1 h and allowed to cool. Few drops of 80% methanol was added to it and then filtered. The clear solution was then measured by an absorbance of 459nm.
- (g) Cyanogenic glycoside determination: Methods described by Piero *et al.* [34] was used: 1g of each plant sample was dispersed in 200ml of warm water and the suspension heated for 3h while stirring continuously. The filtrate and residue were then re-extracted with 100ml acetone and reduced to 10ml on heating. The concentrate was then weighed and Cyanogenic glycoside content calculated.

### Data Analysis

Data obtained from the phytochemical analysis of were arranged in a Completely Randomized Design (CRD) and subjected

to Analysis of Variance (ANOVA) while the Duncan Multiple Range Test (DMRT) was used to separate the means ( $p=0.05$ ).

## RESULTS

### Profile of Parasitized Host Trees/Shrubs and Associated Mistletoe Species

Fifty-two trees/shrubs distributed among seven species were parasitized by mistletoe in the study area. The profile of sampled plants, mistletoe and incidence on host plants and their locations are shown in Table 1.

### Phytochemical Constituents of Leaves of Mistletoe and Twigs of Trees/Shrubs

Results revealed generally higher phytochemicals in the mistletoe than twigs of host plants. Overall, *S. mombin* twig and associated mistletoe were richest in three phytochemicals (saponin, tannins and flavonoid) and were significantly different from the rest plants ( $p<0.05$ ). The mistletoe associated with *M. oleifera* had best results for alkaloid while phenolic content was significantly higher in mistletoe obtained from *C. sinensis*, *M. oleifera* and *C. portoricensis*. Anthraquinone was richest in mistletoe obtained from *P. guajava*, *P. americana* and *C. portoricensis* while Cyanogenic glycoside was highest in mistletoe collected from *M. oleifera* (Table2).

## DISCUSSION

Most mistletoe species recorded were characteristically not selective of, or specific to, any particular host; implying that the hemi-parasites were predominantly generalists. From results, *P. capitata* which parasitized four host trees and shrubs, namely *S. mombin*, *P. americana*, *C. portoricensis* and *P. guajava*, and *T. ogowensis* which infected *M. oleifera* and *P. guajava*. In an investigation conducted in Ghana Overfield *et al.* [35] found '*T. ogowensis* to have infested a wide range of hosts such as *Citrus* spp. (*Rutaceae*), *P. americana* (*Lauraceae*), *T. cocoa* (*Sterculiaceae*) and *C. nitida* (*Sterculiaceae*).

The high incidence of *P. nigrifera* recorded in one of the two orchards of *C. sinensis* encountered during the study portrayed the hemi-parasite as localized or specific. Dibong *et al.* [36] designated *Globimetula opaca* which parasitized *C. sinensis* in an orchard in the Duala region of Cameroon as site- and host-specific. This result is not in agreement with the results of investigations carried out on African mistletoe by several authors who worked in West and Central Africa. For example, in some studies conducted in Cameroon, Ghana and Nigeria [15,36,37] several species of mistletoe, namely *Globimetula* spp. *T. bangwensis*, *T. ogowensis*, *P. capitata*, *P. incana*, *T. globiferus* found in citrus orchards, were classified as 'generalists'.

The above findings aptly confirm that the mode of infestation of host plants by mistletoe was not overly restrictive. For example, the number and type of shrubs encountered during the study as host species (e.g., *C. portoricensis*, *P. alba*, *P. guajava*) and the

**Table 1: List of Parasitized Host plants and associated Mistletoe species**

Scientific name & (Habit of plant)	Common name	Sample plant height (m)	Mistletoe species (and incidence on host plant)	Position of tree/shrub
<i>Calliandra portoricensis</i> (Shrub)	English Powder puff	5.82	<i>Phragmanthera capitata</i> (6)	N06° 24' 22" E005° 37' 24"
<i>Citrus sinensis</i> (Tree)	Sweet Orange	4.36	<i>Phragmanthera nigritana</i> (8)	N06° 23' 51" E005° 37' 35"
<i>Moringa oleifera</i> (Tree)	Moringa	6.44	<i>Tapinanthus ogowensis</i> (3)	N06° 24' 24" E005° 37' 26"
<i>Persea Americana</i> (Tree)	Avocadopear	7.25	<i>Phragmanthera capitata</i> ; <i>Loranthus micranthus</i> (6)	N06° 24' 23" E005° 37' 12"
<i>Plumeria alba</i> (Shrub/Small tree)	Frangipani	4.83	<i>Tapinanthus ogowensis</i> (4)	N06° 24' 22" E005° 37' 10"
<i>Psidium guajava</i> (Shrub/Small tree)	Guava	3.75	<i>Phragmanthera capitata</i> (3) <i>Tapinanthus ogowensis</i> (3)	N06° 24' 18" E005° 37' 36"
<i>Spondias mombin</i> (Tree)	Hog plum	12.93	<i>Phragmanthera capitata</i> (9)	N06° 23' 50" E005° 37' 33"

**Table 2: Assortment of secondary metabolites in leaves of mistletoe species and twigs of host plants**

Phytochemical	Source	<i>P. alba</i>	<i>S. mombin</i>	<i>C. sinensis</i>	<i>M. oleifera</i>	<i>C. portoricensis</i>	<i>P. americana</i>	<i>P. guajava</i>
Alkaloid	H	0.772±0.028 <sup>cd</sup>	1.274±0.033 <sup>a</sup>	1.162±0.037 <sup>b</sup>	1.134±0.001 <sup>b</sup>	0.742±0.001 <sup>d</sup>	0.830±0.002 <sup>c</sup>	0.597±0.033 <sup>e</sup>
	M	1.676±0.048 <sup>c</sup>	1.573±0.001 <sup>d</sup>	1.745±0.001 <sup>b</sup>	1.842±0.001 <sup>a</sup>	1.135±0.001 <sup>g</sup>	1.218±0.003 <sup>f</sup>	1.321±0.002 <sup>e</sup>
Phenol	H	0.431±0.005 <sup>c</sup>	0.416±0.001 <sup>c</sup>	0.532±0.001 <sup>d</sup>	0.629±0.004 <sup>a</sup>	0.527±0.004 <sup>b</sup>	0.646±0.006 <sup>a</sup>	0.559±0.031 <sup>b</sup>
	M	0.682±0.006 <sup>d</sup>	0.755±0.043 <sup>c</sup>	1.076±0.003 <sup>a</sup>	1.034±0.001 <sup>a</sup>	1.012±0.001 <sup>a</sup>	0.859±0.040 <sup>b</sup>	0.820±0.001 <sup>bc</sup>
Tannin	H	0.527±0.068 <sup>e</sup>	1.225±0.001 <sup>a</sup>	1.124±0.001 <sup>a</sup>	0.859±0.042 <sup>e</sup>	0.673±0.001 <sup>d</sup>	1.024±0.001 <sup>b</sup>	1.159±0.025 <sup>a</sup>
	M	1.039±0.004 <sup>f</sup>	1.428±0.003 <sup>a</sup>	1.264±0.027 <sup>c</sup>	1.114±0.001 <sup>d</sup>	0.829±0.003 <sup>f</sup>	1.288±0.025 <sup>c</sup>	1.365±0.001 <sup>b</sup>
Saponin	H	0.285±0.035 <sup>e</sup>	0.732±0.003 <sup>a</sup>	0.412±0.001 <sup>bc</sup>	0.357±0.001 <sup>cd</sup>	0.381±0.032 <sup>bcd</sup>	0.432±0.001 <sup>b</sup>	0.336±0.002 <sup>de</sup>
	M	0.640±0.002 <sup>e</sup>	1.016±0.001 <sup>a</sup>	0.748±0.031 <sup>d</sup>	0.888±0.027 <sup>b</sup>	0.827±0.004 <sup>c</sup>	0.909±0.019 <sup>b</sup>	1.022±0.001 <sup>a</sup>
Flavonoid	H	0.461±0.043 <sup>b</sup>	0.717±0.001 <sup>a</sup>	0.252±0.001 <sup>c</sup>	0.216±0.001 <sup>c</sup>	0.133±0.003 <sup>d</sup>	0.103±0.001 <sup>d</sup>	0.126±0.001 <sup>d</sup>
	M	0.716±0.004 <sup>b</sup>	1.134±0.001 <sup>a</sup>	0.546±0.012 <sup>c</sup>	0.564±0.036 <sup>c</sup>	0.422±0.003 <sup>d</sup>	0.578±0.032 <sup>c</sup>	0.515±0.001 <sup>c</sup>
Anthraquinone	H	0.342±0.048 <sup>d</sup>	0.236±0.001 <sup>e</sup>	0.328±0.001 <sup>d</sup>	0.419±0.001 <sup>c</sup>	0.522±0.001 <sup>b</sup>	0.422±0.003 <sup>c</sup>	0.568±0.002 <sup>a</sup>
	M	0.370±0.024 <sup>d</sup>	0.440±0.006 <sup>c</sup>	0.521±0.005 <sup>b</sup>	0.557±0.035 <sup>b</sup>	1.014±0.001 <sup>a</sup>	1.035±0.001 <sup>a</sup>	1.042±0.005 <sup>a</sup>
Cyanogenic glycoside	H	0.042±0.001 <sup>b</sup>	0.036±0.001 <sup>c</sup>	0.105±0.002 <sup>a</sup>	0.013±0.001 <sup>e</sup>	0.014±0.001 <sup>e</sup>	0.012±0.001 <sup>e</sup>	0.021±0.001 <sup>d</sup>
	M	0.112±0.002 <sup>d</sup>	0.214±0.001 <sup>b</sup>	0.114±0.001 <sup>d</sup>	0.303±0.002 <sup>a</sup>	0.014±0.002 <sup>e</sup>	0.015±0.002 <sup>e</sup>	0.204±0.001 <sup>c</sup>

Means (a, b, c, d, e, f, g) with different superscript along row are significantly different ( $p \leq 0.05$ ); H=Host plant twig; M=Mistletoe

number and occurrence of hemi-parasites in each plant palpably confirm that “for any mistletoe to transform from generalist to specialist and *vice versa*, some salient factors, including abundance/diversity of host species, the dispersing agent’s ability to encounter preferred host, etc. were likely to come into play. Some authors [1,38] have argued that ‘host choice can be considerably influenced by host abundance’; while others [39] asserted that “mistletoe are found to be generalists in places with high host species diversity”.

Several authors [1, 10, 38] have made allusions to the fact that host abundance often exerts a strong influence on parasitization by mistletoe species. But the findings in this study appear to be at variance with the authors’ assertion because two tree species encountered in the study area, *Azadirachta indica* and *Delonix regia*, though abundant, but were not parasitized. Additionally, of the four stands of *C. portoricensis* and *P. alba* assessed, only one stand each was parasitized while infestation incidence was six and four respectively. Downey [40] has argued that host selection favours close physiological adaptations of the mistletoe. But Fadini [9] opined that ‘host specificity depends on host abundance as well as compatibility at the physiological, genetic, mechanical and biochemical levels. While one is at liberty to reason that both *A. indica* and *D. regia*, lacked the physiological adaptation to attract mistletoe that parasitized other host trees;

to what could the absence of mistletoe on the remaining three stands of *C. portoricensis* and *P. alba*, which very close to each other, be ascribed? Some authors [11, 41] have contended that mistletoe specialization on trees is not dependent on frequent encounter of mistletoe seeds and host trees. This may rather be explained by non-random perch preferences of dispersal birds which could result in the concentration of seeds on both most abundant and less abundant trees [42,43].

Some important factors that might have influenced specificity or specialized infestation of hosts by mistletoe in the study area were abundance, diversity and/or assortment of cultivated fruit, ornamental and amenity tree/shrub species. According to Fadini [9], ‘a number of mistletoe species are specialized in living on different hosts due to frequent encounters between mistletoe seeds and commonest plants’. This may result in concentration of mistletoe seeds on either the most abundant trees or the less abundant ones [9]. For example, Burkill [44] found *Tapinanthus bangwensis* on 43 different hosts in 18 plant families; while Pattanayak and Sunita [45] reported that *Dendrophthoe falcata* – the most common of all the mistletoe that occur in India –was associated with about 401 plant hosts.

From study results, plant height and crown size seemed to have significantly influenced the prevalence of mistletoe on some of



the parasitized trees in the study area. For example, the tallest host tree, Hog plum (height: 12.93m) which also had the largest crown recorded the highest incidence/concentration of mistletoe species. This result is in agreement with the findings of Didier *et al.* [46] who reported that “host characteristics such as branch size, age and height and the duration of association between the host and the parasite affect prevalence. Tree height is an important host characteristic that controls mistletoe distribution and abundance; taller trees tend to attract more dispersers of mistletoe compared to shorter trees [42,47]. But Tizhe *et al.* [10] claimed that factors like abundance of host plants and vulnerability (characteristics) of the host plant could play a pivotal role in the parasitization of plant by mistletoe. Overton [48] also reported that ‘characteristics such as branch size, age, and height of a host plant can have a strong influence on mistletoe attachment resulting in size related mistletoe infection patterns’.

The outcomes of phytochemical analyses revealed that mistletoe and twigs of associated host plants had significant amounts of all the phytochemicals investigated. The presence of these chemicals in the host plants might have acted as Haustorium Inducing Factors (HIFs), which aided in attracting these hemiparasites as the presence of HIFs such as phenols, flavonoid, etc. in the host plants may have in turn helped to initiate the development of haustorium [49]. While making reference to earlier findings on ‘correlation between host and chemical profiles of the plant and parasite’ many authors [43,50,51,52] surmised that “the influence of host chemistry on the chemical constituent of the parasite on different hosts might justify why the host is as important as the parasite in ethnomedicine and why the use of mistletoe in the treatment of ailment is often dependent on a particular or specific host”.

Hemi-parasitic plants can trigger salt imbalance leading to a concomitant reduction of salt content in parasitized host branches/twigs. Dibong *et al.* [53] investigated the influence of *P. capitata* on the distribution of sodium and potassium salts in the plant tissues five host species (*Citrus maxima*, *Manikara zapota*, *P. americana*, *P. guajava* and *Theobroma cacao*) and found that *P. capitata* reduced the Na<sup>+</sup> concentrations in the aerial parts of infected host trees and maintained the salt level low in photosynthetic organs. The authors also observed high concentrations of K<sup>+</sup> in the leaves of the non-infected host branches of three hosts, *C. maxima*, *M. zapota* and *P. guajava* and concluded that Loranthaceae could contribute to decrease the content of salt on parasitized host boughs and then on branches bearing fruits. The fact that the mistletoe species found in this study recorded more phytochemicals than the twigs of their hosts might presumably be due to their hemiparasitic characteristics as suggested by Williams [54].

The similarity between phytochemicals contained in mistletoe and associated host plants have been highlighted in several past studies. For example, like its hemi-parasites, *S. mombin* has high extractable phenolic, saponins, tannin and anthraquinone contents [55,56]. The anti-malarial activity of *S. mombin* may be linked to a range of compounds like anthraquinones, berberine, flavonoids, naphthoquinones, sesquiterpenes, quassinoids,

indole and quinoline alkaloids [57]. This is because the efficacy of drugs are thought to be dependent on the host plant(s) and that mistletoes harvested on certain hosts are suited for the treatment or cure of a particular health problem [15]. Ayoka *et al.* [58], citing different authorities, chronicled the medicinal uses of *S. mombin* and by implication the mistletoe associated with it to include: abortifacients [59, 60]; anti-epileptic and anti-psychotic [61]; anti-viral [62]; anti-ageing [63, 64]; anti-bacterial [62,65]; anti-diarrhoea [59]; anti-fertility [66]; anti-helminthic [67]; anti-inflammation [68]; anti-malarial [57]; anti-microbial [56, 62]; anti-oxidant [64] and anxiolytic [69].

*C. portoricensis* leaves contain saponins, tannins, flavonoids and glycosides [70]. Numerous pharmacological activities have been reported, including anti asthmatic [71]; hypolipidaemic, erythropoietic [72], anti-sickling [73], anticonvulsant [74,75] anthelmintic [70], antimicrobial as well as traditional management of sickle cell anaemia and prostate cancer in Africa [71], and analgesic [76,77].

The contention that presence or accumulation of secondary metabolites in mistletoe depend on the season of the year, mode of extraction of secondary metabolites as well as mistletoe - host plant dependency [23, 24] was accentuated as *T. ogowensis* from *M. oleifera* recorded highest amount of alkaloids. Phytochemical studies on *L. micranthus* leaves harvested from six different host trees, namely, *Persia americana*, *Baphia nitida*, *Kola acuminata*, *Pentaclethra macrophylla*, *Azadirachta indica*, and *Irvingia gabonensis*, revealed large quantities of alkaloids in the extracts of *K. acuminata*, *P. americana*, and *I. gabonensis*. Several authors [13, 25] have alluded to the strong relationships and phytochemicals – host dependency influence commanded by some fruit trees/shrubs. A petroleum ether extract of *L. micranthus* leaves parasitic on *P. americana* harvested at different seasons (January, April, July, and November) showed only presence of alkaloids in April and July and proved harvesting period dependency in phytochemicals of *L. micranthus* [26]. In a separate study, Ibrahim *et al.* [52] found no Alkaloids in all 27 mistletoe representing seven different species obtained from different locations spread across eight cities/towns in Nigeria palpably confirms the findings in this study and several others [13,23,24,25,26].

All the mistletoe and host plants encountered in this study were found to have reasonable quantities of alkaloids. Alkaloids, the most valued of all the phytochemicals, are said to be pharmacologically active and their actions are felt in the autonomic nervous system, blood vessels, promotion of diuresis, respiratory system, gastrointestinal tract, uterus, malignant diseases, infections and malaria [78, 79]. Alkaloids are known to possess antimicrobial, anti-HIV and anti-parasitic abilities [80]. It has also been reported that alkaloids have anti-depressant-like actions in rodents [81]. Trease and Evans [82] and Madziga *et al.* [83] surmised that, “plants containing alkaloids do not feature strongly in herbal medicine as they are toxic”.

Results on anthraquinone revealed that the mistletoe, *P. capitata* and *T. ogowensis* from Guava recorded best followed by *P. capitata* and *L. micranthus* from Avocado pear and then

*P. capitata* from English Powder-puff; and they were significantly different ( $p < 0.05$ ) from the rest results. These parasitic plants would be more suitable in the treatment of constipation, malaria and cancer [84,85]. The mistletoe, *P. capitata*, obtained from *Spondias mombin* and *T. brameus* collected from *Plumeria alba* were richest in flavonoid content. Flavonoid have been described as potent water-soluble antioxidants and free radical scavengers, which prevent oxidative cell damage and it does contain strong anticancer activity [86]. They are known to have a wide range of biological and pharmacological activities – anti-inflammatory, anti-oxidant, anti-microbial and anti-cancer properties [87,88]. Halilu *et al.* [89] reported the presence of Flavonoids, Saponins, Cardiac glycosides in the mistletoe *Tapinanthus globiferus* but found anthraquinones and alkaloids absent, although present in the associated *M. oleifera* leaves.

The mistletoe, *Phragmanthera capitata*, collected from *Spondias mombin* during the study had the highest tannin content. Tannins are widely distributed through the plant kingdom which consists of anticancer properties by blocking the production of enzyme required for cancer cell line growth [90], and possess anti-oxidant properties which have the capacity to protect the tissues from the action of free radicals due to the cellular aging processes [91]. Tannins are also known for their anti-cancer properties and their usefulness in the protection of the urinary tract, cardiovascular and immune system [92]. Tannins have stringent properties, and hasten the healing of wounds and inflamed mucous membranes [93]. Tannins are a major phenolic compounds that act as primary antioxidant or free radical scavengers [94].

Saponin, recorded highest in *P. capitata* and *T. ogowensis* collected from *P. guajava*. Saponin is a secondary metabolite having medicinal properties such as anti-tumor properties; helps maintain blood cholesterol level and can be used as adjuvant in vaccines [95,96]. It has the precipitating properties and coagulating properties on red blood cells [86]; lower the cholesterol level and equipped with anti-diabetic and anti-carcinogenic properties [78]. Adodo [14] reported that the mistletoe associated with guava cure cancer.

The mistletoe, *P. nigriflora*, obtained from *C. sinensis* *M. oleifera* and *C. portoricensis* recorded highest phenolic content and were significantly different ( $p > 0.05$ ) from those associated with *P. americana* and *P. guajava*. Asami and Danny [97] asserted that phenol has anti-oxidant protection against heart disease and cancer and it also possesses anti-inflammatory ability. Like other phytochemicals, the phenol recorded in the mistletoe, *P. nigriflora*, was higher than what was obtained from the twig of *Citrus*. These results are in accord with earlier investigations by scientists [98,99] who reported that “high amounts of phenol have long been known to be a phytochemical feature of parasitic flowering plants and they are said to occur at a level that is generally higher than their host plant”.

The *T. ogowensis* obtained from *Moringa oleifera* was richest in cyanogenic glycoside. Cyanogenic glycoside is toxic and poses a potential risk to consumers [100]. Consequently, it has to be consumed in very low quantity, as its intake may lead to growth

retardation especially if the protein intake of an individual is low; this is because detoxification of cyanide in human body requires sulfur donors from sulfur-containing amino acids [101]. Detoxification of cyanide in the human body can only be done if low level of cyanide is generated and this is only possible if a small amount of cyanide is consumed [100]. The fact that Cyanogenic glycoside was richest in *T. ogowensis* obtained from *Moringa oleifera* is an apt signal that the selection and use of host plant and associated mistletoe in ethnomedicine should be done with utmost caution.

## CONCLUSION AND RECOMMENDATION

Mistletoe can be generalists or host specific; but host specificity and presence or accumulation of secondary metabolites and by extension, the potency of mistletoe/host plants as remedy for the treatment of ailments are a function of diverse inherent and external factors, including features of host plant (e.g. abundance, diversity, height, age, diameter and compatibility), season, agent and pattern of dispersal, screening methods, etc. The results of phytochemical composition revealed marked variations in secondary metabolites contained in mistletoe and twigs of parasitized host plants. Caution should therefore be exercised when selecting the species of mistletoe and parasitized host plant to use or prescribe for treatment of ailments. Finally, the basic aspects of mistletoe and host plant ecology including how they speciate should be investigated.

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