Hindawi
Journal of Chemistry
Volume 2018, Article ID 6869294, 9 pages
https://doi.org/10.1155/2018/6869294

Research Article

Nutritional and Biological Evaluation of Leaves of *Mangifera indica* from Mauritius

S. Jhaumeer Laulloo,[1] **M. G. Bhowon**,[1] **S. Soyfoo**,[1] **and L. S. Chua**[2]

[1]*Department of Chemistry, Faculty of Science, University of Mauritius, Réduit, Mauritius*
[2]*Institute of Bioproduct Development, Universiti Teknologi Malaysia, Johor Bahru, 81310 UTM Skudai, Malaysia*

Correspondence should be addressed to S. Jhaumeer Laulloo; sabina@uom.ac.mu and M. G. Bhowon; mbhowon@uom.ac.mu

Received 19 April 2018; Accepted 19 June 2018; Published 1 August 2018

Academic Editor: Sevgi Kolaylı

Mango trees are evergreen plants that are present all around Mauritius. In this study, mango leaves, *Mangifera indica* grown in Mauritius were investigated for their nutritional values involving proximate composition, total flavonoid (TFC), total phenolic (TPC), and mineral content, and phytochemicals as well as its antioxidant and antibacterial properties. The ash, crude fat, neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) of the mango leaves were found to be 12.61, 3.92, 35.32, 34.98, and 12.86%, respectively. The calcium content (2.15%) was above the normal required range, while the phosphorus content (0.12%) and crude protein content (13.60%) were within the normal required range of common fodders. The phytochemical results showed the presence of saponins, alkaloids, phenols, tannins, and flavonoids in the crude, EtOAC, and MeOH extracts. The values of TPC and TFC were higher for the EtOAC extract compared to the MeOH extract. Several secondary metabolites were identified from the leaves of the *Mangifera indica* which include 11 phenols, 4 xanthones, 9 flavanols, 10 benzophenones, 7 terpenoids, and 4 derivatives of gallotannins using UPLC-MS/MS. The presence of these metabolites is responsible for good antioxidant and antibacterial properties. Hence, mango leaves can be exploited for its potential use as a supplementary fodder for ruminants.

1. Introduction

Mauritius relies heavily on importation of meat and dairy products. Local production of cattle and goat is limited and not well organized. Being a small island, pasture fields are limited and are on decline. Small livestock breeders need to look for alternative feed sources for their animals especially during the dry months of the year.

Tree leaves play an important role in the nutrition of grazing animals in areas where few or no alternatives are available [1]. Fodder trees are an important source of supplementary protein, vitamins, and minerals in developing countries and are an alternative source of livestock feeding which have the potential for alleviating some of the feed shortages and nutritional deficiencies for small ruminants, goats, and sheep diets [2].

Mango trees are evergreen plants which withstand dry periods very efficiently, and they can be used as a food supplement. Although a number of studies have been conducted for the different uses of mango fruits, peels, juice, and stem bark, there are limited reports on the importance of *Mangifera indica* leaves and its suitability to be used as fodder. Mango leaves fed at high percentage may cause poisoning for cattle [3]; however, it can form an important part of feed given to ruminants [4]. According to the study conducted in Nigeria, it was shown that when goats were fed on mango leaves together with Guinea grass (*Panicum maximum*), the weight gain recorded was higher compared to when the mango leaves were replaced by *Ficus* (*Ficus thonningii*) or Gliricidia (*Gliricidia sepium*). The study also pointed out that mango leaves were accepted by the goats and are therefore palatable to the animals [5].

Mangifera indica have been the focus of intense research in search of a variety of biomolecules from different parts of the plants such as stems, leaves, fruits, and seed kernels. Its medicinal value is well established and has been used for centuries for the treatment of different kinds of diseases [6]. It is found to possess different pharmacological properties including the antibacterial property [7].

Previous research on the phytochemical screening of the leaves of *Mangifera indica* revealed the presence of phenols, saponins, tannins, steroids, flavonoids, anthraquinone, and glycosides [7–9]. Therefore, the purpose of this work was to investigate the nutritional values in terms of proximate analysis, TPC, TFC, antioxidant, and antibacterial studies and to identify the major secondary metabolites present in the leaves of *Mangifera indica* from Mauritius.

2. Materials and Methods

2.1. General. Ethylenediamine tetraacetic acid disodium salt, ascorbic acid, antimony potassium (+) tartrate, and disodium tetraborate were purchased from BDH (England). Gallic acid monohydrate, sodium dodecyl sulphate, copper acetate, 2, 2-diphenyl-1-picrylhydrazyl (DPPH), quercetin, and Folin–Ciocalteu reagent were obtained from Sigma-Aldrich. *N*-Cetyltrimethyl ammonium bromide (CTAB) and ammonium molybdate were purchased from Techno Pharmachem, India. All other common chemicals and solvents used were of analytical grade.

2.2. Sample Preparation. The fresh leaves of *Mangifera indica* (dauphiné, Mauritian variety) collected from the central region of Mauritius were washed with water and dried in a drying cabinet. The dried leaves were then reduced to a particle size of 1 mm using an electric grinder and stored in a well-closed plastic container for further use.

2.3. Proximate Analysis. The different chemical parameters for proximate analysis were determined based on the Association of Official Analytical Chemists (AOAC), 2005, Official Method [10].

2.3.1. Determination of Crude Protein. The total nitrogen content was obtained from a Eurovector EA 3000 elemental analyzer, and the crude protein content was calculated by multiplying the *N*% by a factor of 6.25.

2.3.2. Crude Fat Content. The fat content was extracted from the ground mango leaves (2 g) using 140 mL of petroleum ether in a Soxhlet extractor for 6 h. After extraction, the solvent was distilled off, the residue was weighed, and the percentage of fat was calculated as a percentage of the sample used.

2.3.3. Ash Content. Two grams of powdered mango leaves was heated on a hot plate in a 50 mL silica crucible which was further heated for 3 h at 525°C in a muffle furnace. The residual ash was weighed and was expressed as percentage of mass of ash with respect to mass of the original sample.

2.3.4. Determination of Fiber Content. The neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined using the Fibertec method according to the method described in Foss Application Note AN 304 (2005) and Foss Application Note AN 3429 (2005), respectively [11].

Determination of NDF involved boiling 1 g of dried powdered leaves (W_1) for 1 h in 100 mL of neutral detergent solution (disodium hydrogen phosphate, sodium tetraborate decahydrate, EDTA disodium salt dehydrate, sodium dodecyl ethoxyethanol, and anhydrous sodium sulphite), in which 1 drop of octan-2-ol and 1 or 2 drops of α-amylase were added. The resulting residue was dried at 105°C in an oven for 3 h and weighed (W_2). The residue was further ashed for 3 h in a muffle furnace, cooled, and weighed (W_3). The percentage of NDF was calculated according to the following equation:

$$\%\text{NDF} = \left[\frac{W_2 - W_3}{W_1}\right] \times 100, \quad (1)$$

where W_1 = mass of the sample, W_2 = mass of crucible + residue, and W_3 = mass of the crucible and ash.

For ADF content, 100 mL of acid detergent solution (conc. sulphuric acid and *N*-cetyltrimethyl ammonium bromide) were added to 1 g of powdered leaves (W_1) in a fritted predried Foss crucible followed by adding 1 drop of octan-2-ol, filtered, dried for 3 h at 105°C in the ovenm, and weighed (W_3). The percentage of ADF was calculated according to the following equation:

$$\%\text{ADF} = \left[\frac{W_3 - W_2}{W_1}\right] \times 100, \quad (2)$$

where W_1 = mass of the sample, W_2 = mass of the crucible, and W_3 = mass of the crucible and residue.

2.3.5. Determination of Acid Detergent Lignin (ADL). Acid detergent lignin (ADL) was determined according to the method described in Foss Application Note AN 3430 (2005) [10]. The ADF residue was soaked in conc. sulphuric acid (72%) for 3 h, ashed for 3 h at 525°C, and weighed. The percentage of ADL was calculated according to the following equation:

$$\%\text{ADL} = \left[\frac{W_3 - W_2}{W_1}\right] \times 100, \quad (3)$$

where W_1 = mass of the ADF residue, W_2 = mass of crucible + ADF residue, and W_3 = mass of crucible + ash.

2.3.6. Determination of Hemicelluloses, Cellulose, and Lignin in the Fiber and Total Digestible Nutrient. The percentage of hemicelluloses, cellulose, and lignin in the fiber was calculated as follows [12]:

$$\begin{aligned}\text{Hemicellulose} &= \%\,(\text{NDF} - \text{ADF}) = (35.32 - 34.98)\\ &= 0.34\%,\\ \text{Cellulose} &= \%\,(\text{ADF} - \text{ADL}) = (34.98 - 12.86)\\ &= 22.12\%,\\ \text{Lignin} &= \%\text{ADL} = 12.86\%.\end{aligned} \quad (4)$$

Total digestible nutrient (TDN) was computed from the following formula:

$$\begin{aligned}\text{TDN} &= 88.9 - (\text{ADF}\% \times 0.7)\\ &= 88.9 - (34.98 \times 0.7)\\ &= 64.41\%.\end{aligned} \quad (5)$$

2.3.7. Determination of Calcium and Phosphorus Content. The ash obtained as mentioned in Section 2.3.3 was double ashed using a mixture of 2 : 1 conc. HCl/HNO_3 (3 mL) in a muffle furnace at 525°C for 3 h. After the double ashing, the residue was dissolved in conc. $HCl : HNO_3$ (3 : 1) and the solution was made up to 200 mL. The obtained filtrate was used for the determination of calcium content using atomic absorption spectrophotometer (AAS) and phosphorus content using UV-Vis spectrophotometer at a wavelength of 650 nm.

2.4. Phytochemicals Screening

2.4.1. Preparation of Extracts. Ten grams of powdered leaves was extracted in the Soxhlet extractor using hexane, EtOAc, and MeOH, successively. The extracts were concentrated and stored at 4°C until further analysis.

0.5 g of each extract was dissolved in 100 mL of methanol, and these diluted extracts were qualitatively tested for the presence of different phytochemicals using standard procedures [8, 13, 14] and antioxidant activity.

2.4.2. Determination of Total Phenolic Content (TPC) and Total Flavonoid Content (TFC). The total phenolic content (TPC) of the extracts was determined using the reported procedure [15]. 0.2 mL of the extract was mixed with 0.2 mL of the Folin–Ciocalteu reagent, and 0.8 mL of sodium carbonate (2%) was added after 5 min. The samples were then incubated at room temperature. The TFC was determined using the aluminium chloride colorimetric method [16]. 0.25 mL of extract in 1.25 mL of distilled water was mixed with a solution containing sodium nitrite (0.075 mL, 5%), aluminium chloride (0.15 mL, 10%), and sodium hydroxide (0.5 mL, 1 M), and the mixture was made up to 2.5 mL with distilled water. The absorbance of solution was read at 650 nm for TPC and 510 nm for TFC using the Biochrom Libra S22 UV-Vis spectrometer. The TPC/TFC extract solutions were expressed as gallic acid/quercetin equivalent (mg/g) of ground leaves.

2.5. Antioxidant Activity. The free radical scavenging capacity of the extracts was determined using the DPPH assay [17]. 2 mL of the methanolic sample with different concentrations, ranging from 100 to 5000 ppm, was added to 2 mL of methanolic DPPH solution (4.5 g/100 mL). The absorbance was measured at 492 nm using a Labsystems Multiskan Ms. EIA reader (Thermo Fisher Scientific/lab system, California, USA) after 30 min of incubation at 37°C. Ascorbic acid and methanol were used as positive and negative controls, respectively. The ability to scavenge the DPPH was calculated using (6), where A_0 and A_s are the absorbances of the control and sample, respectively. The concentration of the sample required to scavenge 50% of DPPH was determined as follows:

$$\%\ \text{scavenging activity} = \left[\frac{A_0 - A_s}{A_0}\right] \times 100. \quad (6)$$

2.6. Antibacterial Activity. Antimicrobial tests were performed against three Gram-positive bacteria *Staphylococcus aureus* (ATCC 29213), *Staphylococcus epidermidis* (ATCC 12228), and *Bacillus cereus* (ATCC 10876) and three gram negative bacteria *Escherichia coli* (ATCC 22922), *Pseudomonas aeruginosa* (ATCC 27853), and *Klebsiella pneumoniae* (ATCC 13883) using agar disk diffusion method [18] at varying concentrations (1, 0.5, 0.25, 0.125, 0.062, and 0.031 mg/mL). The diameter of the zone of inhibition was recorded in mm.

3. Results and Discussion

Availability of proper animal feed and efficient feeding are the foundation of successful livestock production. Proper animal feeding is the supply of a diet balance in all nutrients which increases the animal productivity, quality of product, and animal welfare. The availability of accurate reliable and reproducible analytical data is imperative for proper feed formulation.

3.1. Proximate Analysis. Proximate analysis gives valuable information to access the quality of the plant material as fodder. Proximate analyses were carried out on the leaves of *Mangifera indica* grown locally to evaluate the nutritional value and to investigate whether the leaves can be used as fodder for ruminants. The proximate composition of *Mangifera indica* leaves in terms of ash, crude fat (CF), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) is given as percentage of dry matter in Table 1. The % ash content of major fodder trees has been reported to be in the range of 5–17% [19], and the ash content of locally grown *Mangifera indica* leaves was found to be 12.61% which indicates that locally grown mango leaves have a relatively high mineral content. The CF content was 3.92% which was lower than the value (8%) which is considered to be harmful to ruminants [20].

The CP content was within the range of CP values (12–14%) of top fodder trees which is regarded as the amount required to support the growth of ruminants [19]. Detergent fiber (NDF, ADF, ADL, HC, and CEL) composition of the

Table 1: Proximal composition expressed in % (dry basis) of *Mangifera indica*.

Constituents (%)	*Mangifera indica* (dauphiné Mauritian variety)	*Mangifera indica* (Nigerian variety)	*Mangifera indica* (Laos variety)	Major fodder trees (Nepal)
Ash	12.61	10	—	(5 to 17)
CF	3.92	0.48	—	—
CP	13.60	20.38	6.90	(11 to 22)
NDF	35.32	43.14	50.10	(27 to 75)
ADF	34.98	51.48	—	(20 to 54)
ADL	12.86	2.47	—	(6 to 35)
HC	0.34	8.34	—	(1 to 31)
CEL	22.12	49.01	—	(13 to 33)
Calcium	2.15	—	—	>1
Phosphorus	0.12	—	—	(1.16 to 0.40)

— = not reported.

local mango leaves together with Nigerian [5] and Laos [21] varieties are summarized in Table 1. The range of values of different parameters for the major fodder trees from Nepal are also given in Table 1 [19]. The values of ADF and ADL obtained for local mango leaves were within the range as reported for major fodder trees and therefore considered as acceptable and digestible by the ruminants. In the locally grown mango leaves, the cellulose (CEL) and hemicellulose (HC) levels were quite low. Relatively low CEL and HC have been reported in various fodder tree leaves found elsewhere [19].

3.2. Calcium and Phosphorus Content. Calcium and phosphorus are considered as macrominerals and are needed in gram quantities. In animals a Ca : P ratio above 2 helps to increase the absorption of calcium in the intestine [22]. Fodder with Ca : P ratio of 1.5–1.3 is considered to be good. However, it is reported that fodder which is too rich in calcium may hamper the absorption and assimilation of phosphorus, magnesium, zinc, copper, and other microelements. The normal requirement for calcium and phosphorus contents for ruminants is in the range of 0.19–0.82 and 0.12–0.48%, respectively [23]. The Ca : P ratio in the local mango leaves was 17.9 : 1. However, a few major fodder trees have been reported with higher calcium than that obtained for the mango leaves. High Ca : P ratio can be dangerous for ruminants, and this could be corrected by adding cereal by-products to the fodder which are low in calcium and high in phosphorus.

3.3. Phytochemical Analysis. The phytochemical screening of the crude, hexane, EtOAC, and MeOH extracts indicated the presence of an array of phytochemicals including saponins, alkaloids, phenols, tannins, flavonoids, glycosides, diterpenes and coumarins in the *Mangifera indica* leaves, and these findings were in line with data reported elsewhere [8, 9] (Table 2). Phenols, flavonoids, alkaloids, and tannins were identified mainly in the EtOAc and MeOH extracts.

Therefore, in this study, the EtOAc and MeOH extracts of the *Mangifera indica* leaves were analyzed using UPLC-MS/MS and different metabolites were identified by comparison with reference compounds/literature data/their mass spectral fragments from both the positive- and negative-ion modes.

Table 2: Summary of the phytochemical screening test performed on the different extracts.

Phytochemicals	Crude	Hexane	EtOAc	MeOH
Saponin	+	−	+	+
Alkaloids	+	−	+	+
Phenols	+	−	+	+
Tannins	+	−	+	+
Flavonoids	+	−	+	+
Steroids	+	−	−	+
Starch	−	−	−	−
Glycosides	+	+	−	−
Diterpenes	+	+	+	−
Anthocyanins	−	−	−	−
Amino acids	−	−	−	−
Coumarins	−	−	−	−

3.3.1. Phenolic Compounds. A number of polyphenolic compounds characterized as benzophenone derivatives, flavonols, xanthones, and gallotannins by their molecular weight and mass spectrometric data, and comparison with reported data are given in Tables 3 and 4 (The mass spectra used to support the findings of this study are included within the supplementary information files (available here)).

Seven phenolic compounds were identified in the protonated form, $[M+H]^+$ ions: protocatechuic acid $[C_7H_7O_4]^+$ of *m/z* 155 (entry 1), gallic acid $[C_7H_7O_5]^+$ of *m/z* 171 (entry 2), methyl gallate $[C_8H_9O_5]^+$ of *m/z* 185 (entry 3), di-*tert*-butylphenol $[C_{14}H_{22}O]^+$ of *m/z* 207 (entry 4), and tetrahydroxy sodium benzoate of *m/z* ion 209 (entry 5); based on its fragmentation pattern, ellagic acid $[C_{14}H_7O_8]^+$ (entry 8) and theogallin $[C_{14}H_{17}O_{10}]^+$ (entry 10) were detected at *m/z* 302 and 345. The entry 6 and 7 with an *m/z* value of 235 and 276 were detected in both the MeOH and EtOAc extracts, and based on their mass spectral fragmentations at *m/z* 194 (sodium gallate), 171 (gallic acid), and 153 (procatechuic acid) are most probably derivatives of gallic acid. The compound at *m/z* 329 (entry 9) is most probably a derivative of theogallin (Table 3). Another phenolic compound, sodium gallate (entry 1) was identified as a $[M-H]^-$ deprotonated molecule

Table 3: Compounds determined by UPLC-MS/MS in the EtOAc and MeOH extracts of *Mangifera indica* in the positive-ion mode.

	Identified compounds (name, formula)	Retention time	$[M+H]^+$	Mol. wt.	Fragment ions	EtOAC	MeOH	Reference
Phenolic compounds								
1	Protacatechuic, $C_7H_6O_4$	4.570	155	154	153, 125, 79	√	√	[24, 25]
2	Gallic acid, $C_7H_6O_5$	2.174	171	170	153, 127, 109, 107, 97, 81, 79, 69	√		[25, 26]
3	Methyl gallate, $C_8H_8O_5$	5.612	185	184	153, 125		√	[27]
4	2,5-Di-tert-butylphenol, $C_{14}H_{22}O$	7.148	207	206	189, 174, 159, 147, 119, 105, 91, 95, 79, 77	√	√	[28]
5	Tetrahydroxy sodium benzoate, $C_7H_5O_6Na$	4.211	209	208	191, 173, 167, 163/151/121, 107, 93, 91, 79, 77	√	√	
6	Derivative of gallic acid	18.951	235	234	194, 171, 153, 137, 118, 84, 73	√	√	
7	Derivative of gallic acid	2.022	276	275	258, 230, 212, 194, 153, 112, 97		√	
8	Ellagic acid, $C_{14}H_6O_8$	6.876	303	302	257, 229		√	[29]
9	Derivative of theogallin with one OH missing, $C_{14}H_{16}O_9$	8.121	329	328	271, 249, 235, 225, 192, 176, 153, 107	√		
10	Theogallin, $C_{14}H_{16}O_{10}$	2.257	345	344	327, 299, 209, 153, 137, 125	√	√	[30]
Xanthones								
11	Mangiferin, $C_{19}H_{18}O_{11}$	8.928	423	422		√	√	[31]
12	Mangiferin 3-methyl ether, $C_{20}H_{20}O_{11}$	8.299	437	436		√		[32]
13	Mangiferin-6'-O-gallate, $C_{26}H_{22}O_{15}$	8.875	575	574	465, 455, 439, 423, 327, 313, 303, 285, 261, 193		√	[31]
Flavonols								
14	Quercetin, $C_{15}H_{10}O_7$	9.284	303	302		√	√	[33]
15	Rhamnetin, $C_{16}H_{12}O_7$	12.036	317	316	262, 167, 156, 154, 126		√	[34]
16	Quercetin carboxylic acid, $C_{16}H_{10}O_9$	10.643	347	346	302, 284, 255, 227		√	
17	Quercetin pentoside, $C_{20}H_{18}O_{11}$	8.394	435	434	354, 311, 259, 188, 162	√	√	[31]
18	Quercetin 3-O-rhamnoside $C_{21}H_{20}O_{11}$	9.640	449	448	285, 284, 255	√	√	[31, 34]
19	Epicatechin gallate hexamalonate, $C_{22}H_{18}O_{11}$	13.983	459	458		√	√	[24]
20	Rhamnetin hexoside, $C_{22}H_{22}O_{12}$	14.067	479	478	315, 169	√	√	[30]
Benzophenones								
21	Maclurin, $C_{13}H_{10}O_6$	10.549	263	262	167, 124, 121, 111, 99, 65	√	√	[31]
22	Iriflophenone glucoside derivative	7.860	405	404	369, 351, 327, 313, 298, 273, 253, 99, 81	√	√	
23	Iriflophenone 3-C-β-D-glucopyranoside, $C_{19}H_{20}O_{10}$	7.232	409	408	391, 355, 325, 313, 231, 195, 177, 165, 121	√	√	[35]
24	Maclurin 3-C-(6"-O-p-hydroxybenzoyl)β-D-glucoside, $C_{26}H_{24}O_{13}$	9.011	545	544		√		[36]
25	Iriflophenone-di-O-galloyl-glucoside, $C_{33}H_{28}O_{18}$	9.302	713	712			√	[31, 36]
26	Iriflophenone tri-O-galloyl-glucoside, $C_{40}H_{32}O_{22}$	9.902	865	864		√		[36]
Terpenoids								
27	Lupeol, $C_{30}H_{50}O$	7.819	427	426		√	√	[37, 38]
28	Mangiferonic acid, $C_{30}H_{46}O_3$	9.575	455	454		√		[39, 40]
29	Manglanostenoic acid, $C_{30}H_{48}O_3$	14.444	457	456			√	[40]
30	Cycloart-25-ene-3,24,27-triol, $C_{30}H_{50}O_3$	13.910	459	458		√	√	[38]

Table 3: Continued.

	Identified compounds (name, formula)	Retention time	$[M+H]^+$	Mol. wt.	Fragment ions	EtOAC	MeOH	Reference
31	Cycloartane-3,24,25-triol, $C_{30}H_{52}O_3$	7.013	461	460		√	√	[38]
Other compounds								
32	Unknown	1.216	224	223	219, 201, 183, 155, 127, 99, 81		√	
33	Unknown	1.844	229	230	142, 70, 65		√	
34	Unknown	6.930	240	240	180, 174, 67	√		
37	Unknown	3.502	367	366	349, 332, 258, 229, 202, 188, 156, 144, 118, 91		√	
40	Unknown	5.884	391	392	373, 325, 249, 211, 191, 167, 149, 109, 95	√	√	
42	Unknown	9.041	567	566	549, 436, 322, 209, 114	√		

Table 4: Compounds determined by UPLC-MS/MS in the EtOAC and MeOH extracts of *Mangifera indica* in the negative-ion mode.

Entry	Identified compounds (name, formula)	Retention time	$[M-H]^-$ (m/z)	Mol. wt. (m/z)	Fragment ions	EtOAC	MeOH	Reference
Phenolic compounds								
1	Sodium gallate, $C_7H_5NaO_5$	1.567	191	192	173, 109, 93, 85	√	√	[27]
Xanthones								
2	Mangiferin, $C_{19}H_{18}O_{11}$	7.715	421	422	331, 301, 285, 272, 259, 243, 227, 183, 171, 163	√	√	[34]
3	Isomangiferin, $C_{19}H_{18}O_{11}$	6.999	421	422	331, 301	√	√	[34]
4	Mangiferin, 3-methyl ether $C_{20}H_{20}O_{11}$	9.575	435	436	313, 255, 169, 140, 124	√		[32]
5	Mangiferin, 6-O-gallate $C_{26}H_{22}O_{15}$	3.994	573	574	403, 331, 301		√	[34]
Flavonol								
6	Kaemferol, $C_{15}H_{10}O_6$	11.150	284	286	269, 240, 224, 212	√		[25]
7	Quercetin 3-O-glucoside, $C_{21}H_{19}O_{12}$	10.863	462	463	445, 419, 401, 383, 355, 301, 291, 247, 229, 218, 171, 164	√		[33]
Benzophenones								
8	Maclurin 3-C-β-D-glucoside, $C_{19}H_{20}O_{11}$	8.859	423	424	332, 302, 286, 272, 259, 227, 216, 165	√	√	[41]
9	3-Glucosyl-2,3',4,4',6-pentahydroxybenzophenone, $C_{19}H_{20}O_{11}$	4.423	423	424	314, 169, 109		√	[36]
10	Maclurin 3-C-(6'-O-p-hydroxybenzoyl)β-D-glucoside, $C_{26}H_{24}O_{13}$	9.862	543	544	271, 258, 211, 193, 190, 175, 169, 163, 150	√	√	[36]
11	Maclurin mono-O-galloyl-glucoside, $C_{26}H_{24}O_{15}$	8.574	575	576	231, 283			[31, 42]
12	Maclurin di-O-galloyl-glucoside, $C_{33}H_{28}O_{19}$	9.718	727	728	287, 270, 245, 211, 193, 169	√		[31, 42]
Terpenoids								
	Cycloartane-3,29-diol; 3β-form, $C_{30}H_{52}O_2$	3.242	443	444	237, 189, 95, 59	√		[38]
	3,27-Dihydroxycycloart-24-en-26-oic acid, $C_{30}H_{48}O_4$	12.009	472	472	256	√		[38]
Gallotannins								
	Digalloyl glucoside, $C_{20}H_{20}O_{14}$	11.865	483	484	254, 245, 153		√	
	Tri-O-galloyl glucoside, $C_{27}H_{24}O_{18}$	7.285	635	636	211, 169		√	[27, 43]
	Tetra-O-galloyl glucoside, $C_{34}H_{28}O_{22}$	9.146	787	788	618, 466, 313, 295, 211, 193, 169	√	√	[26, 43]

Table 4: Continued.

Entry	Identified compounds (name, formula)	Retention time	$[M-H]^-$ (m/z)	Mol. wt. (m/z)	Fragment ions	EtOAC	MeOH	Reference
	Penta-O-gallose-glucose, $C_{41}H_{32}O_{26}$	9.577	939	940			√	[25, 44]
Other compounds								
	Ferulic acid hexoside, $C_{16}H_{20}O_9$	12.439	355	356	183	√		[24]
	Unknown	13.582	666	667	311, 282, 267, 226, 159	√		
	Unknown	10.005	681	682	529, 331, 288, 269, 211, 169	√	√	
	Unknown	12.725	815	816	410, 309, 355, 323	√	√	
	Unknown	13.725	817	818	622, 283, 271, 255, 243	√		
	Unknown	13.867	960	961	914, 415, 397, 277, 255, 233	√		

m/z 191 and yielded ion fragments at m/z 173, 109, 93, and 85 (Table 4).

3.3.2. Xanthones. Four xanthone derivatives were detected in the MeOH and EtOAc extracts of the leaves of *Mangifera indica*. The peaks at m/z 421/423, 435/437, and 573/575 corresponded to $[M-H]^-/[M+H]^+$ ions and were attributed to mangiferin, mangiferin-methyl ether, and mangiferin-O-gallate, respectively. The other peak at m/z 421 ($[M-H]^-$ ion) corresponded to isomangiferin. The fragment ions at m/z 331 and 301 corresponded to the loss of C-glycoside phenolic compounds.

3.3.3. Flavonols. In the literature, a number of flavanol and flavanol glycosides have been detected in mango peels, bark, and seed kernels. In the EtOAc and MeOH extracts, nine flavanols have been detected in the protonated or deprotonated form. The peak at $[M-H]^-$ with m/z 285 corresponded to kaemferol while that at $[M-H]^-$ with m/z 463 was attributed to quercetin 3-O-glucoside. The other quercetin derivatives were identified in the positive-ion mode $[M+H]^+$ at m/z 303, 317, 347, 435, 449, 459, and 479 based on their mass fragmentation and by comparison with literature data (Table 3).

3.3.4. Benzophenone Derivatives and Related Compounds. Ten benzophenone derivatives were identified from the EtOAC and MeOH extracts. The compound with a $[M+H]^+$ ion at m/z 263 with a major fragment at m/z 124 was identified as maclurin. Moreover, a number of iriflophenone galloyl glucosides or maclurin glucoside derivatives were identified at m/z 409, 545, 713, and 865 (entry 23–26, Table 3) in the positive-ion mode and 423, 543, 575, and 727 (entry 8–12, Table 4) in the negative-ion mode. The compound having m/z 405 (entry 22, Table 3) in the positive-ion mode is most probably a derivative of iriflophenone glucoside based on its mass fragments.

3.3.5. Gallotannins. In the MeOH extract, a number of gallotannins were identified at m/z 483, 635, 787, and 939 as $[M-H]^-$ ions corresponding to di-O-galloyl, tri-O-galloyl, tetra-O-galloyl, and penta-O-galloyl glucosides as given in Table 4.

Table 5: Total phenolic content and total flavonoid content in crude, EtOAc, and MeOH extracts of *Mangifera indica*.

Extract	TPC (mg/g)	TFC (mg/g)	SC_{50} (μg/mL)
Crude	230 ± 2	131 ± 1	64
EtOAc	186 ± 1	191 ± 2	80
MeOH	99 ± 1	46 ± 1.0	313

3.3.6. Terpenoids. In the positive mode, the peak at m/z 427 corresponded to the terpenoid lupeol. Six cycloartane terpenoids derivatives (Tables 3 and 4) were identified from the leaves of locally grown mango leaves.

4. Biological Properties

4.1. Antibacterial. It has been reported that the phytochemicals present in the leaves of *Mangifera indica* are responsible for antibacterial, antioxidant, and anti-inflammatory activities. Previous research conducted by Doughari and Manzara [7] reported that the extracts of *Mangifera indica* showed a potent antibacterial activity against Gram-positive and Gram-negative bacteria with MIC values ranging from 12.5 to 175 mg/mL. The results of the present study revealed that the crude extract of the leaves exhibited moderate to good antibacterial properties against Gram-positive bacteria with zone of inhibition of 8.6 ± 0.6 mm (*P. aeruginosa*), 10.3 ± 1.5 mm (*B. cereus*), and 11 ± 1.0 mm (*S. aureus*), and against Gram-negative bacteria with zone of inhibition of 10 ± 1.0 mm (*S. epiderdimis*), 16 ± 0.2 mm (*K. pneumoniae*), and 20.3 ± 0.6 mm (*E. coli*) at the conc. of 1 mg/mL. The MIC values were found to be 0.062 for *S. aureus*, *B. cereus*, *E. coli*, *S. epidermis*, and *P. aeroginosa* while for *K. pneumonia,* an MIC value of 0.031 mg/mL was obtained.

Values of TPC and TFC of the crude, EtOAc, and MeOH extracts of the locally grown *Mangifera indica* are given in Table 5. The crude extract exhibited antioxidant potential

which can be related to its high TPC and TFC. The EtOAc extract showed higher antioxidant activity attributed to a more elevated concentration of phenolic and flavonoid contents as compared to the MeOH extract.

5. Conclusion

This study was conducted in a view to understand the nutritional characteristics of leaves from *Mangifera indica* trees grown in Mauritius for their use as fodder for ruminants. It is observed that mango leaves is a good source of mineral elements since it has a high percentage of ash content. The crude fat was low while crude protein was within the range required to support the growth of ruminants. The parameters (NDF, ADF, and ADL) were comparable to values reported for major fodder trees apart from HC which was found to be below the range. The phytochemical studies indicated the presence of various phenolic compounds including xanthones, flavanols, benzophenones, terpenoids, and tannins which can be the source of natural antioxidant and antibacterial agents. These scientific findings would be environmentally and economically sustainable for considering mango leaves as good fodder for feeding ruminants.

Data Availability

The mass spectra in the positive- and negative-ion modes that are used to support the findings of this study are included within the supplementary information files.

Conflicts of Interest

The authors declare that there are no conflicts of interest.

Acknowledgments

The authors thank the University of Mauritius for providing the facilities to carry out the project.

Supplementary Materials

Mass spectra of the secondary metabolites isolated from *Mangifera indica* in the positive- and negative-ion modes. (*Supplementary Materials*)

References

[1] M. Meuret, J. Boza, N. Narjisse, and A. Nastis, "Evaluation and utilization of range land feed by goat," in *Goat Nutrition*, P. Morand Fehr, Ed., Pudoc, Wageningen, Netherlands, pp. 161–170, 1990.

[2] U. B. Cheema, M. Younas, J. I. Sultan, M. R. Virk, M. Tariq, and A. Waheed, "Fodder tree leaves: an alternative source of livestock feeding," *Advances in Agricultural Biotechnology*, vol. 2, pp. 22–24, 2011.

[3] C. Orwa, A. Mutua, R. Kindt, R. Jamnadass, and S. Anthony, *Agro Forestree Database: a Tree Reference and Selection Guide Version 4.0*, World Agroforestry Centre, Nairobi, Kenya, 2009.

[4] J. B. Lowry, R. J. Petheram, and T. Budi, "Plants fed to village ruminants in Indonesia. Notes on 136 species, their composition and significance in village farming system," ACIAR Technical Reports, Australian Centre for International Agricultural Research, Canberra, NT, Australia, 1992.

[5] D. A. Ajayi, J. A. Adeneye, and F. T. Ajayi, "Intake and nutrient utilization of west African dwarf goats fed mango *(Mangifera indica)*, *Ficus (Ficus thonningii)*, *Gliricidia (Gliricidia sepium)* foliage and concentrates as supplements to basal diet of guinea grass *(Panicum maximum)*," *Journal of Agricultural Science*, vol. 1, no. 2, pp. 184–189, 2005.

[6] K. A. Shah, M. B. Patel, R. J. Patel, and P. K. Parmar, "*Mangifera Indica* (Mango)," *Pharmacognosy Reviews*, vol. 4, no. 7, pp. 42–48, 2010.

[7] J. H. Doughari and S. Manzara, "In vitro antibacterial activity of crude leaf extracts of *Mangifera indica* Linn," *African Journal of Microbiology Research*, vol. 2, pp. 67–72, 2008.

[8] O. O. Aiyelaage and M. P. Osamudiamen, "Phytochemical screening for active compounds in *Mangifera indica* leaves from Ibadan, Oyo state," *Journal of Plant Science and Research*, vol. 2, no. 1, pp. 11–13, 2009.

[9] D. O. Somkuwar and V. A. kamble, "Phytochemical screening of ethanolic extract of stem, leaves, flower and seed kernel of *Mangifera indica* L," *International Current Pharmaceutical Journal*, vol. 4, no. 2, pp. 383–389, 2013.

[10] Association of Official Analytical Chemists, *Official Method*, Association of Official Analytical Chemists, Arlington, USA, 18th edition, 2005.

[11] ANON 2005, Foss application note AN304, AN3429 and AN3430, Fibre Determination using Fibertec, 2005.

[12] D. Rivera and J. Parish, *Interpreting Forage and Feed Analysis Reports*, Extension Service of Mississippi State University, Mississippi State, MS, USA, 2010.

[13] P. Tiwari, B. Kumar, M. Kaur, G. Kaur, and H. Kaur, "Phytochemical screening and extraction: a review," *Internationale Pharmaceutica Sciencia*, vol. 1, no. 1, pp. 98–106, 2011.

[14] R. N. S. Yadav and M. Agarwala, "Phytochemical analysis of some medical plants," *Journal of Phytological Research*, vol. 3, no. 12, pp. 10–14, 2011.

[15] R. K. Sharma, S. Chatterji, D. V. Rai et al., "Antioxidant activities and phenolic contents of the aqueous extracts of some Indian medicinal plants," *Journal of Medicinal Plants Research*, vol. 3, no. 11, pp. 944–948, 2009.

[16] X. Liu, M. Zhao, J. Wang, B. Yang, and Y. Jiang, "Antioxidant activity of methanolic extract of emblica fruit (*Phyllanthus emblica* L.) from six regions in China," *Journal of Food Composition and Analysis*, vol. 21, no. 3, pp. 219–228, 2008.

[17] S. B. Moosun, S. Jhaumeer Laulloo, E. C. Hosten, T. I. A. Gerber, and M. G. Bhowon, "Antioxidant and DNA binding studies of Cu(II) complexes of N,N′-(1,1′-dithiobis (phenylene))-bis(salicylideneimine): synthesis and characterization," *Transition Metal Chemistry*, vol. 40, no. 4, pp. 445–458, 2015.

[18] A. W. Bauer, W. M. M. Kirby, J. C. Sherris, and M. Turck, "Antibiotic susceptibility testing by a standardized single disk method," *American Journal of Clinical Pathology*, vol. 45, no. 4, pp. 493–496, 1966.

[19] R. C. Khanal and D. B. Subba, "Nutritional evaluation of leaves from some major fodder trees cultivated in the hills of Nepal," *Animal Feed Science and Technology*, vol. 92, no. 1-2, pp. 17–32, 2001.

[20] N. J. Van Dyke and P. M. Anderson, *Interpreting a Forage Analysis. Alabama Cooperative Extension System*, Alabama A&M University and Auburn University, Huntsville, AL, USA, 1998.

[21] D. Kongmanila and I. Ledin, "Chemical composition of some tropical foliage species and their and digestibility by goats," *Asian-Australasian Journal of Animal Sciences*, vol. 22, no. 6, pp. 803–811, 2009.

[22] O. B. Ajayi, S. F. Akomolafe, and A. Adefioye, "Proximate analysis, mineral contents, amino acid composition, anti-nutrients and phytochemical screening of Brachystegia Eurycoma harms and pipper guineense schum and thonn," *American Journal of Food and Nutrition*, vol. 2, no. 1, pp. 11–17, 2014.

[23] L. R. McDowell, *Minerals for Grazing Ruminants in Tropical Regions, Institute for Food and Agricultural Sciences*, University of Florida, Gainesville, FL, USA, 1997.

[24] H. Palafox-Carlos, E. M. Yahia, and G. A. González-Aguilar, "Identification and quantification of major phenolic compounds from mango fruit (*Mangifera indica*, cv. Ataulfo) by HPLC–DAD–MS/MS-ESI and their individual contribution to the antioxidant activity during ripening," *Food Chemistry*, vol. 135, no. 1, pp. 105–111, 2012.

[25] M. H. Jahurul, I. S. Zaidul, K. Ghafoor et al., "Mango (*Mangifera indica L.*) by-products and their valuable components: a review," *Food Chemistry*, vol. 183, pp. 173–180, 2015.

[26] H. Kim, H. Kim, A. Mosaddik, R. Gyawali, and K. S. Ahn, "Induction of apoptosis by ethanolic extract of mango peel and comparative analysis of the chemical constitutes of mango peel and flesh," *Food Chemistry*, vol. 133, no. 2, pp. 416–422, 2012.

[27] R. H. Rasoanaivo, F. Albrieux, and M. Lemaire, "Chemical constituents of peels, kernels and hulls of fruits of *Mangifera indica* var. Hiesy and their potential valorization," *Journal of Pharmacognosy and Phytochemistry*, vol. 3, no. 4, pp. 225–233, 2014.

[28] H. L. Sani, I. Malami, S. W. Hassan, M. A. Alhassan, E. M. Halilu, and A. Muhammad, "Effects of standardized stem bark extract of *Mangifera indica L.* in Wistar rats with 2,4-dinitrophenylhydrazine-induced haemolytic anaemia," *Pharmacognosy Journal*, vol. 7, no. 2, pp. 89–96, 2015.

[29] J. E. Ramirez, R. Zambrano, B. Sepulveda, and M. J. Simirgiotis, "Antioxidant properties and hyphenated HPLC-PDA-MS profiling of Chilean pica mango fruits (*Mangifera indica L.* Cv. Piqueno)," *Molecules*, vol. 19, no. 1, pp. 438–458, 2014.

[30] E. Dorta, M. González, M. Gloria Lob, C. Sánchez-Moreno, and B. Ancos, de, "Screening of phenolic compounds in by-product extracts from mangoes (*Mangifera indica* L.) by HPLC-ESI-QTOF-MS and multivariate analysis for use as a food ingredient," *Food Research International*, vol. 57, pp. 51–60, 2014.

[31] N. Berardini, R. Carle, and A. Schieber, "Characterization of gallotannins and benzophenone derivatives from mango (*Mangifera indica L.* Cv. 'Tommy Atkins') peels, pulp and kernels by high-performance liquid chromatography/electrospray ionization mass spectrometry," *Rapid Communications in Mass Spectrometry*, vol. 18, no. 19, pp. 2208–2216, 2004.

[32] J. Qin, J. Deng, X. Feng, Q. Wang, and S. Wang, "Quantitative RP–LC Analysis of Mangiferin and Homomangiferin in *Mangifera indica L.* Leaves and in *Mangifera persiciforma* C.Y. Wu et T.L. Ming Leaves," *Chromatographia*, vol. 68, no. 11-12, pp. 955–959, 2008.

[33] S. M. R. Ribeiro, L. C. A. Barbosa, J. H. Queiroz, M. Knodler, and A. Scheiber, "Phenolic compounds and antioxidant capacity of Brazilian mango (*Mangifera Indica* L.) varieties," *Food Chemistry*, vol. 110, no. 3, pp. 620–626, 2008.

[34] A. Schieber, N. Berardini, and R. Carle, "Identification of flavonol and xanthone glycosides from mango (*Mangifera indica L.*) peels by high-performance liquid chromatography-electrospray ionization mass spectrometry," *Journal of Agricultural and Food Chemistry*, vol. 51, no. 17, pp. 5006–5011, 2003.

[35] J. A. Severi, Z. P. Lima, K. Hélio et al., "Polyphenols with antiulcerogenic action from aqueous decoction of mango leaves (*Mangifera indica L.*)," *Molecules*, vol. 14, no. 3, pp. 1098–1110, 2009.

[36] T. Tanaka, T. Sueyasu, G. Nonaka, and I. Nishioka, "Tannins and related compounds. XXI. Isolation and characterization of galloyl and p-hydroxybenzoyl esters of benzophenone and xanthone C-glucosides from *Mangifera indica L*," *Chemical and Pharmaceutical Bulletin*, vol. 32, no. 7, pp. 2676–2686, 1984.

[37] S. Shailajan, S. Menon, S. Kulkarni, and B. Tiwari, "Standardized extract of *Mangifera indica L.* leaves as an antimycobacterial and immunomodulatory agent," *Pharmacognosy Communications*, vol. 6, no. 3, pp. 137–147, 2016.

[38] V. Anjaneyulu and P. Radhika, "The triterpenoids and steroids from *Mangifera indica* Linn. Ind," *Journal of Chemistry*, vol. 39, pp. 883–893, 2000.

[39] H. X. Nguyen, T. N. Van Do, T. H. Le et al., "Chemical constituents of *Mangifera indica* and their antiausterity activity against the PANC-1 Human pancreatic cancer cell line," *Journal of Natural Products*, vol. 79, no. 8, pp. 2053–2059, 2016.

[40] A. H. Ansari, M. Ali, and K. J. Naquvi, "New manglanostenoic acids from the stem bark of *Mangifera indica* var, Fazli," *Journal of Saudi Chemical Society*, vol. 18, no. 5, pp. 561–565, 2014.

[41] Y. Zhang, L. Han, D. Ge et al., "Isolation, structural elucidation, MS profiling and evaluation of triglyceride accumulation inhibitory effects of benzophenone C-glucosides from leaves of *Mangifera indica L*," *Journal of Agricultural and Food Chemistry*, vol. 61, no. 8, pp. 1884–1895, 2013.

[42] J. C. Barreto, M. T. S. Trevisan, W. E. Hullbrito et al., "Characterization and quantitation of polyphenolic compounds in bark, kernel, leaves, and peel of mango (*Mangifera indica L.*)," *Journal of Agricultural and Food Chemistry*, vol. 56, no. 14, pp. 5599–5610, 2008.

[43] B. G. Oliveira, H. B. Costa, J. A. Ventura et al., "Chemical profile of mango (*Mangifera indica L.*) using electrospray ionisation mass spectrometry (ESI-MS)," *Food Chemistry*, vol. 204, pp. 37–45, 2016.

[44] L. S. Jiang, S. He, Y. J. Pan, and C. R. Sun, "Bioassay-guided isolation and EPR-assisted antioxidant evaluation of two valuable compounds from mango peels," *Food Chemistry*, vol. 119, no. 4, pp. 1285–1292, 2010.