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In vitro antidiabetic, antiobesity and antioxidant activities of selected endemic plants from Mount Mayon and Mount Malinao Albay, Philippines

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Abstract

The present study investigated the *in vitro* biological activities of the ethanol extracts of thirteen endemic medicinal plants in Albay against diabetes, obesity and as free radical scavenger. Alpha-glucosidase (AG), porcine pancreatic lipase (PPL) inhibition tests and DPPH radical scavenging assay were used to determine the antidiabetic, antiobesity and antioxidant properties of the extracts. Quantitative phytochemical analysis of the extracts was performed using standard methods. Of the 13 endemics, 6 plants exhibited potent inhibitory activity (>70%) against AG enzyme. *Hydnocarpus alcalae, Merremia peltata*, *Trema orientalis*, *Cascabela thevetia*, *Stachytarpheta jamaicencis*, and *Ficus septica*. *S. jamaicensis* has the lowest computed IC_{50} (0.8ug/ml) indicating highest AG inhibitory activity compared to the other plants and the drug *Acarbose* (1.88ug/ml). A total of 7 plants were found to have strong inhibitory activity of >70% against PPL: *Leea guineensis*, *Solanum torvum*, *Cheilocostus speciosus*, *Melastoma malabathricum*, *Dendrocnide meyeniana*, *Cinnamomum mercadoi*, and *Gmelina arborea*. L. *guineensis* which recorded the highest activity against PPL, contains very high amount of alkaloids (237.49mg/g AE), flavonoids (490.54mg/g QE), and phenols (220.59mg/g GAE) and also exhibited strong antioxidant activity in the DPPH assay. *M. malabathricum* has the lowest IC_{50} suggesting that it has the highest activity against PPL compared to the other plants and the drug *Orlistat* (1.81ug/ml). DPPH radical scavenging assay showed that all plant extracts possessed statistically ($p \leq 0.05$) higher antioxidant activity against ascorbic acid suggesting promising potent source of antioxidants. There is a need to conduct further study on the toxicity of the evaluated plants.

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Introduction

Non-communicable diseases (NCDs), where diabetes mellitus and obesity are leading causes of mortality have now become the main threat to global health (Perk 2017). In the country, six million Filipinos have been diagnosed to have diabetes and this fig. could double to 12 million by 2040 because of undiagnosed diabetes cases as reported by the Philippine Center for Diabetes Education Foundation (2016). Concurrently, 18 million Filipinos are obese and overweight, according to a report released in 2016 by Asia Roundtable on Food Innovation for Improved Nutrition. Statistics likewise shows that 22.3% of Filipino adults are overweight and 6.1% are obese (FNRI, 2011). In the region particularly, there is high incidence of lifestyle related illnesses and metabolic disorders which is further aggravated by expensive medicines and inadequate health services (RDP, 2011). It is therefore necessary to address the health situation of the Albayanos and seek affordable alternative therapies from available natural local sources.

Investigation of natural products is a research field with great potential and is especially important in developing countries possessing great biodiversity, like the Philippines, having 2/3 of the earth's biodiversity and about 70-80% of the world's plant and animal species (CBD, 2009). To date, despite increasing research efforts leading to the discovery of many bioactive compounds from terrestrial plants including flavonoids, terpenoids, carotenoids, phytosterols, isothiocyanates and other phytochemicals (Lynn *et al.*, 2006; Hayes and Eggleston, 2008; Ragasa *et al.,* 2009; Raga *et al.*, 2011; Macabeo *et al.*, 2013), it is understandable that there is still a huge number of plant compounds that are not well investigated pharmacologically in the approximately 310,000 plant species described so far (IUCN, 2015). In the province of Albay, Mount Malinao and Mount Mayon are considered as key biodiversity areas that support endemics, abundant, indigenous and medicinal species. Phytochemical and pharmacological studies of species that are abundant in the locality and with reported medicinal uses are therefore promising areas for exploration.

This research is one of the first assessments of the *in vitro* biological activities of these endemic medicinal plants: (1) *Gmelina arborea* (Lamiaceae); (2) *Solanum torvum* (Solanaceae); (3) *Stachytarpheta jamaicensis* (Verbenaceae); (4) *Cascabela thevetia* (Apocynaceae*);* (5) *Melastoma malabathricum* (Melastomataceae); (6) *Dendrocnide meyeniana* (Urticaceae); (7) *Cinnamomum mercadoi* (Lauraceae); (8) *Cheilocostus speciosus* (Costaceae); (9) *Trema orientalis* (Ulmaceae); (10) *Leea guineensis* (Vitaceae); (11) *Ficus septica* (Moraceae); (12) *Hydnocarpus alcalae* (Achariaceae); and (13) *Merremia peltata* (Convolvulaceae).

In this study, the 13 endemic medicinal plants were evaluated for their phytochemical components, antioxidant property, alpha-glucosidase and lipase inhibition activities using a simple, fast, efficient, and reliable spectrophotometric method. The plant extracts were also compared with standard chemical agents (e.g. Ascorbic acid, Acarbose and Orlistat) in order to assess their potential use as an alternative to these drugs.

Materials and methods

The study conducted comparative *in vitro* screening of the antioxidant, anti-diabetic and anti-obesity activity of thirteen (13) endemic medicinal plants from Mount Mayon and Mount Malinao Albay. The thirteen plants were selected because of their medicinal use, availability and endemicity.

Collection of plant samples and preparation of extracts

Leaves of the thirteen endemic medicinal plants were obtained from Mt. Mayon and Mt. Malinao in Albay. The plants were identified and authenticated at Jose Vera Santos Memorial Herbarium Institute of Biology, University of the Philippines, Diliman Quezon City. Preparation of extracts and all analyses were done at College of Science Bicol University, Legazpi City. Prior to extraction, the different leaf samples were washed, air-dried and reduced to fine particles using Osterizer™ blender. The pulverized samples, soaked in 95% ethanol (1:2 w/v) for 72 hours were filtered and concentrated by evaporation under reduced pressure at 45^{o}C using rotary vacuum evaporator.

The total ethanol extract concentrate yield per gram of dried plant material was determined using the formula: weight (g) of dried extract/dry-weight (g of plant material) x 100

Antioxidant Activity

DPPH assay

To evaluate free radical scavenging activity, sample stock solutions (1mg/ml) were serially diluted to prepare concentrations of 1000µg/ml, 500µg/ml, 250µg/ml, 125µg/ml, 62.5µg/ml and 31.5µg/ml of the extracts. About 50µL various concentrations of plant ethanolic extracts were mixed with 150 µL 0.1mM DPPH-ethanol solution. Ascorbic acid was used as a positive control and blank control was prepared with ethanol and DPPH. After 30 minutes of incubation at room temperature in the dark, the absorbance at 517nm was measured using BIOBASE-EL10B ELISA reader. The DPPH radical scavenging activity of the samples was calculated using the following formula described by Sroka and Cisowski (2005):

DPPH scavenging activity (%) = (Absorbance of sample)/(Absorbance of control) X 100.

Concentrations of 250, 125, 62.5, and 31.25ug/ml were used to determine the effective concentration (EC50) to scavenge the DPPH radical by 50%.EC50 were calculated using linear regression analysis described by Mensor *et al.* (2001) where plots were generated with the abscissa represented the concentration of tested plant extracts and the ordinate the scavenging activity from three independent experiments.

Anti-diabetic Activity

α-Glucosidase inhibition assay

Crude extracts from 13 identified plants were investigated for its α-glucosidase inhibitory activity using a Glucosidase Activity Assay Kit (Sigma Aldrich Co Ltd St. Louis, MO, USA). Samples were dissolved in dimethyl sulfoxide (DMSO) at various concentrations. 20 µl of each sample extracts were transferred at separate wells. The samples were treated with 200 µl Assay buffer, pH 7.0 and 8 µl (4-Nitrophenyl-b-D- glucuronide) a-NPG Substrate. The initial absorbance of the released p-nitrophenol was measured at 405nm using ELISA microplate reader (Biobase EL10B). Samples were incubated at 37 °C and final absorbance was measured at 405nm after 20 minutes. The inhibition of □-glucosidase activity in the sample was calculated based from the formula described by Hyun *et al.,* (2015)

$$\text{Inhibition rate (\%)} = 1 - \frac{\text{Abs sample - Abs blank}}{\text{Abs control}} \times 100$$

Where Abs sample represents the absorbance of the extracts after 20 minutes, Abs blank denotes the absorbance of water added with calibrator, and Abs control represents the absorbance of the water (control).

Anti-obesity Activity

Lipase inhibition assay

The inhibition of lipase by the ethanolic extracts of the selected plant species was determined using a Lipase Activity Assay Kit (Sigma Aldrich Co Ltd St. Louis, MO, USA). Lipase activity was measured using a coupled enzyme reaction, which results in a colorimetric (570nm) product proportional to the enzymatic activity present. One unit of lipase is the amount of enzyme that will generate 1.0mmole of glycerol from triglycerides per minute at 37°C. Plant samples were diluted in DMSO and centrifuged at 10,000 x g for 10 minutes to remove insoluble materials. The inhibitory activities of the plant extracts and Orlistat were measured at concentrations of 1000, 500, 250, 125, 62.5 and 31.25ug/ml. Samples and standard solutions were adjusted to final volume of 50ul with Lipase Assay Buffer into a 96 well plate. 100ul of the reaction mix was added to each well by pipetting. After the mixture was incubated at 37°C for 2-3 minutes, the initial measurement was read at 570nm using ELISA EL10B. The mixture was further incubated until it reached the final measurement. The final absorbance measurement for calculating the enzyme activity was the penultimate reading at 30 minutes. The plate was protected from light during the incubation. The measurements were performed in triplicate. The inhibitory activity (%I) was calculated according to the following formula (Dechakhamphu and Wongchum, 2015), where A is the activity of the

enzyme initial absorbance, and a is the 0 blank control initial absorbance; B is the activity of the enzyme final absorbance, and b is the 0 blank control final absorbance:

$$1\% = (1 - \frac{B - b}{A - a}) \times 100$$

The half maximal inhibitory concentration (IC_{50}) determination

The IC50 value of the extracts was determined at a concentration of 1000, 500, 250, 125, 62.5, and 31.25ug/mL. *Acarbose* and *Orlistat* were used as a positive control. IC50 value was calculated by the following formula (Dechakhamphu and Wongchum, 2015), where Low Inh%/HighInh% signify% inhibition directly below/above 50% inhibition, and Low Conc/High Conc are the corresponding concentrations of extract.

$$IC50 = (\frac{50\% - \text{low Inh}\%}{\text{high Inh}\% - \text{low Inh}\%}) \times (\text{high conc} - \text{low conc}) + \text{low conc}$$

Phytochemical Screening

Determination of Total Alkaloids

Spectrophotometric method was used to determine the total alkaloids using a Perkin Elmer Lambda 35 Double-beam UV/Vis Spectrometer. A 1000 ppm atropine stock solution was prepared to determine the standard calibration curve. Samples were prepared by dissolving 0.1 g of dried extract with 10mL 200-proof ethanol solvent. Sample, standard (20-150µg/mL) and blank (ethanol) solutions were prepared by mixing 1mL aliquot with 5mL phosphate buffer (pH 4.7) and 5mL 0.1mM Bromocresol Green solution. The mixture was transferred to a funnel and three successive portions of 5, 3, then 2mL chloroform were used for solvent extraction. The chloroform portions were combined to make up a 10mL solution. Absorbance of each solution in quartz sample cell was read against the blank at 470nm. Concentrated plant extract was prepared for sample with absorbance below the linear range of the calibration curve. The total alkaloids are reported with mean values of three replicates in milligrams atropine equivalent (AE) per gram of dried extract.

Determination of Total Flavonoids

The total flavonoid contents of the extracts were measured by the aluminum chloride colorimetric assay as adopted from Sulaiman and Balachandran (2012). Briefly, a 1000 ppm quercetin stock solution was prepared for the standard calibration curve. Samples were then prepared by dissolving 0.1 g of dried extract with 25mL 200 proof ethanol solvent. Sample, standard (10-80µg/mL) and blank (ethanol) solutions were added with 4mL of distilled water and 300µL 5% $NaNO_2$. After five minutes, 300 µL 10% $AlCl_3$ was added to the mixture. For another 5 minutes, 2mL 1M NaOH was added to the mixture and diluted to 10mL with distilled water. Absorbance was read at 550nm using Biobase EL10B Microplate Reader. The total flavonoids are reported as milligrams quercetin equivalent (QE) per gram of dried extract.

Determination of Total Phenols

The total phenolic composition of the ethanolic extracts was determined using the Folin-Ciocalteu reagent as first described by Singleton *et al.* (1999). Briefly, a 1000ppm gallic acid stock solution was prepared to determine the standard calibration curve. Samples were then prepared by dissolving 0.1 g of dried extract with 25mL 200-proof ethanol. Sample, standard (10-50µg/mL) and blank (ethanol) solutions were added with 0.4mL Folin-Ciocalteu reagent. After 5 minutes, 4mL 7% Na_2CO_3 was added to the mixture. The mixture was incubated for 90 minutes before diluting to 10mL with distilled water. Sample solutions were further filtered before reading the absorbance at 550nm using Biobase EL10B Microplate Reader. Solutions not within the linear range of the curve were either diluted or a concentrated. Total phenols are reported as milligrams gallic acid equivalent (GAE) per gram of dried extract.

Statistical Analysis

Statistical analysis of the data was performed using the SPSS 21.0 program. Data are expressed as mean ± SEM. Statistical comparisons were assessed by the Mann–Whitney U test. Values of p□0.05 were considered significant.

Results and discussion

Phytochemical screening

Preliminary phytochemical analysis of the ethanolic extracts of the 13 endemic medicinal plants was determined by qualitative analyses. Phytoconstituents namely alkaloids, flavonoids and phenols were detected in the extracts. Results of the present study reflected that the quantity of alkaloids, flavonoids and phenols vary from one plant to another, and this finding is in agreement with previous studies (Srivastava *et al.,* 2012 and Liu 2008).

The data obtained after analysis of total alkaloids as shown in Table 1 was largely variable among the plants. The highest alkaloid content was found in *Stachytarpheta jamaicensis* (344.50mg/g) followed by *Trema orientalis*, *Hydnocarpus alcalae, Leea guineensis, Solanum torvum, Cinnamomum mercadoi* and *Gmelina arborea* with the values being 316.80, 271.06, 237.49, 154.23, 109.19 and 97.89mg/g, respectively.

Comparison of the flavonoid content of the 13 plants revealed that the leaves of *L. guineensis* were found to have maximum flavonoid content followed by leaves of *T. orientalis, M. peltata, H. alcalae, M. malabathricum, C. mercadoi, C. thevetia, G. arborea, D. meyeniana, C. speciosus, S. torvum, S. jamaicencis* and *F. septica*, with the minimum flavonoid content. Measurement of the total phenolic composition of the extracts suggested that the maximum phenolics (ug/ml) was found in the leaves of *C. thevetia* (237.80) followed by *L. guineensis* (220.59), *S. jamaicensis* (187.13), *M. peltata* (141.41), *D. meyeniana* (104.68), *C. speciosus* (94.53), *S. torvum* (89.48), *G. arborea* (86.16), *C. mercadoi* (70.43), *M. malabathricum* (63.90), *H. alcalae* (57.12). Relatively low level of phenols was detected in the leaves of *T. orientalis* and *F. septica*.

Table 1. Extraction yield, total alkaloids, flavonoids and phenols content of leaf extracts of endemic medicinal plants in Albay.

Scientific Name	Family	Extraction yield (EtOH)	Total Alkaloids	Total Flavonoids	Total Phenols
		% Yield (W/W)	AE (mg/g)	QE (mg/g)	GAE (mg/g)
Gmelina arborea	Lamiaceae	4.32	97.89±5.61	202.58±1.36	86.16±4.49
Solanum torvum	Solanaceae	9.9	154.23±5.0	127.02±1.82	89.48±6.51
Stachytarpheta jamaicensis	Verbenaceae	1.16	344.50±44.54	104.49±0.79	187.13±7.83
Cascabela thevetia	Apocynaceae	5.23	53.86±6.46	203.69±3.59	237.80±6.06
Melastoma malabathricum	Melastomataceae	2.49	31.12±12.67	267.87±2.88	63.90±1.58
Dendrocnide meyeniana	Urticaceae	7.85	46.80±3.98	201.22±1.71	104.68±7.73
Cinnamomum mercadoi	Lauraceae	7.11	109.19±3.00	221.28±1.33	70.43±1.05
Cheilocostus speciosus	Costaceae	3.1	18.63±2.14	156.74±3.12	94.53±2.52
Trema orientalis	Ulmaceae	10.67	316.80±41.19	391.39±5.53	35.32±0.62
Leea guineensis	Vitaceae	4.79	237.49±21.17	490.54±1.52	220.59±1.87
Ficus septica	Moraceae	2.99	62.78±6.57	77.11±4.11	5.01±0.02
Hydnocarpus alcalae	Achariaceae	1.73	271.06±2.68	329.90±1.14	57.12±1.87
Merremia peltata	Convolvulaceae	10.02	62.36±13.33	338.24±1.44	141.41±0.73

AE – Atropine equivalent, QE – Quercetin Equivalent, GAE – Gallic Acid Equivalent All result are reported as mean ± SEM in milligrams per gram of dried extract, N = 3

Antioxidant activity

The ethanol extracts of 13 endemic medicinal plants showed statistically higher antioxidant activity towards the DPPH radical compared with the standard ascorbic acid at higher concentrations (1000ug/ml-250ug/ml) and comparable to ascorbic acid at lower doses (Table 2). At high dose (1000ug/ml) higher scavenging activity was observed in *M. peltata* followed by *S. jamaicensis* and *G. arborea*. For EC50, lowest EC50 was observed in *M. peltata* followed by *C. speciosus, S. jamaicensis, C. mercadoi, G. arborea, D. meyeniana, M. malabathricum, T. orientalis, C. peruviana, S. torvum, F. septica, L. guinensis,* and *H. alcalae* with the highest EC_{50}. An EC_{50} value is the concentration that is required to scavenge 50% of the free radicals in

the system that is inversely proportional to the antioxidant activity (Sahu, Kar and Routay, 2013). Thus, *M. peltata* with the lowest EC_{50} is shown to possess the highest activity compared to other plants. To the authors' knowledge, this is the first report of the antioxidant activity of *M. peltata* collected in Bicol region or in the Philippines. Antioxidants compounds such as alkaloids, flavonoids and saponins were observed in *M. peltata* collected in Iligan, Philippines (Perez *et al.*, 2015).

Table 2. DPPH scavenging activity of 13 Endemic Medicinal plants of Albay at different concentration.

Plant extracts	1000ug/ml	500ug/ml	25ug/ml	125ug/ml	62.5ug/ml	31.25ug/ml	*EC50±SEM (ug/ml)
G. arborea	293.00±29.00*	227.52±37.68*	181.12±32.66*	98.83±16.42*	57.51±8.23*	34.28±4.93	95.07±60.04
S. torvum	194.72±55.01*	174.1±55.87*	123.53±40.30*	72.45±19.16	43.13±7.96	22.79±1.51	243.03±129.71
S. jamaicensis	299.25±12.02*	212.08±23.13*	123.10±7.99*	74.89±4.26*	45.49±3.00*	31.35±2.21*	74.77±11.09
C. thevetia	185.38±30.12*	128.88±25.36*	73.29±10.19*	42.79±4.00*	28.74±2.39	23.35±1.33	193.46±66.41
M. malabathricum	241.06±19.16*	133.02±13.31*	75.89±5.15*	50.42±2.45*	38.70±1.52*	33.32±1.34	127.71±23.09
D. meyeniana	164.91±42.24*	140.42±22.29*	64.21±14.02*	37.92±6.20	36.53±1.17	39.06±4.62	113.38±70.68
C. mercadoi	162.38±39.55	129.21±17.04*	62.97±11.25*	43.16±5.38*	37.18±2.93	41.73±3.75	76.92±33.12
C. speciosus	131.85±37.81*	161.57±33.38*	49.89±8.16*	35.23±2.00	33.22±4.74	41.10±10.65	73.18±74.27
T. orientalis	179.53±31.17*	203.21±28.69*	83.01±5.98*	53.59±2.01	62.26±9.45*	48.94±10.09	157.17±61.75
L. guineensis	213.93±32.83*	131.28±31.59*	102.79±9.89*	134.32±28.80	99.93±25.54*	74.08±17.29	274.51±168.01
F. septica	180.84±31.18*	95.14±15.50*	54.91±5.73*	35.75±3.01	26.85±2.00	17.49±2.54*	255.40±79.65
H. alcalae	179.47±36.61*	125.41±40.82*	95.32±30.52*	52.29±14.60	34.14±7.79	25.95±4.69	312.69±135.17
M. peltata	312.69±3.87*	229.22±15.07*	128.13±10.16*	76.46±4.14	53.52±2.56	37.76±2.87*	61.99±12.64
Ascorbic acid	29.79±6.40	25.89±2.60	27.18±3.34	28.23±3.24	27.03±2.89	30.23±3.44	

*significant to ascorbic acid

*values obtained from regression lines with 95% of confidence level

Anti-diabetic activity

The alpha-glucosidase (AG) inhibitory activity of the plant extracts was determined using Acarbose as the standard drug (Table 3.) A total of six (6) plants exhibited potent inhibitory activity (>70%) against alpha-glucosidase enzyme. *Hydnocarpus alcalae* belonging to Family Achariacea showed the highest inhibitory activity (91.67%), the other plants include: *Merremia peltata* belonging to Family Convulvolaceae (89.76%); *Trema orientalis* belonging to Family Ulmaceae (84.56%); *Cascabela thevetia* from Family Apocynaceae (79.59%); *Stachytarpheta jamaicencis* from Family Verbenaceae (75.08%); and *Ficus septica* from Family Moraceae (73.93%). As per the results of the phytochemical screening, all these plants contained high amount of total flavonoids (391.39 to 77.11mg/g QE) together with the other phytoconstituents considered in the study, alkaloids and phenols at varying concentration. Interestingly, *H. alcalae* has a total flavonoid content of 329.9mg/g QE and total alkaloid value of 271.06mg/g AE.

Moderate AG inhibition was observed in the extracts of *Cheilocostus speciosus* (67.66%) and *Solanum torvum* (52.45%). Weak inhibitory activity of <50% against AG was recorded in the leaf extracts of *Leea guineensis* (44.11%), *Cinnamomum mercadoi* (32.57%), *Melastoma malabathricum* (21.03%), and *Gmelina arborea* (1.91%). Negative AG inhibitory effect was detected from the extracts of *Dendrocnide meyeniana*, notwithstanding the presence of high amount of total flavonoids (201.22mg/g QE) and total phenols (104.68mg/g GAE) quantified in the phytochemical test.

The different concentration of the ethanol extracts were measured for IC_{50} at a concentration of 1000, 500, 250, 125, 62.5 and 31.25ug/ml. *S. jamaicensis* has the lowest IC_{50} value of 0.8ug/ml suggesting that it has the highest activity compared to the other plants and the drug Acarbose with IC_{50} value of 1.88ug/ml.

Table 3. α-Glucosidase inhibitory effects of 13 selected medicinal plants.

Scientific Name	Family	Inhibition (%)
Gmelina arborea	Lamiaceae	1.91±6.55*
Solanum torvum	Solanaceae	52.45±14.19
Stachytarpheta jamaicensis	Verbenaceae	75.08±1.53
Cascabela thevetia	Apocynaceae	79.59±1.0
Melastoma malabathricum	Melastomataceae	21.03±14.67
Dendrocnide meyeniana	Urticaceae	-0.76±50.84*
Cinnamomum mercadoi	Lauraceae	32.57±14.66
Cheilocostus speciosus	Costaceae	67.66±3.97
Trema orientalis	Ulmaceae	84.56±7.64
Leea guineensis	Vitaceae	44.11±3.05
Ficus septica	Moraceae	73.93±1.07
Hydnocarpus alcalae	Achariaceae	91.67±0.61
Merremia peltata	Convolvulaceae	89.76±1.90
Acarbose		99.47±0.27

*P <0.05 compared to *Acarbose*, data were presented as mean ± SEM (n = 3). The final concentration of the extracts used in this experiment was 125ug/mL.

Anti-obesity activity

The porcine pancreatic inhibitory (PPL) activity of the plant extracts was determined using Orlistat as the standard drug (Table 4). Among the 13 endemics, 7 plants were found to have strong inhibitory activity of >70% against porcine pancreatic lipase (PPL): *Leea guinnensis* (89.67%), *Solanum torvum* (89.07%), *Cheilocostus speciosus* (86.15%), *Melastoma malabathricum* (85.36), *Dendrocnide meyeniana* (85.12%), *Cinnamomum mercadoi* (81.35%), and *Gmelina arborea* (71.49%). The strong PPL inhibitory activity is associated with the presence of flavonoids, phenols and alkaloids in these plants, consistent with the results of the phytochemical test. Likewise, study results revealed that, *L. guineensis* which recorded the highest activity against PPL, contains very high amount of alkaloids, flavonoids and phenols and also exhibited strong antioxidant activity in the DPPH assay (Table 2).

Table 4. Lipase inhibitory effects of 13 selected medicinal plants.

Scientific Name	Family	Inhibition (%)
Gmelina arborea	Lamiaceae	71.49±7.52
Solanum torvum	Solanaceae	89.07±4.58
Stachytarpheta jamaicensis	Verbenaceae	57.48±8.14
Cascabela thevetia	Apocynaceae	15.30±20.41*
Melastoma malabathricum	Melastomataceae	85.36±8.50
Dendrocnide meyeniana	Urticaceae	85.12±11.32
Cinnamomum mercadoi	Lauraceae	81.35±2.94
Cheilocostus speciosus	Costaceae	86.15±6.35
Trema orientalis	Ulmaceae	18.28±8.47*
Leea guineensis	Vitaceae	89.67±1.11
Ficus septica	Moraceae	53.00±16.09
Hydnocarpus alcalae	Achariaceae	42.99±2.055*
Merremia peltata	Convolvulaceae	49.90±0.69*
Orlistat		96.97±17.81

*P <0.05 compared to *Orlistat*, data were presented as mean ± SEM (n = 3). The final concentration of the extracts used in this experiment was 125ug/mL.

Previous studies supported the positive correlation between alkaloid, flavonoid and phenolic contents and PPL inhibition activity (Chedda *et al.,* 2016; Adnyana 2014; Gupta *et al.,* 2012). Moderate PPL inhibition was observed in the extracts of *Stachytarpheta jamaicensis* (57.48%), and *Ficus septica* (53%). The extracts of *Merremia peltata and Hydnocarpus alcalae,* recorded <50% inhibition of PPL. The 2 plants: *Trema orientalis* and *Cascabela thevetia* exhibited weak inhibitory activity of <20% against porcine pancreatic lipase. Surprisingly, these 2 plants contained sufficient quantity of the major phytoconstituents (Table 1) present in combination of either: high alkaloids/flavonoids and low phenols or high flavonoids/phenols and low alkaloids. The findings of the study propose that high phenolic or alkaloid content and strong antioxidant activity is not necessarily connected with relevant anti-lipase activity. Moreover, plants contain other bioactive components including polyphenols, terpenes, and saponins.

Recent phytochemical studies revealed the presence of many saponins (Marrelli *et al.*, 2016; Hwang *et al.*, 2013; Hernandez-Carlos *et al.*, 2012) and terpenes (Bustanji *et al.*, 2011) with anti-lipase properties.

The different concentration of the plant extracts were measured for IC_{50} at a concentration of 1000, 500, 250, 125, 62.5 and 31.25ug/ml. The IC_{50} is defined simply as the inhibitor concentration that decreases the biotransformation of a substrate at a single, specified concentration by 50% (Pharmacology, 2009). The extracts of *M. malabathricum*, *D. meyeniana*, *C. mercadoi*, *G. arborea, C. speciosus, S. torvum,* and *L. guineensis* had IC_{50} values of 1.03, 1.07, 1.19, 1.2, 1.31, 1.35, and 2.14u g /ml, respectively. Whereas, *Orlistat* had IC_{50} value of 1.81ug/ml. *M. malabathricum* has the lowest IC_{50} suggesting that it has the highest activity compared to the other plants and the drug *Orlistat*.

Conclusion and recommendations

The thirteen (13) endemic medicinal plants collected from Mount Mayon and Mount Malinao Albay showed strong *in vitro* antioxidant capacity consistent with the results of the phytochemical screening. The general assessment of the analytical results for the plant extracts definitely shows the individual specificity of each plant sample and a broad range of alkaloid, flavonoid and phenolic compounds. To the best of our knowledge, the plants studied in this paper have not been screened earlier for *in vitro* biological activities. Results of the study propose that all the plant extracts can be considered as good source of natural antioxidants. Six plants are potent alpha glucosidase inhibitors and seven plants are strong inhibitors of porcine pancreatic lipase.

Conflict of interest

The authors declare that there was no conflict of interest

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