PHYTOCHEMICAL SCREENING OF SELECTED INDIGENOUS EDIBLE PLANTS FROM THE TOWNS OF ISABELA, PHILIPPINES

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ABSTRACT

Present investigation conducted phytochemical screening of indigenous edible plants collected from the local markets of the six towns of Southern Isabela Philippines namely: Echague, San Isidro, Jones, San Agustin, Santiago and Cordon. Standard procedures of chemical screening were performed on the selected indigenous food plants scientifically known as: Diplazium esculentum, Amaranthus viridis Linn, Artocarpus altilis, Phaseolus ilocanus Blanco, Amorphophallus campanulatus Blume, Syzygium lineatum (DC.) Merr. & L.M. Perry, Lycopersicon esculentum var. cerasiforme, Schizophyllum commune, Colocasia esculenta Lin, Momordica chinensis, Phyllanthus acidus (L.) Skeels, Sesbania grandiflora (Linn.) Pers.), Cajanus cajan (Linn.) Mill, Phaseolus vulgaris and Psophocarpus tetragonolobus. Wide range of chemical classes was found in the fifteen plant extracts as revealed by the appearance of certain color or precipitate by the chemical tests applied in the phytochemical screening. Such extracts revealed dominantly the occurrence of saponins (10 plants); tannins and flavonoids (9 plants); phenolics and alkaloids (8 plants). Triterpenes (5 plants); glycosides and sterols (3 plants) were also detected. Anthraquinones, diterpenes and anthocyanins were found to be present in two plants for each metabolite. The current findings revealed the richness of the indigenous edible plants with different groups of active compounds that need to be studied for different applications to achieve nutritional security.

Keywords: Indigenous edible plants, secondary metabolites, phytochemical screening.

INTRODUCTION

Climate change and population growth in many developing countries impede progress toward achieving food and nutritional security (Tirado, et al 2013)[1]. Meanwhile, diversified diets, based on a range of crop species which includes fruits and vegetables, are essential for nutritional security. In spite of the growing body of evidence which highlights the protective effect of fruits and vegetables, their intakes are still inadequate in many low and middle-income countries.

The Philippines' produce consumption of 60 kg per person per year in 2007 was one of the lowest in Asia, according to the World Health Organization (WHO). Vegetable expenditures are allocated over the following vegetable commodities: cabbage, water spinach, horse radish tree leaves, Chinese white cabbage, bitter gourd, eggplant, okra, tomato, hyacinth bean, mungbeans, string beans and other vegetables. These commodities are the most commonly-used vegetables among all households in the Philippines and they account for 78% of total vegetable expenditures (Mutuc, M. et al, 2006)[2].

A nutritious and varied diet is a critical means by which good health can be maintained. Thus, World Health Organization (WHO, 2004)[3] recommends 400 grams per day or 146 kg per year of vegetables and fruits to help prevent various non-communicable diseases such as gastrointestinal cancer, heart disease and stroke. Consumption of less than 200 g of vegetables per person per day in many countries today is common and this low amount, often in conjunction with poverty and poor medical services, is associated with unacceptable levels of mortality and malnutrition in preschool children and other vulnerable groups (Hall et al, 2009)[4].

An increase in the availability, affordability and consumption of nutrient-dense vegetables is one way malnutrition may be substantially reversed. Vegetables or food plants are an excellent source of vitamins and micronutrients; increasing consumption of these can help alleviate malnutrition from imbalanced diets wherever it occurs, in developing countries as well as in developed countries. The contribution of local plant foods in reducing health risks has always been recognized as part of the local knowledge which forms a greater part of the complex cultural system. Research has shown that many edible plants are rich in specific constituents, referred to as phytochemicals that may have health promoting effects

Food plants are high value cash crops that generate employment and income, and contribute to gender equity and better livelihoods. Many of these, including indigenous edible plants in particular – have high levels of micronutrients and could significantly contribute to nutritional security if eaten as part of the daily diet. Many of the plants adapt easily to degraded soils and to drought-prone, flooded, or saline land, and can be more resilient to extreme climatic events. Despite the perceived importance of the indigenous plants in combating malnutrition and poverty, and despite the wealth of traditional knowledge about these species, many are still poorly studied and understood by the community because their value is not appreciated, particularly in regions where the plants are not native.

Keeping this in view, selected indigenous edible plants found in the local market of Isabela Philippines were subjected to phytochemical screenings to gather relevant information regarding the chemical constituents that the edible plants contain to achieve a better understanding of the indigenous resources promoting both their sustainable utilization and their protection.

MATERIALS AND METHODS

Sampling Area and Collection of Samples

The samples were collected from the public markets of six (6) towns of District IV of the province of Isabela, Philippines. The sampling site is shown in Figure 1 encircled by the violet line. The towns include Echague, San Isidro, Santiago, Jones, San Agustin and Cordon. Samples were collected during the designated market days for the town.

Informal interview was used to collect information from the vendors about when, how and where they obtained the plants they sell in the market. Local names of the plants, edible parts, cooking and eating methods, knowledge of health effects, sources, seasonal availability and the approximate prices for retail at the market (Oselebe et al 2013)[5] were also obtained. Whenever possible, elderly vendors were selected for the interview, the perceived efficacy of the remedies was mostly asked directly. Photographs and actual sample of the plants on sale were taken.

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Figure 1. Map showing the sampling area

Plant Selection and Identification

Identification of the plant samples used for analysis was done with the use of references such as Merrill (2013)[6] Quisumbing (1978)[7], Rummel (2009)[8], Pelser (2011)[9] and International Plant Name Index (IPNI) [10]. Plants included in the priority list of Department of Agriculture-Bureau of Agricultural Resources (DABAR) program were selected for analysis including other indigenous plants with ethnomedical information provided by the vendors specifically on the type of disease that the indigenous plants can heal.

Preparation of Plant Extracts

The edible parts as described by the vendors were used in the analysis to mimic as closely as possible the traditional preparations (Fabricant and Farnsworth 2001)[11]. The parts used for the analysis includes the young shoots for *Diplazium esculentum, and Amaranthus viridis* Linn; the pulp from immature fruits for *Artocarpus altilis*, the seeds of *Phaseolus ilocanus* Blanco, *Phaseolus vulgaris*; the ripe fruits of *Lycopersicon esculentum var. cerasiforme*, whole fruits of *Momordica chinensis* and *Psophocarpus tetragonolobus*; ripe fruits of *Phyllanthus acidus* (L.) Skeels, seeds of *Cajanus cajan* (Linn.) Mill, and ripe fruits of *Syzygium lineatum (DC.) Merr. & L.M. Perry*; stalks were used for *Amorphophallus campanulatus* Blume; whole plants for *Schizophyllum commune*; root runners for *Colocasia esculenta* Lin; and flowers for *Sesbania grandiflora* (Linn.) Pers.).

The edible parts were isolated from the whole plant, washed, cut and air dried in a room with circulating fan for continuous air flow. The dried samples were then ground finely and kept in air tight container for further processing. The processed samples were soaked separately in sufficient amount of 95% ethyl alcohol (Njume et al 2011[12]; Masoko et al 2010[13]) using a conical flask, then allowed to stand for three days at room temperature then filtered off . Extraction was repeated to completely extract the active constituents. The filtrates were

concentrated using a rotary evaporator, at temperature between 45^0C to 50^oC. The crude extracts were then transferred into clean petri dishes and placed over a water bath at a temperature of 40^0C to remove traces of solvent. The extracts obtained were kept aseptically in an airtight container to avoid loss of volatile principles until the extract's further use.

Phytochemical Screening

Phytochemical screening was performed using standard procedures as described below.

1. ***Screening for Alkaloids***. Meyer's reagent (potassium mercuric iodide) 1.36 gm of mercuric chloride was dissolved in 60 ml of distilled water and 5 gm of potassium iodide was dissolved in 10 ml of water. These two solutions were mixed and diluted to 100 ml with distilled water. To 1 ml of the extract, a few drops of reagent were added. Formation of white or pale precipitate showed the presence of alkaloids.

2. ***Screening for Anthraquinones***. A 0.5 g of the extract was boiled with 10 ml of sulphuric acid (H_2SO_4) and filtered while hot. The filtrate was shaken with 5 ml of chloroform. The chloroform layer was pipette into another test tube and 1 ml of ml of dilute ammonia was added. The resulting solution was observed for color changes

3. ***Screening for Flavonoids***. In a test tube containing 0.5 ml of extract, 5 to 10 drops of diluted HCl and small piece of $ZnCl_2$ or magnesium were added and the solution was boiled for few minutes. The appearance of reddish pink or dirty brown color indicates the presence of flavonoids.

4. ***Screening for Phenolics.*** To 1 ml of the extract 3 ml of distilled water followed by few drops of 10% aqueous Ferric chloride solution was added. Formation of blue or green color indicates the presence of phenols

5. ***Screening for Tannins***. In a test tube containing about 5 ml of the extract, a few drops of 1% solution of lead acetate was added. A yellow or red precipitate indicates the presence of tannins.

6. ***Screening for Saponins.*** In a test tube containing about 5 ml of the extract, few drops of sodium bicarbonate was added. The mixture was shaken vigorously and kept for 3 minutes. A honeycomb like froth was formed and it showed the presence of saponins.

7. ***Screening for Steroids***. To 2.0 ml of extract, 1.0 ml of concentrated sulphuric acid was added carefully along the sides of the test tube. A red color produced in the chloroform layer shows the presence of steroids.

8. ***Screening for Diterpenoids***. To 0.5g of the extract 2ml of CHCl3 was added. 3ml of concentrated H_2SO_4 was carefully added to form a layer. A reddish brown coloration of the interface indicates the presence of terpenoids.

9. ***Test for Terpenoids (Salkowski Test).*** To 0.5 g each of the extract, was added 2 ml of chloroform. Concentrated sulfuric acid was carefully added to form a layer. A reddish coloration of the interface indicates the presence of triterpenes.

10. ***Screening for Cardiac Glycosides.*** To 0.5 g of extract diluted to 5 ml in water, 2 ml of glacial containing one drop of ferric chloride solution This was underlayed with concentrated sulphuric acid. A brown ring at the interface indicated the presence of a deoxysugar characteristic of cardenoloids. A violet ring may appear below the brown ring, while in the acetic acid layer a greenish ring may appear below the brown ring just above the brown ring and gradually spread throughout this layer.

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11. ***Screening for Anthocyanins.*** Two ml of aqueous extract is added to 2 ml of 2N HCl and ammonia. The appearance of pink-red turns blue-violet indicates the presence of anthocyanins.

Accordingly, the appearance of certain color or precipitate was taken as a presumptive evidence of chemical detection at the end of each testing process, based on characteristics of the different metabolites groups. The data were then tabulated.

RESULTS AND DISCUSSION

Profile of the Plant Samples

The scientific and local names of the plants analyzed as well as the family to where these plants belong are summarized in table 1. This table also contains information on marketers' opinion on the plant part consumed either due to their nutritional value or claimed therapeutic effect.

Table 1. Profile of the plant samples considered for the study

Scientific Name	Family	Local Name(s)	Edible parts
Diplazium esculentum	Athyriaceae	Pako (Tag, Ilk.)	Young shoots
Amaranthus viridis Linn.	Amaranthaceae	Kalunai (Ilk), Kolitis (Tag)	Young shoots
Artocarpus camansi Blanco	Moraceae	Pakak (Ilk), Kamansi (Tag) Bulai-patani (Tag)	Immature pulp of fruit
Phaseolus lunatus Linn	Leguminocea	Palpadi (Ilk.) Patani (Ilk,Tag)	Seeds
Amorphophallus campanulatus Blume	Araceae	Tiggi a magmanto (Ilk) Apong-apong (Tag)	Stalk
Syzygium lineatum (DC.) Merr. & L.M. Perry	Myrtaceae	Malibado (Ilk.)	Fruit
Lycopersicon pimpinellifolium	Solanaceae	Botbotines (Ilk) Kamatis (Tag)	Whole fruit
Schizophyllum commune	Polyporaceae	Kudit (Ilk)	Whole plant
Colocasia esculenta Linn	Araceae	Daludal (Ilk)	Young roots
Momordica chinensis Spreng.	Cucurbitaceae Juss	Ampalayang ligaw (Tag)	Whole fruit
Cicca acida (Linn.) Merr	Euphorbiaceae	Karamay (Ilk), Iba (Tag)	Fruit
Sesbania grandiflora Linn	Fabaceae	Katurai (Tag), Katudai (Ilk)	Flowers
Cajanus cajan (Linn.) Mill	Leguminocea	Kardis (Ilk)	Seeds
Phaseolus vulgaris	Leguminocea	Tudo (Ilk)	Seeds
Psophocarpus tetragonolobus	Leguminocea	Wing Bean(Eng), Sigarilyas (Tag)	Whole fruit

Common ailments like bronchitis, stomach related ailments and some skin diseases were mentioned by the marketers as uses of the plants they sell. The information obtained from the vendors conformed to the study undertaken by Hossain, et al (2011)[15] agreeing with the

traditional use of plants as anti diarrhea, wound healing, stomach ache, to name a few**.** Furthermore, information about plant usage of the indigenous plant species indicated that young leaves, flowers, and tubers are the plant parts used for consumption to provide nutrition and for the claimed healing effect (Dlamini et al 2010)[16] .

The study validated the observations that members of the community tend to use preferably the plants that are easily available to them excluding those that are toxic. As was affirmed by Turner (2005)[17], the more common a plant species is in an area, the greater is the probability of its popular usage.

Phytochemical Screening

Qualitative phytochemical analysis of the plant sample was done by using color forming and precipitating chemical reagents to generate preliminary data on the constituents of the plant extract. The results of preliminary qualitative phytochemical analysis are tabulated in table 2.

The chemical tests revealed some differences in the constituents of the fifteen different plants analyzed. The secondary metabolites are also called plant defensive compounds since these have been evolved to deter pathogens or herbivores such as insects and mammals. The plants growing on low nutrient soil or in harsh conditions are often more dependent on evolved chemical defenses and thus could contain secondary metabolites with a wide range of interesting activities.

Table 2a. Phytochemical screening of secondary metabolites of different plant extracts

Secondary Metabolites Evaluated	*Result*						
	Pako	*Kalunay*	*Pakak*	*Bulai patani*	*Tiggi*	*Malibado*	*Botbotines*
Alkaloids	+	+			+		+
Anthraquinones	+					+	
Flavonoids			+		+	+	+
Phenolics	+		+		+		+
Tannins					+	+	
Saponins	+	+		+	+	+	
Sterols	+				+		
Diterpenes	+		+				
Triterpenes	+					+	
Glycosides						+	
Anthocyanins	+						+

Note: + indicates presence of the secondary metabolites evaluated

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Table 2b. Phytochemical screening of secondary metabolites of different plant extracts

Secondary metabolites evaluated	*Result*							
	Kudit	*Daludal*	*Ampalayang ligaw*	*Karamai*	*Katurai*	*Kardis*	*Tudo*	*Sigadillas*
Alkaloids	+	+	+		+			
Anthraquinones								
Flavonoids		+		+	+	+		+
Phenolics	+	+	+					+
Tannins		+	+	+	+	+	+	+
Saponins		+		+	+	+	+	
Sterols			+					
Diterpenes								
Triterpenes		+				+	+	
Glycosides			+		+			
Anthocyanins								

Note: + indicates presence of the secondary metabolites evaluated

Wide range of polar chemical classes was found in the fifteen plant extracts. Such extracts revealed dominantly the occurrence of saponins (10 plants); tannins and flavonoids (9 plants); phenolics and alkaloids (8 plants). Triterpenes were also detected in five (5) plants while three (3) plants were found to contain glycosides and the same number of plants (3) were positive to sterols. Anthraquinones, diterpenes and anthocyanins were found to be present in two plants for each metabolite respectively.

Phytochemical compounds were found to be interesting because they may inhibit bacterial growth by mechanisms different from presently used treatment regimens, and could therefore be of clinical value in the treatment of resistant bacteria. Tannins, flavonoids, alkaloids, essential oils and many phenolic compounds serve as plant defense mechanisms against predation by insects, herbivores and infection by microorganisms (Cowan, 1999)[18]. It is therefore not surprising that these compounds have been found to exhibit profound antimicrobial activities *in-vitro* against a wide array of organisms as claimed by the traditional practice in the community.

These phytochemicals have the potential to be incorporated into foods or food supplements as nutraceuticals. In addition to its contribution to the nutraceutical industry, it could potentially be used to identify concepts and products for different markets, such as edible plants, as sources of new and natural colorants and flavourants, nutritional/herbal supplements, as well as sweeteners and for the control of hunger (van Rensburg *et al*., 2007)[19]. It has been generally stated that the health promoting effects of nutraceuticals and other functional foods

are likely due to biochemical and cellular interactions, which together promote the overall health of an individual (Dillard and German, 2000)[20].

CONCLUSION AND RECOMMENDATION

In this study, although there are variations in the chemical constituents found to be present, the result gave comparative information about the main classes of secondary metabolites found in the indigenous edible plants evaluated. Therefore, the current findings revealed the richness of the indigenous edible plants in different groups of active compounds that need to be studied for different applications may help achieve nutritional security.

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