Phytochemical Screening, Nutritional Values and Antimicrobial Activity of *Eclipta Alba* (L.) Hassk (Kyeik-Hman)

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Abstract

The present work is phytochemical screening, nutritional values and antimicrobial activity of *Eclipta alba* (L.) Hassk. The sample was collected from Sittway Township. The phytochemical investigation of the plant sample observed the presence of alkaloids, α-amino acids, carbohydrates, glycosides, reducing sugars, starch, saponins, steroids, flavonoids and phenolic compounds. Tannins and cyanogenic glycosides were absent in the plant sample. The nutritional values were examined by AOAC method, the percentage of fat, fiber, moisture, protein, ash and carbohydrate 1.78, 10.99, 7.58, 24.14, 14.13 and 41.38. The energy value of *Eclipta alba* (L.) Hassk was found to be 278.1 kcal/100g. The antimicrobial activity of ethyl acetate, ethanol, methanol and watery extracts of *Eclipta alba* (L.) Hassk were investigated against six strains of microorganisms such as *Staphylococcus aureus, Bacillus subtilis, Pseudomonas aeruginosa, Bacillus pumilus, Escherichia coli and Candida albicans* by agar well diffusion method. Ethanol extract of selected sample showed more potent antimicrobial activity with inhibition zone diameter ranges between 12 mm-20 mm against all tested organisms.

Keywords: *Eclipta alba* (L.) Hassk, phytochemical constituents, nutritional values, antimicrobial activity

Introduction

Eclipta alba (L.) Hassk. (also known as *Eclipta prostrate Roxb*.) belongs to the Asteraceae family and is commonly known as false daisy in English and bhringoraj or bhringraj in Bangladesh and India. Plants have always been exemplary sources of drug and many of the currently available drugs have been directly or indirectly obtained from plants. Recently, the search for appropriate hypoglycemic agent has been focused on plants used in traditional medicine as plant natural products that involved in traditional medicine leads to better treatments than currently used drugs. The World Health Organization (WHO) has recommended that traditional plant treatments for diabetes warrant further evaluation (FAO/WHO.1973).

Materials and Methods

Sampling of Plant Materials

In this research, the whole plant of Kyeik-Hman was collected from Sittway Township, Rakhine State, Myanmar. The scientific name of this plant was identified by authorized Botanist at the Department of Botany, Sittway University. The sample was washed with water and air dried at room temperature. The dried sample was cut into small pieces and ground into powder by grinding machine. The dried powdered sample was stored in the air-tight containers to prevent moisture changes and contamination (Trease and Evans, 1980; Robinson, 1983; Vogel, 1996).

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(a) Plant (b) flower

Figure 1. Photographs of *Eclipta alba* (L.) Hassk (Kyeik-Hman**)**

Phytochemical Examination of *Eclipta alba* (L.) Hassk (Kyeik-Hman)

Phytochemical screening of *eclipta alba* (L.) was carried out by the standard method (Harbone, 1993)

Determination of Nutritional Values

Nutritional values such as moisture, ash, fiber, protein, fat and carbohydrate contents was carried out by appropriate reported methods (AOAC., 2000).

Determination of moisture content

Moisture content was determined by using oven drying method. In this method, the principle of azeotropic distillation between moisture and toluene was utilized (AOAC., 2000).

Determination of ash content

Ash content was determined by AOAC which is also the method given in Food Analysis. The ash value of the food staff represents the inorganic residue after the organic has been burnt away.

Determination of crude fiber content

Crude fiber content was determined by the method given in “The Chemical Analysis of Food”.

Determination of protein content

Protein content was determined by using Macro-Kjeldahl’s method.

Determination of fat content

Fat content was determined by the Soxhlet extraction method given in AOAC (AOAC., 2000).

Determination of carbohydrate content

Carbohydrate content was determined by subtraction method. The total carbohydrate content of sample can be obtained as the difference between 100 and the sum of the percentage of ash, fat, fiber, moisture and protein. The carbohydrate content of sample was given in Table 3.

Screening of antimicrobial activity of *Eclipta alba* (L.) Hassk

Antibacterial activity of various crude extract such as ethyl acetate, ethanol, methanol and watery extracts were studied by agar well diffusion method at Department of Development Centre of Pharmaceutical and food Technology, Yangon (Finegold, S. M., and W. J. Martin, 1982).

Screening by agar well diffusion method

The agar well diffusion method was used to test the antibacterial action of the extracts on 24 hours broth culture of the organisms used.

The extracts of ethyl acetate, ethanol, methanol and watery, (1 g each) were dissolved in 1cm^3 of their respective solvent. 1 mL each of the bacterial suspension of 34 hours of nutrient agar was streaked evenly onto the surface of trypticase soy agar plates with sterile cotton swab. Immediately after hardening of the agar well were made with a 10 mm sterile corn borer from each seeded agar. After removing the agar, the wells were filled with the drug extract to be tested the plates were incubated at 37 °C for 18-24 hours. The diameters of the inhibition zone were measured and recorded in mm.

Table 1. Types of Microorganisms and Their Effects

Organisms	Gram	Shape	Effect	Types
Bacillus subtilis	+	Rod	causes conjunctivitis, food poisoning	Bacteria
Staphylococcus aureus	+	Spherical	Causes pneumonia, abscesses, wound sepsis, burns, food poison, soft tissue infection, bone and joint infections, gastronintestinal	Bacteria
Pseudomonas aeruginosa	-	Rod	chronic lung, ear infection, burn infections, surgical wounds, ocular infection	Bacteria
Bacillus pumilus	+	Rod	eye infection, soft tissue infection, food poison	Bacteria
Candida albaican	+	Capsule	skin infection, cardiac infection, bowel disorders, sinus irritation, intense itching, sored, ringworm	Fungi
Escherichia coli	-	Spherical	urinary tract infection, bloody diarrhea, appendix abscess, gastroenteritis	Bacteria

RESULTS AND DISCUSSION

Phytochemical Constituents of *Eclipta alba* (L.) Hassk (Kyeik- Hman)

After preparing the sample, the whole plant of *Eclipta alba* (L.) Hassk (Kyeik-Hman) observed the presence of alkaloids, α-amino acids, carbohydrates, glycosides, reducing sugars, starch, saponins, steroids, terpenoids, flavonoids and phenolic compounds but tannins and cyanogenic glycosides were absent in the plant sample. The results were shown in Table 2 and Figure 2.

Table 2. Results of Phytochemical Investigation of Plant Sample (Kyeik-Hman) by Standard Method

No.	Type of compound	Extracts	Test reagent	Observation	Remark
1.	Alkaloids	1 % HCl	(1) Mayer reagent (2) Dragendroff reagent (3)Wagner reagent (4) Sodium picrate	White ppt. Orange ppt. Reddish brown ppt. Yellow ppt.	+ + + +
2.	α-amino acid	H_2O	Ninhydrin	Pink spot	+
3.	Carbohydrate	H_2O	10 % α-naphthol & conc. H_2SO_4	Red ring	+
4.	Glycosides	H_2O	10 % lead acetate	White ppt.	+
5.	Phenolic compounds	H_2O	10 % $FeCl_3$	Deep blue color	+
6.	Starchs	H_2O	Iodine solution	Deep blue color	+
7.	Saponins	H_2O	Distilled water	Frothing	+
8.	Tannins	H_2O	2 % NaCl, gelatin	No brown ppt.	-
9.	Cyanogenic Glycosides	H_2O	Conc: H_2SO_4 and Sodium picrate	No color change	-
10.	Flavonoids	EtOH	Conc. HCl and Mg turning	Pink color	+
11.	Steroids	PE	Acetic-anhydrate & conc. H_2SO_4	Green color	+
12.	Terpenoids	$CHCl_3$	Benedict's solution	Reddish brown	+
13	Reducing Sugars	Dilute H_2SO_4	Benedict's solution	Brick red ppt.	+

(+) = Present, (-) = Absent, (ppt.) = Precipitate

Figure 2. Phytochemical constituents of *Eclipta alba* (L.) Hassk (Kyeik- Hman) by standard method

Nutritional Values of *Eclipta alba* (L.) Hassk (Kyeik- Hman)

Moisture content can be determined by using oven drying method. The moisture content of sample was found to be 7.58 %.The ash content may be determined by heating the sample in a porcelain crucible to 600 °C. The ash content of plant sample was 14.13 %. Crude fiber is the insoluble and combustible organic residue which remains after the sample has been treated with petroleum ether. The yield percent of the crude fiber was 10.99 % in plant sample. Fat was determined by Soxhlet extraction method using petroleum ether (b.pt 40-60 °C). The result of the fat

content was found to be 1.78 % in plant sample. In plants, proteins are found in all living parts but usually in relatively small quantities except in the seed. The result of the protein content was 24.14 % in plant sample. The energy value was found to be 278.1 kcal/100 g. The results of nutritional values were shown in Table 3.

Table 3. Nutritional Values of *Eclipta alba* (L.) Hassk (Kyeik- Hman)

No.	Parameters	Content (%)
1	Moisture	7.58
2	Ash	14.13
3	Protein	24.14
4	Fiber	10.99
5	Fat	1.78
6	Carbohydrate	41.38
	Energy Values (kcal/100g)	278.10

Antimicrobial Activity of *Eclipta alba* (L.) Hassk (Kyeik- Hman)

Screening of antimicrobial activity of ethyl acetate, ethanol, methanol and watery extracts of *Eclipta alba* (L.) Hassk has been done by agar well diffusion method. In this investigation, the sample was tested on six species of microorganisms such as *Bacillus subtilis, Staphylococcus aureus, Pseudomonas aeruginosa, Bacillus pumilus, Candida albicans* and *Escherichia coli* species. According to the results presented in Table 4 and Figure 3.

It was found that watery extract of Kyeik- Hman showed no activity on all tested microorganisms. The ethanol extract of Kyeik-Hman showed higher antimicrobial activity than other extracts on these microorganisms. Therefore, ethyl acetate, ethanol and methanol extracts of the sample can be effective in the formulation of medicine for treatment of various diseases.

Table 4. Antimicrobial Activity of crude Extracts on Different Species of Microorganisms by Agar Well Diffusion Method (Kyeik- Hman)

No	Type of Microorganisms	Inhibition Zone Diameter (mm)			
		H_2O	MeOH	EtOAc	EtOH
1.	*Bacillus subtilis*	_	13 (+)	_	13 (+)
2.	*Staphylococcus aureus*	_	14 (+)	12 (+)	14 (+)
3.	*Pseudomonas aeruginosa*	_	_	_	20 (++)
4.	*Bacillus pumilus*	_	12 (+)	11 (+)	12 (+)
5.	*Candida albicans*	_	12 (+)	_	12 (+)
6.	*Escherichia coli*	_	12 (+)	12 (+)	13 (+)

Agar well diameter – 10 mm
10 mm - 14 mm (+)
15 mm - 19 mm (++)
20 mm above (+++)

MeOH = Methanol
EtOH = Ethanol
EtOAc = Ethyl acetate
H_2O = Watery

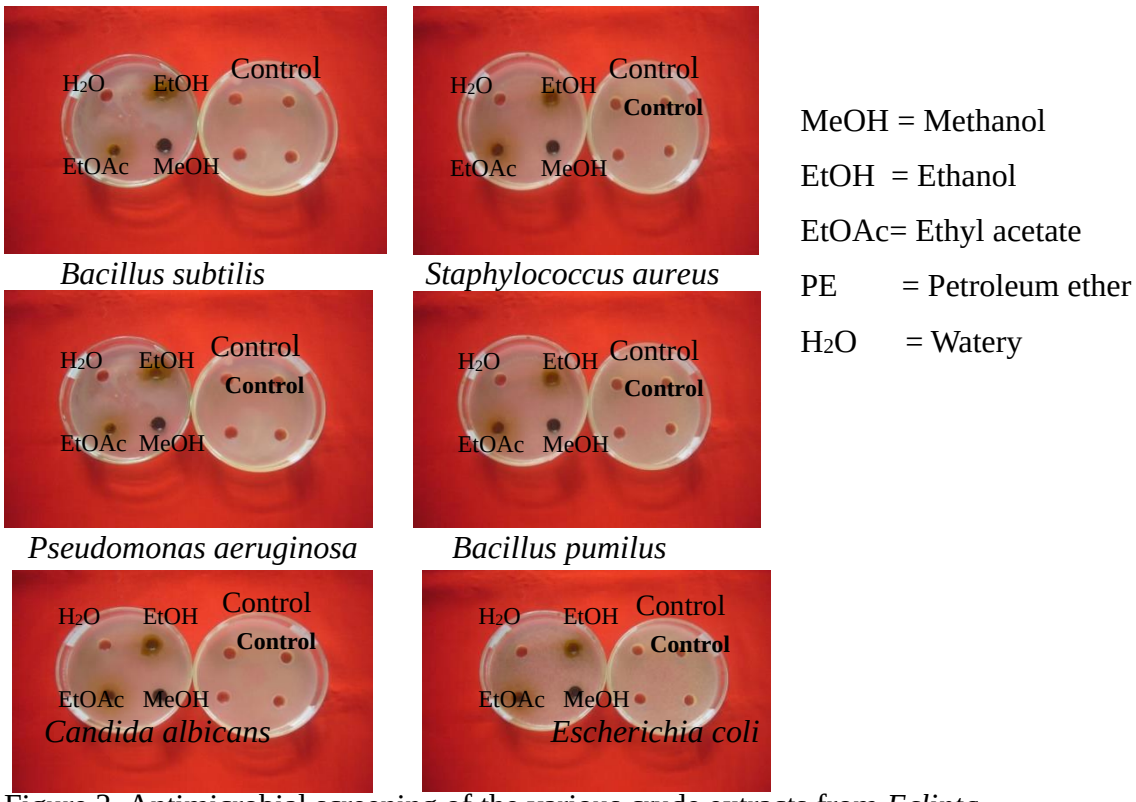

Figure 3. Antimicrobial screening of the various crude extracts from *Eclipta alba* (L.) Hassk (Kyeik- Hman)

CONCLUSION

The sample of *Eclipta alba* (L.) Hassk (Kyeik- Hman) observed the presence of alkaloids, α-amino acids, carbohydrates, glycosides, reducing sugars, starch, saponins, flavonoids and phenolic compounds. Tannins and cyanogenic glycosides were absent in plant sample.

Nutritional values of moisture content (7.58 %), ash content (14.13 %), protein content (24.14 %), fiber content (10.99 %), fat content (1.78 %), carbohydrate (41.38 %) and energy value (278.1 kcal/100 g) were observed. The determination of nutrient values show higher content of carbohydrate than the others such as protein, fat and fiber.

The antimicrobial activity of ethyl acetate, ethanol and methanol extracts of Kyeik-Hman was investigated against six strains of microorganisms such as *Staphylococcus aureus, Bacillus subtilis, Pseudomonas aeruginosa, Bacillus pumilus, Escherichia coli and Candida albicans* by agar well diffusion method. Ethanol extract

of plant showed more potent antimicrobial activity with inhibition zone diameter ranges between 12mm-20 mm against all tested organisms.

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References

AOAC. (2000). *Official and Tentative Methods of Analysis, Association of Official Analytical Chemists*, Washington D.C, 11th Ed., 978

FAO/WHO. (1973). *Energy and Protein Requirements*, Rome, Repeat of a Joint FAO/WHO Ad Hoc Expert Committee, Food and Agriculture Organization of the United Nation, FAO and WHO

Finegold S. M., and W. J. Martin, (1982) "*Diagnostic Microbiology*," 6th Edition, C.V. Mosby Co. St. Louis, Toronto, London

Harborne, J. B. (1993). *Methods in Plant Biochemistry*, London, Academic Press, **8,** 397

Robinson, T. (1983). *The Organic Constituents of Higher Plants*, North America, 5th Ed., Cordus Press

Trease, G. E. and W. C. Evans. (1980). *Pharmacognosy*, London, 1st Ed., Spotteswoode Ballantyne Ltd., 108-518

Vogel, A.I. (1996). *A Text Book of Practical Organic Chemistry*, London: 3rd Edn., Language Book and Longman Group Ltd.,45