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PHYTOCHEMICAL SCREENING AND ANTIMICROBIAL ACTIVITY OF PLANT EXTRACTS OF SEMALU PLANT (*Mimosa pudica* L.)

NUR SYAFIQAH BINTI ABDULLAH HISHAM

PERPUSTAKAAN UNIVERSITI MALAYSIA SABAH

DISSERTATION SUBMITTED IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE DEGREE OF BACHELOR OF AGRICULTURE SCIENCE WITH HONOURS

> CROP PRODUCTION PROGRAM SCHOOL OF SUSTAINABLE AGRICULTURE UNIVERSITY MALAYSIA SABAH 2013



DECLARATION

I hereby declare that this dissertation is based on my original work except for citations and quotations which have been duly acknowledged. I also declare that no part of this dissertation has been previously or concurrently submitted for a degree at this or any other university.

Nur Syafidah Binti Abdullah Hisham BR09110048 23 January 2013



1. Madam Devina David SUPERVISOR

DEVINA DAVID Penpyarah/ Penasihat Akademik Sekolah Pertanian Lestari Universiti Malaysia Sabah

Ham

PROF. MADYA. DR. HARPAL SINGH SAINI PROFESOR MADYA/PENASIHAT AKADEMI SEKOLAH PERTANIAN LESTARI UNIVERSITI MALAYSIA SABAH

BOSMAH MURDAD Lecturer / Academic Advisor School Of Sustainable Agriculture Universiti Maleysia Sabah

monst

DR. SITTI RAEHANAH MUHAMAD SHH... DEKAN SEKOLAH PERTANIAN LESTARI UMS KAMPUS SANDAKAN



2. Prof. Madya Dr. Harpal Singh Saini EXAMINER 1

3. Madam Rosmah Murdad EXAMINER 2

4. Dr. Sitti Raehanah Binti Muhamad Shaleh DEAN SCHOOL OF SUSTAINABLE AGRICULTURE

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ABSTRACT

A study was conducted on different parts of Mimosa pudica L which are leaves, flower and stem that collected around UMS, Sandakan campus. The purpose of this study is to screen phychemical content and investigate antimicrobial activities on different plant parts of *M. pudica*. The study was conducted in the School of Sustainable Agriculture laboratory and starting in April 2012 until October 2012. In this study, preliminary phytochemical by qualitative test and antimicrobial activity towards different parts of M. pudica was conducted by using ethanol as solvent extraction. Phytochemical screening result was obtained from visual observation revealed the presence of glycosides, saponins, flavonoids, tannins and phenolic compound. The antimicrobial activity of different plant parts were investigated by disc diffusion method against bacterial pathogen E. coli, S. aureus, S. enteritis, C. freundii. The zone of inhibition of crude extracts was compared with standard antibiotic streptomycin (5mcg/disc) and diameter of inhibition zone was observed after 24 hours incubation period. According to this study, in terms of bacterial activity (zone of inhibition) as whole showed a significant difference mean values (p < 0.05) for all crude extract. Among the three different parts, crude extracts of leaves showed higher inhibition zone of 9.8+2.98mm. This study proceeds with minimum concentration for inhibitory zone and was found that lowest concentration of leaves extract at 5mg/ml against E. coli, while S. aureus and S. enteritis showed inhibition zone at the lowest concentration of crude extract at 10mg/ml and bacteria C. freundii showed lowest inhibition zone at concentration 40mg/ml. The lower concentration of the crude extract that can inhibit bacteria, the higher the potential of the plant can be used in medical field hence through this study the abilities of M. pudica extracts inhibit bacteria at low concentrations with the higher amount of crude extract produced prove that this plant has potential as an effective antimicrobial agent.

Key words: *Mimosa pudica*, antimicrobial activity, phytochemical screening, disc diffusion method, minimum inhibitory concentration.



KAJIAN FITOKIMIA DAN AKTIVITI ANTIMIKROBIAL TERHADAP EKSTRAK TUMBUHAN POKOK SEMALU (*Mimosa pudica* L.)

ABSTRAK

Satu kajian telah dijalankan ke atas ekstrak tumbuhan Mimosa pudica L. iaitu daun, bunga dan batang yang tumbuh di sekitar kawasan UMS, kampus Sandakan. Tujuan kajian ini adalah untuk mengekstrak sebatian fitokimia dan mengkaji ekstrak bahagian tumbuhan yang berbeza terhadap aktiviti bakteria. Kajian ini telah dijalankan di makmal Sekolah Pertanian Lestari dalam tempoh masa kajian yang bermula pada April 2012 sehingga Oktober 2012. Etanol digunakan sebagai pelarut untuk menghasilkan ekstrak. Keputusan ujian saringan fitokimia secara kualitatif telah diperolehi secara pemerhatian visual untuk mengkaji kehadiran glikosida, saponin, flavonoid, tanin dan sebatian fenolik. Aktiviti antimikrob terhadap ekstrak bahagian tumbuhan yang berbeza telah dijalankan melalui kaedah penyebaran cakera terhadap bakteria patogen E. coli, S. aureus, S. enteritis, C. freundii. Zon perencatan ekstrak tumbuhan dibandinakan dengan antibiotik streptomycin (5mcg/disc) dan diameter zon perencatan diperhatikan selepas 24 jam tempoh pengeraman. Berdasarkan kajian ini, dari segi aktiviti bakteria (zon perencatan) secara keseluruhannya menunjukkan nilai purata kesemua ekstrak mempunyai perbezaan yang signifikan (p <0.05). Hasil kajian ini juga menunjukkan bahawa diantara tiga bahagian tumbuhan yang berbeza, ekstrak daun menunjukkan perencatan zon yang tertinggi sebanyak 9.8+2.98mm. Kajian ini diteruskan dengan kepekatan perencatan minima dan didapati bahawa pada kepekatan ekstrak daun secara minima iaitu 5mg/ml terhadap bakteria E. coli, manakala S. aureus dan S. enteritis menunjukkan perencatan minima pada 10mg/ml dan bakteria C . freundii menunjukkan perencatan minima pada 40mg/ml. Semakin rendah kepekatan ekstrak yang dapat merencat bakteria, maka semakin tinggi potensi tumbuhan tersebut digunakan dalam bidang perubatan dan melalui kajian ini kebolehan ekstrak M. pudica menghalang bakteria pada kepekatan yang rendah disamping menghasilkan ekstrak tumbuhan yang banyak membuktikan bahawa tumbuhan ini mempunyai potensi sebagai agen antimikrobial.

Kata kunci: Mimosa pudica L., penyaringan fitokimia, kaedah cakera resapan



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LIST OF SYMBOLS, UNITS AND ABBREVIATIONS

% °C / µg µl ANOVA CFU g HCI mg MIC min ml mm SEM	percentage degree celcius per microgram microliter Analysis of Variance colony forming unit gram hydrochloric acid milligram minimum inhibitory concentration minute milliter milimeter standard error of mean
	Statistical Package for Social Science
SPSS	Statistical Fackage 101 Social Science



CHAPTER 1

INTRODUCTION

1.1 Introduction

Weed is any plants that grow and gives competition to cultivated crops and often causing yield losses. Weed is also classed as pest because it competes for nutrients, water and light. However, weeds also give some benefits such as protecting from soil erosion and also increase plant diversity, some provide good wildlife habitat because many weed species are native and adapted to specific microclimates and local conditions (Smith *et al.*, 2000). Usually farmers use herbicide to suppress the weeds. But, actually weeds give various beneficial to human as medicinal purpose.

Recently, there are thousand weed species in our agro-ecosystems, many of which are invasive. The paths of invasion of weeds in our agro-ecosystems are largely unknown. Management of invasive weed species in Malaysian agro-ecosystems are very much herbicide-based, integrated with other control measures including cultural practices, prescribed burning, animal grazing, and to certain extent, followed by manual and mechanical roughing. Successful management of noxious invasive in our ecosystems will require the development of a long-term strategy incorporating prevention program, extension and educational activities, and sustainable and educational multi-year integrated approaches that prevent reinvasion or encroachment by other noxious invasive weed species. Invasive weed species impact on public awareness, legislation, conservation biology, agriculture, forestry, soil and water resources, and recreational and other related activities in the Malaysian agriculture and waterways management (Baki, 2004).



According to Chowdhury *et al.* (2008), the trend of using natural based products have been increased and new discoveries by using plant extract products which attracted a great attention as therapeutic potential of antioxidant in controlling disease and antimicrobials. Several plant extract such as *M. pudica* plant have been found by using different classes of phytochemicals that contain antioxidant activity.

In this study focus on three different parts of *M. pudica* L. such as leaves, flower, and stem that have antimicrobial properties and some phytochemical content that can give beneficial to human consumption especially in traditional medicine. According to John (2001), the extract of *M. pudica* plant can give some functional purpose towards human such as can promote regeneration of nerves and antidepressant activity while roots extracts has been reported to be a strong emetic.

1.2 Justification

Weed is a plant that grows in places where it's not supposed to. Weed is not deliberately cultivated plants, but appears in itself or carried by some vector without realizing it especially area to be cleared for agricultural land or development. Farmers usually faced this problem because to suppress the weeds they need to spend high cost for buying the herbicide and cause economic losses to land managers. In agriculture, it is important for controlling the weed from spreading throughout the farm. In previous study by Azly (1988) stated that weeds can provide minimal structural and habitat diversity which may reduce or limit the ability for yield to grow well and consumed sufficient water, nutrients and space.

According to McCarty and Murphy (1992), weeds always are being avoiding and people categorize the weed as troublesome especially weeds invading the road area that can block the eyesight for driving. Even though weeds can cause several allergies and poisonous to human and livestock, it is not at all actually. Moreover, some weeds can be categorized as herbs and give some benefit for human to cure illness and diseases. Traditional medicines are more prefer to use plant or herbs as medicine to cure sickness (Keating, 1994).

Thus, the aim of this study was to evaluate the potential antimicrobial properties towards three different plant parts of *M. pudica* which is leaves, flower and



stem against different groups of bacteria. Moreover, the plant extracts were subjected to preliminary phytochemical screening and analyze the possible antimicrobial activities they contain. The study provides scientific evidence on the use of these plants which are being utilized traditionally as herbal medicines.

1.3 Objectives

Purpose of this study is to:

- i. To screen phytochemical activity of ethanolic extract of three different plant parts (leaves, flower and stem) of *Mimosa Pudica* L.
- ii. To investigate antimicrobial properties of ethanolic extract of three different plant parts (leaves, flower and stem) of *Mimosa Pudica* L.



CHAPTER 2

LITERATURE REVIEW

2.1 Weeds

Weed is a plant growing where it is not desired. Weed also known as plant that is undesirable at not particular time or space. It can grow anywhere on land that been cleared such as land for agriculture purpose. In general, weed can be divided into three categories. The first is a weed pest, second is weed that has economic uses and the third is weed that is neutral. However, the quality of the properties of this weed as either a pest, beneficial or neutral varies according to circumstances.

In agriculture, weed always is a problem for farmers in their crops cultivation. Most weed grown there are pest and can affect yield growth. Weeds give problematic losses of yield and give adverse affect to human and animals. This adverse effect may be seen either directly or indirectly. Negative consequences of such weeds are harmful effects on agriculture, the effect of applied herbicides on humans and animals and also to environment. But, weeds not always gives harmful effect, it also give some benefit to human and land. Beneficial of the weed can be seen when it gives role for reducing erosion. The weeds are encouraged or allowed to grow in area that is exposed by erosion. For example weeds growing on the slope of the hills along the highway function as cover crop that can reduce soil erosion (Noormaizurah, 1999).

Most weed germinate and become established relatively quickly. They also produce viable seed even though under unfavourable soil condition. Moreover, weed can remain viable over 40 years or more buried in the soil. In most cases, majority seeds only exist in few years due to germination, predator feeding, decomposition or some external factors. Weeds also have some mechanism for dispersal of seed and special structures to fly, cling or float (Baki, 2004)



2.2 Mimosa pudica L.

Mimosa pudica is a creeping annual or perennial herb that comes from family of Fabaceae and sub family of Mimosoideae. Their leaves fold inward and droop when touched or shaken and re-opening minutes later. This type of motion has been termed nyctinastic movement. *Mimosa pudica* is native in South America but it is also a pantropical weed. Common name of mimosa pudica are shameplant, sleeping grass, *malu-malu*, or *semalu*. *Mimosa pudica* has erect stem but will becomes creeping or trailing with age. Their stem is slender, branching and growing until length of 1.5 m.

Mimosa leaves are bipinnate with one or two pairs and about 10 to 26 leaflets per pinna. Inflorescences have many florets with pale pink or purple heads. Every pod contains about one to four seeds with 2.5 mm long. The fruits are prickly on the margins. Moreover, some researchers consider this plant a woody herb. This short-lived shrub is a small, prostrate and ascending. The reddish-brown woody stems are sparsely or densely armed with curved prickles. *Mimosa pudica* has taproot system and extensive fibrous roots with nodules. This plant has fine and flexible twigs that support leaves with one or two pinnae (Azly, 1988).

Mimosa pudica has pink flower head and clustered in globose heads. Inflorescense a dense pink to liliac globose spike about 10 to 13mm long borne on a 10 to 30mm hairy peduncle, 1 to 5 heads appearing from a leave axil. Length of calyx about 0.2mm while corolla about 2.3mm long and the petals joined for more than half of their length and almost colourless. Mimosa has four stamens with about 0.3mm anthers borne on 8mm pink filaments. Pod length of *M. pudica* plant is about 20mm long with 3 to 5mm wide. It has flattened shape while the upper and lower margins intended between each seed. The margins provided with coarse, spreading 2 to 3mm bristles, dark brown in colour and breaking up when ripe that containing about four seeds.

Seeds size about 3mm long with pear and flattened shaped and stem diameter about 1 to 2.5mm that usually can grow until four feet or more. Seed coloured dark red-brown to almost black with finely granular surface. It has alternative thorns on the stem usually 2 to 3mm long. Stem is tall, prickly and appears to have 10 to 26 leaflets



per stem. Stems appear to branch out in different directions. *Mimosa pudica* prefers full sunlight, dislikes root disturbance and over wintering (Gurcharan, 2004).

2.2.1 Origin

Mimosa pudica L. is originate from the tropical America. It distribution is widespread in tropic, subtropic and temperate area around the world. *Mimosa pudica* considered as a pan-tropical weed with distribution throughout Africa, Asia, Australia and the Pacific islands. In Malaysia, *M. pudica* grows best during the rainy season. The seed can germinate within two weeks. This herb grows in full sunlight or light shade in warm climates.

Mimosa pudica cannot tolerate in frost condition. It can thrives and become the ground cover in poor soil because it has ability to fix nitrogen. This sensitive plant predominantly grows indoors as an annual and groundcover. In middle England, this herbal plant is commonly grown indoors as a houseplant. While in Malaysia this plant can be found along the road, in the backyard or vigorously grow on the wet land (Kinsey, 2012).

Mimosa pudica is evergreen shrubs that can reaches five feet in height and can produces small globe shape flowers. *Mimosa pudica* has a reaction known as seismonatic movement that responds to the physical reaction to shock. It has feature fern-like leaves close and fold up when touched. During night, mimosa fold up that give responds to the absences of light known as nyctonastic movement (Srivastava *et al.*, 2012).

2.2.2 Secondary metabolites in *M. pudica* plant

Several reports have show that extracts of *M. pudica* plant have various phytochemical compounds. Phytochemical screening of mimosa leaves has been studied in India that prove the presence of some chemical constituent such as alkaloids, saponins, phytosterols, phenols, tannins, flavonoids and diterpenes by using different test (Kaur *et al.*, 2011). *Mimosa pudica* plant can also give some functional purpose towards human such as can promote regeneration of nerves and antidepressant activity while roots extracts has been reported to be a strong emetic (John, 2001).



According to Mukesh and Smita (2010), several plants and herb species used traditionally have potential antimicrobial and antiviral properties. Previous study showed that mimosa plant has some antimicrobial activities towards different microorganisms. The antimicrobial activities were measured by using zone of inhibition and the result could explain the rationale for the use of plant in the treatment of the various conditions in traditional medicinal practice.

Mimosa pudica has been categorized as traditional medicinal herbs in India and used in human and animal health care. Herbal drug formulation been recorded in Ayurveda and the pharmaceutical industries are largely based on about hundred plant species. Research on herbal plants used in traditional veterinary practice that useful in establishing standard dosage for investigating their toxicity (Rajeshwari and Ragunath, 2012).

According to the Rajeshwari and Ragunath (2012), the leaves extract show positive present of various phytochemical constituents like alkaloids, glycosides, flavonoids and saponin and also poses anti-microbial, anti-diabetic and anti-histaminic activity. The freshly collected plant material was dried and extracted with various solvents such as n-hexane, chloroform, ethyl acetate and methanol. Thin layer chromatography technique was carried out to confirm the presence of phytoconstituents. According to the preliminary test, result showed that n-hexane extract give positive result on carbohydrate, saponin and steroids. While methanol extract showed positive result on alkaloids, glycosides, carbohydrate, steroids, flavonoids and phenols. The different solvent extracts give different result of phytoconstituents and conclude that methanol extract as the best solvent among the other three solvents.

Furthermore, Sriram *et al.* (2011) investigated on GC-MS study and phytochemical profiling of *Mimosa pudica* Linn. Gas chromatography-mass spectrum was carried out on methanol extract of *M. pudica* for identification of phytochemicals in the plant. Gas chromatography-mass spectrum was being used as direct analysis of components existing in medicinal plants and has proved to be a method for analysis of several compounds (Jie and Choi, 1991; Sriram *et al.*, 2011). The phytochemical size screening on leaves extract showed the presence of some phytochemicals like



terpenoids, flavonoids, glycosides, alkaloids, quinines, phenols, tannins, saponins and coumarin.

Gas chromatography-mass spectrum analysis on the methanol extract of *M. pudica* showed that 19 compounds were identified of which 10 were found to have proven therapeutic values such as glycerine, myo-inositol, phytol, vitamin E and squalene. In this investigation, a variety compounds have been detected including glycerine for the presence of alcohol and myo-inositol for the presence of aromatic compound in *M. pudica*. Through this analysis revealed the presence of alkaloids, terpenoids, saturated and unsaturated fatty acids in *M. pudica* plant.

2.2.3 Ethnomedicinal uses

As ethnomedicinal uses, *M. pudica* is the plant that posses antimicrobial, hyperglycemic, anti-oxidant, anti-cancer and anti-diabetic. According to the Unani system of medicine, root is resolvent, alternative useful in diseases arising from blood impurities and bile, bilious fevers, piles, jaundice. In Ayurveda, the uses of *M. pudica* in traditional medicine are for inflammation, fatigue, asthma, diarrhea, and for treating skin disease leprosy (Pankaj, 2004).

Every part of the *M. pudica* such as root, flower and leaves give different functional purpose in medicinal. Root is bitter and acrid function as cooling, vulnerary, fatigue, asthma and treating disease of the blood. While juice of mimosa leaves used in sinus, sores, piles and fistula paste applied to glandular swelling (Rajeshwari and Raghunath, 2012). Other than that, *M. pudica* can also grow in garden herbs as useful green manuring, fixes nitrogen and can be used as fodder. It can form nodules that are inhabitable by nitrogen fixing bacteria that are able to convert atmospheric nitrogen (Srivastava *et al.*, 2012).

Khare (2004) stated that, fresh tissue of mimosa plant gave nor-epinephrine Dpinitol and beta-sitosterol and found that leaves part contain alkaloids. In his study resulted decoction of leaves of *M. pudica* in rats and dogs exhibit diuretic activities. Traditional uses in South American applied *M. pudica* on cuts and wounds also for childbirth and infertility. While bath with plant decoction can relieves insomnia. In mexico, aqueous extracts of dried leaves are employed to alleviate depression. While in Philippines, the leaves soaked in coconut oil used to treat ulcer (Koh *et al.*, 2009).



According to previous study by Rekha and Ekambaram (2010), leaves of the plant have been reported to contain an alkaloid mimosine and mucilage. The leaves are used in the treatment of piles and fistula. Leaves paste is applied to hydrocele and cotton impregnated with juice of leaves is used for dressing sinus. Plants also been used in the sore gum treatment and for blood purifier. Through this study by using chloroform extract revealed that *M. pudica* leaves contains flavonoids, alkaloids and glycosides. *Mimosa pudica* leaves are boiled in water for a half hour and drank hot or cold for the treatment of diabetes and obesity. The boiled leaves also used to treat sore throat, hoarseness and urinary complaints (Backmann *et al.*, 1997)

2.3 Phytochemical contents

Some research had been proved that *M. pudica* plant has several phytochemical contents in leave extract. According to Mukesh and Smita (2010), their research on the phytochemical and pharmacological screening of combined *M. pudica Linn* and *Tridax procumbens* for in vitro antimicrobial activity give positive result of some chemical content. Extraction is done by using mimosa plant including flower heads with various solvents such as hexane, ethyl acetate and methanol. Alkaloid, flavonoid, tannins, saponins and phenolic give positive result that show present of these phytochemical in that plants while negative resulted for amino acid and glycoside test.

2.3.1 Glycosides

Glycosides are substances produced in plants by metabolic processes and are usually bitter tasting that are important in human and animal nutrition. Glycosides have both toxic and beneficial properties and have abilities converting toxic materials to none or less toxic. Presence of glycoside compound in *M. pudica* extracts showed that the plant contains potential that may be use for the development of phytomedicine (Rekha *et al.*, 2009).

Glycosides are water soluble compounds and insoluble in organic solvents but some glycoside soluble in alcohol. Function of glycosides is transferring water soluble substance by using monosaccharide, source of energy and storing harmful product such as phenol. Some glycosides have beautiful colour and give protection to the plants from bacteria and diseases (Joseph *et al.*, 2005)

2.3.2 Saponins

Saponins exist as secondary metabolite that occurs in plant species. Saponins stored in plant cells as active precursor but are readily converted into biologically active antibiotics by plant enzymes in response to pathogen attack. According to Ayoola *et al.* (2008), screening of phytochemical compound in *M. pudica* extract showed positive reaction that presence of saponin compound.

Saponins have hemolytic, anti-inflammatory and immune stimulating activity that demonstrate antimicrobial properties particularly against fungi, bacteria and protozoa. This phytochemical is a complex compounds that are composed of a saccharide attached to steroid or triterpene. Their molecules have considerable commercial value and being processed as drugs, and medicine, foaming agents, sweeteners, taste modifiers and cosmetic (Hostettmann and Marston, 1995; Figen, 2006)

2.3.3 Flavonoids

Flavonoids are plant pigments that are synthesized from phenylalanine that can be found in almost plant based food and beverages. Besides their relevance in plants, flavonoids are important for human health because of their high pharmacological activities as radical scavengers. Flavonoids known as agents with strong antioxidant properties and enhance the effect of vitamin C (Lubica, 2011).

Previous study by Doss *et al.* (2011) the antimicrobial screening assay of leaves *M. pudica* plant in flavonoid extract gave relatively wide inhibition zone against the test strains compound and from the result obtained possible that flavonoids may be used as natural antimicrobial substance to replace antibiotics for controlling bacterial infections. This is due to their ability with extracellular and soluble proteins and bacterial cell wall.

Flavonoid is biologically active against liver toxins, tumours, viruses and microbes, allergies and inflammation. Hesperidin content in flavonoids give raises blood levels of the cholesterols and lowers the high cholesterols. While tangeretin induces programmed cell death in cancer cells such as leukemia but it does not affect normal healthy cells (Yao *et al.*, 2004).



2.3.4 Tannins

According to Elsa *et al.* (2011), tannins refer to heterogenous group of polymeric phenolic compound that usually present in plants. Tannins possess about 16 phenolic groups and 5 to 7 aromatic rings per 1000 units of relative molecular mass. However, tannins function to human nutrition is not clearly been reported and need more research is required.

According to Kaur *et al.* (2011) the preliminary phytochemical screening of *M. pudica* extract showed the presence of various bioactive components including tannins. From the study, it is concluded that traditional plants may represent new source of biologically active components that can establish a scientific base for the use of plants in modern medicine.

Tannins are likely to have effect in gut bacteria and the environment of the bowel which may link to short chain of fatty acid synthesis and protection against cancer. Legumes, tea and wine are good source of tannins since tannin has antioxidant properties and can also inhibit cancer activation (Barry, 2000)

2.3.5 Phenolic compound

Phenolic compounds are widely found in edible plants especially in secondary products of medicinal plants. Phenolic posses aromatic ring bearing with one or more hydroxyl constituents that have potential as antioxidant and free radicals scavengers (Singh *et al.*, 2007; Cespedes *et al.*, 2008).

Previous study by Chowdhury *et al.* (2008), showed that various solvent of crude extract of *M. pudica* plant presence of phenolic compound that may be responsible for antioxidant properties of many plants. However, many studies have described those phenolic posses biological properties such as anticarcinogenic, antiinflammation and cardiovascular protection. This shows that more phenolic content in the herbal medicine good for antioxidant activity (Han *et al.*, 2007; Ali *et al.*, 2008).

2.4 Solvent extraction

Solvent can be defined as a substance that dissolves in another substance to form a liquid. Solvents can be classified as either aqueous which is water-based or organic



hydrocarbon-based. In this research focused on the used of ethanol that act as the solvent extract. Organic solvents are widely used to dissolve and disperse fats, oils, waxes, pigments and many other substances (Fasihuddin, 1993).

Some characteristic of organic solvent is volatility. Ethanol tends to evaporate and inhalation exposure is an important exposure pathway to be considered when assessing the health hazard that solvents may present. Density of organic solvent also needs to be considered. As density increase the rate at which the solvent dissipates will decrease. Solvents from different chemical groups can differ markedly in their characteristic.

According to previous study by Sivakamasundari and Jeeva (2012), screening of methanol, diethyl ether and acetone extracts of medicinal plant *M. pudica* for antibacterial activity showed that among the three extracts, methanol extract was more effective than diethyl ether and acetone. Another study also reported that alcoholic extract of plants is better than aqueous extract. Moreover, ethanol extract of *M. pudica* was much effective against bacteria and extraction of crude extract of plant materials is largely dependent on the type if solvent used (Parekh and Chanda, 2006; Aktar *et al.*, 2010).

2.5 Streptomycin

Streptomycin is an antibiotic agent that usually applied in scientific study of antimicrobial activities of plant extracts. There has been considerable interest recently in antibiotic agents streptomycin and always been used as positive control to test growth of bacteria in the media.

Streptomycin is a water soluble aminoglycoside that derived from *Streptomyces griseus* and it is marketed as the sulphate salt of streptomycin. Streptomycine sulphate with molecular formula is $(C_{21}H_{39}N_7O_{12})_2$ -3H₂SO₄ and has molecular weight 1457.41. Usually people take streptomycin to treat or prevent infections that are proven suspected to be caused by bacteria. This antibiotic also functioning as reducing the development of drug-resistance bacteria and maintain the effectiveness of streptomycin and other antibacterial drugs.



REFERENCES

- Aktar, A., Neela, F. A., Khan, M., Islam, M. S. and Alam, M. F. 2010. Screening of ethanol, petroleum ether and chloroform extracts of medicinal plants, *Lawsonia inermis* L. and *Mimosa pudica* L. for antibacterial activity. *Indian Journal Pharmacy Science* 72: 388-92
- Ali, S. S., Kasoju, N., Luthra, A., Singh, H., Sharanabasava, A., Sahu, and Bora, U., 2008. Indian medicinal herbs as source of antioxidants. *Journal of International Food Research* **41**: 1-15
- Ayoola, G. A., Coker, H. A. B., Adesegun, S. A., Adepoju-Bello, A. A., Obaweya, K., Ezennia, E. C., Atangbayila, T. O. 2008. Phytochemical screening and antioxidant activities of some selected medicinal plants used for malaria therapy in southwestern nigeria. *Tropical Journal of Pharmaceutical Research* 7(3): 1019-1024
- Azly b. Mohd Yusof. 1988. *Rumpai panduan berilustrasi*. Ampang, Selangor: Dewan Bahasa dan Pustaka
- Backman, P. A., Wilson, M. and Murphy, J. F. 1997. Bacteria for biological control of plant diseases. *Environmentally Safe Approaches to Crop Disease Control*. Pp. 95-109
- Baki hj Bakar. 2004. Invasive weed species in Malaysian agro-ecosystems. *Malaysian Journal of Science* **23**: 1-4
- Barry, T. N. 2000. Polyphenols in human health. *Tannin in Livestock and Human Nutrition* **92**: 4-6
- Bauer, A. W., Kirby, W. M., Sherris, J. C., and Turck, M. 1966. Antibiotic susceptibility testing by a standardized single disc method. Am. J. Clin. Pathol **45**: 493-496
- Cardoso, M. O., Ribeiro, A. R., Luciana, R. S., Fernando, P., Hamilton, L. S., Carlos, T. P., Silvio, L. and Vladimir, P. 2006. Antibiotic resistence in Salmonella Enteritidis isolated from broiler carcasses. *Brazilian Journal of Microbiology* **37**:368-371
- Cespedes, C. L., El-Hafidi, M., Pavon, N., and Alarcon, J. 2008. Antioxidant and cardioprotective activities of phenolic extracts from fruits of Chilean blackberry *Aristotelia chilensis* (Elaeocarpaceae), Maqui. *Food Chemistry* **107**: 820-829
- Chowdhury, S. A., Jannatul Islam, Md. Mahfujur Rahaman, Md. Mostafizur Rahman, Nowshin Nowaz Rumzhum, Rebeka Sultana, Most. Nazma Parvin. 2008. Cytotoxicity, antimicrobial and antioxidant studies of the different plant parts of *Mimosa pudica. Stamford Journal of Pharmaceutical Sciences* **1**(2): 80-84
- Datta, S., Costantino, N., Zhou, X. and Donald, L. C. 2008. Identification and analysis of recombineering functions from gram-negative and gram-positive bacteria and their phages. *Proceeding of National Academy of Science* **105**(5): 1629



- Doss, A., Vijayasanthi, M., Parivuguna, V. and Anand, S. P. 2011. Evaluation of antibacterial properties of ethanol and flavonoids from *Mimosa pudica* linn and *Panicum maximum* jacq. *Plant sciences feed* **1**(2): 39-44
- Elsa, L., Harshadrai, R., Florian, J. S., Fernando, C., Ana, F., Ana, R. C., Celia, A., Andre, M. A., Ana, V., Coelho, and Elvira, S. B. 2011. The effect of Tannins on Mediterranean Ruminant Ingestive Behavior: The Role of the Oral Cavity. *Open* access Molecules 16: 2766-2784
- Fasihuddin Ahmad Hasmah Raji. 1993. Kimia hasilan semulajadi dan tumbuhan ubatan. Dewan Bahasa dan Pustaka. Malaysia
- Figen, M. T. 2006. Saponins versus plant fungal pathogens. *Journal of Cell and Molecular Biology* **5**: 13-17
- Gandhiraja, N., Sriram, S., Meenaa, V., Kavitha, S. J., Sasikumar, C., and Rajeswari,
 R. 2009. Phytochemical screening and antimicrobial activity of the plant
 extracts of *Mimosa pudica* L. against selected microbes.
 Ethnobotanical Leaflets 13:618-24
- Girish and Satish, S. 2008. Antibacterial activity of important medicinal plants on human pathogenic bacteria-a comparative analysis. *World Applied Sciences Journal* **5**(3): 267-271
- Gurcharan, S. 2004. Plant systematics and integrated approach. Science publishers. Enfield, United states of America.
- Han, X., Shen, T., and Lou, H. 2007. Dietary polyphenols and their biological significance. *International Journal of Molecular Science* 8: 950-988
- Hostettmann, K. A. and Marston, A. 1995. Saponins. *Chemistry and pharmacology of natural products*. Cambridge University Press, Cambridge, United Kingdom.
- Jie, M. S. F. and Choi, C. Y. C. 1991. Chemistry. *Journal International Federal Clinical*. 3: 122
- John, A. P. 2001. Healing Plants of Peninsular India. USA; CABI publishing, pp: 557 563
- Joseph, M. L., Noe, R. P., Ilia, A. G., Michael, H. F. & Thorson, J. S. 2005. *Enhancing the anticancer properties of cardiac glycosides by neoglycorandomization*. The Scripps Research Institute, La Jolla, CA
- Kaur, P., Nilesh, K., Shivananda, T. N. and Gagandeep, K. 2001. Phytochemical screening and antimicrobial activity of the plant extracts of *Mimosa pudica* L. against selected microbes. *Journal of Medicinal Plants Research* 5(22): 5356-5359
- Keating, B. 1994. The power of neem. The Miraculous Healing Herbs. 1-2
- Khare, C. P. 2004. Indian herbal remedies. Society for new age herbals. New Delhi, India



- Kinsey, B. T. 2012. Hawaiian plants and tropical flowers. *A guide to the flowers and plants of Hawaii*
- Koh, H. W., Chua, T. K., Tan, C. H. 2009. *Mimosa pudica* L. (Leguminosae). *A guide to medicinal plant.* 99-100
- Kokate, C. K., Purohit, A. P., Gokhale, S. B. 1995. Pharmacognosy. 3rd edition. Nirali Prakashan, Prune
- Lubica, H. 2011. Flavonoids in prevention of diseases with respect to modulation of Capump function. *Interdisciplinary Toxicol* **4**(3): 114–124
- McCarty, B. L. and Murphy, T. R. 1992. Control of turfgrass weeds. 1-2
- Mukesh, C. S., and Smita, S. 2010. Phytochemical and pharmacological screening of combined Mimosa pudica Linn and Tridax procumbens for in vitro antimicrobial activity. *International Journal of Microbiological Research*. 1(3): 171-174
- Mukeshwar, P., Dipali, S., Manoj, K., Vyas, Shobit, G., Abhishek, S., Pooja, S., Vinay,
 K., Surendra, K. C. 2012. Antibacterial evaluation of plant extracts: an insight into phytomedicine. *International Journal of Phytomedicine* 4(1): 470
- Neil, C. B. 1999. 7 Alkaloids. Cambridge, United Kingdom
- Noormaizurah Omar. 1999. Rumpai kebaikan, keburukan & pengawalan. Serdang, Selangor: Universiti Kebangsaan Malaysia
- Pande, M. and Pathak, A. 2009. Aphrodisiac activity of roots of Mimosa Pudica Linn ethanolic extract in mice. *International Journal of Pharmaceutical Science and Nanotechnology* **2**(1):447-485
- Pankaj, O. 2004. Chhui-Mui or Lajwanti (*Mimosa pudica* Linn.) Society for Parthenium Management (SOPAM). Raipur, India
- Parekh, J. and Chanda, S. 2006. Screening of aqueous and alcoholic extracts of some Indian medicinal plants for antibacterial activity. *Indian Journal Pharmacy Science*, **68**: 835-842
- Rajeshwari, S. and Raghunath, P. 2012. GCMS studies of *Mimosa pudica*. International Journal of PharmTech Research **4**(1): 93-98
- Rekha, R. 2009. Preliminary phytochemical analysis & anti-bacterial activity of *Mimosa pudica* linn leaves. *Journal of global pharma technology* **1**(1): 76-81
- Rekha, R. and Ekambaram, K. 2010. Hypolipidemic activity of chloroform extract of *Mimosa pudica* leaves. *Avicenna Journal Medicine Biotechnology* **2**(4): 215 221
- Saifuddin, K., Jinesh, K. S., Suresh, D. K., Rajnish, K. S., Narasimha, R., Sunil, K. 2011. Evaluation of anti-diarrhoeal potential of ethanolic extract of *Mimosa pudica* leaves. *International Journal of Green Pharmacy* **5**(1): 75-78



- Santiago, F. E., Thomas, S. W., Cynthia, L. W., Margaret, A. R., and Richard, E. L. 2005. Genomic divergence of Escherichia coli strains: evidence for horizontal transfer and variation in mutation rates. Research Article International Microbiology 8: 271-278
- Singh, R., Singh, S., Kumar, S. and Arora, S. 2007. Evaluation of antioxidant potential of ethyl acetate extract/fractions of *Acacia auriculiformis* A. Cunn. *Food Chemistry Toxicol* **45**:1216-1223
- Sivakamasundari, S. and Jeeva, S. 2012. Screening of methanol, diethyl ether and acetone extracts of medicinal plant *Mimosa pudica L*. for antibacterial activity against *Enterobacter aerogenes*. *Drug Invention Today* **4**(3): 355-357
- Smith, R., Thomas, W., Gaskell, M., Mitchell J., Steven, T., Fouche, C. 2000. Weed management for organic crop. Organic Vegetable Production In California Series, 1-2
- Sofowora, E. A. (1982). Medicinal Plants and Traditional Medicine in African. Chichester, John Willey and Sons, London. pp. 109-111.
- Sriram, S., Meenaa, V., Kavitha, V. and Agnel, A. J. N. 2011. GC-MS study and phytochemical profiling of *Mimosa pudica* Linn. *Journal of Pharmacy Research* 4(3): 741-742
- Srivastava, V., Sharma, A. and Alam Imran. 2012. A review on ethnomedical and traditional uses of *Mimosa pudica*. *International Research Journal of Pharmacy* **3**(2): 41-44
- Stojek, N. M., Szymanska, J. and Dutkiewicz, J. 2008. Gram-negative bacteria in water distribution systems of hospitals. An Agriculture Environment Medicine 15:135 142
- Suresh, K., Deepa, P., Harisaranraj, R. and Vaira, A., V. 2008. Antimicrobial and phytochemical investigation of the leaves of *Carica papaya* L., *Cynodon dactylon* (L.) Pers., *Euphorbia hirta* L., *Melia azedarach* L. and *Psidium guajava* L. *Ethnobotanical Leaflets* **12**: 1184-91
- Trease, G. F. and Evans, W. C. (1978). A Textbook on Pharmacgnosy. Eleventh Edition, Baillere Trindal London. pp. 22-40
- Ukoha, P. O., Egbuonu, A. C., Cemaluk, O. L., Namdi and Ejikeme P. M. 2011. Tannins and other phytochemical of the *Samanaea saman* pods and their antimicrobial activities. *African Journal of Pure and Applied Chemistry* 5(8):237-244
- Yao, L. H., Jiang, Y. M., Shi, J., Tomas, B. F. A., Datta, N., Singanusong, R. and Chen. S. S., 2004. Flavonoids in food and their health benefits. *Plant Foods for Human Nutrition* 59(3): 113-122
- Zaika, L. L. 1988. Spices and herbs: Their antimicrobial activity and its determination. Journal Food safety **9**: 97-11

