Preliminary phytochemical screening of different solvent extracts of flower and whole plant of *Wedelia biflora*

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Abstract. In this study, comparison of preliminary phytochemical screening of *Wedeliabiflora*was conducted using the n-hexane and ethyl acetate solvents extracts. The *W.biflora* samples were grouped into flowers andleaves. The different solvents of flowers extracts exhibited secondary compound viz. flavonoid, alkaloid, fenol, tannin, steroid, and saponin. While the extracts of leaves revealed flavonoid, alkaloid, fenol, tannin and steroid without saponin. Thus, the present of various bioactive components in the *W. biflora*extract might bepotential for responsible on the pharmacological activities including antifungi, antibacterial, antioxidant, anti-inflammatory and anticancer.

1. Introduction

The genus *Wedelia* is widely distributed throughout the world specifically in tropical and warm temperature regions. It consists of approximately 65 – 70 species[1].*Wedelia biflora* is a very attractive plant due to its nearly constant and prolific blooming. Besides, *W.biflora* produces primary and secondary constituents that are responsible in therapeutic activities, for instance antibacterial, anti-mutagenic, anti-viral, anti-inflamantory, antioxidant. Anticonvulsant and anti-cancer effects [1; 2; 3; 4; 5].

W. *biflora*is called by different names such as sernai/ seruni(Indonesia), Wedelia kuning (Malaysia), Atiat (Puluwat), Bay Biscayne creeping oxeye, creeping daisy, creeping wedelia, rabbit's paw, Singapore daisy, gold cup, water zinnia, trailing daisy and wild marigold (English), America hama-guruma (Japan), hansenfuss (Geramany), di jinhua (China), Singapore-madeliefie (Africa), kradum tong (Thailand), ampelkrage (Sweden), arnica-do-mato, pseudo-arnica, vedelia (Brazil), and uttelia (Marshall Islands), and [6].

Moreover, *W. biflora* is a member of the Aster family, Asteraceae. The taxonomic position of this plant is as follows:

Kondom – Plantae
Division - Magnoliophyta
Class – Magnoliopsida
Order – Asterales
Family – Asteraceae
Genus – Wedelia
Species – *W. biflora*[7].

Wbiflora was first identified by Georg Wolfgang Wedel (1645 – 1721), Professor of Botany at Jena, Germany. The morphological description of this plant is a creeping or climbing habitoften creates a dense ground cover approximately 20 cm tall and occasionally upto 70cm tall that crowd out the growth of other species. The stemis rounded, green and coarsely hairy with grow up to 2m long and regularly develop roots (adventitious roots) at its nodes. Short, semi-upright (ascending), flowering branches are produced of these creeping stems. The leave is attractive bright shiny green and oppositely arranged.Its leaves have long and wide up to 9 cm and 5 cm, acute at the apex and winged at the base, irregularly toothed (serrated) margins. They are glossy in appearance, mostly hairless (glabrous).The flower is single attractive bright-yellow, the head are daisy-like in appearance and are borne on the end of terminal and axillary stalks (peduncles) 2-9 cm long. Each flower-head has 8-13 yellowish "petals" that are 6-15 mm long with 1- 3 finely with chaffy bracts toothed tips. In the centre of these flower-heads there are numerous tiny yellow tubular disc florets 4-5 mm long. The ray and disc florets are both yellow. The base of each flower-head (capitulum) in enclosed in a row (involucre) of narrow (lanceolate) green bracts (1 cm long). Flowering occurs throughout the year, but is most common from spring to autumn (Figure 1)[6].

Ethno medically, *Wedelia* is used in traditional medicine in the Caribbean and Central America against bronchitis, colds, abdominal pains, dysmenorrhoeal and fertility enhancer [8]. The Miskito Indians of eastern Nicaragua use leaves for treatment of kidney dysfunction, cold, stingray wounds, snakebite, purge and amenorrhea [9]. The Tamil Nadu, India employs this plant for treatment of wounds, ulcers, sore throat, varicose of veins, skin diseases, headache and stomach ache. The flower is said to be violent urgative[2].Hong Kong developed *W. trilobata* for the dealingwith the common cold, hepatitis, indigestion and infections. In Trinidad and Tobago, it against reproductive problems, amenorrhea, and dysmenorrheal.Vietnam consumed Wedelia for the treatment of fewer and malaria[6]. Mishr*a et al*. [3]identified that the *W.chinensis* had anticonvulsant activity due to the presence of glycocides, alkaloid, flavonoid, and steroid. Moreover, Manjamalai and Grace[1] and Tsai *et al*. [10] developed the positive effect of essential oil of *W.chinensis* against lung and prostate respectively.

According to literature above, *W. biflora* that is very closed to species of *W. Chinensis* and *W. trilobata* could be considered has great potential in pharmacological activities instead the research about this species in medicine is not yet developed. Therefore, the objective of the present study is to explore the present of secondary metabolites in the different solvents (n-hexane and ethyl acetate) of*W. biflora* extracts and compare to fresh samples. The finding of this preliminary study would be developed as phytomedicine modern against breast cancer.

Figure 1. *Wedelia biflora*

2. Materials and Methods

2.1. Plant Materialand Chemicals

Sample of *W. biflora* was collected from Langsa, Aceh-Indonesia. The plant material was dried at room temperature to retain their fresh colour and also prevent the decomposition of active compounds. The dried plant materials were cut into thick slices (±0,3 cm)and stored in air tight container. The chemicals used were analytical grade and purchased from C.V. Multikreasi Bersama, Medan Indonesia including n-hexane, ethyl acetate, and aquaeous.

2.2. Methods

2.2.1. Extraction Process

The coarse materials offlower and leavesof *W. biflora* were extracted with various sequential solvents of n-hexane and ethyl acetate. The different extracts were filtered by filter paper (Whatman No.1) to removed debris and concentrated under rotary vacuum evaporator. The extraction yield was calculated based on equation (1) and this dried residue was stored in air tight container and subjected to qualitative phytochemical screening for the identification of the phytoconstituents.

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$$\text{Extraction yield (\%)} = \frac{mass\ of\ extracted\ product}{mass\ of\ raw\ material} x\ 100 \text{ ……(1)[11].}$$

(a) (b) (c)

Figure 2. Flowers(a), leaves of *W.biflora* (b), extraction process (c)

2.2.2. Phytochemical Screening

Analysis of secondary compounds in *W. biflora* was carried by following procedures mentioned:

2.2.2.1 Detection of Alkaloids

Wagner's Test: ±10 mg of the each extract was taken and was dissolved in 2 ml of the Wagner's reagent. After dissolving the both, the appearance of reddish brown colour precipitates confirms the presence of alkaloids in the plant extract.

Mayer's test: Filtrate was treated with Mayer' test reagent (Potassium Mercury Iodide). Formation of a yellow colourer precipitate indicates the present of [12].

2.2.2.2 Detection oft Flavonoids

About 10 mg of the each extract was taken and few drops of diluted NaOH were added to the each. The appearance of yellow colour which disappears or become colourless after adding few drops of diluted H_2SO_4 confirms the presence of flavonoids in the plant extract[13].

2.2.2.3 Detection of Saponins

About 2g of the samples extractwas mixed with 10ml of distilled water and shaken vigorously (±12 min) for a stable persistent froth. Formation of froth on top of the test tube shows the presence of saponins in the plan extract [2].

2.2.2.4 Detection of Steroids

About 10 mg of the each extract and 1ml of concentrated H_2SO_4 was added to the each by the side walls of the test tube. Appearance of dark reddish green colour confirms the presence of steroids in the plant extracts[12].

2.2.2.5 Detection of Tannins

About 10 mg of the each extract was dissolved in 45% of the ethanol. The test tubes were then boiled for 5 min and 1 ml of 15% ferric chloride solution was added to each. The appearance of greenish to black colour confirms the presence of tannins in the plant extract[14].

3. Results and Discussions

In this study, n-hexane and ethyl acetate solvents were used for extraction process. Based on calculated extraction yields, it is clear observed that each type of solvent can pull out the chemicals from W. biflora differently and or has influence to extraction yield (Fig. 1). In the solvent extraction method, the yields of Leaves extract obtained fromn-hexane and ethyl acetate solvents were 0.26 and 0.36% respectively.The finding is accordance with Sriariyanun*et al*. [11] that stated ethyl acetate solvent possessed high extraction yield because this solvent can extract both polar and non-polar compounds. Unlike, n-hexane can extract only non-polar compounds.

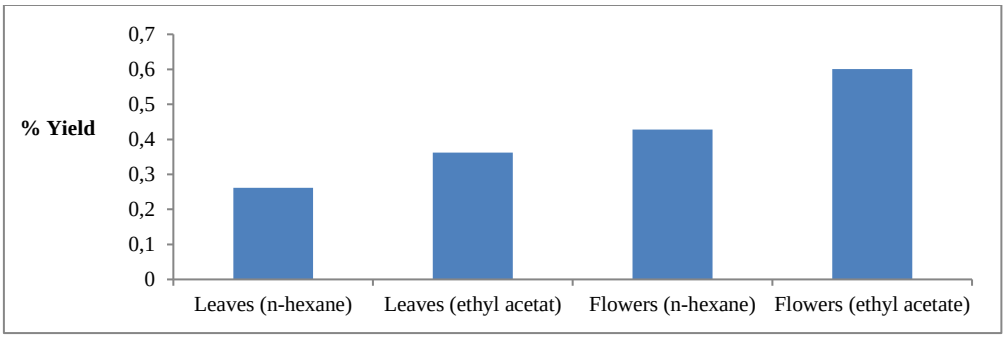

Figure 3. Extraction Yield of *W. biflora* from different solvents

Further study was preliminary phytochemical investigation. This step to be a key in developing new drugs since it provides the basic source for establishment of several pharmaceutical industries such as detection of alkaloids, flavonoid, phenolic compounds, tannin, saponin, steroid etc which have pharmalogical activities. The constituents present in a plant play a significant role in the identification of crude drug. The knowledge of bioactive compounds of this plant could be needed in designing new, safe and effective formulation of nutriceuticals and phytotherapeutics for humans containing medicinal parts of this plant[15]. In this study, the n-hexane and ethyl acetate extracts of *W. biflora* (flowers and whole plants) showed the presence of phytochemical constituents namely alkaloid, flavonoids, tannin, saponins, steroids and absence of terpenoid, as tabulated in Table 1.This initial phytochemical screening test might be helpful in the screening of the bioactive compounds and eventually might help to detection, development and designingof new drugs, safe and effective formulation of nutriceuticals and phytotherapeutics for humans containing medicinal parts of this plant[15].In addition, these tests make easy its qualitative separation and quantitative estimation of pharmacologically active chemicals compounds [16].

Flavonoids and tannin are phenolic compounds that are acting asantioxidants or free radical scavengers. Tannins and alkaloids may play role as anti-hyperglycaemic and anti-inflamantory activities [17], alkaloid alone has led to the invention of powerful pain killer medications [18]. Steroid is to be an important cardio tonic activities posse's antimicrobial property and also used in herbal medicines and cosmetics The terpenoids have also been revealed to decrease blood sugar level in animal studies [19]. Saponin act as bioactive antibacterial agents in plants and also used to treat hypercholesterolemia, hyperglycemia and obesity [20].

Table 1. Phytochemicals screening for fresh and flowers and whole plant extract of Wedelia biflora

Chemicals Compounds	Solvents					
	Extract Flowers		Fresh Flowers	Whole plants		Fresh Plants
	n-Hexane	Ethyl acetate	Distilled water	n-Hexane	Ethyl acetate	Distilled water
Alkaloids	+	+	-	+	+	+
Flavonoids	+	+	+	+	+	+
Fenol	-	+	+	-	-	+
Saponin	-	-	+	-	-	-
Steroid	-	+	+	+	+	-
Tanin	-	+	-	-	-	+

(+) = present and (-) =absent

4. Conclusion

The study investigated the various secondary compounds that were existing in the different solvents extraction of *W. biflora*. The results publicized that the flower extract contain alkaloids, flavonoids, fenol, steroids, tannin, and saponin. On the other hand, in the leaves extract of *W. biflora* is absence of saponin. This preliminary screening would be valuable for further studies in the scope of farmacology [21].

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