

Preparation Of Siam Weed Extracts

Nor Shamsiah Mohamed, Koh Ching Theng
Faculty of Mechanical and Manufacturing Engineering,
University Tun Hussein Onn Malaysia, 86400, Parit Raja,
Johor, Malaysia.

* ctkoh@uthm.edu.my

ABSTRACT

Siam Weed is a perennial shrub in the Asteraceae family that spreads throughout tropical and subtropical areas of the world. Siam weed has been used as a traditional herbs for ailments such as fever and soft tissue wounds. Previous studies show potential utilization of Siam weed extracts in medical areas. This study is aimed to produce Siam weed extracts to be used to further exploit its application in medical products. We first repeated step of preparation for Siam weed crude extract and then modified the steps to develop a new way of preparation of Siam weed aqueous extract that is suitable for further development of medical application. The chemical properties of the aqueous extract were investigated using Fourier transform infrared spectroscopy (FTIR). This characterization results revealed that the Siam weed aqueous extract was successfully prepared and its chemical properties are similar to that of crude extracts. By maintaining similar chemical properties, Siam weed aqueous extract can be useful to further exploitation of Siam weed extracts in medical applications.

Keywords: *Siam weed crude extract, Siam weed aqueous extract, FTIR.*

Introduction

Siam Weed (*Chromolaena odorata* (L.) King and Robinson) is a perennial scandent or semiwoody shrub in the Asteraceae family. It is native to Central and South America and also spreads throughout the tropical and subtropical areas of the world, including Thailand and Malaysia. Siam weed has been used as a traditional herbs in many countries for a variety of ailments such as fever, influenza, cold, cough, diabetes, malaria, bleeding, soft tissue wounds, inflammation and wound healing [1,2]. It also used as an antispasmodic, antiprotozoal, antitrypanosomal, antibacterial, antifungal, antihypertensive, astringent, diuretic and hepatotropic agent [3,4]. Oladure B. Taiwo et al. [5] proved the existence of anti-inflammatory as well as antipyretic activity of methanolic extract of Siam weed. The investigation also extended to the effects of the extract on intestinal transit of charcoal meal and castor oil-induced diarrhea. The extract also produced significant reduction in rectal temperature in mice by yeast suspension. The extract produced antimotility and antidiarrhoeal effects in intact mice.

Numerous studies demonstrated that Siam weed extract (SWE) accelerates hemostatic and wound healing [6, 7]. Investigation on the wound healing effect of aqueous leaf extracts of Siam weed in rabbits was evaluated by Biswal PR et al. [8]. The extension research also on the antibacterial effect in different isolates of bacteria. Studies also determined that different amount of extract can also affect Siam weed effectiveness. Phan Toan Thang et al. [9]

evaluated that the total ethanolic extract at 400 and 800 µg/ml showed maximum and persistent protective cellular effect on oxidant toxicity at low or high doses of oxidants.

Significant protective effects on fibroblast against hydrogen peroxide was also showed by the total ethanolic extract. While for keratinocytes, the dose amount of the total ethanolic extract only affects the hydrogen peroxide but the protective action correlated with oxidant dosage. On the other hand, the significant inhibition of collagen and contraction by eupolin extract at 50-200 µg/ml in various concentrations of collagen was evaluated by Thang T. Phan et al. [10]. In addition, in previous research of Chakraborty et al. [11], the ethanolic extract of Siam weed showed maximum analgesic activity at a dose of 300 mg/kg. The comparison was done between petroleum ether extract and chloroform extract, showing moderate activity at the same dose. While Jitendra Patel et al. [12] studied the anthelmintic activity of Siam Weed where the ethanolic extract exhibited a large amount of anthelmintic activity at highest concentration of 100 mg/ml. These previous studies proved that different amount and different type of extracts from Siam weed can affect its functions. Siam weed is found to be a highly efficacious medicinal herbs according to the indigenous and complementary remedial systems [13]. However, the preparation steps of Siam weed extracts are not universal as there are many ways of preparing the extracts [10,14]. Previous study also showed that different type of extracts can affect the effectiveness. In this study, we modified existed step and developed a new way of preparation of Siam weed aqueous extract that is suitable for further developing in medical application. We evaluated and compared the chemical structures of the Siam weed extracts with previous research to assure the similar composition of the extracts.

Materials and Methods

Plant materials

Siam weed plants were collected at 1.9816° N, 102.8784° E, Parit Sulong 85300 Johor Darul Takzim, Malaysia. The collected plants were washed with running tap water, air dried and stored in air-tight bottles between collection and extract preparation.

Preparation of Siam weed crude extracts

Steps of preparation of Zhao et al., [14] was followed to produce Siam weed crude extract. Siam weed stem Acetosolv lignin (AL), was delignified by 93% acetic acid–water (v/v) solution with 0.1% w/w HCl as catalyst at boiling point temperature (~107 °C) for 3 h with the liquid-to-solid ratio of 10:1 (v/w). The mixture was filtered using Whatman No.1 filter paper (Merck, Germany) into a clean beaker and concentrated to dryness using double boil technique for 3 hours. Then, seven volumes of water were poured into the concentrated solution in order to precipitate the dissolved lignin. The crude Acetosolv lignin was separated by centrifugation and washed with deionized water [14].

Preparation of Siam weed aqueous extracts

The plants were dried in hot air oven at 60°C for 24 hours. The dried leaves were grounded with a 110V fine grinding planetary ball mill machine (Deco, China) to produce moderate Siam weed powder. Siam weed powder of 100 g was weighed using a weighing balance (Citizen, India). The weighed sample was soaked in 600mls of distilled water contained in a beaker and stirred for 12 hours. The mixture was filtered using Whatman No.1 filter paper (Merck, Germany) into a clean beaker and concentrated to dryness using double boil technique for 3 hours. The extract was stored in an air-tight bottle at 4°C until

testing. Figure 1 illustrated the flowchart of the preparation of Siam weed aqueous and crude extracts.

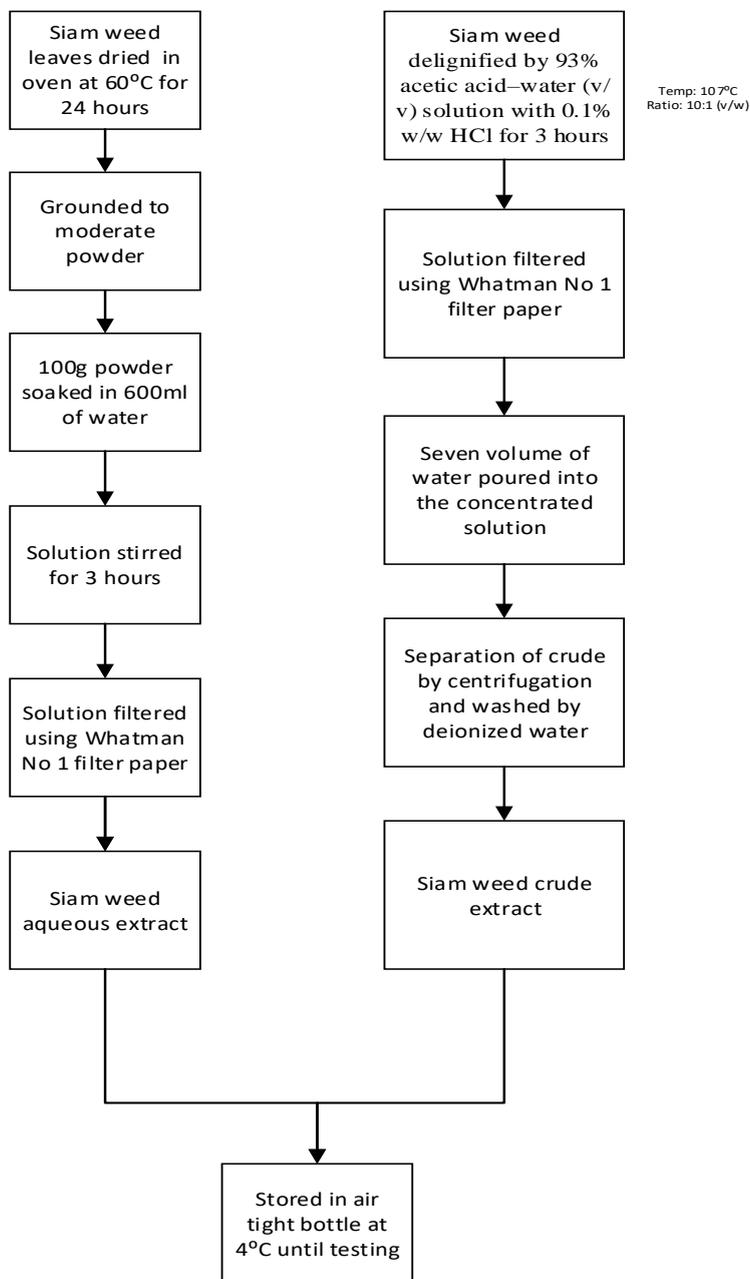


Figure 1: Flowchart of preparation of Siam weed aqueous extract (left) and crude extract (right).

Characterization

A Spectrum 100 FTIR (Perkin Elmer, United States) was used to determine infrared spectra of the extract. The spectra were recorded in the absorption

band mode in the wavelength range of 4000–600 cm^{-1} . All the spectra were baseline corrected and normalized using Perkin Elmer Spectrum software.

Results and Discussion

Table 1: Location of FTIR spectra bands and corresponding assignments of Siam weed crude and aqueous extracts.

Band (cm^{-1})	Assignment	Crude extract band location (cm^{-1})	Aqueous extract band location (cm^{-1})
3500-3300	O–H stretching	3430	3280
~2940	C–H asymmetric stretching in methyl and methylene group.	2940	2938
2850-2830	C–H symmetric stretching in methyl and methylene group.	2840	2850
1720-1680	C=O stretching in unconjugated ketone, carbonyl, and ester groups	1735	1720
1660-1640	C O stretching in conjugated p-substituted aryl ketones	1650	1638
1610-1590	Aromatic skeletal vibrations plus C O stretching	1600	1600
1515-1505	Aromatic skeletal vibrations	1510	1516
1470-1460	C–H deformations (asymmetric in –CH ₃ and –CH ₂ –)	1463	1451
~1420	Aromatic skeletal vibrations combined with C–H in-plane deformations	1423	1423
1365-1370	Aliphatic C–H stretching in CH ₃ (not in OMe) and phenolic OH	1373	1372
1235-1210	C–C plus C–O plus C O stretching (Gcondensed >Getherified, typical of G units)	1234	1252
1166	Typical for HGS lignins	-	-
1130-1120	Typical of S units; also C O stretching and symmetric stretching of C–O–C	1126	1128
1035-1030	Aromatic C–H in-plane deformation (G > S) plus C–O deformation in primary alcohols plus C O stretching (unconjugated)	1035	1026
875-870	C–H out of plane in positions 2, 5 and 6 (G units)	873	871

In this report, we compared the chemical properties of two extracts of Siam weed, aqueous and crude extracts as showed in Figure 2(a) and 2(b)

respectively. Siam weed aqueous extract prepared in this report showed similar bioactive compounds to Siam weed crude extract characterization from Zhao et al [14].

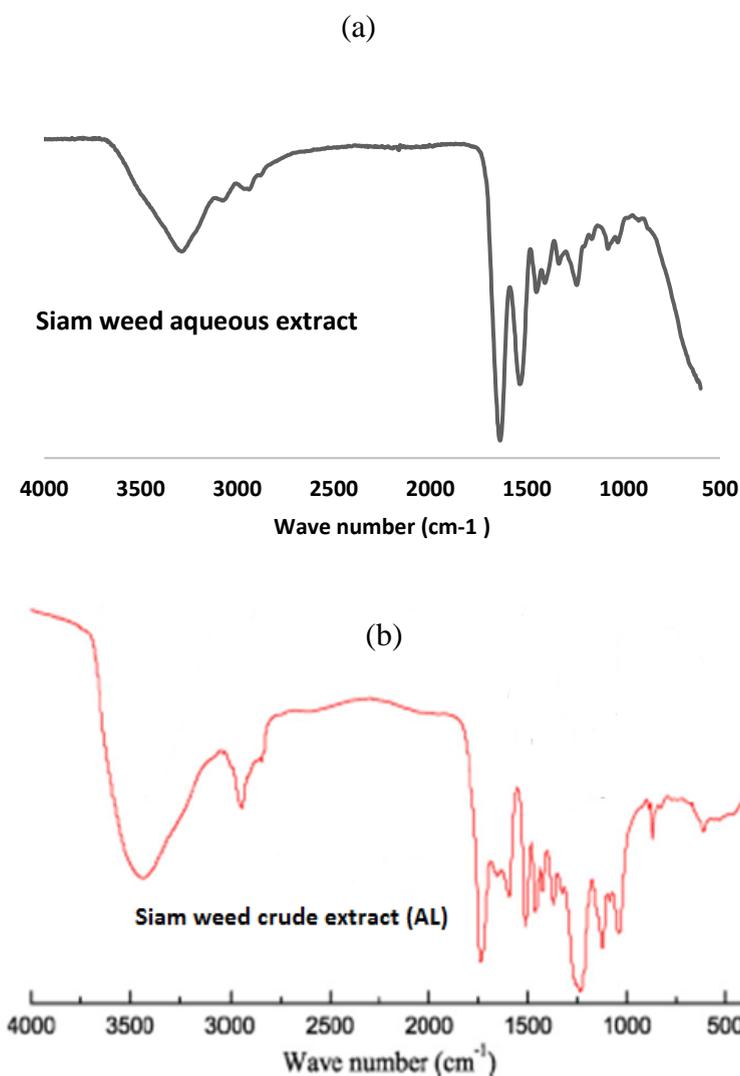


Figure 2: Comparison of FTIR spectra of the Siam weed aqueous extract (a) and Siam weed crude extract taken from Zhao et al., [14](b).

The reaction progress was investigated through FT-IR spectroscopy. For Siam weed aqueous extract, the band at 1510cm⁻¹ (aromatic skeletal vibrations) was used as a reference to estimate the relative intensities of the other bands. The band at ~3280.44 cm⁻¹ was assigned to the stretching of O–H groups similar to Siam weed crude extract band at ~3430 cm⁻¹, inclusive of R–OH and Ar–OH. Apparent characteristic bands for the aromatic skeletal vibrations at ~1600, ~1516 and ~1451cm⁻¹ were observed while Siam weed crude extract recorded ~1600, ~1510 and ~1463cm⁻¹. There was also obvious signal at 1372cm⁻¹ which was due to aliphatic C–H stretching in CH₃ close to 1373cm⁻¹ band of comparison. Further evidence for more carbonyl moieties in Siam weed extract is that it had strong intensities at ~1252 and ~1026cm⁻¹ while ~1234 and ~1035cm⁻¹ recorded by Zhao et al [14] respectively. Our FT-IR results are in good agreement with the report of Zhao et al., [14].

Table 1 illustrated the location of FTIR spectra bands and corresponding assignments of Siam weed crude extract [14] and Siam weed aqueous extract. It was proven that the corresponding band and assignments between the crude extracts from Zhao et al [14] and Siam weed aqueous extracts were similar. This characterization results clearly revealed that the Siam weed aqueous extract was successfully prepared and identified as the existed biomaterials and its reaction progress were similar to the past researches. There were many reports evaluating the chemical composition of different types of Siam weed extracts thus many ways of preparing the extracts were developed. This indicated that the new developed way of preparing the Siam weed aqueous extract in this paper can be useful to further analysis of Siam weed extracts.

Conclusion

This work shows a preparation method in producing Siam weed aqueous extract. The successfulness of the preparation of the Siam weed aqueous extracts is crucial in order to maintain elemental components of Siam weed in medical products. The experimental results of FTIR indicated similar chemical properties of Siam weed aqueous extract as compared to that of Siam weed crude extract in literature. Such Siam weed extracts will be added to various medical products such as burn relief patch and tissue engineered graft in future study.

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