Phytochemical Screening of the Solvent Extracts of the Dried Leaves of the Mixture of *Chromolaena odorata* L. and *Sida acuta* Burm. F.

Benjamin Sarfo Appau [1*], Yaw Ameyaw [2], Benjamin Obeng Konadu [3], Nawal Nuhu [4]
[1] Department of Science, Assin Manso Senior High School, Assin Manso, Ghana
[2] Department of Biology Education, University of Education, Winneba, Ghana
[3] School of Teaching and Learning, Illinois State University, USA
[4] Department of Science, Ghana Senior High School, Koforidua, Ghana

* Corresponding Author: **Benjamin Sarfo Appau**

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Abstract
Herbal medicines are affordable and cause minimal side effects and this has contributed to the high patronage of herbal medicine recently. *Chromolaena odorata* L. is also traditionally used for the treatment of various ailments in humans and animals such as wound healing.
Sida acuta is also useful in treating diseases such as headache, cold, fever, skin diseases, urinary diseases, ulcer, snake bite, facial paralysis. The researcher used *Chromolaena odorata* L. and *Sida acuta* Burm. F. that are found in Assin Andoe in the Assin South District. The researcher picked 15g each of the powdered leaves of *C. odorata* and *S. acuta* making 30g and mixed them in a beaker. Distil water, 70% ethanol, 30% ethanol and 30% methanol were the solvent used to perform maceration extraction of the mixture. The researcher ran a qualitative test to assess the phytochemical compounds present in the various extracts of the herbal mixture. It was revealed in the experiment that saponins, flavonoids, alkaloids, tannins, steroids, terpenoids, phenol, phlobatannin, gum and mucilage are the phytochemicals present in the various extracts of the mixture of *Chromolaena odorata* and *Sida acuta* leaves. Glycosides and anthraquinones on the other hand were absent in all the extracts.
Again, it was concluded that 30% methanol (v/v) and distil water are most effective in extracting *Chromolaena odorata* L. and *Sida acuta* Burm. It was recommended that quantitative assessment of the phytochemicals present in the herbal mixture should be performed to investigate the relative abundance of the phytochemicals present.

Keywords: Phytochemical Screening, Solvent Extracts, Dried Leaves, *Chromolaena odorata* L., *Sida Acuta* Burm. F

1. Introduction

Many plants have been used traditionally to treat wounds due to their high efficiency in the healing process (Santos-Buelga *et al*., 2014) [27]. The reason is that herbal medicines are affordable and cause minimal side effects (Ekor, 2014) [12]. From Nagori *et al*. (2011) [22], extensive research has been done in the field of wound healing and wound management through plant-based medicines recently. Many plants contain antioxidants which are beneficial and are of therapeutic use in several ailments that relate to potential pathologic actions of oxidants as well as wound healing (Yeoh, 2000) [36]. Since the introduction of *Chromolaena odorata* into Ghana since 1970 according to Ghanaweb (2020) [14], the herb has been used to stop bleeding, it is efficient in healing wounds, treatment of snake poison when it is immediately administered after a bite. The herb is also used to treat stomach aches, bilharzia and also to preserve dead bodies. People in most rural areas in Ghana mash and apply the mashed leaves of *Chromolaena odorata* (Acheampong) on a fresh cut to clot blood in few seconds after the treatment (Ghanaweb, 2020) [14]. *Chromolaena odorata* extract has been shown to stimulate haemostasis and wound healing management. The phytochemical substances in the leaf of *Chromolaena odorata* were used for antibacterial, antifungal, anti-inflammatory, anticancer, antidiabetic, antidiarrheal and hepatoprotective activities (Sirinthipaporn & Jiraungkoorskul, 2017) [30].

Sida acuta has been shown to have wound healing potentials. The methanolic extract of *S. acuta* produced significant healing in wounds treated with it. However, *Sida acuta* has been traditionally used for thousands of years and is used for treating various ailments, including malaria, bacterial infections, ulcers (Tcheghebe *et al*., 2017) [31]. In the Ghanaian society, a decoction of the leaves of the *Sida acuta* is applied on wounds to stop bleeding (CSIR, 2023) [10].

Several studies have investigated the effect of *Chromolaena odorata* and *Sida acuta* on wound healing. For example, Ezeja *et al*. (2012) [13] conducted a study to investigate the effect of *Chromolaena odorata* on wound healing in rats.Thus the present study determined the phytochemical screening of the solvent extracts of the dried leaves of the mixture of *Chromolaena odorata* L. and *Sida Acuta* Burm. F.

2. Literature Review

Classification of *Chromolaena odorata* L.

C. odorata L. is in Kingdom Plantae, Subkingdom Viridiplantae, Infrakingdom Streptophyta, Superdivision Embryophyta, Division Tracheophyta, Subdivision Spermatophyta, Class Magnoliopsida, Superorder Asteranae, Order Asterales, Family Asteraceae, Genus *Chromolaena*, Specific epithet *odorata* (King & Rob, 2016).

Health benefits of *Chromolaena odorata* L.

According to Ikuenobe and Analiefo (2003) cited in Tiamiyu and Okunlade (2020) [32], *Chromolaena odorata* L. is a perfect fallow plant as it met the expected properties of species for fallow plant which include easy establishment, large biomass, fast rate of decomposition and weed suppression. Research done in Nigeria by Ikuenobe and Analiefo showed that infestation of weeds was lower in plots which have been cultivated by *C. odorata* than plots that are modified by natural bush fallow. This is to say *C. odorata* is a very good weed suppressor (Tiamiyu & Okunlade, 2020) [32].

Research done by Kanmegne *et al*. (1999) [17] in central-southern Cameroon indicated that, *C. odorata* significantly improved the soil quality by exchanging potassium with a sandy developed on granites and a sandy – clayey soil developed on gneiss. Approximate constituent analysis of *C. odorata* by Tiamiyu and Okunlade (2020) [32] are 9.26% moisture, 15.28% fiber, 3.56% fat, 18.86% protein and 41.28% Carbohydrate. It was concluded that the possession of these secondary metabolites could be ascribed to its medicinal and nutritional usefulness.

Chromolaena odorata L. is also traditionally used for the treatment of various ailments in humans and animals. (Vijayaraghavan *et al*., 2017). The herb possesses insecticidal properties and is used as a green manure. It is also used for the preservation of dead bodies (Ukwueze *et al*., 2013) [34]. The fresh leaves of *C. odorata* or the decoction has been used by practitioners of traditional medicine for the treatment of human burns, soft tissue wounds, ulcerated wounds, burn wounds, post – natal wounds and also for the treatment of leech bites, indigestion and skin infections (Panyaphu *et al*., 2011).

Plate 1: *Chromolaena odorata* L.

Classification of *Sida acuta* Burm. F.

This perennial shrub belongs to Kingdom: Plantae, Division: Angiospermophyta, Class: Dicotyledonae, Order: Malvales, Family: Malvaceae, Genus: *Sida*, Specific epithet: *acuta* (Senthilkumar *et al*., 2018) [29].

Health benefits of *Sida acuta* Burm. F.

Senthilkumar *et al*. (2018) [29] highlighted some usefullness of wire weed. The whole plant contains anthelmintic, antiemetic, diuretic, aphrodisiac, stomachic and antipyretic properties.

The plant was observed inhibiting the activities of *Bacillus subtilis* and *Escherichia coli*. The aqueous extraction of the leaves also showed moderate anti-bacterial activity against *Staphylococcus aureus* and *Pseudomonas aeruginosa.*

The secondary metabolites present in the plant is useful in treating diseases such as headache, cold, fever, skin diseases, urinary diseases, ulcer, snake bite, facial paralysis.

A research work done in Nigeria by treating methanolic extracts of the leaves of *Sida acuta* Burm. F. of different dosages (3000 mg, 4000 mg and 5000 mg mixed in 1 Kg paraffin oil each) on the wounds excision on guinea pigs confirmed that methanolic extracts of the leaves of *Sida acuta* is efficient in healing wounds (Oduwegwu *et al*., 2017) [24].

The phytochemical analysis of the aqueous and ehtanolic extracts of *Chromolaena odorata* L. done by Anyasor *et al*. (2011) [6] is summarized in Table 1.

Plate 2: *Sida acuta* Burm. F.

Table 1: Phytochemical analysis of aqueous and ethanolic extracts of *Chromolaena odorata* leaf

Bioactive compounds	Relative abundance	
	Aqueous	Ethanolic
Terpenoids	-	+ +
Tannin	+	+ +
Saponin	+ +	+
Phlobatannin	+ +	-
Cardiac glycoside	-	+ +
Flavonoids	-	-
Cardenolides	-	-
Anthraquinones	-	+
Phenol	+ +	+
Alkaloid	+ +	-
Volatile oil	-	-

Key: + + = abundant, + = trace, - = absent. Source: Anyasor *et al*., (2011) [6].

Phytochemicals present in aqueous and ethanolic extraction of *Sida acuta*

In the preliminary photochemical screening of the aqueous extraction of the leaves, it was found that alkaloids, steroids, flavonoids, phenols, terpenoids and cardiac glycosides were present. On the other hand, tannins, saponins, anthroquinones and phlobatannins were absent (Senthilkumar *et al*., 2018) [29]. In the ethanolic extraction of 80% ethanol concentration, *Sida acuta* is found to contain alkaloids, flavonoids, saponins, steroids, tannins, phlobatannins, terpenoids and cardiac glycosides (Adeniyi *et al*., 2010) [2].

3. Methodology

The study area

Through unstructured observation by the researcher, the researcher found out that Assin Andoe is one of the many towns with the largest Siam weed and wire weed infestation in the Assin South District. The study used *Chromolaena odorata* L. and *Sida acuta* Burm. F. that are found in Assin Andoe in the Assin South District. Assin Andoe is in the Southern part of Ghana, which lies in the rain forest belt along Cape Coast to Kumasi highway (Fig. 1).

The town is marked by different forms of plant species, ranging from perennating trees to annual crops. Among the herbs in the town are *Sida acuta, Chromolaena odorata, Phyllantus amarus, Tuja nana, Sporobolus pyramidalis* etc. The quality of the soil supports agriculture as well as weeds, hence the infestation of *Chromolaena odorata* L. and *Sida acuta* Burm. F.

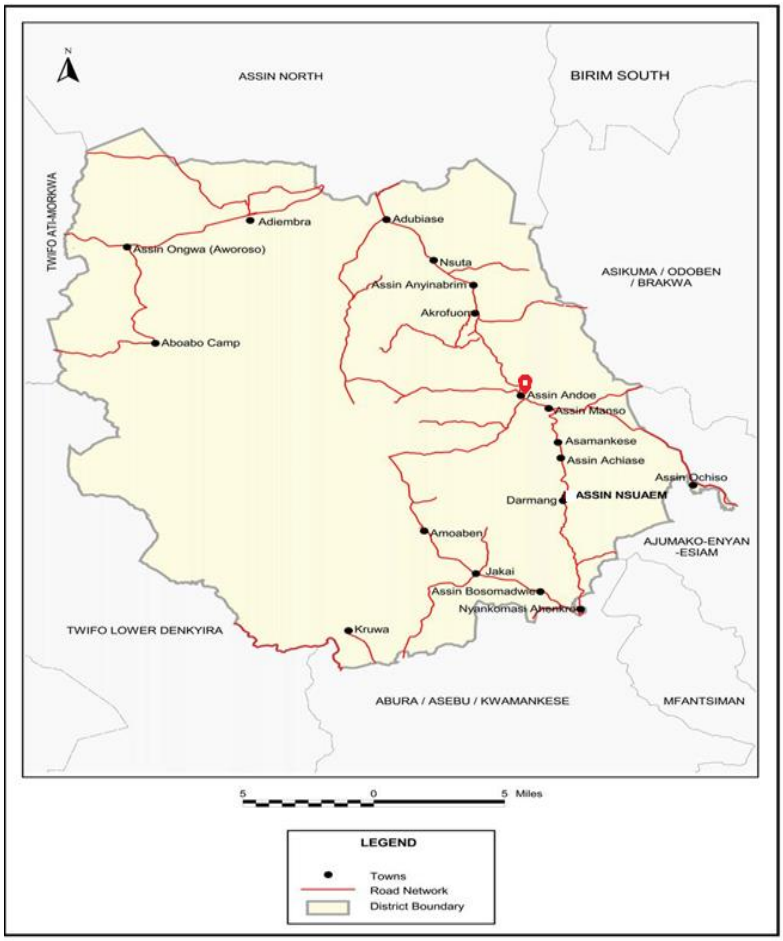

Fig 1: A map to Assin Andoe

Plant materials collection and processing

Fully matured dark green leaves of *C. odorata* and *S. acuta* were collected in Assin Andoe near the vicinity of the Andoe new market. The plant species were identified by verifying the pictures of the species on google lens as well as by following the description and identification of the characters of the species by eye inspection. The leaves were thoroughly washed with water. The leaves were dried under shade separately for four weeks. The dried leaves were powdered by using an electric blender. A sieve with size 1.0 mm was used to sieve the pulverized leaves. The sieved leaves were kept in two plastic containers separately.

Extraction procedure

The instruments used for the extraction are balance, stirrer, spatula, four funnels, measuring cylinder, wash bottle, four plastic containers, one beaker and four conical flasks. The glassware and the plastic containers were washed with distil water. The extraction method adopted is maceration. The researchers picked 15g each of the powdered leaves of *C. odorata* and *S. acuta* making 30g and mixed them in a beaker. The mixture was introduced into one plastic container. Using the measuring cylinder, 500mL of distilled water was added to the mixture. The mixture was then stirred followed by vigorous shaking to form a uniform solution.

70% ethanol was prepared by picking 350mL of absolute ethanol and diluted with 150mL of distilled water. 15g of *S. acuta* and 15g of *C. odorata* leaves were dissolved in the 70% (v/v) ethanol in another plastic container. The same procedure was repeated for 30% ethanol and 30% methanol. The four solutions were duly labelled and were left for three days with intermittent agitation to aid in the extraction procedure.

Whatman no. 1 filter paper was used to filter the solution after three days. On the other hand, the aqueous extract was filtered after two days and was kept in a freezer to prevent it from going bad. The ethanolic and methanolic extracts were concentrated by using a vaporizer followed by heating the water bath. The aqueous extract was concentrated by heating it over the water bath.

Phytochemical screening

The researchers ran a qualitative test to assess the phytochemical compounds present in the various extracts of the dried leaves. The methods used were adopted by Dhawan and Gupta (2017) [11] and other authorities.

- **Test for saponins**

10 mL of the extracts of each the solvents were transferred into four different test tubes labelled SC1, SC2, SC3 and SC4; 70% ethanolic extract, 30% ethanolic extract, 30% methanol extract and aqueous extract are all transferred into SC1, SC2, SC3 and SC4 respectively. The extracts were diluted with water and were shaken for 15 mins. The extracts that formed foam on the supernatant indicated the presence of saponins.

- **Test for flavonoids**

10 mL of each of the different extracts was transferred from the stock into four different test tubes: SC1, SC2, SC3 and SC4. Few drops of dil. NaOH were added to each extract. There was the appearance of yellow colour. The yellow colour changes to colourless upon the addition of a few drops of dil. H_2SO_4. The disappearance of the yellow colour confirms the presence of flavonoids.

- **Test for alkaloids**

10 mL of each of the different extracts was transferred into four different test tubes. 2 mL of the Wagner's reagent was added to each test tube. The appearance of reddish – brown precipitate confirmed the presence of alkaloids.

- **Test for tannins**

10 mL of each of the different extracts was taken and dissolved in 45% of the ethanol in four different test tubes. The test tubes were boiled for 5 mins and 1 mL of 15% ferric chloride solution was added to each. The formation of greenish to black in colour confirmed the presence of tannins in the leaf extracts.

- **Test for steroids**

10 mL each of the extract were taken and 1 mL of conc. H_2SO_4 was added to each of the test tube. Appearance of dark reddish – green colour confirmed the presence of steroids.

- **Test for Terpenoids**

This method was adopted by Prabhavathi *et al*. (2016) [25]. To 10 mL of each extract, 0.5 ml of chloroform was added followed by few drops of concentrated sulphuric acid. There was formation of reddish – brown precipitate.

- **Glycoside**

3 mL of chloroform was added to all the extracts followed by 10% ammonia. Formation of pink colour indicated the presence of glycoside (Roghini & Vijayalakshmi, 2018) [26].

- **Phenol**

A few drops of distilled water were added to the extracts followed by a few drops of 10% ferric chloride. Blue or green colour indicated presence of phenol (Roghini & Vijayalakshmi, 2018) [26].

- **Anthraquinone**

5 ml of chloroform was added to 0.5 g of the extracts. The resulting mixture was shaken for 5 mins after which it was filtered. The filtrate was then shaken with equal volume of 10 % ammonia solution. The presence of a bright pink colour in the aqueous layer indicated the presence of free anthraquinones (Ajayi *et al*., 2011) [3].

- **Phlobatannin**

Few drops of 2% hydrochloric acid were added to 1ml of each of the extracts. Appearance of red colour precipitate indicated the presence of phlobatannins (Roghini & Vijayalakshmi, 2018) [26].

- **Gum and mucilage**

10 mL of distilled water was added to the extracts followed by 2 mL of absolute alcohol. White or cloud precipitate formed indicated the presence of gum and mucilage (Banu & Cathrine, 2015) [7].

4. Results

Results from the qualitative analysis of the extracts

During the qualitative analysis of the chemical constituents of the mixture of *Chromolaena odorata* L. and *Sida acuta* Burm. F. leaves, the following observations were made:

- **Saponins**

All the extracts in the test tubes SC1, SC2, SC3 and SC4 contain 70% ethanolic extract, 30% ethanolic extract, 30% methanol extract and aqueous extract respectively, upon the dilution with water and shaken for 15 minutes, there was formation of foam on the supernatant. The presence of foam indicated the presence of saponin in all the extracts.

- **Flavonoids**

Few drops of dil. NaOH were added to 10 mL of each of the extracts in the test tubes. There was formation of yellow

precipitate in all the extracts. Upon addition of few drops of dil.H_2SO_4, the yellow precipitate disappeared in all the extracts which confirmed the presence of flavonoids in all the extracts.

- **Alkaloids**

2mL of the Wagner's reagent was added to each extract in the test tubes. The straw colour of the Wagner's reagent formed reddish – brown precipitate in test tubes SC3 and SC4. The reddish-brown precipitate was absent in SC1 and SC2. The appearance of reddish - brown precipitates confirmed the presence of alkaloids.

- **Tannins:**

When 10 mL of each of the different extracts was taken and dissolved in 45% ethanol, there was formation of brown precipitate in all the extracts. They were boiled for 5 mins and 1 mL of 15% ferric chloride solution was added to each extract. There was formation of black precipitate in all the extracts which confirmed the presence of tannins.

- **Steroids**

Few drops of 1 mL of conc. H_2SO_4 were added to 10 mL of each extract. It was observed that dark reddish – green colour appeared in all the extracts which confirmed the presence of steroids.

- **Terpenoids**

0.5 mL of chloroform was added to 10 mL of each extract. Cloudy precipitate was formed at the bottom of SC1, brown precipitate was formed on the supernatant of SC2, SC3 and SC4. Few drops of concentrated sulphuric acid were added and there was formation of reddish - brown precipitate in all the extracts which indicated the presence of terpenoids.

- **Glycoside**

3mL of chloroform was added to all the extracts followed by 10% ammonia. The cloudy precipitate formed did not change colour to pink. Absence of pink coloration indicated absence of glycoside in all the extracts.

- **Phenol**

A few drops of distilled water were added to the extracts followed by a few drops of 10% ferric chloride. There was formation of dark green colour in SC1 and dark blue colour in SC3 and SC4. This confirmed the presence of phenol in SC1, SC3 and SC4.

- **Anthraquinone**

5 ml of chloroform was added to 0.5 g of the extracts. The resulting mixture was shaken for 5 mins after which it was filtered. The filtrate was then shaken with equal volume of 10 % ammonia solution. The cloudy precipitate formed did not change colour to pink which confirmed the absence of anthraquinone in all the extracts.

- **Phlobatannin**

Few drops of 2% hydrochloric acid were added to 1ml of each of the extracts. Brownish-red precipitate was formed in SC2, SC3 and SC4 but not in SC1. The precipitation indicated the presence of phlobatannin.

- **Gum and mucilage:**

10 mL of distilled water was added to the extracts followed by 2 mL of absolute alcohol. There was formation of cloudy precipitate which confirmed the presence of gum and mucilage in all the extracts. Table 2 is the summary of the phytochemical constituents in the various extracts.

Table 2: Phytochemical compounds present in the mixture of *Chromolaena odorata* L. and *Sida acuta* Burm. F. leaves extract

Chemical compound	SC1	SC2	SC3	SC4
Saponins	+	+	+	+
Flavonoids	+	+	+	+
Alkaloids	-	-	+	+
Tannins	+	+	+	+
Steroids	+	+	+	+
Terpenoids	+	+	+	+
Glycoside	-	-	-	-
Phenol	+	-	+	+
Anthraquinone	-	-	-	-
Phlobatannin	-	+	+	+
Gum and mucilage	+	+	+	+

Key: + presence of the phytochemical, – absence of phytochemical

Discussion of results from the qualitative analysis of the extracts

From Table 2, Saponins, flavonoids, alkaloids, tannins, steroids, terpenoids, phenol, phlobatannin, gum and mucilage are the phytochemicals present in the various extracts of the mixture of *Chromolaena odorata* and *Sida acuta* leaves. Glycoside and anthraquinone on the other hand were absent in all the extracts. And this agrees to the work done by Tiamiyu *et al*., (2019) [].

A phytochemical screening of *Chromolaena odorata* L. leaves extract by Tiamiyu *et al*., (2019) [32] showed that alkaloids, tannin, phlobatannin, saponin, flavonoids, steroids, terpenes, phenol and cardiac glycosides are present whereas anthraquinone, cardenolides are absent.

According to Anyasor *et al*. (2011) [6], the following phytochemicals are present in the ethanolic extract of the *Chromolaena odorata* leaves; terpenoid, tannin, saponin, cardiac glycoside, anthraquinone, phenol whereas the following were absent; phlobatannin, flavonoids, cardenolides, alkaloid and volatile oil.

Again, research by Adeniyi *et al*. (2010) [2] on the phytochemical analysis of 80% ethanolic extract of *Sida acuta* leaves showed that alkaloids, flavonoids, saponins, steroids, tannins, phlobatannins, terpenoids and cardiac glycosides are present.

When the two-research works were married, the following phytochemicals were expected to be present in the ethanolic extract of the mixture of *Chromolaena odorata* and *Sida acuta* leaves; terpenes, tannins, saponin, phlobatannin, cardiac glycoside, flavonoids, anthraquinone, phenol, alkaloids and steroids.

From Table 2, both SC1 and SC2 form ethanolic extracts of the mixture of *Chromolaena odorata* and *Sida acuta* leaves. Thus, the results from the two extracts include saponins, flavonoids, tannins, steroids, terpenoids, phenol, phlobatannin and gum and mucilage. The results do not agree with the results from Adeniyi *et al*. (2010) [2] and Anyasor *et al*. (2011) [6]. This is because alkaloids and anthraquinones are absent according to Table 2. This may be because of the difference in concentrations of the ethanol used in the extraction method by the researcher.

Again, according to Anyasor *et al*. (2011) [6], the following phytochemicals are present in the aqueous extract of *Chromolaena odorata* leaves; tannin, saponin, phlobatannin, phenol and alkaloid whereas terpenoid, cardiac glycoside, flavonoids, cardenolides, anthraquinone and volatile oil were absent. The aqueous extract of *Sida acuta* leaves according to Senthilkumar *et al*. (2018) [29] showed the following chemical

constituents; alkaloids, steroids, flavonoids, phenols, terpenoids and cardiac glycosides. When the two works were married, the following phytochemicals were expected: terpenoids, tannin, saponins, phlobatannin, cardiac glycosides, flavonoids, phenols and alkaloids.

From Table 2, the aqueous extract showed the following chemical constituents; saponins, flavonoids, alkaloids, tannins, steroids, terpenoids, phenol, phlobatannin, and gum and mucilage. The results of the aqueous extracts agree with the work by Senthilkumar *et al*. (2018) [29] and Anyasor *et al*. (2011) [6] since the phytochemicals in aqueous extract are similar to that from Table 2.

From Table 2, nine out of eleven phytochemicals were successfully extracted from the mixture of *Chromolaena odorata* and *Sida acuta* leaves in SC3 and SC4 while seven out of eleven phytochemicals were successfully extracted in SC1 and SC2. This means 30% methanol (v/v) and distilled water are most effective in extracting *Chromolaena odorata* L. and *Sida acuta* Burm. F. leaves which agree to Truong *et al*. (2019) [33] that methanol was most effective in extracting *Severinia buxifolia* and Abdullahi and Haque (2020) [1] which states that water is capable of dissolving wide range of chemicals.

Effect of the phytochemicals in *C. odorata* and *S. acuta* leaves on wounds

1. **Flavonoids (Fig. 2):** these phytochemicals are used in traditional medicine as anti – inflammatory, pain relieving, promoting healing and anti – allergens among others. They also react with free radicals, can chelate metals, increase enzymatic reactions and have action on adenosine receptors and influence biological membranes. The main molecular structure of these chemicals is two aromatic rings connected by three carbon bridges. All flavonoid compounds contain phenol groups which in general stimulate an antioxidant activity (Maver *et al*., 2017) [21]. Flavonoids are mainly water-soluble compounds but can be extracted by 70% ethanol. They are mostly present in plants as mixtures, and it is scarcely found in isolation. Flavonoids are present in all vascular plants (Harborne, 1998)[16].

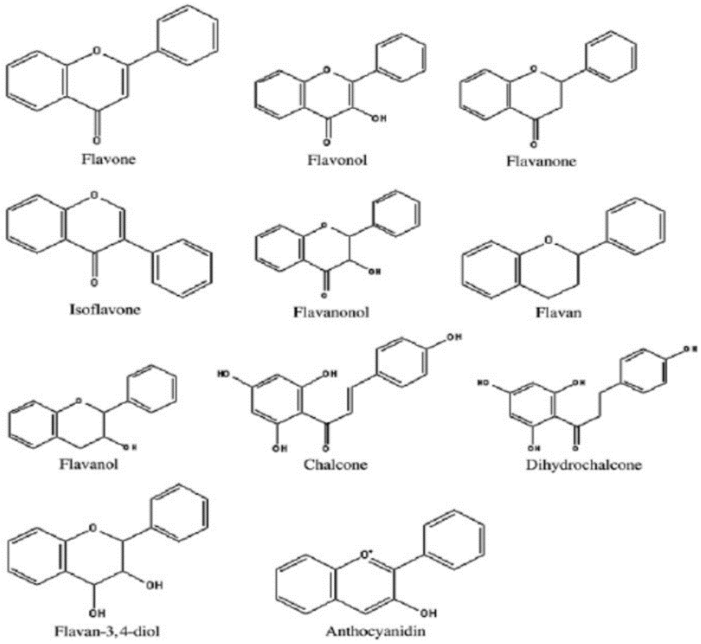

Fig 2: Flavonoids

2. **Alkaloids (Fig. 3):** they are heterocyclic compounds that have nitrogen atom in at least one of the heterocyclic compounds. They have bitter tastes; this accounts for why *C. odorata* leaf tastes bitter. Alkaloids have potential effects on wound healing. They stimulate bone marrow leucocytes which modulate the inflammation phase of wound healing (Maver *et al*., 2017) [21]. Harborne (1998) [16] reports that alkaloids are either absent or infrequently occur in lower plants.

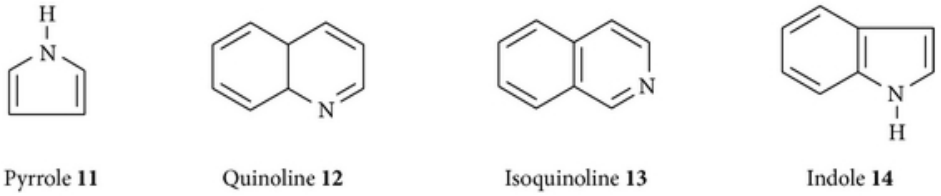

Fig 3: Alkaloids

3. **Monoterpenes (Fig. 4):** these are chemicals compounds with a core of ten carbons. Many of these compounds exist in the form of essential oils because of their low molecular weight. Monoterpenes have wound – healing ability, antibiotic activity and anti-inflammatory activity by limiting leukocyte migration etc. (Barreto *et al*., 2014) [8].

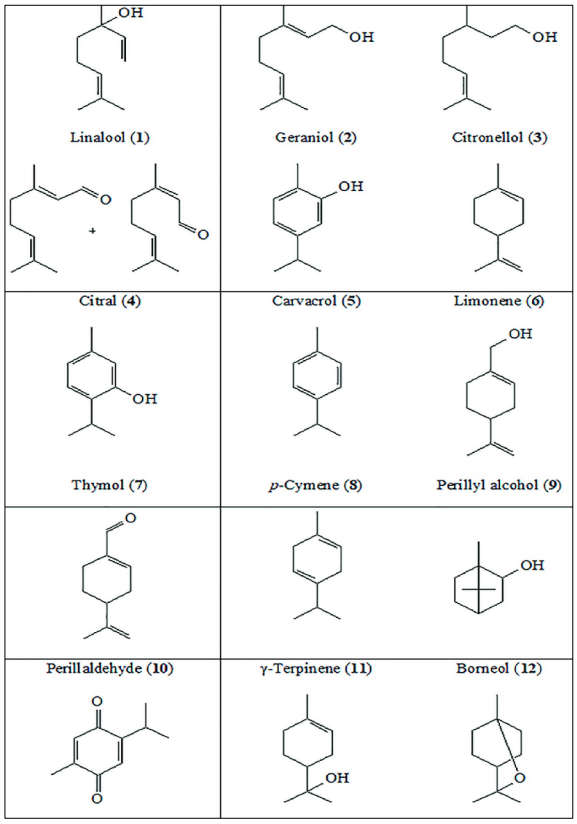

Fig 4: Monoterpenes

4. **Saponins (Fig. 5):** they are phytochemicals that can accelerate numerous biological activities involving hemolytic, anti-bacterial, anti-viral and anti-oxidative functions. They do not only promote re-epithelization of wounds but also efficiently prevent inflammatory reactions. They also promote matrix synthesis throughout the wound healing process. Based on this, saponins are effective in healing incisional skin wounds (Kim & Bae, 2011) [18].

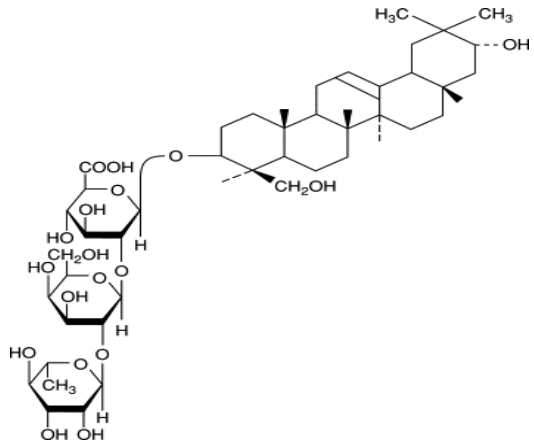

Fig 5: Saponins

5. **Steroids (Fig. 6):** Corticosteroids augments risk of wound infection and delay healing of wounds. Steroids do that by interfering with inflammation, collagen synthesis and degradation, angiogenesis, wound contraction and re – epithelialization (Anstead, 1998) [5]. In effect, steroids are used on doctor's advice, and they are used for skin conditions but areas on the skin that have cuts, scrapes and burns should be exempted.

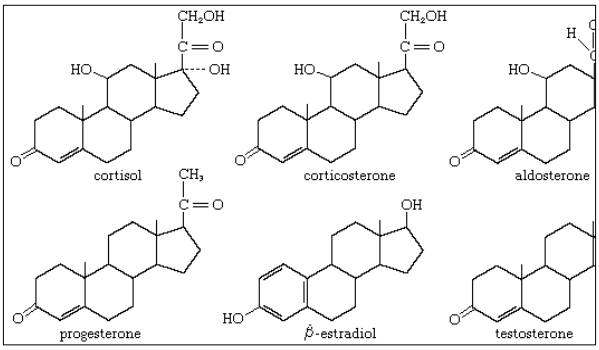

Fig 62: Steroids

6. **Tannins (Fig. 7):** According to Harborne (1998) [16], tannins mostly occur in vascular plants and are usually associated with proteins that helps in converting hide into leather in leather industry. Tannins are detastable hence animals tend to avoid eating tanning – containing plants.

Advantages of these phytochemicals are relief of pain, inhibition of secondary infection, prevention of loss of plasma and enhancement of epithelialization. Tannins also improve wound healing and reduction in scar tissue formation by preventing the formation and elimination of reactive oxygen substances (Chokotho & Hasselt, 2005).

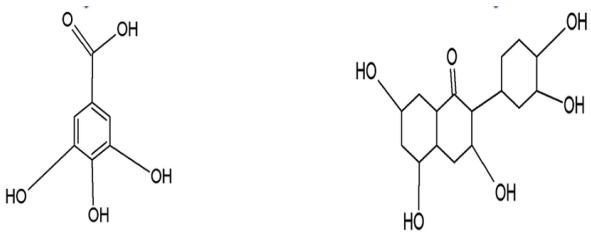

Fig 7: Hydrolysable tannins Condensed tannins

7. **Anthraquinone (Fig. 8):** This compound is a phenolic compound which is a special type of quinone in plants. Other groups of quinones include benzoquinones, naphthaquinones and isoprenoid (Harborne, 1998) [16].

Anthraquinones have been found to exhibit anti-inflammatory properties by suppressing the release of inflammatory mediators and reducing inflammation at the wound site (Kshirsagar *et al*., 2014) [20]. This can help in the control of excessive inflammation during wound healing and again, it has been found to increase angiogenesis and collagen synthesis (Zhao *et al*., 2020) [37].

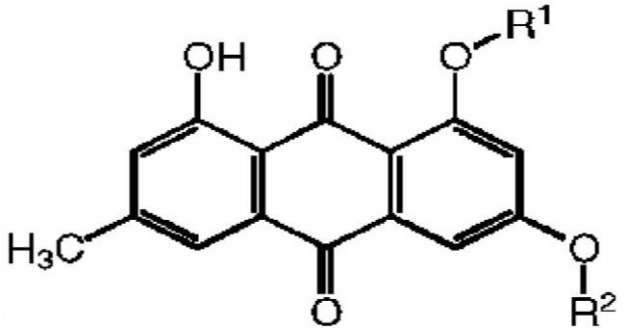

Fig 8: Anthraquinone

8. Gum and mucilage

Gum is a viscous substance that is derived from the sap or resin of certain plant species. In wound healing, gum acts as a protective barrier to prevent the wound from becoming infected. Gum provides a moist environment for the wound to heal, which helps to promote cell growth and tissue regeneration. Gum also contains natural antimicrobial properties that can help to prevent infection (Gutierrez-Reyes *et al*., 2023) [15].

Gum acts as a protective barrier to prevent the wound from becoming infected. It provides a moist environment for the wound to heal, which helps to promote cell growth and tissue regeneration. Gum also contains natural antimicrobial properties that can help to prevent infection. Mucilage (Fig. 9), on the other hand, promotes wound healing by its wound-healing, moisturizing, soothing, and anti-inflammatory effects.

Mucilage is also known to promote granulation tissue formation, which is an essential step in the healing process (Valizadeh *et al*., 2015) [35].

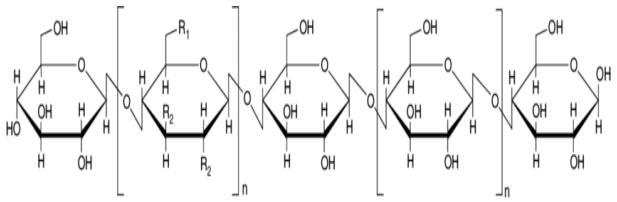

Fig 9: Molecular structure of mucilage in yellow mustard

Glycosides (Fig. 10)

Glycosides is a type of organic compound that consists of a sugar molecule (glycone) attached to a non-sugar moiety (aglycone or genin) through a glycosidic bond (Nandi *et al*., 2021) [23]. The sugar molecule in a glycoside is typically a monosaccharide, such as glucose, fructose, or galactose, while the non-sugar component can vary and may include phenols, flavonoids, alkaloids, or terpenoids (Alamgir, 2018) [4]. They serve important biological functions, such as storage of carbohydrates, defense against pathogens and herbivores, and attraction of pollinators. In addition, many glycosides have medicinal properties and are used in traditional medicine and pharmaceutical applications (Sayed, 1980) [28].

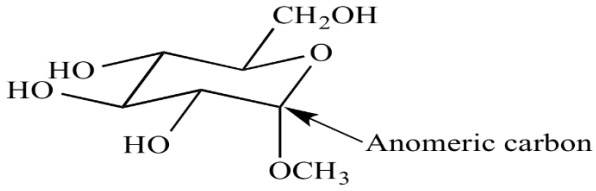

Fig 30: Glycosides

5. Limitation

Compound degradation could impact the presence of the phytochemical during the extraction procedure.

6. Implications of The Study

The study forms the basis for identifying the secondary metabolites present in the extracts of the mixture of dried leaves of *Chromolaena odorata* and *Sida acuta* which have medicinal properties. The findings add to the research of understudied traditional herbs and further identify the scientific benefits of traditional herbs- *Chromolaena odorata* and *Sida acuta*.

7. Conclusion

The qualitative analysis of the extracts from the mixture of *Chromolaena odorata* and *Sida acuta* leaves revealed the presence of several phytochemicals including saponins, flavonoids, alkaloids, tannins, steroids, terpenoids, phenol, phlobatannin, and gum and mucilage. However, glycoside and anthraquinone were absent in all the extracts. The results were consistent with previous studies on the individual plants.

Again, it was concluded that 30% methanol (v/v) and distil water are most effective in extracting *Chromolaena odorata* L. and *Sida acuta* Burm.

8. Recommendation

Quantitative assessment of the phytochemicals present in the herbal mixture should be performed to investigate the relative abundance of the phytochemicals present in the solvent extracts of the dried leaves of the mixture of *Chromolaena odorata* L. and *Sida acuta* Burm. F.

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