

## Screening for Bioactive Compounds of Solvent Extracts of Some Selected Vegetables

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**Abstract**

Bioactive compounds in selected vegetables (wild lettuce, fireweed, fluted pumpkin and tree spinach) were qualitatively identified in water, methanol, ethyl acetate and chloroform extracts of the vegetables. The vegetables were obtained, washed, cut in smaller pieces, air-dried at room temperature and sieved. A known amount was weighed and soaked with different solvent (water, methanol, ethyl acetate and chloroform) in ratio 1:10 for 72 h inside reagent bottles which were intermittently shaken. It was filtered and the filtrates were evaporated using rotary evaporator. The extractive values of the solvents were calculated and each extract was screened for ten different phytochemicals. The phytochemicals considered were flavonoid, phenol, saponin, tannin, volatile oil, anthraquinone, steroid, glycoside and reducing sugar. The extractive values were highest with fluted pumpkin leaves ranged from 3.083 – 11.737% and lowest in fireweed ranged from 2.302 – 5.733% while the extractive values for wild lettuce and tree spinach leaves were ranged from 2.857 – 9.049% and 1.329 – 6.896% in all the four solvents used. It was found out that methanol and chloroform had the highest solvent potency in extracting phytochemicals from the vegetables considered while water had the lowest solvent potency. Fluted pumpkin and tree spinach leaves were richer in phytochemicals than fireweed and wild lettuce leaves.

**Keywords:** Phytochemical; Solvent; Extracts; Vegetables.

**Introduction**

Bioactive components also known as phytochemical compounds comprise of various chemical substances that are gotten from plants in little quantity which have vital roles in prevention of oxidative stress caused by imbalance between some free radicals and antioxidant. Phytochemicals prevents the occurrence of some diseases such as cancer, diabetic (Arawande et al., 2021a) and they also anti-inflammatory properties. They control the chemical messengers known as hormones in the body (Joao, 2012; Abiodun et al., 2017). And they also serve as prerequisite in the production of synthetic drugs (Sahira & Cathrine, 2015).

Vegetables are important sources of some food nutrients like minerals, vitamins, proteins and fiber which are necessary in daily diet and can be eaten raw by adding it to salad or by cooking it with soup or sauces (Joao, 2012). It has a scavenger potency against free radical damage making it a source of antioxidant (Arawande et al., 2021b), and they also serve as a source of protection against different forms of diseases (Abiodun et al., 2017).

*Epilobium agustifolium* belong to the family of *Onagraceae* with a synonymous name *Chamaenerion angustifolium*. This vegetable is called fireweed in North America, great willow herb in Canada and Rosebay willow herb in Britain and Ireland (Wikipedia, 2021). The vegetable is useful in treating some diseases like stomach upset, wounds or skin problem due to it pharmacological effect on the body. It prevents cancer if eaten regularly (Dreger et al., 2020). The presence of secondary metabolites and therapeutic properties in fireweed makes them a source of antioxidant, anti-inflammatory, antimicrobial and anti antrogenic (Anna et al., 2021; Adnortey et al., 2019; Igor et al., 2009).

*Cindosculus chayamansa* popularly called Chaya, which is from the family of *Euphorbiaceae* (Chidi et al, 2013) and is commonly cultivated in Mexico especially in a broad-leaf forest area. Its English name is tree spinach. It is planted through stem cutting; when cut, it produces a milky sap and it can grow to a height of 4-5 cm (Kuri-Garcia et al., 2017).

Chaya is used for medicinal purpose in curing some chronic diseases and oxidative related diseases due to the abundance of phenolic compounds and antioxidant properties present in them (Chidi et al., 2013; Kuri-Garcia et al., 2017). It is also good for consumption being a source of protein, potassium, vitamins,  $\beta$ - carotene, calcium, ascorbic acid and minerals by adding it to soup or mixing with vegetable dishes; daily consumption can prevent nutrient deficiency and it can also be used as ornamental plant (Chidi et al., 2013; USAID, 2013).

*Launaea taraxacifolia* is a common leafy vegetable with greenish colors, which germinated mostly on a rocky soil in clusters or singly form (Bello et al., 2018). It is a common vegetable in African especially the western part of Nigeria and it is referred to as African lettuce or wild lettuce as the English name. The western part of Nigeria called it “Efo yanrin or Odundun odo” while in the northern part of Nigeria called it “Namijin dayri, Nomen barewa and Nonan barya”. Aside from Nigeria, some parts of Africa have also planted this vegetables plant locally, countries like Senegal, Ghana, Dahomey and Sierra Lorne (Bello et al., 2018). *Launaea taraxacifolia* has a medicinal capability (Borokini & Labumi, 2017) as a result of its phytochemical constituents, and pharmacological potency which is useful in eliminating migraine, in treating conjunctivitis (Sakpere & Aremu, 2008). African lettuce helps to treat urinary tract problem, diabetes mellitus (Adinortey et al., 2018), in preventing whooping cough and to increase the production of milk by feeding the lactating cows (Bello et al., 2018). It can be incorporated in our daily diet either by mixing it with salad or cooking with sauce or soups (Sakpere & Aremu, 2008).

*Telfairia occidentalis* is a leafy vegetable with a tender stem and edible seed, which are commonly cultivated and consumed in the eastern part of Nigeria. Its English name is fluted pumpkin. The Igbo tribe in Nigeria referred to it as “Ugu” and the Yorubas called it “Apiroko” and Efik referred to the same plant as “Ubong” (Akoroda, 1990). The leaves, the seed and the shoot of *Telfairia occidentalis* has a great importance to the human health due to its high protein content (Akoroda, 1990). It is used locally in curing some illness like convulsion, anemia and is used for making soup (Wikipedia, 2020).

There is so much literature on the cultivation, medicinal usefulness and antioxidant, proximate and mineral contents on these vegetables. However, there are little or no information on the effectiveness of solvents to extract bioactive compounds from these vegetables. Consequently, this research is aimed at investigating the extractive values of four different solvents (methanol, water, ethyl acetate and chloroform) and identifying the phytochemicals present in each of the solvent extracts of the vegetables under consideration so as to establish suitable and appropriate solvent for isolating a particular phytochemical.

## Materials and Methods

Fresh leaves of fluted pumpkin (*Telfairia occidentalis*), fire weed (*Epilobium angustifolium*), wild lettuce (*Laurea taraxacifoliab*) and tree spinach (*Cnidocolus chayamansa*)

were obtained from powerline area of Ajagbale, Oka, Ondo City, Ondo State, Nigeria. And analytical chemicals used were 99.5% pure and they were purchased from Lioxy K Chemicals in Akure, Ondo State.

## Preparation and extraction of *Telfairia occidentalis*, *Epilobium angustifolium*, *Laurea taraxacifoliab* and *Cnidocolus chayamansa* leaves

The leaves of each plant were cut into small pieces, washed and air dried at room temperature. The dried leaves were grounded into fine particles using electric blending machine and it was then sieved with 70 x 140 mm mesh size. Each of the powdered samples was packed into a transparent air tight container prior to extraction and it was labeled appropriately. A cold extraction process was carried out by adding each solvent (water, methanol, ethyl acetate and chloroform) to each of dried powdery samples weighed inside different dried container at ratio 10:1 (that is 100 mL of each solvent were used to soak 10 g of each powdery sample) for 72 h and it was shaken intermittently using shaking orbit machine (Arawande et al., 2021a). The mixture was then filtered using vacuum filtration technique. The filtrate was kept inside a fume cupboard for easy evaporation of the solvent and the dried extract were preserved inside a refrigerator at 4°C (Arawande et al., 2021a) and percentage yield of each extract was calculated using weight of extract obtained as described by Arawande & Abitogun, 2009).

## Phytochemical screening of solvent-extracts of the plant sample

The qualitative phytochemical evaluation was carried out on the extract using standard methods described by (Sofowora et al., 2013 and Sofowora, 2008).

### Test for flavonoids

To about 0.2 g of the extract, dilute sodium hydroxide was added, which led to the formation of yellow color that changed to colorless on addition of concentrated Hydrochloric acid and this indicated the presence of flavonoid.

### Test for phenol

0.5 g of extract was mixed with 0.5 mL of 10% ferric chloride. The formation of deep bluish green solution showed the presence of phenol

### Test for saponin

About 0.2 g of the extract was mixed together with 5 mL of distilled water and it was shaken vigorously. The appearance of foam which persisted for some minutes indicated the presence of saponin.

### Test for glycosides

0.2 g of the extract was hydrolyzed with 5% v/v hydrochloric acid solution and neutralized with 5% v/v sodium hydroxide solution and then five drops of Fehling solution A and B were added. Appearance of red precipitate indicated the presence of glycosides.

### Test for reducing sugar

Four drops of Fehling solution A and B were added to 0.2 g of extract and boiled. The appearance of orange red precipitate indicated the presence of reducing sugar.

### Test for steroids

To 0.5 g of the extract, 3 mL of acetic anhydride was added. Two drops of concentrated sulphuric acid was added down the side of the test tube. Formation of color ranges from violet to blue or green in some plant samples confirmed the presence of steroids.

### Test for tannin

5 mL of distilled water was added to 0.2 mL of extract and filtered. 1 mL of filtrate was mixed with three drops of 10% ferric chloride. Formation of blue black or blue green color showed the presence of tannin.

### Test for alkaloids

To about 0.2 g of the extract, 2 mL of 1% hydrochloric acid was added and six drops of drops of Dragendoff reagent was added. Orange red precipitate/turbidity indicated the presence of alkaloids.

### Test for anthraquinone

0.5 g of the extract was boiled with 10% hydrochloric acid for five minutes on water bath; it was filtered and allowed to cool. Then chloroform was added to the filtrate and four drops of 10% ammonia solution were also added to the mixture and heated. The formation of pink, red, violet color in ammoniacal phase indicated the presence of free anthraquinone.

### Test for volatile oil

0.5 g of the extract was dissolved with 90% ethanol and five drops of ferric chloride were added. The appearance of green color indicated the presence of volatile oil.

## Results and Discussion

**Table 1:** Extractive value (% yield) of some selected vegetables using different solvents.

Solvents	Percentage yield of extract (%)			
	W i l d lettuce	Fireweed	F l u t e d pumpkin	Tree spinach
Water	9.049	5.733	11.737	3.278
Methanol	4.770	3.883	15.603	6.896
Ethyl acetate	3.327	2.490	3.328	4.624
Chloroform	2.857	2.302	3.082	1.729

The extractive value (% yield) of some selected vegetables in using different solvents is presented in Table 1. The vegetables considered for this research work were wild lettuce, fireweed, fluted pumpkin and tree spinach while the solvents used are water, methanol, ethyl acetate and chloroform. It was conspicuously observed that water and methanol gave the highest extractive values while chloroform gave the lowest extractive values in all the vegetables except in water for tree

spinach. Fluted pumpkin gave the highest yield of extract in all the solvents except in ethyl acetate. The second highest yield of extract was wild lettuce in all the solvents apart from ethyl acetate and methanol. In water and chloroform, tree spinach had the lowest yield of extracts.

**Table 2:** Qualitative phytochemical analysis of solvent extracts of wild lettuce leaves.

Phytochemicals	Solvent extracts			
	Water	Methanol	Ethyl acetates	Chloroform
Flavonoid	-	-	-	-
Phenol	-	-	-	+
Saponin	-	+	-	-
Tannin	-	-	-	-
Alkaloid	-	-	-	-
Anthraquinone	-	-	-	-
Volatile oil	-	-	-	+
Steroid	-	+	-	+
Glycosides	-	-	-	-
Reducing sugar	-	-	-	-
% phytochemical extractable	0	20	0	30

(-) = absent (+) = present

Qualitative phytochemical analysis of different solvent extracts of wild lettuce leaves is shown in Table 2. Water, methanol, ethyl acetate and chloroform were solvents used for the extraction. Flavonoid, phenol, saponin, tannin, alkaloid, anthraquinone, volatile oil, steroid, glycosides and reducing sugar were the phytochemicals screened for in the plant solvent extracts. There was no phytochemicals in the water and ethyl acetate extracts of wild lettuce leaves. While only saponin and steroid were detected in the methanol extract of wild lettuce leaves. In chloroform extract of wild lettuce leaf, it was found that phenol, volatile oil and steroid were detected. The percentage extractable phytochemical in methanol and chloroform extracts of wild lettuce leaves were 20% and 30% respectively.

**Table 3:** Qualitative phytochemical analysis of solvent extracts of fluted pumpkin leaves.

Phytochemicals	Solvent extracts			
	Water	Methanol	Ethyl acetates	Chloroform
Flavonoid	-	+	-	+
Phenol	+	+	+	+
Saponin	+	+	-	+
Tannin	-	-	-	-
Alkaloid	-	+	-	-
Anthraquinone	-	-	+	-
Volatile oil	-	+	+	+
Steroid	-	-	+	+
Glycosides	-	-	-	-
Reducing sugar	-	-	-	-
% phytochemical extractable	20	50	40	50

(-) = absent (+) = present

The qualitative phytochemical analysis of solvent extracts of fluted pumpkin leaves is presented in Table 3. Out of ten phytochemical screened for in solvent extracts of fluted pumpkin leaves, it was observed that only phenol and saponin were present in water extract, flavonoid, phenol, saponin, alkaloid and volatile oil were detected in methanol extract, phenol, anthraquinone, volatile oil and steroid were present in ethyl acetate extract while in chloroform extract, there were flavonoid, phenol, saponin, volatile oil and steroid. The percentage phytochemical extractable in fluted pumpkin leaves were 20%, 50%, 40% and 50% in water, methanol, ethyl acetate and chloroform respectively. It was observed that fluted pumpkin leaves were richer in phytochemicals than wild lettuce leaves in all the solvent extracts considered. Phenol was detected in all the solvent extracts of fluted pumpkin leaves. Saponin was detected in all the solvent extracts except ethyl acetate. Also volatile oil was detected in all the solvent extracts apart from water extract.

**Table 4:** Qualitative phytochemical analysis of solvent extracts of fireweed leaves.

Phytochemicals	Solvent extracts			
	Water	Methanol	Ethyl acetates	Chloroform
Flavonoid	-	-	-	-
Phenol	-	-	+	+
Saponin	-	+	-	+
Tannin	-	-	-	-
Alkaloid	+	+	+	-
Anthraquinone	+	-	-	-
Volatile oil	-	+	-	-
Steroid	-	-	+	-
Glycosides	-	-	-	-
Reducing sugar	-	-	-	-
% phytochemical extractable	20	30	20	20

(-) = absent (+) = present

The qualitative phytochemical analysis of solvent extracts of fireweed leaves is depicted in Table 4. The percentage phytochemical extractable in water, ethyl acetate and chloroform extract in fireweed leaf was 20% each but it was 30% in methanol extract of fireweed leaves. Alkaloid and anthraquinone were detected in water extract of fireweed leaves while it was saponin, alkaloid and volatile oil that were present in methanol extract. Ethyl acetate extract of fireweed contained alkaloid and steroid, and phenol and saponin were detected in the chloroform extract of fireweed leaves. Alkaloid was detected in all the solvent extract except chloroform.

**Table 5:** Qualitative phytochemical analysis of solvent extracts of tree spinach leaves.

Phytochemicals	Solvent extracts			
	Water	Methanol	Ethyl acetates	Chloroform
Flavonoid	-	-	-	-
Phenol	+	-	+	+
Saponin	+	+	+	-
Tannin	-	-	-	-
Alkaloid	-	+	-	+
Anthraquinone	+	-	-	-
Volatile oil	-	-	-	+
Steroid	+	+	-	-
Glycosides	-	-	-	-
Reducing sugar	-	-	-	-
% phytochemical extractable	40	30	20	30

(-) = absent (+) = present

The qualitative phytochemical analysis of solvent extracts of tree spinach leaves is presented in Table 5. Water extract of tree spinach leaves contained phenol, saponin, anthraquinone and steroid and methanol extract contained saponin, alkaloid and steroid. Only phenol and saponin were detected in ethyl acetate extract of tree spinach leaves. Phenol, alkaloid and volatile oil were detected in chloroform extract of tree spinach leaves. Phenol was detected in all the solvent extracts excluding methanol extract, and saponin was also present in all the solvent extracts except chloroform extract. The percentage phytochemical extractable in tree spinach leaves were 40%, 30%, 20% and 30% in water, methanol, ethyl acetate and chloroform respectively.

Considering results obtained in Table 2 to Table 5, fluted pumpkin and tree spinach leaves contained more bioactive compounds than wild lettuce and fireweed leaves. The overview of solvent effectiveness in all the vegetables showed that methanol and chloroform were better solvents in extracting bioactive compounds than water and ethyl acetate. Although for tree spinach leaves, water ranked best and ethyl acetate ranked second for fluted pumpkin leaves.

### Conclusion

Water and methanol has the highest percentage yield extract due to the polarity of the solvent. Fluted pumpkin and tree spinach leaves had more bioactive compounds and wild lettuce leaves had the least bioactive compounds in all the vegetables examined. Chloroform and methanol served as best solvents for the extraction of bioactive compounds in all the selected vegetables. It can also be concluded that high percentage yield of extracts in solvents is not depended on the quantity of phytochemical present in such an extract. Further research like checking for the quantitative phytochemical constituents, it antioxidant capacity can be carried out on these selected vegetables.

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