



## Effect of aqueous extract of *Colpomenia sinuosa* on some secondary products in lupine

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**Abstract** *Colpomenia sinuosa* was gathered, and the location was near the Red Sea coast of Egypt at Hurghada.. Seeds of *Lupinus termis* were primed in distinctive concentrations (0, 50, 100, 150, 200, 250 ml/L) of aqueous extract from *C. sinuosa* followed by growing in Hogland's solution. The different bioactive substances, such as total phenols, total flavonoids, total flavonols, and total tannins, were identified using lupine leaf extracts. The findings show that, based on the concentration of the examined *C. sinuosa* extract, the four bioactive components were elevated at varying rates. The results suggest priming of plant seeds in seaweed extracts during germination to enhance the antioxidant compounds which have various biological activities particularly the antioxidant activity.

**keywords:** *Colpomenia sinuosa*, Lupine, secondary metabolites, phenolics, flavonoids, flavonols.

### 1. Introduction

Plants produce two different kinds of metabolites through their metabolic pathways: (i) plant secondary metabolites (PSMs) and (ii) plant primary metabolites (PPMs) [1]. Massive production of PPMs is necessary to sustain such fundamental plant functions as growth and development, respiration, and photosynthesis [2]. Proteins, lipids, carbohydrates, amino acids, and hormones are a few examples of significant PPMs.

The secondary metabolites in plants are produced in minute amounts and they don't significantly contribute to the fundamental processes of plant life but they are essential for the plant ability to adapt to challenging conditions [3]. Flavonoids, alkaloids, anthocyanins, quinones, peptides, phenolics, terpenoids, and amines are some of the secondary metabolites of plants that are mostly utilized as food additives, bio-pesticides, and fragrances [4].

One of the primary classes of secondary metabolites present in plants are phenols, which are notable for the numerous advantages they provide to both plants and other living things. It has been proven that they serve to shield the plants. This role is exemplified by preventing

damage effect of UV rays exposure, excessive transpiration, and other unfavorable environmental variables, as well as by avoiding the invasion of tissues by fungus, bacteria, or viruses [5]. Because of their numerous pharmacological properties, phenolic substances like zingerone, curcumin, raspberry ketone, and magnolol are well-known in both traditional and modern medicine [6,7].

Plants produce tiny molecules called flavonoids, which have a variety of biological actions. They can participate in how plants interact with other species and respond to environmental challenges because of their structural and metabolic characteristics. Their potent antioxidant abilities account for most of their actions [8,9].

Polyphenols called tannins are abundantly present in plants. For a long time, Plant tannins are utilized in animal husbandry. [10]. Tannins include ellagic tannins and partially hydrolyzed polyol residues in hydrolyzed tannins. [11]. In the food and medicinal industries, tannins' antioxidant abilities are widely used. Tannins possess antioxidant properties that can help prevent cancer, osteoporosis, and cardiovascular disease [12].

A vital and significant type of legume field crops is lupine. Due to its superior amino acid profile and highest protein contents of any legume, lupine have very good nutritional qualities. Lupine can contribute significantly to the future food and protein needs of the growing world population in the ensuing decades. [13].

Numerous bioactive substances, including polyphenols, sterols, flavonoids, alkaloids, proteins, tannins containing vital amino acids, and polyunsaturated fatty acids, are abundant in seaweeds. In addition to offering seaweeds protection, these bioactive chemicals have a high nutritional value and various advantages for people [14].

A globally distributed species of brown macroalgae is *Colpomenia sinuosa*. Humans take *C. sinuosa* because it is an excellent source of phenols, vitamins, folic acid, and amino acids, and because it has antioxidant properties. *C. sinuosa* has a high ash content and is a good source of lysine, fucosterols, stigmaterols, palmitic acid, and unsaturated fatty acids [15].

Consequently, the current study sought to determine how *C. sinuosa*'s aqueous extract affected a few lupine secondary products.

## **2-Materials and methods**

### **Collection and preparation of *C. sinuosa* extract**

The collection site was on Egypt's Red Sea shoreline near Hurghada during autumn 2022. Excess salt and debris were removed using distilled water. In addition, the samples were sliced to the proper size and stored for 24 hours at 40°C in the oven. Subsequently, the samples were ground into an extremely fine powder. The powdered seaweed was collected in an airtight container and kept at -20°C for further analysis [16].

### **Seeds of *Lupinus termis* L and plant growth**

The Egyptian Ministry of Agriculture was the source of the lupine seeds, which were surface sterilized for 10 minutes in 2% sodium hypochlorite. Following a week of seed priming in different concentrations (0, 50, 100, 150, 200, 250 mL/L) of *C sinusa* extract, the seedlings were moved to Hoagland's solution and left to grow for a period of 21 days [17].

### **Preparation of lupine leaf extract**

The leaves of 21-day-old lupine seedlings were gathered, cleaned, dried at 40°C for 48 hours, and then processed in a grinder to a fine powder. A range of leaf extract concentrations (0, 50, 100, 150, 200, and 250 ml/L) were created with distilled water.

### **Determination of total phenolic content in lupine leaf extract**

The overall phenolic amount of the lupine leaf extract was measured using the colorimetric method reported in [18] using the Folin-Ciocalteu reagent. A 10-fold diluted Folin-Ciocalteu reagent, one milliliter of leaf extract, and one milliliter of sodium carbonate solution were mixed. A 30-minute incubation period at 30°C allowed for the colorimetric determination of the entire mixture at 500 nm.

### **Determination of total flavonoids in lupine leaf extract**

The amount of each flavonoid was calculated using the AlCl<sub>3</sub> method [19]. 1 mL of leaf extract was combined with 1.5 mL of methanol, 2.8 mL of distilled water, 0.1 mL of 10% (w/v) AlCl<sub>3</sub>, and 0.1 mL of 1 M potassium acetate. After 30 minutes of incubation at 30°C, the mixture's absorbance at 415 nm was measured using a wavelength spectrophotometer. A catechin standard curve was used to calculate the whole amount of flavonoid.

### **Determination of total flavonols in lupine leaf extract**

The total flavonols technique was established in accordance with [21, 22]. Two milliliters of ethanol-produced AlCl<sub>3</sub>, one milliliter of extract, and three milliliters of 50 g/L sodium acetate were utilized. solution were put into a centrifuge tube. Following a thorough blending using a vortex mixer, this was incubated for one hour. Then, at 440 nm, absorbance was measured using spectrophotometry.

### **Determination of total tannins content in lupine leaf extract**

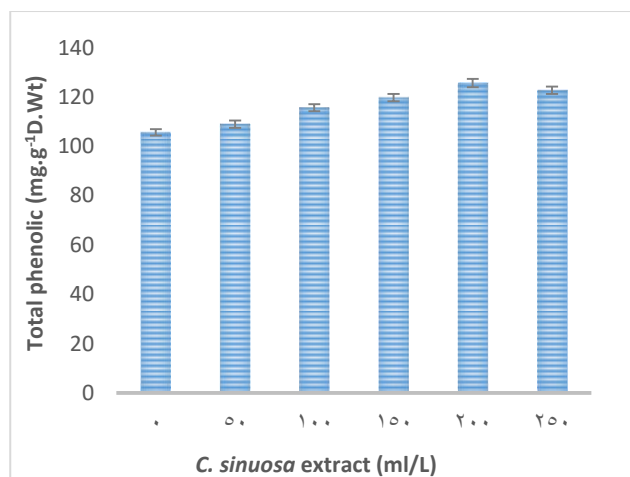
The entire level of tannin was ascertained using the Folin-Ciocalteu technique. A 10-milliliter volumetric flask was loaded with a 0.1 mL extract sample [23], 7.5 mL of distilled water, 0.5 mL of Folin-Ciocalteu phenol

reagent, 1 mL of 35% Na<sub>2</sub>CO<sub>3</sub> solution, and 10 mL of distilled water for dilution. After giving the mix a firm vibrato, it was held at room temperature for 30 minutes. Spectrophotometric measurements of the absorbance were made at 725 nm. The tannin concentration was given as mg of GAE/g of extract.

### 3-Results

#### Effect of aqueous extract of *C. sinuosa* on total phenols in lupine leaves

The influence of algal extract of *C. sinuosa* on total phenols in lupine was tested at distinct concentrations (50, 100, 150, 200 and 250 ml/L). The results in Fig. 1 Demonstrate that the seaweed extract resulted in the increase of total phenols continuously up to 200 ml/L where 125.6 mg.g<sup>-1</sup> representing 118.9%, after which the total phenols declined at 250 ml/L.



**Fig. 1:** Effect of aqueous extract of *C. sinuosa* on total phenols in lupine leaves.

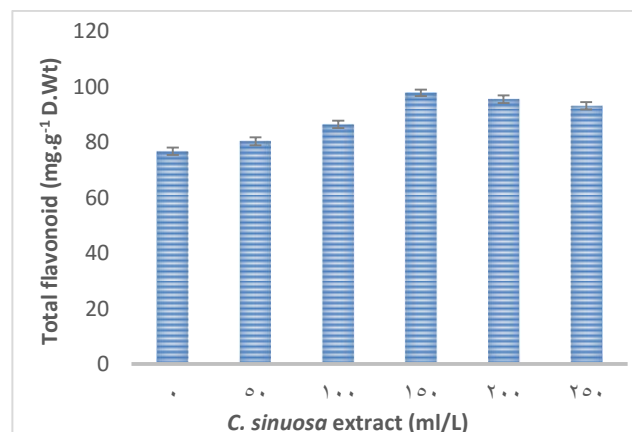
#### Effect of aqueous extract of *C. sinuosa* on total flavonoids in lupine leaves

The effect of various concentrations of *C. sinuosa* extract (50, 100, 150, 200 and 250 ml/L) on total flavonoids content on lupine leaves was looked into. The outcomes shown in Fig. in Fig. 2 indicate that there was a consistent rise in in total flavonoids up to 150 ml/L where the total flavonoid content was 97.9 mgg<sup>-1</sup>, representing 127.5% of the control value, then the content declined at the higher concentrations

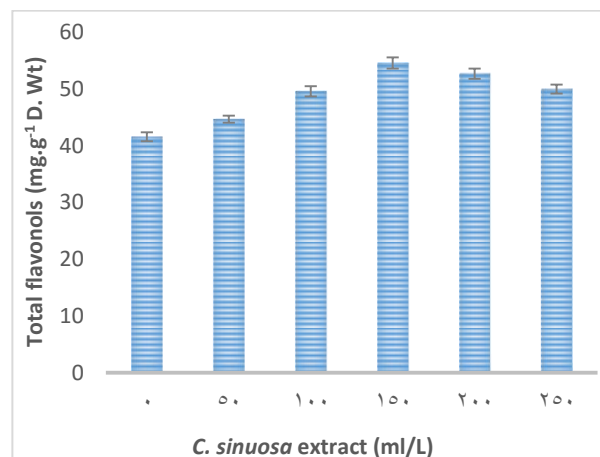
#### Effect of aqueous extract of *C. sinuosa* on total flavonols in lupine leaves

The effect of aqueous extract of *C. sinuosa* on total flavonols in lupine was investigated at

different concentrations (50, 100, 150, 200 and 250 ml/L). The results in Fig. 3 indicate that seaweed extract induced total flavonols in a concentration-dependent manner up to 150 ml/L where the highest content was 54.5 mg g<sup>-1</sup> representing 136.1% of the control value followed by reduction of the content at higher concentrations (200 and 250 ml/L).



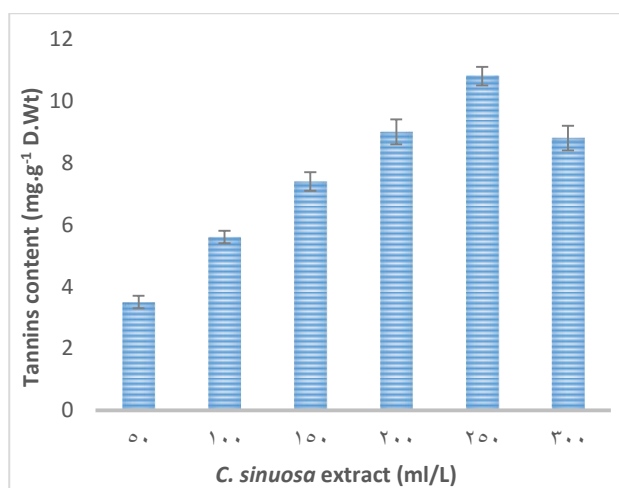
**Fig 2:** Effect of aqueous *C. sinuosa* extract on total flavonoids in lupine leaves.



**Fig. 3:** Effect of aqueous *C. sinuosa* extract on total flavonols in lupine leaves.

#### Effect of aqueous extract of *C. sinuosa* on total tannins content in lupine leaves

In this experiment the impact of extract of *C. sinuosa* on tannins content in lupine leaves was investigated at different concentrations (50, 100, 150, 200, 250, 300 ml/L). The results in Fig. 4 show that increasing in the *C. sinuosa* aqueous concentrations resulted in increasing of the total tannins content until it reached 10.8 mgg<sup>-1</sup> D.Wt at 250 ml/L concentration then declined at 300 ml/L.



**Fig. 4:** Effect of *C. sinuosa* extract on total tannins content in lupine leaves.

Since seaweeds are generated by biomass in response to exposure to both intrinsic and extrinsic environmental factors—for example age, length, and tissue type as well as external factors like herbivory, salinity, light, depth, and seasonality, they can be used as a source of antioxidants [24, 25, 26].

The present results indicate an increase in total phenolic compounds under treatment with *C. sinuosa* extract and the increase was dependent on the concentration. Simple phenols, such as phenolic acids, such as, caffeic, hydroxycinnamic acids, , ferulic, and sinapic acid, and hydroxybenzoic acids, such as gallic, p-coumaric, vanilic, , protocatechuic, syringic, and gentisic acid, 4-hydroxybenzoic are examples of phenolic compounds [24, 27]. Polyphenols, on the other hand, consist of flavonoids and non-flavonoids.

The extract of *C. sinuosa* increased the amount of flavonoids in lupine leaves. The concentration of seaweed extract determined how much this content was enhanced. Polyphenolic substances known as flavonoids are categorized into flavones, flavonols, flavanones, isoflavones, anthocyanidins, chalcones, and catechins based on their molecular structure [28]. Flavonoids have the capacity to raise glutathione levels, another potent antioxidant, which helps in protecting the cells [29]. Through a variety of processes, including the scavenging of free radicals including superoxide, hydroxyl, and peroxy radicals as well as the inhibition of the enzymes that produce free radicals, flavonoids demonstrate their antioxidant capacity [30].

Different ROS, such as superoxide anion, peroxy radicals, and hydroxyl radicals, are scavenged by flavonoids. They may also serve as singlet oxygen suppressors [31].

Following treatment with *C. sinuosa* extract, the flavonol content showed the same increase as the total flavonoids. Commonly present in plants, total phenol and total flavonoid have been associated with a number of biological functions, comprising the ability to function as antioxidants. When seaweed extracts were applied to lupine seeds, the quantity of total phenols and total flavonoids increased. As evidence, it has been observed that treating cabbage and spinach with seaweed boosted their phenolic content [32]. Seaweed extract treatment enhanced the amount of phenolic and flavonoid components in broccoli and cabbage, according to similar studies [33].

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