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Detection of the Components of the Root of *Saussurea Costus* and Study of the Effect of the Aqueous Extract of Its Roots on the Kidneys of Male Rats Treated with Chlorpromazine

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Keywords: Chemical composition, Saussurea Costus, Histological Structure, Kidneys, Chlorpromazine.

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Abstract:

Saussurea costus (Falc.) Lipsch plant belongs to the Asteraceae family. Its root has traditionally been used to treat a variety of ailments, including strep throat, dysentery, ulcers, and asthma. Studies have shown that S. costus root has a wide range of therapeutic effects, including antioxidant, antitumor, hepatoprotective and anti-inflammatory effects. The preliminary phytochemical screening of *S. costus* root revealed the presence of resins, flavonoids, alkaloids, saponins, and several other components. Twenty-five Swiss albino rats divided into five groups to study the effects of the extract. The first group represented the negative control group, while the second group was administered Chlorpromazine alone at a concentration of 2 mg/kg once daily. In the third, fourth, and fifth groups, alcoholic extracts of Indian costus root were administered at concentrations of 1.25 mg/kg, 2.4 mg/kg, and 4 mg/kg, respectively. The study showed that costus extract acted as a protective agent against the side effects of Chlorpromazine, particularly in safeguarding the kidneys from symptoms such as infiltration and tissue damage that were caused by the drug used in the treatment.

Keywords: Chemical composition, *Saussurea costus*, Histological structure, kidneys, Chlorpromazine.

الكشف عن مكونات جذور نبات *Saussurea costus (Falc.) Lipsch* ودراسة تأثير المستخلص المائي لجذورها على كلى ذكور الفئران المعالجة بالكلوربرومازين"

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الخلاصة:

ينتمي نبات *Saussurea costus (Falc.) Lipsch* إلى عائلة *Asteraceae*. حيث تم استخدام جذره تقليديا لعلاج مجموعة متنوعة من الأمراض، بما في ذلك التهاب الحلق والدوسنتاريا والقرحة والربو. أظهرت الدراسات أن جذر *S. costus* له مجموعة واسعة من التأثيرات العلاجية، بما في ذلك تأثيرات مضادة للأكسدة ومضادة للأورام وواقية للكبد ومضادة للالتهابات. كشف الفحص الكيميائي النباتي الأولي لجذر *S. costus* عن وجود الراتنجات والفلافونويدات والقلويدات والصابونين والعديد من المكونات الأخرى. تم تقسيم خمسة وعشرين فأرًا سويسريًا أبيض إلى خمس مجموعات لدراسة تأثيرات المستخلص. مثلت المجموعة الأولى مجموعة السيطرة السلبية، بينما تم إعطاء المجموعة الثانية الكلوربرومازين وحده بتركيز ٢ ملغم/كغم مرة واحدة يومياً. في المجموعات الثالثة والرابعة والخامسة، تم إعطاء المستخلصات الكحولية لجذور القسط الهندي بتركيزات ١.٢٥ ملجم/كجم، ٢.٤ ملجم/كجم، و٤ ملجم/كجم، على التوالي. وأظهرت الدراسة أن مستخلص القسط يعمل كعامل وقائي ضد الآثار الجانبية للكلوربرومازين، وخاصة في حماية الكلى من أعراض مثل الارتشاح وتلف الأنسجة التي يسببها الدواء المستخدم في العلاج.

الكلمات المفتاحية: التركيب الكيميائي، نبات القسط الهندي، التركيب النسيجي، الكلى، الكلوبرومايزين.

1. Introduction:

Chlorpromazine is considered one of the drugs that cause infertility, as it causes reproductive problems by causing oxidative and nitrous stress in the testicles [1], and it also hurts hormones, especially tissues that contribute to the formation and secretion of hormones that are related to sexual activity. Chlorpromazine mainly affects the central nervous system; it can have some effects on other organs, including the kidneys. The drug affects the blood flow in the kidneys by decreasing the blood flow in the kidneys, which may lead to decreased perfusion in the kidneys. This effect may be due to alpha-adrenergic blocking properties, which can cause blood vessels to dilate [2]. It is related to electrolyte imbalances. Chlorpromazine can upset the balance of electrolytes in the body, including sodium, potassium, and calcium. These imbalances can indirectly affect kidney function and may lead to complications. Urinary retention Chlorpromazine can cause anticholinergic effects, including urinary retention [3]. When urine

is not adequately eliminated from the body, it can increase Chlorpromazine has been associated with the development of interstitial nephritis, an inflammation of the kidney tissue. Symptoms of interstitial nephritis may include fever, rash, blood in the urine, and impaired kidney function [4]. The severity of these effects can vary depending on factors such as dose, duration of use, and an individual's susceptibility to infection.

Saussurea costus (Falc.) Lipschitz, also known as *Saussurea lappa* C.B. Clark, belongs to the Asteraceae family. It is a tall perennial herb that can grow up to 1-2 meters in height. The flowers are clustered and typically measure around 3-5 cm. The rosettes and heads of the flowers can vary in color [5], ranging from dark bluish to black. The plant blooms during July and August. Its fruits are approximately 3mm long, curved, arched, and compressed. The roots can grow up to 60 cm in length. One distinctive feature of this plant is its strong aromatic scent. The dried roots have a bitter taste [6]. *Saussurea costus* is a well-known and highly valued medicinal plant used in various indigenous medicinal systems to treat a wide range of ailments including asthma, inflammatory diseases [7], ulcers, and stomach problems. Sesquiterpene lactones have been identified as the primary bioactive constituents present in this plant species [8].

Northwest Himalayas, which is rich in medicinal terms, grows as a medicinal herb where it is used in the treatment of a variety of ailments such as carminative, expectorant, anti-inflammatory, antiseptic, aphrodisiac, analgesic and anthelmintic without any noticeable adverse effects [9], the root extract of S. costus root can significantly reduce the nephrotoxicity in rats. Because of its valuable effects, we recommend that Costus be added to the diet, and manufacturers and consumers should be more aware of the toxic effects of ethephon [10,11] ,It contains many biologically active phytochemical components. Substances used as traditional medicines are extracted from these plants. S. costus contains biologically active substances such as anti-tumor [12], anti-bacterial, and anti-inflammatory and anti-aging [13-16].

The study aimed to detect the active substances and some nutritional mineral elements present in the root extract of Indian Costus plant, and its effect on the histological structure of the kidneys treated with the drug Chlorpromazine.

2. Material and methods:

2.1. Preparation of plant materials: The healthy, disease-free roots of the S. costus plant root were acquired from the local markets in Ramadi city, dried, and crushed using an electric grinder. The plant powder was then stored in appropriate containers until usage, roots of S. costus were chopped and ground through a sieve of 150 microns In a water bath for 72 hours [17] 1000 ml of distilled water is pulverized into a very fine powder The mixture is stirred frequently for hours, then filtered Bypassing it through a piece of cotton gauze was the remnant

Re-washed three times with the same amount of moisturizing vent. The filter was also re-filtered using Whatman No. 1 filter paper and was the final nomination Oven dries completely. At a temperature of 4 °C, do not store the product in the refrigerator.

2.2. Phytochemical and mineral measurement methods: The plant was determined some phytochemical compound using standard methods by GC-MSsuch as (Glycoside, resin, phenolic compounds, flavonoids saponin alkaloids and tannins [18], Phosphorus was measured using spectrophotometer at wavelength 650 nm, while Calcium, Iron, Sodium, Potassium and Manganese and the other elements were measured by atomic absorption meter type [19] in College of Technical sciences laboratories.

2.3. Experiment design: Thirty-six adult male white rats were obtained from the animal house of the College of Agriculture at Baghdad University. In the College of Education for Women /Anbar University's animal home, animals were put in plastic animal cages. The cages were covered with sawdust, with attention to the cleanliness of the cages, and the sawdust was changed three times a week. Six rats were placed in each cage when rats were distributed. The rats were kept in suitable conditions in laboratories for ventilation, temperature, and lighting while receiving a constant supply of water and a standard dietary requirement. The experimental rats were assigned to five groups in the following manner:

1-Negative control group received only water.

2-Positive control group received oral chlorpromazine at a concentration of 2 ml per day, administered as a single dose. The chlorpromazine was diluted in 500 ml of distilled water to achieve the desired concentration of 2 ml, which was used to induce sterility in laboratory animals [19].

3-The groups receiving Costus root extract were divided based on the concentration of the extract. There were three groups, with each group receiving a daily dose of the extract at concentrations of 1.25 ml, 2.4 ml, and 4 ml respectively, for a period of 30 days.

2.4. Histological examination: Using the technique described in [20], histological preparations were produced after the testicle was removed and put in plastic containers containing 10% formalin .

3. Results and Discussion:

The phytochemical composition of root portions extracted from S. costus has characterized Seven components. The major constituents were, Phenolic compound 0.56 %, Glycoside 0.65% , Alkaloids 0.76 % ,Resins 40% Saponin 4.7% ,Tannins 12.5 % and Flavonoids 48%, as shown in Table 1.

Table 1: phytochemical analysis of f S. costus root extract

Compound	Amount%

Phenolic compound	0.56
Glycoside	0.65
Alkaloids	0.76
Saponin	4.7
Tannins	12.5
Resins	40
Flavonoids	48

Mineral concentration in root of S. Costus reveals six components of minerals as presented in **Table 1**, including potassium (K) (0.67), Calcium (Ca) (0.49), Copper (Cu) (0.65%), Magnesium(Mg) (0.33%%), Phosphorus (P) (0.3), Manganese (Mn) (0.1), and Lead (Pb) (0.5) showed in **Figure 1**.

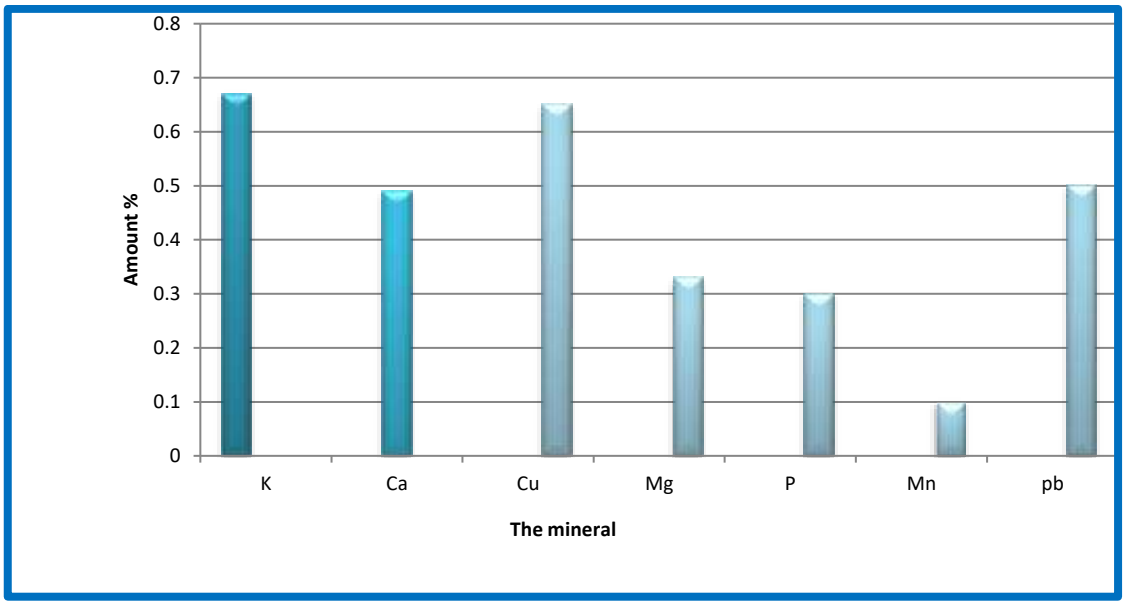

Figure 1: Mineral concentration in S. costus root (%)

In S. costus lignans, flavonoids, steroids, glycosides, triterpenes, sesquiterpenes, lactones, etc., were extracted and fractionated. Among the sesquiterpenes found in S. Costus are costunolide, α-cyclocostunolide,β-cyclocostunolide,iso-zaluzanin-C, ehydrocostuslactone ,ialantolactone, isoallierantolactone **[21]**, and sesquiterpenes from S. Costus have anticancer properties against breast, liver, colon, prostate, ovarian **[22]**.

Various pharmacological studies A variety of in vitro and in vivo models have provided compelling evidence to support the notion that it can be anti-inflammatory, antiulcer, anticancer, and hepatoprotective effects, supporting therationale for many of itstraditional uses **[23]**. The coylidene, dehydrocoylidene, and cynarin isolatedfromthisplanthavebeenidentified as potential bioactive molecules. Due to thesignificantbiologicalactivity of high-quality *S. costus*anditsconstituents, it is appropriateto developthem as medicinalproducts **[24]**,*Costus Saussurea's* efficacy in treating cancer, ulcers

and protecting the liver has been proven through numerous clinical trials, supporting plants are associated with a range of traditional applications and practices **[25,26].** The exceptional pharmacological properties of *Saussurea Costus* make it a preferred alternative for the discovery of new drugs. Although the information available to date indicates safety and potential efficacy, the bioactive ingredients, physiological mechanisms, pharmacokinetics, accessibility, and health impact remain unclear.

The histological examination results revealed significant pathological tissue changes in the kidneys of animals treated with Chlorpromazine compared to the control group. Administration of chlorpromazine to mice at a weekly dose for 30 days showed major alterations in the kidney tissue structure. These alterations included kidney tissue damage caused by the drug, characterized by acute vascular congestion, infiltration of inflammatory cells in multiple locations, desquamation of the proximal and distal tubular lining, acute tubular dilatation, edema, and tubular swelling with amyloid deposition in the glomerulus accompanied by thickening of Bowman's capsule. These renal tissue damages were attributed to the nephrotoxic effects of Chlorpromazine in comparison to the control group.

However, when examining the kidneys of animals that were orally administered 4 mg/kg of ethanol extract from the cayenne root for 30 days, along with oral administration of chlorpromazine, the histological diagnosis revealed that the kidney tissue tended to resemble the typical structure of Bowman's cyst dilatation, which is considered normal, meanwhile, the histological examination results of the kidneys in animals orally treated with an alcoholic extract of Indian costus root at a concentration of 1.4 mg/kg, along with simultaneous oral administration of the drug, revealed tubular edema in some renal tubules, glomerular congestion, expansion of Bowman's capsule, Henle's loop dilation, and desquamation of the superficial lining of the tubules. **Figure 2**

The histological diagnosis results indicated significant tissue alterations in the kidneys of the group treated solely with Chlorpromazine. The observed renal tissue damage and changes were attributed to the side effects of the drug, including inflammatory responses in multiple locations, desquamation of the proximal and distal tubular lining, tubular edema and swelling, and thickening of Bowman's capsule, consistent with the findings **[27]**, which indicated the effects of Chlorpromazine on liver and blood parameters. The results matched the study by **[28]**, which indicated the presence of these enzymes in the research sources and represented acute vascular congestion with infiltration of cells in the distal tubules due to daily exposure to Chlorpromazine drug, which negatively affected the distal tubules.

The protective effect of the costus plant against renal toxicity has been demonstrated based on the histological and biochemical analysis results. These findings are consistent with the

results of [29] which showed that administration of the alcoholic extract stimulated complete protection of the kidneys against toxins. Other plant extracts have also been shown to have a significant impact on tissue protection. Researchers [30] demonstrated that *S. costus* can reduce cellular changes and programmed cell death resulting from the effects of chemical substances and drugs. Administration of cinnamon extracts to experimental animals provided good protection against renal injury caused by aromatic hydrocarbons, resulting in a noticeable restoration of the pathological tissue structure of the kidney with minimal cellular damage due to the rich antioxidant, anti-inflammatory, and anti-apoptotic properties of costus extract, as well as its free radical scavenging activities [31,32].

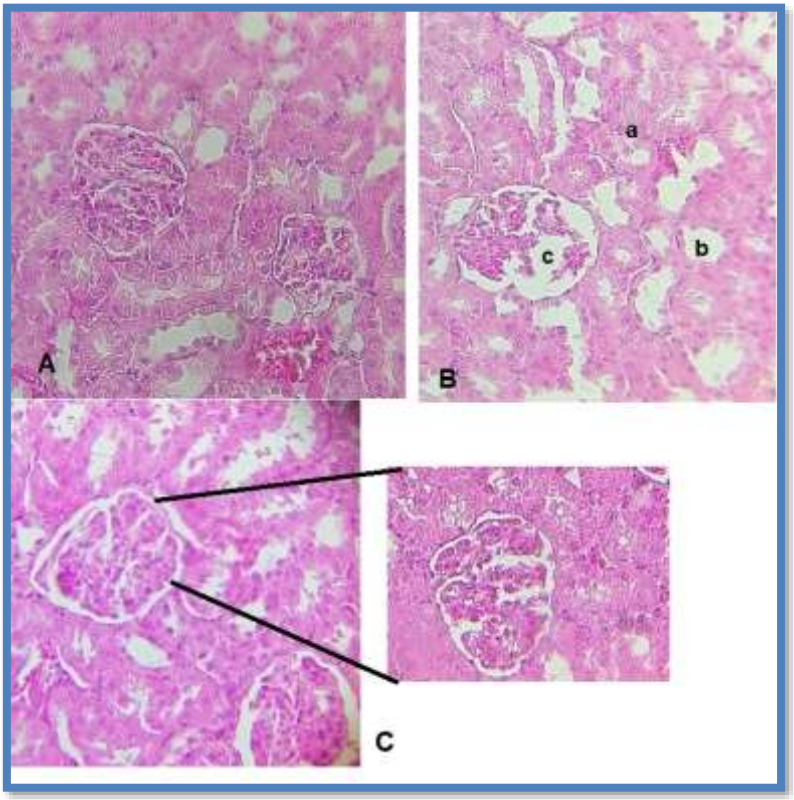

Figure2: A:The group treated with S. costus alcoholic extract at a concentration of 0.4 mg/kg and Chlorpromazine showing the histological appearance close to normal with Bowman's capsule dilated (40x),B: Histological section of a mouse kidney from the group Chlorpromazine only at concentration (a) shows acute blood congestion in the blood vessels. (b) Infiltration of inflammatory cells. (c)-The severe hydronephrosis of the tubules (40x), C: The group treated with alcoholic extract of S. costus 40x

Figure 3 indicates that there are significant variations in the weights between the groups, with a significance level of $P \leq 0.05$ were treated with Chlorpromazine and the groups that were treated with the alcoholic extract of premium and the drug together, the control group, where there was a significant increase in the values of their average weights in the group that was found. That and a wall were found in the parts that led to an increase in weight [33].

Experiments have shown that the weight of the kidneys increases when exposed to a cancerous or toxic substance, and the reason may be due to the tissue change and the accumulation of water [34,35].

A study showed higher concentrations of total phenols, total flavonoids, and antioxidant effects using 70% ethanol extract of Cassia root. It is the most effective antioxidant extract against sodium nitrite toxicity in the kidneys of male rats and can be safely used as a natural additive in the food industry [36] . Histological studies showed more pathological changes in the liver and kidneys compared to the negative control group. Ethanol extract from S. lappa protects liver and kidney cells from drug damage [37].

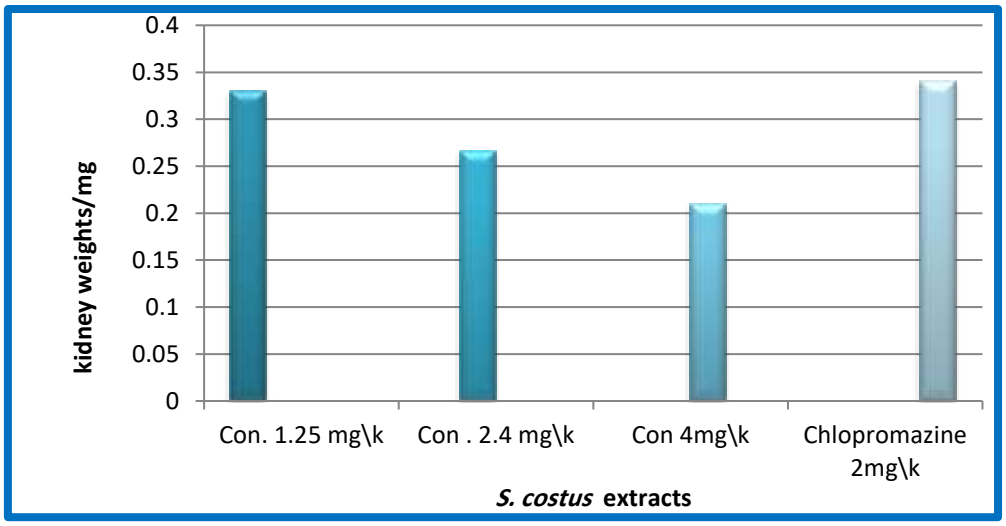

Figure3: Effect of S. costus extracts and the chlorpromazine on kidney weights/mg.

5. Conclusions

Research has focused on *Saussurea* premium and its potential therapeutic effects. *Saussurea costus*, a member of the Asteraceae family, has a long history of traditional use in treating various ailments. The study showed that the alcoholic extract of Costus root acted as a protective agent against the side effects of chlorpromazine, specifically in maintaining kidney health. Preliminary phytochemical examination revealed the presence of several biologically active components in the plant, such as resins, flavonoids, alkaloids, and saponins. *Costus* extract mitigates the adverse effects of chlorpromazine, particularly in protecting the kidneys from effusion and tissue damage caused by the drug alone. These results indicate that the extract from *Saussurea* root may have potential therapeutic effects in protecting against chlorpromazine-induced kidney damage. However, further research is necessary to elucidate the underlying mechanisms and confirm these findings in human studies.

Overall, this study provides valuable insights into the protective effects of Costus sagittarius extract on kidney tissues and highlights its potential as an adjunctive therapy to reduce renal side effects associated with chlorpromazine treatment.

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