EVALUATING THE ANTIMICROBIAL AND ANTIOXIDANT PROPERTIES OF *DUCHESNEA INDICA* FRUIT EXTRACTS

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ABSTRACT

The perennial herb *Duchesnea indica*, which belongs to the rosaceae family, is found all over the world. An essential component of medication research and development is ethnobotanical studies. The study of medicinal plants used in traditional medicine is justified by the increasing need to develop alternatives for the treatment of chronic degenerative disorders, including metabolic syndrome, diabetes, and hypertension. Several *Duchesnea indica* fruit extracts were used in a study for several tests. The fruits' substantial fiber and protein and oil concentrations were discovered via chemical analysis. Extracts also include a sizable amount of flavonoids, alkaloids, tannins, saponins, and phenols. Fruit showed the highest antioxidant capacity when the scavenging activity of the fruit extracts was assessed using a variety of solvent-based tests. The leaf extracts' antibacterial properties were evaluated against both and found to be effective against microorganisms. Higher inhibitory efficacy against gram-negative bacteria was demonstrated by the methanolic extract. Fruit extracts from *Duchesnea indica* have been shown to have the potential to offer pharmaceutical raw materials to the industry for the production of antibacterial and anticancer medications, among other uses, and to be a good source of natural antioxidants.

Key words: *Duchesnea indica*, secondary metabolites, anti-oxidant, anti-microbial potential.

INTRODUCTION

Pakistan holds a special place on the map. It has a wide range of climatic conditions with significant potential for medicinal plant varieties (Khan and Gul, 2007). Significant amounts of compounds are produced by plants, such as optional and necessary metabolites, which are used extensively in modern medicine. Indigenous people typically rely on traditional medications made from medicinal plants. These plants are capable of absorbing a wide range of harmful substances. Basic responses may occur if such plants are used for medicinal purposes. on the toxicological theory of medicinal plants, an attempt has been made to examine the fundamental components of Pakistani ethnotherapeutically important plants and to draw attention to the present gaps in knowledge on the safety and effectiveness of traditional natural remedies.

Because of their genotype, plants differ in their chemical and phytonutritional makeup; hence, a chemical molecule that is active in one plant may not be present in another. The soil in which plants are grown and the kind of fertilizer that is given to them both affect the chemical makeup of plants.

Consuming strawberries has been shown to reduce the incidence of several diseases since they are a high source of polyphenols, which also contribute to fruit shading and plant disease resistance. The polyphenolic structure of commercial strawberries (Fragaria x ananassa) has been extensively studied, but little is known about the polyphenols in wild strawberries (*Duchesnea indica*) (Archbold,2016)

Duchesnea indica is a member of the Rosacea family. Although it appears to be the separate development of a comparable fruit type, it is quite similar to the actual strawberry, Fragaria ananassa. *Duchesnea indica* is native to Eastern and Southern Asia, although it is widely utilized as a decorative plant elsewhere in the world. In traditional medicine, the herb is used as an anticoagulant, an antiseptic, and to lower fever. Fruit is used to treat skin conditions, blossoms are good for blood circulation, and leaves are used to treat swellings. Additionally, the plant

combination is used to treat burns, boils, abscesses, and other conditions (Duke et al., 1985). Flavonols, hydroxybenzoic acid and hydroxycinnamic acid derivatives, ellagitannins, ellagic acid and ellagic acid glycosides, and others are being found in D. indica (Zhu et al., 2015). Because natural medicines are more effective and have fewer side effects, there has been a recent surge in interest in them. The chemicals known as antioxidants prevent other molecules from oxidizing. Free radicals are created during the process of oxidation, which involves the loss of electrons. Thus, by creating more free radicles, the generated free radicle starts a chain reaction. Numerous illnesses, such as cancer, heart disease, brain problems, Alzheimer's disease, and cognitive impairment, can be brought on by free radicals (Kinnula and Crapo, 2004; Singh and Jialal, 2006; Sas et al., 2007; Smith et al., 2000; Guidi et al., 2006). The two main types of antioxidant systems that protect the body against the generation of free radicals are the enzymatic and non-enzymatic systems. Antioxidants can also obtained through antioxidant supplements and a variety of foods that are high in antioxidants if the body is unable to produce them. Elderly members of the community are fully aware of ethnotherapeutic conventional understanding on the uses of native medicinal herbs for various human illnesses. The younger generation pays relatively little attention to these important traditional treatment methods (Ejaz et al., 2015). Antibiotics heal a lot of microbiological disorders. However, long-term usage of antibiotics causes drug resistance, which lowers the medicine's effectiveness.

These plant-based medications are safe, effective, affordable, and seldom cause adverse effects. Among other things, research is focused on plant-derived antimicrobials due to their broad antimicrobial range and ease of accessible. The antibacterial qualities that reduce the likelihood of microbial resistance development are typically attributed to a combination of substances with distinct target locations (Al-Zahrani et al., 2016). Scientists are creating new antibiotics as a result of the emergence of several drug-resistant bacteria. Bacterial and fungal infections are become increasingly prevalent over time. Antimicrobial compounds are naturally produced by *Duchesnea indica* as a defense against plant pathogens. sa According to Rosa et al. (2003), these substances naturally stop bacteria and fungi from growing on plants. As a result, this study has goals and

objectives.

(1) Compositional study of *Duchesnea indica* fruit.

(2) Evaluation of different bioactive components in fruit extracts.

(3) Evaluation of the fruit of Duchesenea indica for bioactivity.

MATERIALS AND METHODS

Collection of Samples

In 2023, samples of Duchesne indica leaves and fruit will be gathered from the mountainous regions around Islamabad. Samples of leaves and fruit will be gathered in delicate plastic bags that are properly labeled with the sample's name, date, and collecting location. Expert taxonomists will correctly identify the samples, and they will be registered as specimens for future use.

Getting Plant Samples Ready

The samples of leaves and fruits will be sun-dried and shadow-dried before being oven-dried overnight at 60 degrees Celsius. The dried samples will be stored in plastic bags for later use after being processed using an electrical grinder and an 80 msh sieve.

Preparation of plant Extracts

Using 50 grams of plant extracts each, methanolic and ethaolic extracts were made using the Soxhlet apparatus and rotary evaporator procedures. The samples were shaken overnight and then filtered.

PROXIMATE ANALYSIS

Moisture, crude fat, crude protein, crude fiber, and carbohydrates were the characteristics identified for proximate analysis. All of these analyses were carried out using the methodology described by Rao et al. (2007).

QUALITATIVE ANALYSIS OF PHYTOCHEMICALS

Preparation of plant Extracts

200 milliliters of refined water and five grams of finally dried and powdered plant are placed in a container. For 20 minutes, heat on a hot plate while constantly stirring at between 30 and 40 degrees Celsius. Channel paper was used to sift the water extract, and the filtrate was then used for the phytochemical analysis. For subsequent tests, the water extract must be kept in the refrigerator (Sofowra, 1993; Trease et al., 1989).

Test for Proteins

Ninhydrin test: crude extracts heated in two milliliters of water. The presence of amino acids was detected by violet coloring and 2% of Ninhydrin solution.

Test for Reducing Sugars

Fehling's test: Two milliliters of the Fehling A and Fehling B reagent solution were added to the crude extract, and the mixture was then boiled. The presence of reducing sugars is indicated by the brick-red precipitate.

Test for Carbohydrates

a) **Benedict's test:** Two milliliters of Benedict's reagent were added to the concentration and heated until it began to boil. Carbohydrates are present when the precipitate appears reddish-brown.

Test for Phenol and Tanins

Crude extract was combined with a 2% NaOH solution. The presence of phenols and tannins was suggested by an overabundance of yellow that became colorless when a few drops of weak acid were added.

Test for Flavonoids

2 milliliters of a 2% NaOH solution were added to the crude extract. When a few drops of diluted acid were added, the strong yellow hue that had developed went colorless, indicating the presence of flavonoids.

Test for Saponins

In a test tube, add the crude extract and 5 milliliters of distilled water, then shake well. The presence of saponins is indicated by the development of foam.

Test for Glycosides

Liebermann's test

Two milliliters of acetic acid and two milliliters of chloroform had been mixed with crude extract. After cooling on ice, the mixture was carefully mixed with concentrated H2SO4. The existence of a steroidal nucleus, or the glycine part of glycosides, was identified by a color shift from violet blue to green.

Test for Steroid

Two milliliters of chloroform were mixed with crude extracts, and H2SO4 was added to the glass tube's walls. The presence of steroids was detected by the red hue that was created in the decreasing chloroform layer.

Test for Terpenoids

After mixing 2 milliliters of chloroform with crude extract, the mixture was evaporated until it was completely dry. Two milliliters of concentrated H2SO4 were then added, and the mixture was heated for two minutes. Terpenoids are indicated by the grayish tint.

Test for Alkaloids

After blending the crude extract with two milliliters of 1% HCl, it is gently heated. The combination was then exposed to Mayer's and Wagner's reagents. The resulting precipitate's turbidity is interpreted as proof that alkaloids are present.

QUANTITATIVE ANALYSIS OF PHYTOCHEMICALS

Determination of Alkaloids

10% acetic acid (20 ml acetic acid + 180 ml ethanol) was mixed with 5 g of the sample. For four hours, cover and stand. Next, After filtering the solution, set the filtrate on a water bath and let it evaporate for a quarter. After that, gradually add concentrated ammonium hydroxide to the

solution and let it settle. Filter the solution once again. Use diluted ammonium hydroxide to wash the precipitates that have been collected on filter paper. Weigh the filter beforehand. Dry in an oven and use the weight difference to get the percentage (Haborne, 1973).

Determination of Saponins

100 milliliters of 20% aqueous ethanol are mixed with around 10 grams of the test. Put them in a water bath at 55 degrees Celsius and let them there for four hours while continuously mixing. Transmit the arrangement. Use 200 milliliters of 20% ethanol to re-extract. After a 90°C water shower, the consolidated concentration decreased by up to 40 milliliters. The transferred concentrate was placed in a separate funnel. Add 20 milliliters of di-ethyl ether and shake vigorously. Two layers were obtained; the aqueous layer was recovered, and the ether layer was discarded. The cleaning process was repeated repeatedly. Two rounds of washing were performed on the consolidated concentrate using 60 milliliters of n-butanol and 10 milliliters of 5% sodium chloride water. Heat the sodium chloride layer in a water shower to disperse the concentration after transferring it to pre-measured recepticles. After the sample was dried in an oven, the weight and percentage of saponins were calculated (Obadori and Ochuko, 2001).

Determination of Flavonoids

At room temperature, 10g of the material was extracted using 80% methanol. Pre-weight falcon tubes were used to transmit the filterate. In a water bath, evaporation takes place. The extracted material was calculated in percentage (Bohn and Kocipai, 1994)

Determination of Tanins

A colorimetric technique was used to assess tannins. For 12 hours, 0.125g of the test was extracted using 25ml of 70% acetone on a mechanical shaker. After the arrangement was separated, 2.5 ml of Na2CO3 and 0.5 ml of Folin-Ciocalteu reagent were added. A spectrophotometer was used to detect absorbance at 725 nm. Tannic corrosive was used as a standard (mg/g of removed

component), and tannins in concentrate were assessed using conventional bend findings. Three separate investigations were conducted (Pearson, 1976).

Determination of Total Phenolic Contents

The Folin-Ciocalteu reagent was used to quantify the amount of phenols in the concentrate. 200 milliliters of purified water and five grams of powder test were mixed together. On a hot plate, warming and mixing were done for 20 minutes at 30 to 40 degrees Celsius. After filtering the solution, 1 milliliter of concentrate, 2.5 milliliters of 10% Folin-Ciocalteu reagent, and 2 milliliters of 2% Na2CO3 were added. For fifteen minutes, the mixture was allowed to sit at room temperature. A spectrophotometer was used to measure the test absorbance at 765 nm by distinguishing between gallic destructive (1 mg/ml), which was used as a clear. Gallic acid equivalent (mg/g of isolated component) was used as a benchmark to quantify the concentrate's total polyphenol content. Three separate examinations are conducted. Harborne (1973).

ANTIOXIDANT ASSAY

Four techniques are employed to determine the antioxidant activity of *Duchesnea indica* fruit and leaves.

Assay for DPPH Radical Scavenging Activity

The technique explained by Chew et al. (2008) is to determine the test's antioxidant effect. Individual solvents were used to break up 1 mg of the unprocessed concentration. Using the stock, completely unique foci were created. Two milliliters of DPPH (2.4 milligrams of DPPH plus 100 milliliters of methanol) were added to a tube containing one milliliter of each focus. At a wavelength of 517 nm, absorbance was measured after 30 minutes of hatching at room temperature. As anticipated, water-soluble vitamins were used. Because DPPH is lightweight and fragile, the experiment should be conducted in a dimly lit environment. The following requirement determines the example's ability to construct the DPPH structure:

$$Scavenging\ effect(\%) = \frac{(Control - Sample)}{Control}$$

ABTS Assay

The ABTS test is performed using the procedure described by Re et al. (1999). Potassium persulfate and ABTS were added to distilled water at concentrations of 2.45 mM and 7 mM, respectively. In order to provide the ABTS radical (ABTS•+), these two configurations were combined, and the combination was allowed to sit at room temperature for 16 hours before being used. The ABTS radical arrangement was weakened with refined water to an absorbance of 1.00 at 734 nm in order to investigate phenolic intensifies. The absorbance reading was then collected six minutes after mixing using the spectrophotometer after the ABTS arrangement was introduced to the test. The results show that phenols may scavenge 50% of the free radical ABTS • + (IC50). Every determination was made three times.

Reducing Power Assay

Cutting back Utilizing the approach described by (Ullah et al., 2013), the concentrates' power was determined. 4 mg was dissolved in 1 ml of several solvents to create the stock arrangement. From the stock, distinctive fixations were removed. The response blend contained 400 µl of phosphate cradle (0.2M, pH 6.6) and 500 µl of potassium ferricyanide (1%). After 30 minutes of incubation at 50ºC, add 500µl of 10% trichloric acid to the response blend. At 700 nm, absorbance was measured. As a positive control, ascorbic acid is used.

ANTIMICROBIAL ACTIVITY

Preparation of Extract For Antimicrobial Activity

Solvents such as methanol and ethanol were used to extract the powdered material. The material was extracted by shaking it for 24 hours and then extracting it again. The sample is then allowed to dry. Crude extract was produced after the solvent evaporated.

Microorganisms Tested

Using the agar well diffusion technique, antimicrobial activity was evaluated against Salmonella typhi, Pseudomonas aeruginosa, Stephylo aureus, and gram-negative Escherichia coli. In various vials, bacterial inoculums were produced in autoclaved Lauria-Bertini medium gL^{-1}. For

optimal development, the inoculum was incubated for 24 hours at 37°C.

Statistical analysis

Data obtained after analysis was for subjected to further statistically analyzed

RESULTS AND DISCUSSION

PROXIMATE ANALYSIS

The five fundamental components of a biological sample—moisture or water content, crude protein, crude fat, crude fiber, and total carbohydrates—are examined using proximate analysis. These tests also provide us with a low-cost indication of the sample's nutritional composition. However, the nutition panel has to be verified.

The related tables and figures display the phytochemical analysis's findings. Although fruit has a higher percentage of crude protein and carbs (14.2 and 18.16 percent, respectively), it has a lower percentage than leaves. Fruit samples have a low moisture level and a high carbohydrate content, as seen by their 5.5% moisture content, 6.32% crude fat, and 6.32% crude fiber.

Table 1. Proximate Analysis (%) of Leaves and Fruits of *Duchesnea indica*

	Fruits
Moisture	5.5± 1.3
Crude Protein	1.2 ± 0.01
Crude Fat	0.001±0.08
Crude Fibre	3.0±0.005

Carbohydrates	8.0±0.02

Result are Mean ± Standard deviation (n=3)

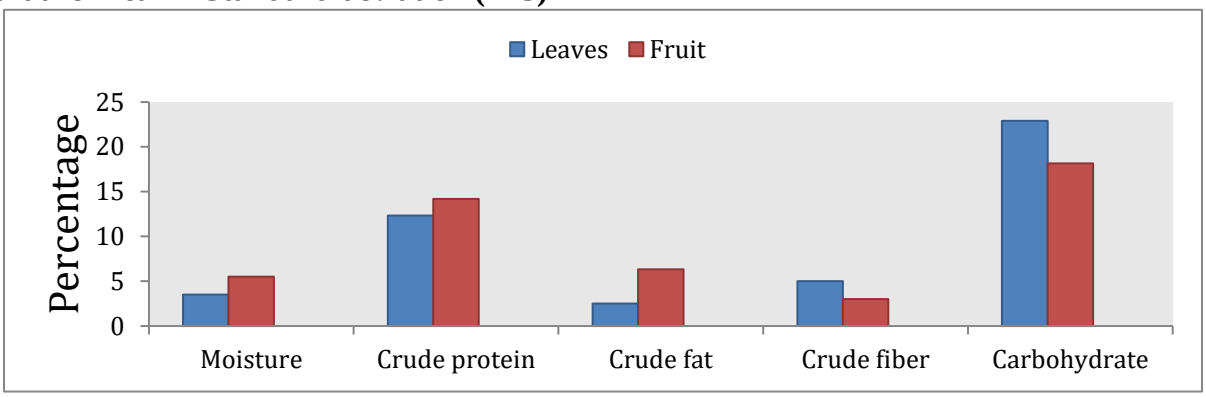

Figure 1. Proximate analysis of leaves and fruit of *Duchesnea indica*

QUALITATIVE ESTIMATION OF PHYTOCHEMICALS

Both the leaves and the fruit of *Duchesnea indica* are rich sources of important bioactive components, such as flavonoids, tanins, saponins, alkaloids, and phenols, according to a phytochemical investigation. Its fruit and leaves have therapeutic value, as evidenced by the presence of several bioactive substances. Because of its high phenol content, the fruit can be utilized to boost immunity against a number of illnesses. Table 2 displays the presence and absence of several bioactive substances. relatively high tannin content when compared to fruit. Each 100g sample contains 26.56±4.5 mg of leaves and 1.22±0.084 mg of fruit. Tannic acid served as the standard for this investigation. The following figures display the tannic acid calibration curve.

Table 2. Qualitative estimation of phytochemicals of *Duhesnea indica* **extracts fruits**

S.No	Phytochemicals	Fruits
1	Alkaloids	+

2	Carbohydrates	+
3	Saponins	+
4	Flavonoids	+
5	Protein and amino acids	+
6	Reducing sugar	+
7	Phenol and tanins	+
8	Cardiac glycosides	+
9	Steroids	+
10	Terpenoids	+
11	Glycosides	+

+ Present – Absent

QUANTITATIVE ESTIMATION OF PHYTOCHEMICALS

To determine a plant's nutritional and therapeutic value, quantitative phytochemical assessment is essential. The most prevalent phytochemicals include alkaloids, flavonoids, phenols, saponins, and tannins. Although phenols have a variety of pharmacological properties, their antioxidant properties are the most significant. The phytochemical content of *Duchesnea indica*,

expressed in milligrams per 100 grams of sample, is displayed in Table 3. Flavonoids are found in fruit at 42.8±1.05 mg/100g of sample and in leaves at 20.56±0.67 mg/100g of sample. Using gallic acid as a reference, the method described by Harborne (1973) was used to determine the total phenols. Gallic acid conventional curve. According to this, *Duchesnea indica* leaves have a lower total phenol content than other leaves, at 84.4±8.3 mg per 100g of sample, however fruit has a greater total phenol content, at 517±214.3 mg per 100g of sample. Both human and animal tissue and cell development are significantly influenced by phenols. Phenols also strengthen the immune system.

Glucosides are saponins. mostly present in herbs, beans, and vegetables. Additionally, saponins can strengthen immunity. Research also shows how they affect anticancer activity, bone loss, and blood cholesterol levels. Comparatively speaking, *Duchesnea indica* leaves had more saponins than fruits, with leaves having 20.4±4.5 mg and fruit having 0.78±0.1 mg per 100 g of sample. In contrast, a greater amount of alkaloids (5.1±0.42) mg per 100 g of sample was found in leaves compared to fruit (4.8±0.2) mg. Because they are phenolic compounds, tannins have the ability to exhibit antioxidant properties. They also have antimutagenic and anticancer properties since they are antioxidants. Upon analyzing the tannin content of *Duchesnea indica*, it was shown that the leaves had.

Table 3. Quantitative estimation of phytochemicals (mgGAE/100g) in *Duchesnea indica* **Leaves and Fruits**

	Flavonoid	**Phenol**	**Saponins**	**Tanins**	**Alkaloids**
Leaves	20.56±0.67	84.4±8.3	20.4±0.53	26.56±4.5	5.1±0.42

Fruit	42.8±1.05	517±214.3	0.78±0.1	1.22±0.084	4.8±0.2

Values expressed as mean ± Standard Deviation (n=3)

ANTIOXIDANT ASSAY

DPPH radical scavenging activity assay

The test most frequently employed to assess a sample's antioxidant activity is DPPH. At 517 nm, it provides an excellent band for absorption. For this experiment, ascorbic acid was the standard. *Duchesnea indica* fruit has been found to be a stronger source of antioxidants than leaves, while methanolic extracts of both the fruit and leaves perform better than ethanolic extracts. The IC50, or the lowest concentrations needed to achieve 50% activity, is a way to explain the DPPH finding. The leaf methanolic extract's IC50 value was 46.2±0.3) µg/ml, whereas the leaf ethanolic extract's was 54±0.5) mg/ml. Additionally, the ethanolic fruit extract's IC50 value was 50.3-0.44 µg/ml, whereas the methanolic fruit's was 38±0.8 µg/ml. The specific findings of *Duchesnea indica* leaves and fruit are presented. The above tables and figures show the percentage of scavenging activity of the leaves and fruits in relation to the scavenging activity of standard ascorbic acid. The methanolic fruit extract was shown to have the highest scavenging activity of all the leaf and fruit extracts, with an IC50 of 38µg/ml. The above figures compare the IC50 values of the fruit and leaves with the standard.

Table 4. DPPH Free radical scavenging potential of leaves and fruits of *Duchesnea indica* **at 517nm Values expressed as mean ± Standard Deviation (n=3)**

	Sr No	**Leave Extract with different solvents**	DPPH scavenging effect (%) at different concentration (µg/ml) ± STDEV					
			20µg/ml	**40µg/ml**	**60µg/ml**	**80µg/ml**	**100µg/ml**	**IC50 µg/ml**

	1	**Standard Ascorbic acid**	83.62±0.54	86.4±0.45	93.52±0.5	94.3±0.28	94.72±0.26	39.92±0.4
LEAVES	**2**	**Methanolic extract of D.I**	75.6±0.5	77.01±1.0	79.5±0.5	80.3±0.4	80.7±0.3	46.2±0.3
	3	**Ethanolic Extract of D.I**	23.5±0.5	52.3±0.3	65.3±0.3	73.3±0.2	82.5±0.4	54.0±0.5
FRU	**4**	**Methanolic extract of D.I**	92.2±0.3	94.5±0.5	96.3±0.5	96.8±0.7	97.4±0.5	38.0±0.8
IT	**5**	**Ethanolic Extract of D.I**	25.1±0.3	50.5±0.5	78.6±0.5	81.5±0.5	82.5±0.4	50.3±0.4

ABTS Assay

2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid), or ABTS, is a source of free radicals that may be used to assess the antioxidant capacity of different materials. In order to create free radicals, ABTS powder is dissolved in distillation water and left in the dark for 12 hours. Light at 734 nm is absorbed by the blue-green radicle cations of ABTS. The radical cation reacts with antioxidants such vitamins, thiols, and phenolics. The blue-green color of ABTS became colorless upon reaction. Ascorbic acid served as the assay's standard. The IC50 value for standard ascorbic acid in this experiment was 39.9±0.51µg/ml. The ethanolic extract of *Duchesnea indica* fruit exhibited the highest activity and, as a result, the lowest IC50 value, measuring 37.2±0.47µg/ml. With an IC50 value of 37.7±0.67, the methanolic extract of leaves likewise shown a high capability for

scavenging, indicating that *Duchesnea indica*'s fruit and leaves are both excellent sources of antioxidants and, as such, have significant therapeutic benefit.

Table 5. ABTS free radical scavenging activity and IC50 of Leaves and Fruits of *Duchesnea indica* **(Absorbance 734nm)**

	Sr No	**Leave Extract with different solvents**	ABTS scavenging effect (%) at different concentration (µg/ml) ± STDEV					
			20µg/ml	**40µg/ml**	**60µg/ml**	**80µg/ml**	**100µg/ml**	**IC50 µg/ml**
	1	**Standard**	89.7±0.7	90.3±0.3	91.2±0.3	92.4±0.6	93.2±0.3	39.9±0.5
LEAVES	**2**	**Methanolic extract of D.I**	93.5±0.5	94.5±0.4	95.2±0.3	96.2±0.3	99.3±0.4	37.7±0.7
	3	**Ethanolic extract of D.I**	2.4±0.3	3.2±0.3	4.5±0.4	14.4±0.4	15.2±0.3	64.9±0.7
FRUIT	**4**	**Methanolic extract of D.I**	95.5±0.5	96.1±0.2	96.7±0.3	97.0±0.3	98.6±0.6	38.5±0.5
	5	**Ethanolic extract of D.I**	96.5±0.5	97.7±0.4	98.0±0.5	98.9±0.4	99.5±0.3	37.2±0.5

Values expressed as mean ± Standard Deviation (n=3)

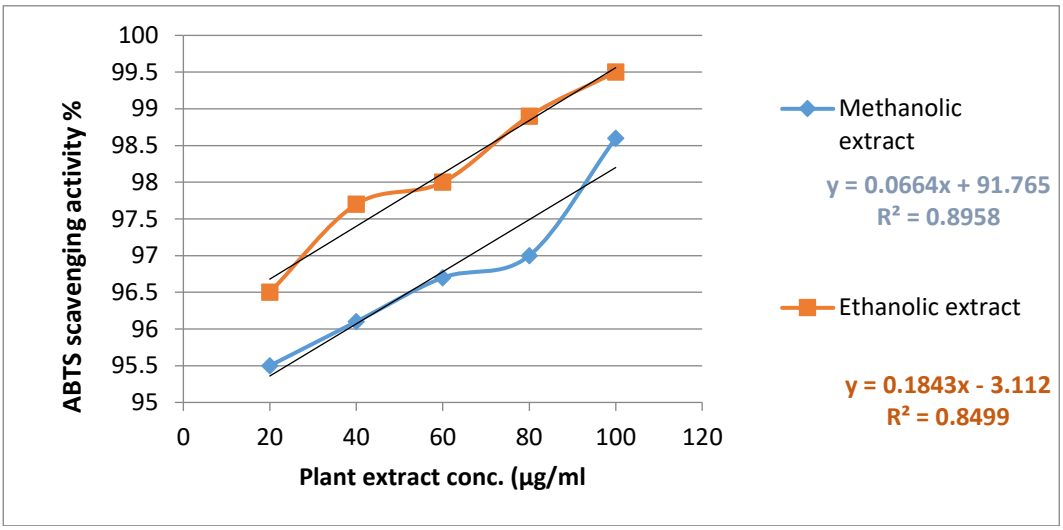

Figure 2. ABTS scavenging activity comparison Methanolic extract of leaves and Ethanolic extract of leaves

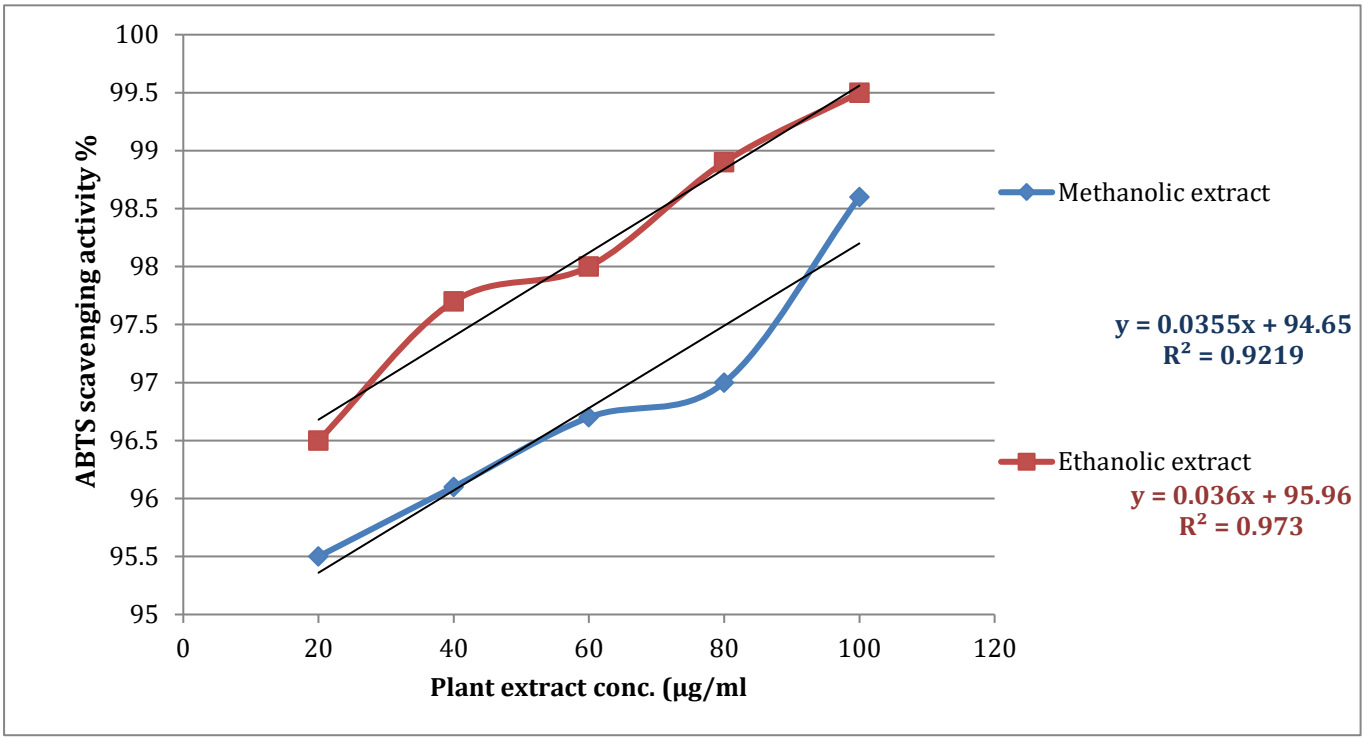

Figure 3. ABTS scavenging activity comparison Methanolic and Ethanolic fruit extracts

Figure 4. Comparison of IC_{50} values of ABTS assay.

Reducing power assay

Antioxidants in the samples would cause Fe3+ to be reduced to Fe2+ in the reducing power test. As sample concentration rises in a reducing power assay, absorbance will rise as well. This indicates that the rate at which Fe3+ is converted to Fe2+ is increasing, which indicates an increase in antioxidant activity. Ascorbic acid is the standard used in the reducing power test. Methanolic and ethanolic extracts were the two distinct extracts used in the test. Specific values of how absorbance rises as sample concentration rises.

Table 6. Reducing power assay of leaves and fruit extract of *Duchesnea indica* **(Absorbance 700nm)**

	Sr No	**Leaves Extract with different solvents**	Reducing power assay scavenging effect (%) at different concentration (µg/ml) ± STDEV				
			20µg/ml	**40µg/ml**	**60µg/ml**	**80µg/ml**	**100µg/ml**

	1	**Standard**	0.25±0.03	0.30±0.003	0.35±0.05	0.42±0.03	0.45±0.05
LEAVES	**2**	**Methanolic extract**	0.18±0.02	0.22±0.01	0.25±0.04	0.29±0.03	0.30±0.02
	3	**Ethanolic extract**	0.41±0.03	0.48±0.03	0.53±0.04	0.54±0.03	0.57±0.03
FRUIT	**4**	**Methanolic extract**	0.11±0.3	0.13±0.01	0.15±0.04	0.18±0.02	0.2±0.02
	5	**Ethanolic extract**	0.01±0.01	0.12±0.03	0.17±0.05	0.18±0.07	0.2±0.10

Values expressed as mean ± Standard Deviation (n=3)

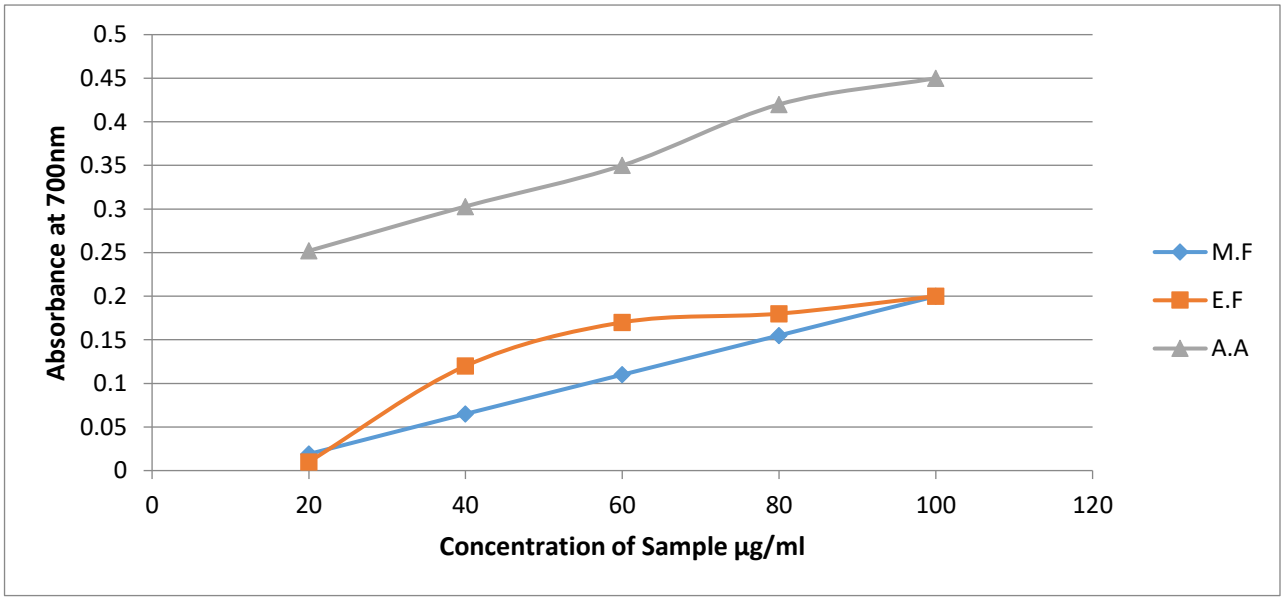

Figure 5. Reducing power of Methanolic fruit and Ethanolic fruit of *Duchesnea indica* and

Ascorbic acid as standard

ANTIMICROBIAL ACTIVITY

Medicinal herbs are widely accessible, affordable, and have few or no negative effects. The majority of them have antibacterial qualities. According to recent research, medicinal plants from all over the world can offer a wealth of antibacterial properties.

The antibacterial properties of several *Duchesnea indica* fruit and leaf extracts were evaluated. Levofloxacin is a common medication. Based on that, the leaves' ethanolic extract exhibited the highest effectiveness against the Escherichia coli inhibition zone, measuring 10.33±5.03 mm. There was no effect of the leaf ethanolic extract on *Salmonella typhi* or *Staphylococcus aureus*. While a 9±1.56 mm zone was seen in the case of the ethanolic extract, the methanolic extract of leaves also shown no efficacy against *Pseudomonas aeruginosa* Table 5.

The methanolic fruit extract exhibited the highest inhibition zone against Pseudomonas aeruginosa (16.82±1.28), but the ethanolic extract had no action against the same strain. Additionally, there was no activity of fruit ethanol extract against Escherichia coli. MIC µg/ml is the minimal extract concentration needed to suppress antibacterial activity. The above tables and figures display the MIC values for the methanolic and ethanolic extracts of *Duchesnea indica*'s leaves and fruit.

Table 7. Antibacterial activity of leaves of *Duchesnea indica*

Test organism	**Diameter of zone of inhibition (mm)**		
	Methanolic extract of leaves 200µg/dicsc	**Ethanolic extract of leaves 200µg/disc**	**Standard drug (Levofloxacin) 200µg/disc**
Escherichia coli	7.2±0.8	10.3±5.03	20.9±0.4

Staphylococcus aureus	7.2±1.9	N.A	23.5±0.5
Pseudomonas aeruginosa	N.A	9.0±1.6	21.0±0.00
Salmonella typhi	8.8±0.8	N.A	22.0±1.5

Values expressed as mean ± Standard Deviation (n=3)

Table 8.Antibacterial activity of fruit of *Duchesnea indica*

Test organism	**Diameter of zone of inhibition (mm)**		
	Methanolic extract of fruit 200µg/dicsc	**Ethanolic extract of fruit 200µg/disc**	**Standard drug (Levofloxacin) 200µg/disc**
Escherichia coli	11.0±4.6	N.A	20.9±0.4
Staphylococcus aureus	10.7±2.5	13.6±0.9	23.5±0.5
Pseudomonas aeruginosa	16.8±1.3	N.A	21.0±0.0
Salmonella typhi	8.7±1.3	11.6±2.1	22.0±1.5

Values expressed as mean ± Standard Deviation (n=3)

Table 9. Minimum inhibitory concentration (MIC) of leaves of *Duchesnea indica*

Test Organism	**MIC value of methanolic extract (µg/ml)**	**MIC value of ethanolic extract (µg/ml)**	**MIC value of standard levofloxin (µg/ml)**
Escherichia coli	50	20	5
Staphylococcus aureus	50	N.A	3.5
Pseudomonasaeruginosa	N.A	20	5
Salmonella typhi	20	N.A	3.5

Table 10. Minimum inhibitory concentration (MIC) of fruit of *Duchesnea indica*

Test Organism	**MIC value of methanolic extract (µg/ml)**	**MIC value of ethanolic extract (µg/ml)**	**MIC value of standard levofloxin (µg/ml)**
Escherichia coli	10	N.A	5
Staphylococcus aureus	15	10	3.5
Pseudomonas aeruginosa	5	N.A	5
Salmonella typhi	20	15	3.5

Since many substances that are effective against diseases are found naturally in plants, using

them to treat illnesses is a traditional practice. Thus, plants are very important in medicine. The therapeutic value of the plant *Duchesnea indica*, sometimes referred to as the mock strawberry or wild strawberry, was covered in the current study. Different forms of traditional Chinese medicine have made use of this plant. It has depurative, febrifuge, anticoagulant, and antimicrobial properties. Additionally, boils and abscesses, ringworm, eczema, stomatitis, laryngitis, acute tonsillitis, snake and bug bites, and traumatic injuries are also treated with it. However, the significance of this study is limited to its antibacterial and antioxidant properties. According to this study, *Duchesnea indica* has a high content of phenolic compounds, indicating the plant's significance as an antioxidant source. For the analysis of phytochemicals 200 milliliters of water are used to make 5 grams of leaf and fruit extract. Numerous phytochemicals, such as alkaloids, carbohydrates, protein, and others, are present, according to the qualitative examination. The purpose of the quantitative study was to determine if 100g of dry extract included alkaloids (L=5.1 mg, F=4.8 mg), flavonoids (L=20.56 mg, F=42.8 mg), total phenols (L=84.4 mg, F=517 mg), tanins (L=26.56 mg, F=1.22 mg), and saponins (L=20.4 mg, F=0.78 mg). Crude extract was made from 10g of the sample in 100g of solvent (methanol and ethanol) for antibacterial and antioxidant examination. The final combination was examined using several antioxidant assays. The IC50 values for leaves (M.E= 46.2µg) and fruit (M.E= 38µg) and fruit (E.E= 58.3µg) in the DDPH photometric assay were found. Parallel to this, the ABTS assay's IC50 value was also noted and reported as (M.E=37.7µg), (E.E=64.9µg), and in fruit (M.E=38.5, E.E=37.2). The reducing power assay, the last test used to measure antioxidant activity, also demonstrates that plants are a strong source of antioxidants due to the fact that absorbance increases as sample concentration raises. The results of *Duchesnea indica*'s antibacterial activity showed that the plant also has a lot of promise for combating microorganisms. When leaves were tested against Escherichia coli, the greatest results were seen in a 10.33mm zone. With a minimum inhibitory concentration of 20µl of sample, activity against various microbiological agents was also noted, including a 7.16 mm zone against Staphylococcus aureus, a 9 mm zone against Pseudomonas aeruginosa, and an 8.8 mm zone against Salmonella typhi. With an inhibitory zone of 16.82 mm for Pseudomonas aeruginosa, 13.57 mm for Staphylococcus aureus, 11.63 mm for Salmonella typhi, and 11 mm for Escherichia coli, the

fruit sample exhibited the strongest antibacterial activity. According to this study, *Duchesnea indica* has significant medical value since it is a rich source of antioxidants. Further research on this topic can aid in the development of safer and more effective antioxidant and antibacterial chemicals.

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