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Proximate composition, minerals analysis and antibacterial potential of Syzygium cumini L. leaves, flower and bark extracts against foodborne pathogens

Javid Ali*¹, Arshad Hussain², Muhammad Siddique³, Inayat ur Rehman⁴, Anwar Zeb⁵

^{1,2}Microbiology and Mycotoxin Research Section, Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories Complex Jamrud Road, Peshawar, 25120, PAKISTAN

³Medicinal Botanic Centre, Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories Complex Jamrud Road, Peshawar, 25120, PAKISTAN ^{4,5}Environmetal Research Section, Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories Complex Jamrud Road, Peshawar, 25120, PAKISTAN

PAKISTAN

*Corresponding author's E-mail: javedali_14@yahoo.com, Tel. +92091-9221398-400, Fax: 091-9221395 ORCID iDs: http:/orcid.org/0000-0002-3530-6427 (Ali); http:/orcid.org/0000-0001-5501-8011 (Rehman)

Abstract

The current study was carried out to evaluate the proximate compositions, minerals assessment and antimicrobial activities of leaves, bark and flower of *Syzygium cumini L*. Proximate compositions were carried out using standard methods. The mineral analyses were assessed by Flame Photometer and Atomic Absorption Spectrophotometer. Antibacterial potential were assayed using Well-diffusion assay for three extracts (decoction, infusion and tincture). The proximate composition of *Syzygium cumini L*. leaves, flower and bark were found in the range i.e. moisture 3.5-05%, ash 05-07%, crude fat 0.4-2.5%, crude fiber 08-23%, protein 02-04%, carbohydrate 64.1-78.9% and energy 268-335Kcal/100 g dw. Similarly the minerals quantification (mg/Kg) range were calculated as Sodium 1300-1600, Potassium 3250-5000, Calcium 12500-24000, Magnesium 5000-9600, Zinc 30-60, Iron 140-500 and Manganese 30-100. The tincture observed the most efficient extract followed by infusion and decoction. While bark showed the least antibacterial potential against the pathogenic bacteria as compared to leaves and flower. The results commonly observed that generally the high opposing were observed by Gram-ve microbes to the *Syzygium cumini L*. extracts as compared to Gram-positive bacteria. These findings observed that *Syzygium cumini* leaves, flower and bark are a connection of health promoting major mineral elements and nutrition ingredients. Therefore, it can be a latent resource of natural minerals, fiber, protein and antibacterial agents in food, feed and medicine. This study exemplifies the latent of this plant as a substitute feed and food supplement to make stronger food security.

Keywords: Decoction, Gram-negative microbes, Gram-positive microbes, human health minerals, infusion, proximate composition, tincture.

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1. Introduction

Foodborne ailments have been observed as severe warning to the community health all over the globe for many decades (Mehdi *et al.*, 2014). The hazard of foodborne ailments could be minimizing by applying various easy safety measures like hygienic practices and avoiding cross contamination. One of the great reasons of various outbreaks is considered to be shortage of or

inadequate cleaning and disinfection of surfaces and equipment in food concerned environments. As results, various foodborne ailments could be stopped by marked disinfection in the mentioned field. Various kinds of disinfectants exist with diverse characteristics, the appropriate disinfectant should be choosing cautiously for precise utilization to gain the requisite antimicrobial result (Grinstead, 2009; Cogan *et al.*, 1999; Cogan *et al.*, 2002). There is augmented requirement for enhanced disinfection techniques, because of the coming out of microorganisms, opposing to various antibiotic compounds. As a result, latest technologies have been applied for competent microbial control and disinfection.

For accurate function and good health of the body, the roles of minerals are very vital (Ahmed *et al.*, 2020). Minerals play very critical function jobs in various biological functions as ingredient of enzymes, body fluid and skeleton (Sartaj *et al.*, 2013). The body requires six (06) nutrient for the overall health and proper functioning. These comprised minerals, vitamins, water, fats, protein and carbohydrates. In various budding countries, undernourishment and starvation are increasing due to soaring food costs, scarcity of productive soil and population explosion (Mohammad *et al.*, 2023).

Plants bioactive compounds have been of enormous attention to human being for a lengthy period because of its therapeutic relation. A huge number of global populations, particularly in the bidding countries depend on the conventional medicine system for numerous diseases. Herbal products are as well an origin of very powerful and potent drugs that have proved the test of period and modern chemistry have unable to discard much of these (Shylaja et al., 2011). Syzygium cumini L. generally recognized as "Jamun" is broadly dispersed in subtropical and tropical regions. It has a great value in Unani and Ayurveda medical system for holding various curative characteristic, which is broadly applied in traditional medicine for the handling of numerous ailments (Ravi et al., 2014). The earlier research recognized that the Jamun bark has possessed numerous characteristics such as antimicrobial, stomachic, constipation, febrifuge, anti-helminthic, digestive, diuretic, carminative, refrigerant and astringent activities (Saravanan and Pari, 2008). The diseases such as high blood sugar, pharyngitis, spleenopathy, urethrorrhea, and ringworm infection treated by using seeds and fruits of Syzygium cumini (Saravanan and Pari, 2008). The leaves are antimicrobial and exercised to make stronger the gums and teeth. The leaves have been also expansively utilized to cure to inhibit blood discharge in the feces, dermopathy, strangury, gastrpoathy, fever, stomachalgia, leucorrhoea, constipation and diabetes. The seeds, leaves and barks extracts of Syzygium cumini L. have been recognized to hold anti-diarrhea, antimicrobial and anti-inflammatory effects. The pulp, stem, bark and leaves possessed efficient anti diabetic activities (Md. Rashedul et al., 2012). The dietary importance could be evaluated by nutrient analysis and proximate composition testing of vegetables and fruits which act a critical role in evaluation their dietary meaning. The benefits of the medicinal plants might be assessed for their dietary importance, as various therapeutic based plants are utilized as food besides their therapeutic values. So keeping the medicinal and food values of Syzygium cumini L. flower, leaves and bark were assayed to quantify their proximate composition, elemental content and antimicrobial activities against the food spoilage contamination microbes of Gram-negative and gram-positive.

2. Material and Methods

2.1. Plant materials

Syzygium cumini L. leaves, flower and bark were obtained from Experimental Research Farm of Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories Complex Peshawar, Khyber Pakhtunkhwa-Pakistan. The plant was identified by a plant taxonomist Dr. Hina Fazal, Senior Scientific Officer, Head of Plant Taxonomy Section-PCSIR, Peshawar-Pakistan. The collected plant parts were washed carefully through tape H_2O to discard the mud, dust and any soil constituent part. The flower, bark and leaves were separated and kept for drying in a dimness room for ten days. The dried materials were grinded into powder form with the help of Waring® Commercial Laboratory Blender USA and store in an air tight glass bottle for further uses.

2.2 Proximate analysis

The moisture, ash, crude fat, crude fiber and crude protein of *Syzygium cumini L*. leaves, flower and bark were quantified according to the protocol (AOAC; 2006; Ali *et al.*, 2011). The moisture content was assayed using a technique of dehydration applying 100-110°C using Electric Oven (Memmert- Germany) till to receive stable weight. The difference between the initial weight and a final constant weight was the moisture value. The ash value was calculated using Electric Furnace (Modal JSMF-120 H, South Korea, Ambient-1100 °C) for five to six hour time to obtained the steady weight keeping the temperature 550 °C. Crude fat was estimated using Soxhlet apparatus (Quickfit, England) connected with Heating Mantle, Electrothermal (England) by petroleum ether extraction method for approximately six to eight hours. Crude fiber was quantified by alkali digestion (NaOH 1.25%) and acid digestion (H₂SO₄ 1.25%) technique. Crude protein was determined by Kjeldhal method using Micro Kjeldahl (Bloc Digest, Spain) and the value was obtained through multiply the Nitrogen (N) value by a 6.25 factor.

The Carbohydrates values were estimated using the following formula (Ulfat *et al.*, 2016) in the *Syzygium cumini L*. leaves, flower and bark powder.

Carbohydrate (%) = 100 - (moisture + ash + fiber + fats + protein).

Total energy was estimated using the bellow formula (Tania et al., 2017) in the Syzygium cumini L. leaves flower and bark powder;

Energy (kcal/100 g) = 4 (g proteins + g carbohydrates) + 9 (g fat).

2.3 Elemental Analysis

The plant material five gram (5g) was weigh in a crucible of silica, warm the samples in an Electric Oven (DAIHAN Scientific Co.

Ltd. South-Korea) at 110 °C to remove moisture and then the samples after charring was heated in a muffle furnace (Model; HD-230-Hobersal-Barcelona-Spain) till no smoke was produced at 400 °C. At room temperature the crucible was cooled in desiccator and the ash residue was wet with 0.5mL concentrated sulphuric acid. The crucible was kept on Hot Plate (PCSIR-Pakistan) and warmed continuously unless the sulphuric acid smoke was completely finished. The sulphated ash in the crucible was getting warmed in Muffle Furnace (Model; HD-230-Hobersal-Barcelona-Spain) with 600°C until a steady weight of the crucible material become achieved. The resulted ash was cool down; suspended in 5mL of 6N hydrochloric acid and then permitted to stand for thirty minutes. The mixture filtration was carried out using Whatman® No. 1 (England) and then 50mL deionized water was used to make up the volume. This final solution was applied to determine the elemental quantification through a Hitachi Zeeman Japan Z-8000, Atomic Absorption Spectrophotometer, which used air acetylene flames and radiation source as standard hallow cathode lamp. Whereas, Flame Photometer-Jenway, PFP7-UK (Ali *et al.*, 2016).

2.4 Crude extract preparation

Decoction was prepared by boiling finely five-gram (5) ground powder of flower, stem bark and leaves separately in 100 ml water (100°C) for 20 min. **Infusion** was made to the addition of water boiled to the five-gram herbal powder 5g/100 ml and left it standing for 20 min. **Tinctures** were synthesized in a closed flask by the addition of 10g Jamun powder to 200mL ethanol (96%) for seven days incubation at 37C with moving condition forty cycle per minute. After cooling, three preparations were vacuum filtered through Whatman® No. 1-England using a Buchner funnel, and filtrate was concentrated to dryness in Rotary-Evaporation Unit (Heating Bath B-490- Buchi with Rotavapor R-200- Switzerland) at 40°C, and was weighed. All the extracts were kept in a cooled incubator at 4°C waiting to utilization in the experiments.

2.5 Microorganisms used

Antibacterial activities of various solvent obtained from Jamun leaves, flower and bark were assayed against the Gram-positive bacteria i.e. *Staphylococcus aureus, Enterococcus faecalis, Bacillus cereus, Bacillus subtilis* and Gram-negative i.e. *Escherichia coli, Salmonella Typhimurium, Pseudomonas aeruginosa, Proteus mirabilis* and *Klebsiella pneumonia*. These bacteria were procured from Food Microbiology Laboratory of PCSIR Laboratories Complex Peshawar-Pakistan. The bacterial stains were maintained on Nutrient Agar slant and Nutrient Broth at 37°C. The slants were preserved in cooled incubator until use.

2.6 Well-diffusion assay

The antibacterial activity of Jamun parts extracts were calculated using the Agar well diffusion technique (Bashir and Javid 2013) with some modification. The Nutrient Agar 15mL were poured into sterilize Petri dishes and then mixed with 100μ L of bacterial population suspension having the microbial load 10^8 CFU/mL of tested bacterial strains. The inoculated media in the Petri dishes were then solidified and cool for thirty minutes. Aseptically drilling well of 6 mm diameter into the inoculated culture medium Petri dishes with help of sterile cork borer. Then add 100μ L (500mg/mL) dissolved in 10% Dimethyl sulfoxide (DMSO) of each extract (tincture, infusion and decoction) were added to the wells. Gentamycin (10mg/mL) was applied as positive reference control and 10% DMSO was a negative control for the bacterial activities. The plates were stand for two hour to permit diffusion at 04°C, and then incubated at 37°C for 24 hour. The antimicrobial activities were generally calculated inhibition zones around the bore wells. The zones inhibition diameters were calculated in millimeter (mm).

3. Statistical analysis

The obtained results were presented as means \pm standard deviations (SD) of three trials of experiments. The confidence level were adjust at *P* <0.05. The SD was not go beyond 5% for the maximum results find.

4. Results and Discussion

4.1. Proximate Composition

Nowadays the uses of therapeutic herbs and protection have held significant quantity of magnitude. It was applied world widely by the local and It was used globally by the marginal and indigenous societies to manage numerous ailments. These herbal plants are frequently utilized as nutraceutical supplements along with its decoction orally. But, slight have been carried out consequently far-flung to confirm application and utilization in this view. The current study was carried out in this regard.

The results of proximate composition of *Syzygium cumini L*. leaves, flower and bark are shown in Table 1. This finding revealed that proximate composition of *Syzygium cumini L*. revealed that the maximum moisture content was found $05 \pm 0.5\%$ in leaves followed by flower and bark. The maximum ash and fiber was noted in the bark $07\pm0.2\%$ and $23\pm02\%$ respectively. While crude fat and protein was increases in the order leaves>flower> bark. Carbohydrate and energy value are high in flower as compared to leaves and bark.

It was recognized that fungi and bacteria that give confidence to food contamination prosper good in foods which have maximum moisture quantity, thus minimizing the shelf life. Though, minimizing the water quantity in food might provide a maximum shelf life and as well easiness of shipping of Jamun samples (Aletan and Kwazo 2019).

The quantity of ash in any plant samples represented elemental fraction of the herbs. Its high quantity could be due the occurrence of various elements in maximum amount, therefore s in greater quantity, thus fulfill the requirement of the elemental nutrition in

the body (Sajjad *et al.*, 2014). The disadvantage of the utilization of vegetables in man diet is their maximum fiber quantity, which may cause irritation in intestine and minimizing diet bioavailability. The recommended dietary fiber allowance quantity for breast-feeding mothers, pregnant, adults and children are 29%, 28%, 21-38% and 19-25% respectively (Ali *et al.*, 2011). The maximum fiber quantification of the botanicals could be accountable of the occurrence of cellulose in the succulent branches or leaves. The grater fiber quantification is useful for fine absorption in the digestive canal way of the foodstuff materials (Sajjad *et al.*, 2014). Plants based amino acids and protein at a standstill remains consider a chief origin of diet nutritional ingredients in the developing countries (Aletan and Kwazo 2019). The *Syzygium cumini* stem, flower and leaves have found a good quantity of protein, which make it better fit to reflect it as better herbal amino acid supplement. As the *Syzygium cumini* leaves have good carbohydrates quantity, its utilization should gave energy and fuel to the body that is essential for exercise and daily activities (Aletan and Kwazo 2019). Sufficient carbohydrate is also essential for optimal function of the immune system, digestion, nervous, heart and brain while its shortage leads to weakening of the body tissues (Aletan and Kwazo 2019).

| Parameters | Leaves | Flower | Bark | | | |
|------------------------|---------|--------------|--------------|--|--|--|
| Moisture % | 05±0.5 | 4.5±0.1 | 3.5±0.2 | | | |
| Ash % | 06±01 | 05±0.3 | 07±0.2 | | | |
| Crude Fat % | 2.5±0.2 | 01±0.2 | $0.4{\pm}00$ | | | |
| Crude Fiber % | 11±0.4 | 08 ± 0.7 | 23±02 | | | |
| Protein % | 04±0.5 | 2.6±0.1 | 2.0±0.3 | | | |
| Carbohydrate % | 71.5±01 | 78.9±01 | 64.1±01 | | | |
| Energy (Kcal/100 g dw) | 324.5 | 335 | 268 | | | |
| | | | | | | |

Table 1. Proximate Composition of Syzygium cumini.

Each value is a mean of $03\pm$ SD

The Jamun seeds possessed 49.50% and 53.49% moisture, 0.62-0.79% fat, the Jamun pulp contained 85.94% and 90.95% moisture, 0.32-0.40% fat. The protein content of the pulp was 4.38% and for seed it was 6.57%. The ash content of seed was lower as compared to pulp (Ahmed *et al.*, 2020). In the current we used the bark, flower and leaves and the result showed that these parts could be a precious origin of nutritional fibre in human being nourishment. Because these plant parts are no commonly used.

4.2 Mineral Analysis

Table 2 shows the minerals composition of *Syzygium cumini L*. The maximum minerals quantity (mg/Kg) i.e. Sodium was found in the bark 1600 ± 12 , Potassium 5000 ± 10 , Calcium 24000 ± 20 , Magnesium 9600 ± 04 , Zinc 60 ± 01 , Iron 500 ± 04 and Manganese 100 ± 07 were found in leaves. These results observed that Na, K, Ca, Mg, Zn, Fe and Mn were calculated in high amount as recommend daily allowance (Bill, 1995).

| Elements | Leaves | Flower | Bark | RDA (Bill, 1995) | | |
|----------|----------|----------|----------------|------------------|---|--|
| | | | | Men | Women | |
| Na | 1500±05 | 1300±10 | 1600±12 | 500 mg | 500 mg | |
| Κ | 5000±10 | 3250±15 | 4850±10 | 2,000 mg | 2,000 mg | |
| Ca | 24000±20 | 12500±13 | 14000 ± 04 | 800 mg | 800 mg, for pregnant/nursing 1200 mg | |
| Mg | 9600±04 | 5000±14 | 5300±06 | 350 mg | 280 mg, 355 mg (nursing), 320 mg (pregnant) | |
| Zn | 60±01 | 30±01 | 40±04 | 15 mg | 12 mg, pregnant (15 mg), nursing (19 mg) | |
| Fe | 500±04 | 140±03 | 150±05 | 10 mg | 15 mg, for pregnant 30 mg | |
| Mn | 100±07 | 30±02 | 32±02 | 2.0-5.0 mg | 2.0-5.0 mg | |
| Cr | ND | ND | ND | 50-200 mcg | 50-200 mcg | |
| Cu | ND | ND | ND | 1.5-3.0 mg | 1.5-3.0 mg | |
| Pb | ND | ND | ND | - | - | |
| Ni | ND | ND | ND | - | - | |
| Cd | ND | ND | ND | - | - | |

Table 2. Minerals Analysis (mg/Kg) of Syzygium cumini.

ND= Not detected, Each values is a mean of $03\pm$ SD, RDA= Recommended Dietary Allowances.

The overall results showed that all the plant parts contained all minerals in appreciable amount. The higher Na, K, Ca and Mg quantification observed that *Syzygium cumini* may have useful source of these elements. The zinc, iron, and manganese appreciable amount calculated that the therapeutic potential of his plant might be also associated with its elemental ingredients. The Cu, Pb, Ni, Cd and Cr were not found in *Syzygium cumini*, it is very appreciable because generally these metals are toxic to human health in high concentration.

Minerals are non-organic compounds, occurred in all body fluids and tissues. Their possession is necessary to retain various chemicals and physical reactions which are vital for life (Aletan and Kwazo 2019). The K and Na neutralize acid base balance of the body fluid and osmotic pressure (Bashir and Ali 2013). Calcium is very important for milk production, blood clotting, bones, teath health, heart muscles and nerve impulses (Muhammad et al., 2013). The deficiencies of calcium have cause the osteoporosis in which the bones weaken and finally the bone fracture risk starts. If the phosphorus quantity in the diet is elevated than calcium, the cell will begin to get calcium from the bones to fulfill the deficiency. But a short period of time or over long time, it's very dangerous to the bones and creates negative effects on bones. It is consequently needed that a good quality origin of calcium is utilized to harmonize the leaves of Syzygium cumini in food (Aletan and Kwazo 2019). Zinc is necessary for brains health, behavioral development, normal insulin secretion, normal growth and wound healing (Muhammad et al., 2013). Further Zinc is generally concerned in regulation of body processes, cell shape synthesis and catalysis (Rose et al., 2023).

Manganese is a part of some enzyme and improves various enzyme efficiency (Naseem et al., 2012). Iron is piece of hemoglobin and acts a function in the biological degradation protein, carbohydrates and lipids. Its scarcity produces a anemia (Naseem et al., 2012). The high mineral quantification of Jamun skin and seed point out their implication to be utilized in animal feeds and food supplementations (Sartaj et al., 2013). The results showed (Sartaj et al., 2013) a wealthy elemental quantification Jamun study parts, but, elevated quantities were calculated in seed followed by skin and pulp. Along with diverse mineral elements K, P, Ca, Mg and Na followed by Fe were assayed in maximum quantities, whereas Na concentration was small as compared to other elements, therefore, Syzygium cumini is measured as a small Na fruit (Bhutani et al., 1989).

4.3 Antimicrobial assessment

The antibacterial activities of Syzygium cumini L. leaves, flower and bark decoction extracts are shown in Table 3.

| Bacteria | Zone of Inhibition | | | | | |
|------------------------|--------------------|----------|---------|------------|---------|--|
| Gram-positive | Leaves | Flower | Bark | Gentamycin | 10%DMSO | |
| Staphylococcus aureus | 19±0.5mm | 16±0.1mm | 12±01mm | 23±00mm | NZI | |
| Enterococcus faecalis | 18±0.3mm | 14±0.2mm | NZI | 25±00mm | NZI | |
| Bacillus cereus | 17±0.1mm | 16±0.1mm | 13±00mm | 27±01mm | NZI | |
| Bacillus subtilis | 16±0.2mm | 15±0.5mm | 12±00mm | 24±00mm | NZI | |
| Gram-negative | | | | | | |
| Escherichia coli | 13±0.2mm | 12±0.1mm | NZI | 22±00mm | NZI | |
| Salmonella Typhimurium | 12±0.1mm | 10±0.2mm | NZI | 20±00mm | NZI | |
| Pseudomonas aeruginosa | 11±0.2mm | NZI | NZI | 23±01mm | NZI | |
| Proteus mirabilis | 10±0.3mm | NZI | NZI | 25±00mm | NZI | |
| Klebsiella pneumoniae | 15±0.2mm | 10±00mm | NZI | 20±00mm | NZI | |

Table 3. Antibacterial Activities of Svzvgium cumini Decoction Extract.

NZI= No Zone of inhibition, Each value represents average of $03 \pm$ Standard Deviation.

The results observe that maximum zone of inhibition among plant parts is leaves decoction extracts against Staphylococcus aureus 19±0.5mm. While flower extract maximum zone of inhibition are calculated against Staphylococcus aureus and Bacillus cereus is 16±0.1mm. The bark extract shown weak antibacterial activities against tested Gram-Positive bacterial, however no zone of inhibition are recorded by bark extract against Gram-Negative. The antibacterial activities of Syzygium cumini L. leaves, flower and bark infusion extract is shown in Table 4.

| Bacteria | Zone of Inhibition | | | | | |
|------------------------|--------------------|----------|----------|------------|---------|--|
| Gram-positive | Leaves | Flower | Bark | Gentamycin | 10%DMSO | |
| Staphylococcus aureus | 21±0.5mm | 17±0.5mm | 11±0.2mm | 23±00mm | NZI | |
| Enterococcus faecalis | 20±0.2mm | 15±01mm | 10±00mm | 25±00mm | NZI | |
| Bacillus cereus | 19±0.4mm | 18±01mm | 12±00mm | 27±01mm | NZI | |
| Bacillus subtilis | 18±0.5mm | 16±02mm | 13±00mm | 24±00mm | NZI | |
| Gram-negative | | | | | | |
| Escherichia coli | 15±0.1mm | 12±0.4mm | NZI | 22±00mm | NZI | |
| Salmonella Typhimurium | 13±0.1mm | 11±00mm | NZI | 20±00mm | NZI | |
| Pseudomonas aeruginosa | 12±00mm | 13±00mm | NZI | 23±01mm | NZI | |
| Proteus mirabilis | 14±0.5mm | 11±00mm | NZI | 25±00mm | NZI | |
| Klebsiella pneumoniae | 16±01mm | 10±00mm | NZI | 20±00mm | NZI | |

NZI= No Zone of inhibition, Each value represents average of $03 \pm$ Standard Deviation.

The results shows that maximum activities (zone of inhibition) are depicted by leaves against *Staphylococcus aureus* 21 ± 0.5 mm, followed by 20 ± 0.2 mm, 19 ± 0.4 mm and 18 ± 0.5 mm against Enterococcus faecalis, Bacillus cereus and Bacillus subtilis respectively. The flower extract shows moderate activities and bark extracts (infusion) show the minimum zone of inhibition. It is also noted that again bark infusion extracts against Gram-Negative bacteria shows no zone of inhibition.

| Bacteria | Zone of Inhibition | | | | | |
|------------------------|--------------------|----------|---------|------------|---------|--|
| Gram-positive | Leaves | Flower | Bark | Gentamycin | 10%DMSO | |
| Staphylococcus aureus | 21±0.2mm | 17±0.1mm | 16±01mm | 23±00 | NZI | |
| Enterococcus faecalis | 22±0.2mm | 16±0.5mm | 14±01mm | 25±00 | NZI | |
| Bacillus cereus | 23±0.2mm | 18±01mm | 16±01mm | 27±01 | NZI | |
| Bacillus subtilis | 20±0.2mm | 16±01mm | 15±01mm | 24±00 | NZI | |
| Gram-negative | | | | | | |
| Escherichia coli | 15±0.2mm | 13±0.1mm | 11±00mm | 22±00 | NZI | |
| Salmonella Typhimurium | 13±01mm | 14±00mm | 10±00mm | 20±00 | NZI | |
| Pseudomonas aeruginosa | 12±0.5mm | 12±00mm | NZI | 23±01 | NZI | |
| Proteus mirabilis | 14±0.2mm | 11±00mm | 11±00mm | 25±00 | NZI | |
| Klebsiella pneumoniae | 16±01mm | 11±01mm | 10±00mm | 20±00 | NZI | |

Table 5. Antibacterial Activities of Syzygium cumini Tincture Extract.

NZI= No Zone of inhibition, Each value represents average of $03 \pm$ Standard Deviation.

The antibacterial activities of *Syzygium cumini* leaves, flower and bark tincture extracts are shown in Table 5. The highest inhibitory effect (zone of inhibition) of tincture leaves extract is shown against *Bacillus cereus* is 23 ± 0.2 mm, followed by 22 ± 0.2 mm, 21 ± 0.2 mm and 20 ± 0.2 mm against Enterococcus faecalis, Staphylococcus aureus and Bacillus subtilis respectively. The reference antibiotics have found the highest zone of inhibition against all the tested bacteria as compared it with the plant extracts. It is because that these antibiotics have prepared using state of the art latest techniques, while the plant extracts have obtained in crude shape using the ordinary laboratory condition. It is generally consider that these antibiotics have very costly, side effects, create microbial resistance and not easily approachable for the poor and remote areas of the developing countries.

Generally, the *Syzygium cumini* is effective against Gram +ve and Gram –ve bacteria and more effective against Gram +ve organisms when compared to Gram –ve. The antibacterial activity increases by extraction techniques in the order tincture> infusion> decoction. While in parts point of view increases leaves>flower> bark.

The variation in the antimicrobial assayed of plant extracts from the similar origin when obtained with various solvents have confirmed that not every one phyto-compounds that are accountable for antibiotic potential are dissolve in a sole solvent. Therefore solvents of various polarity could be applied i.e. non-polar such as petroleum ether, ethyl acetate; polar such as ethanol, acetone and water. Successive and sequential solvent soaking and pulling process is a better choice for good solubility of various bioactive compounds of herbs. But, it is as well essential to identify the bioactive compounds, isolated by every representative solvent as a result to keep away from the addition of needless solvents through the isolation procedures. This is further vital to know the character of every solvent in the soaking and drawing out of a group and individual phytochemicals (Ali *et al.*, 2016).

In actuality, the activities of plants depend on their phytochemicals. While the various condition and ecosystems act a significant role in the biological production of phytochemicals, so the phytochemicals are varied. Ecosystems factors are consequently vital in the synthesis of therapeutic compounds of the herbal medicines. Factors like altitude, light intensity, humidity and temperature, which predict the weather of the location, influence the accretion the phytochemicals (Davise, 1994). It has been recognized that the variation degree of active compounds in herbal medicine are influence by weather. Though, the distribution and accumulation of phytochemicals is not uniform and the dissimilarities in the study's results might be due to the diversity in weather influence of various disparities of plants (Koohsari *et al.*, 2015).

In outlook of the findings acquired (Firas, 2009), methanol was observed only efficient against Gram +ve microbes. It has regularly been recognized that Gram +ve microbes are more vulnerable to essential oils as compared to Gram –ve microbes. The acceptance of gram –ve microbes to essential oils has been attributed to the occurrence of hydrophilic outer membrane which chunks the diffusion of hydrophobic essential oils into marked cell membrane (Ghalbane, 2011). Additionally, numerous phenomena and means of antibiotic opposing are gladly extend to a diversity of microbial genera. Initially, the microbes may obtain genes translating enzymes, like as β -lactamases, which demolish the antimicrobial compound prior to it could have an act. Further, microbes may obtain efflux pumps which extrude the antimicrobial compounds from the cell prior to it could arrive to its object location and apply its outcome. Lastly, microbes might attain various genes for a metabolic pathway which eventually synthesizes distorted microbial cell walls that have no longer possessed the attaching spot of the antibacterial compounds or microbes might obtain mutations that minimize entrance of the antibacterial compounds to the intracellular marked location through down-regulation of porin genes (Tenover, 2006).

In reality, Gram-Positive microbes are extra susceptible to plant extracts as compared to Gram-Negative microbes. This might be due to the composition, nature of plants and inherent tolerance of Gram-Negative microbes. According to the findings, the cell wall of Gram-Positive microbes contrast with Gram-Negative microbes, are additional susceptible to various antimicrobial chemical substances (Kittika *et al.*, 2007) and yet numerous botanical medicine (Sharifa *et al.*, 2008). Periplasmic space and

lipopolysaccharides layer of Gram-Negative microbes are background of the relative opposing of Gram-Negative microbes (Koohsari *et al.*, 2015). It was recognized that Gram +ve microbes are more susceptible to herbs extracts and oil as compared with Gram -ve microbes (Sandasi *et al.*, 2010; Mahboubi and Haghi 2008; Mimica-Dukic *et al.*, 1999; Cosentino *et al.*, 1999; Karaman *et al.*, 2003; Sahin *et al.*, 2002). The *Syzygium cumini L*. extracts were most effectual against Gram +ve bacteria than Gram –ve bacteria (Shylaja *et al.*, 2011). The plants extracts data regarding MBC and MIC observed maximum antimicrobial potential against Gram +ve microbes as compared to Gram –ve microbes. This divergence could be explained towards the bigger complication nature the cell wall of Gram-negative (Italo *et al.*, 2017). The differences level of sensitivity of the microbial strains might be because of the combination of phytochemicals possessed in the extracts, nature of the microbes and intrinsic tolerance of the microbes (Basheer and Abdullah 2013).

The extract of *Syzygium cumini* holds wealthy accessibility of bioactive compounds like flavonoids, tannins, phenols and carbohydrates (Shylaja *et al.*, 2011). The water extracts of seeds and alcoholic extracts of leaves were observed to hold very efficient antibiotic potential against a broad level of Gram-negative and Gram-positive microbes (Shylaja *et al.*, 2011). The Syzygium cumini leaves water-alcoholic extracts possessed phenolic ingredients and tannins which are accountable for the antibacterial activities. It has a capacity against multi opposing and reference bacteria such as *Pseudomonas aeruginosa* and *Staphylococcus aureus* might be investigated in order to expand a theme antibacterial agent to enhance wound skin therapeutic (Mayuri *et al.*, 2019).

The Syzygium cumini is recognized to be extremely wealthy in ellagic and Gallic acid. The antimicrobial activities against *S. aureus*, *P. aeruginosa*, *B. subtilis* and *E. coli* and inhibition activity on glucoamylase of ethanol extracts separated at various temperature dorm seeds of Jamun was examined. The leaves ethanol extracts and seeds water extracts were reported very efficient antibacterial activities for a broad spectrum of Gram-Positive and Gram-Negative microbes (Amol *et al.*, 2017). The Jamun seeds, bark and leaves are high-quality origin of tannins, oxalic and malic acid which are accountable for antimicrobial properties (Amol *et al.*, 2017). The present study gives scientific data on the disparity and comparison of the antimicrobial activities, nutritional ingredients and minerals composition of the Syzygium cumini parts utilized for the industries to occupy the greatest approach in attain marketable products. The current study limitation includes low sample size, only three extraction techniques; well diffusion antimicrobial assay used and plant parts.

5. Conclusion

The present study results revealed that *Syzygium cumini* have a fine origin of minerals and dietary composition such as fiber, carbohydrate, fats and proteins and could be utilized as compounds scarcity in any of these nutrients. Whereas the antimicrobial activities outcomes could be pointed out that its extracts might be utilized in food as organic preservatives opposing the recognized contributory microbes of food spoilage and food borne ailments. As an outcome, herbal origin natural products act a significant role in curing the ailments and in taking into account the augmented pathogenic microbial antibiotic resistance, explore for antimicrobial medicine being of highest significance.

It is recommended and way forward that additional research of regarding mechanism, metabolism and another solvent extractive system of the active substances derived from this plant may go ahead to novel and additional effectual product and drugs formulation for the indigenous manufacturing units.

So, we believe that different parts of Jamun tree should be extracts and isolated substances could be utilized for small and medium enterprises and finally on large scale in the food and feed industries to improve livelihoods and address food security. Future research should focus on the purification and isolation of substances from leaves, flower and bark to treat different ailments. Additional prominently, medical trials are necessary to build up inexpensive drug with a small therapeutic index.

References

- Ahmed, S., Ahmed, K.S., Hossain, M.S., Azam, M.S., Rahman, M. and Hoque, M.M. 2020. Proximate composition and antioxidant activity of *Syzygium cumini* fruit grown at different regions in Bangladesh. *Food Research*. Vol 4, No. 5, pp. 1693 -1699
- Aletan UI., Kwazo HA. 2019. Analysis of the proximate composition, anti-nutrients and mineral content of *Maerua Crassifolia* Leaves. *Nigerian Journal of Basic and Applied Science*. Vol 27, No. 1, pp. 89-96
- Amol CD., Komal NP., Ashok DT., and Rajkumar RA. 2017. Jamun (Syzygium cumini) Skeels: A traditional therapeutic tree and its processed food products. Int. J. Pure App. Biosci. Vol 5, No. 5, pp.1202-1209. https://doi.org/10.18782/2320-7051.4011.
- Association of Official Analytical Chemist (AOAC), 2006. Official Methods of Analysis, 18th ed. Association of Official Analytical Chemists, Arlington, VA, USA
- Ali A. 2011. Determination and comparison of potential nutritive values and mineral elements of three important food edible plants from southern part of Iran. *Croatian Journal of Food Technology, Biotechnology and Nutrition*. Vol 6, No.3-4, pp. 148-151.
- Ali J., Abbas S., Khan FA., Rehman SU., Shah J., Rahman ZU., Rahman IU., Paracha GMU., Khan MA., Shahid M. 2016. Biochemical and antimicrobial properties of citrus peel waste. *Pharmacologyonline*. Vol 3, pp. 98-103
- Basheer AA., Abdullah AA. 2013. Antimicrobial activity of the aqueous extract of mint plant. Science Journal of Clinical Medicine. Vol 2, No. 3, pp. 110-113.

- Bashir A., Javid A. 2013. Physiochemical, minerals, phytochemical contents, antimicrobial activities evaluation and fourier transform infrared (FTIR) analysis of *Hippophae rhamnoides L*. leaves extracts. *African Journal of Pharmacy and Pharmacology*. Vol 7, No. 7, pp. 375-388. https://doi.org/10.5897/AJPP12.1246.
- Bhutani VP., Taoshi VK., Chapra SK. 1989. Mineral composition of experimental fruitwines. Food Sci. Technol. Vol 26, pp.332.
- Bill Gottlieb. New Choices in Natural Healing. Rodale Press, Inc. St. Martin's Press in Emmaus, Pennsylvania, (New York), 1995: pp. 146-149.
- Cogan TA., Bloomfield SF., Humphrey TJ. 1999. The effectiveness of hygiene procedures for prevention of cross-contamination from chicken carcases in the domestic kitchen. *Letter of Applied Microbiology*. Vol 29, No. 5, pp. 354–8. https://doi.org/10.1046/j.1472-765x.1999.00656.x.
- Cogan TA., Slader J., Bloomfield SF., Humphrey TJ. 2002. Achieving hygiene in the domestic kitchen: the effectiveness of commonly used cleaning procedures. *Journal of Applied Microbiology*. Vol 92, No. 5, pp. 885–92. https://doi.org/10.1046/j.1365-2672.2002.01598.x
- Cosentino S., Tubereso CIG., Pisano B., Satta M., Mascia V., Arzedi EPF. 1999. In vitro antimicrobial activity and chemical composition of Sardinian Thymus essential oils. *Letters in Applied Microbiology*. Vol 29, pp. 130-135.
- Davise FS, Albrigo LG, Citrus AB. Citrus. CAB, Wallington: International Press; 1994. p. 9814.
- Firas AA. 2009. Isolation and identification of antimicrobial compound from *Mentha longifolia* L. leaves grown wild in Iraq. *Annals of Clinical Microbiology and Antimicrobials*. Vol 8, pp. 20-25
- Ghalbane, I., Belaqziz, R., Ait, S.L., Oufdou, K., Romane, A., El Messoussi S. 2011. Chemical composition, antibacterial and antioxidant activities of the essential oils from *Thymus satureioides* and *Thymus pallidus*. *Natural Product Communications*. Vol 6, No. 10, pp. 1507 – 1510
- Grinstead D. 2009. Cleaning and sanitation in food processing environments for the prevention of biofilm formation and biofilm removal. In: Fratamico PM, Annous BA, Gunther NW, editors. Biofilms in the Food and Beverage Industries. Oxford: Woodhead Publishing.
- Italo DRDA., Nayara CDA., Andreza CVDAG., Renato FDAJ., Renata MA., Raimundo FDAJ., Kleber JSF., Jose VF., Vania SA. 2017. Chemical composition and evaluation of the antibacterial and Cytotoxic activities of the essential oil from the leaves of Myracrodruon urundeuva. BMC Complementary and Alternative Medicine, Vol 17, pp. 419-429.
- Karaman I., Sahin F., Gulluce M., Ogutcu H., Sengul M., Addiguzel A. 2003. Antimicrobial activity of aqueous and methanol extracts of *Juniperus oxycedrus* L. *Journal of Ethnopharmacology*. Vol 85, pp. 231-235.
- Kittika N., Natta L., Orapian K. 2007. Antibacterial Effect of Five Zingiberaceae essential oils. *Molecules*. Vol 12, No. 8, pp. 2047–2060. https://doi.org/10.3390/12082047
- Koohsari H., Ghaemi EA., Sadegh SM., Jahedi MM., Zahiri MM. 2015. The investigation of antibacterial activity of selected native plants from North of Iran. *Journal of Medicine and Life*. Vol 8, No. 2, pp. 38–42. Corpus ID: 11806453.
- Mahboubi M., Haghi, G. 2008. Antimicrobial activity and chemical composition of *Mentha pulegium* L. essential oil. *Journal of Ethnopharmacology*. Vol 119, pp. 325-327.
- Mayuri AP., Nibha DB., Dipalini DW. 2019. Role of Syzegium cumini (Jamun) in cosmetic. International Journal of Scientific Development and Research. Vol. 4, No. 6. pp.193-201
- Md. Rashedul, A., Akib BR., Md. Moniruzzaman, Mohammad FK., Md. Ahsanul, Mohammad H., Md. Ratan RA. 2012. "Evaluation of antidiabetic phytochemicals in *Syzygium cumini* (L.) Skeels (Family: Myrtaceae). *Journal of Applied Pharmaceutical Sciences*. Vol 2, No. 10, pp. 094-098. https://doi.org/10.7324/JAPS.2012.21019
- Mimica-Dukic N., Popovic M., Jakovljevic V., Szabo A., Gasic O. 1999. Pharmacological studies of *Mentha longifolia* phenolic extracts II. Hepatoprotective activity. *Pharmaceutical Biology*. Vol 37, pp. 221-224.
- Mehdi Z., Amirhesam J., Elahe K. 2014. Antibacterial effect of silver nanoparticles against four foodborne pathogens. Jundishapur Journal of Microbiology. Vol 7, No. 1, pp. e8720. https://doi.org/10.5812/jjm.8720.
- Mohammad P., Hussain F., Ahmad B., Ali J. 2013. Euphorbia granulata forssk as a source of mineral supplement. American-Eurasian Journal of Agriculture and Environmental Sciences. Vol 13, No. 8, pp. 1108-1113. https://doi.org/10.5829/idosi.aejaes.2013.13.08.11017
- Mohammad S., Rakesh KS., and Sumitha T. 2023. "Proximate Composition and Nutritional Values of Selected Wild Plants of the United Arab Emirates". *Molecules* Vol 28, No. 3, pp. 1504. https://doi.org/10.3390/molecules28031504.
- Naseem U., Ali J., Khan FA., Khurram M., Hussain A., Rahman I., Rahman Z., Shafqatullah. 2012. Proximate composition, minerals content, antibacterial and antifungal activity evaluation of pomegranate (*Punica granatum L.*) peels powder. *Middle-East Journal of Scientific Research*. Vol 11, No.3, 396-401.
- Ravi SBE., Raman SI., Sundara RS., Richard SA., Bhadrapura LD. 2014. Antioxidant activity of *Syzygium cumini* leaf gall extracts. *BioImpacts*, Vol 4, No. 2, pp. 101-107. https://doi.org/10.5681/bi.2014.018
- Rose C., John MO., Lydia WN., and Vincent OM., 2023. Physicochemical and nutritional properties of *Syzygium cumini* (L.) skeels fruits grown in varied microclimates in Kenya. *African Journal of Pure and Applied Chemistry*. Vol 17, No. 1, pp. 1-9. https://doi.org/10.5897/AJPAC2022.0896
- Sahin F,. Karaman I., Gulluce M., Ogutcu H., Sengul M., Adiguzel A. 2002. Evaluation of antimicrobial activities of Satureja hortensis L. Journal of Ethnopharmacology, Vol 87, pp. 61-65

- Sajjad Z., Inam UK., Habib U., Safdar S., Zahid H., Shahida B., Alam Z., Abdul M. 2014. Mineral profile of different medicinal plants and their quantitative analysis collected from north west of Pakistan. *Pakistan Journal of Weed Science Research* Vol 20, No. 2, pp. 145-154.
- Sartaj A, Tariq M., Kashif SA., Amjed A., Azhar H., 2013. Some compositional and biochemical attributes of jaman fruit (*Syzygium cumini* L.) from Potowar region of Pakistan. *Research in Pharmacy*, Vol 3, No. 5, pp. 01-09.
- Saravanan G., Pari L. 2008. Hypoglycaemic and Antihyperglycaemic Effect of Syzygium cumini Bark in Streptozotocin-Induced Diabetic Rats. Journal of Pharmacology and Toxicology. Vol 3, pp. 1-10. https://doi.org/10.3923/jpt.2008.1.10
- Sharifa AA., Neoh YL., Iswadi MI., Khairul O., Abdul HM., Jamalubin M., Mohamed AAB., Hing HL. 2008. Effects of methanol, ethanol and aqueous extract of *Plantago majoron* gram positive bacteria, gram negative bacteria and yeast. *Annals of Microscopy*. Vol 8, pp. 42–44. Corpus ID:101588943.
- Sandasi M., Leonard CM., Van SFV., Viljoen AM. 2011. Peppermint (*Mentha piperita*) inhibits microbial biofilms in vitro. South African Journal of Botany. Vol 77, No. 1, pp. 80-85. https://doi.org/10.1016/j.sajb.2010.05.011.
- Shylaja P., Gothandam KM., Karthikeyan S. 2011. Phytochemical and antimicrobial properties of *Syzygium cumini* an ethanomedicinal plant of Javadhu hills. *Research in Pharmacy*, Vol 1, No. 1, pp. 22-32. Corpus ID: 85769122.
- Tania, C.S.P.P., Maria, I.D., Lillian, B., Isabel, C.F.R.F. 2017. Nutritional and chemical characterization of edible petals and corresponding infusions: Valorization as new food ingredients. *Food Chemistry*. Vol 220, pp. 337–343.
- Tenover F. 2006. Mechanisms of antimicrobial resistance in bacteria. Amer J Med. Vol 119, pp. S3-S10
- Ulfat S., Muhammad I., Fazal H. 2016. Nutritional analysis of some selected fodder plants of Darazinda area, F.R. D.I. Khan, Subtropical Range, Pakistan. *American-Eurasian Journal of Agriculture and Environmental Sciences*. Vol 16, No.3, pp. 543-549.

Biographical notes

Dr. Javid Ali, Senior Scientific Officer (SSO) currently working as Section In-charge of Microbiology and Myctoxin Research Section at Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories Complex-Peshawar-Khyber Pakhtunkhwa-Pakistan.

Dr. Arshad Hussain, Principal Scientific Officer (PSO) working as Head of Food Technology Center at Pakistan Council of Scientific and industrial Research (PCSIR) Laboratories Complex-Peshawar-Khyber Pakhtunkhwa-Pakistan.

Mr. Muhammad Siddique, Principal Scientific Officer (PSO) working as Officer In-charge of Pharmaceutical Pilot Plant at Pakistan Council of Scientific and industrial Research (PCSIR) Laboratories Complex-Peshawar-Khyber Pakhtunkhwa-Pakistan.

Dr. Inayat ur Rehman, Senior Scientific Officer (SSO) currently working as Section In-charge of Environmental Research Section at Pakistan Council of Scientific and Industrial Research (PCSIR) Laboratories complex-Peshawar-Khyber Pakhtunkhwa-Pakistan.

Mr. Anwar Zeb, working as Technician in Environmental Research Section at Pakistan Council of Scientific and industrial Research (PCSIR) Laboratories Complex-Peshawar-Khyber Pakhtunkhwa-Pakistan.