

Article

Comparative Insights into the Antimicrobial, Antioxidant, and Nutritional Potential of the *Solanum nigrum* Complex

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Highlights:

- (1) Against the seven bacteria tested, the five taxa studied had varying antibacterial activity.
- (2) Plant extracts were discovered to be as efficient as antibiotics in certain circumstances.
- (3) All plants had moderate DPPH free radical scavenging activity, but *S. chenopodioides* performed the best.
- (4) In the ABTS experiment, *S. retroflexum* had the highest Trolox equivalent antioxidant capacity, with a TEAC of 33.88 mM/100 g of dry weight.
- (5) The therapeutic potential of specific plants may therefore be used to evaluate their functioning and chemical makeup.



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Abstract: *Solanum nigrum* is a traditional medicinal plant renowned as a cure for many diseases due to the presence of bioactive compounds. The *Solanum nigrum* complex refers to a group of more than 30 closely related but morphologically distinct taxa. Five indigenous taxa of this complex were investigated for their medicinal potential by using methanolic extracts. The efficacy of each plant was different for each of the seven bacteria studied. On comparing the MIC values, *S. americanum* was found to be most potent against *Bacillus licheniformis* (34 µg/mL), *S. chenopodioides* against *Escherichia coli* (78 µg/mL), *S. nigrum* against *Bacillus licheniformis* (49 µg/mL) and *Escherichia coli* (49 µg/mL), *S. retroflexum* against *Escherichia coli* (30 µg/mL), and *S. villosum* against *Proteus mirabilis* (45 µg/mL). The extracts were also subjected to six antioxidant assays. Moderate scavenging activity was observed

by all plants in the DPPH free radical assay, but *S. chenopodioides* was the most effective. The total phenolic contents of the five plants were comparable, but the gallic acid equivalents of *S. americanum* and *S. nigrum* were the highest (26.58 mg/100 g GAE). The highest Trolox equivalent antioxidant capacity was observed for *S. retroflexum*, with the ABTS assay giving a TEAC value of 33.88 mM/100 g of dry weight. Metal-chelating activity against Fe^{2+} was observed to be highest for *S. chenopodioides* (70.37%). The FRAP value of *S. nigrum* was the highest (8.5 mM $FeSO_4 \cdot 7H_2O$) among all taxa. The lipid peroxidation trend was very similar for all five samples. The results suggest the specified medicinal use of different members of the *Solanum nigrum* complex, which will also have significant nutritional value.

Keywords: *Solanum nigrum* complex; herbal medicine; methanol extract; antibacterial potential; antioxidant potential; nutritional value

1. Introduction

Nightshades are a variety of plant species that belong to the genus *Solanum*, including many morphological variants collectively known as the *Solanum nigrum* complex [1,2]. Being part of the family Solanaceae, all plants of the *S. nigrum* complex are famous for their historical use in herbal medicine. Traditionally, they have been applied for the treatment of many ailments, including dysentery, joint and muscle pains, ulcers, infections, fevers, and eye infections, and are reported to have laxative, diuretic, sedative, emollient, and tonic effects [3–6]. These medicinal properties are attributed to the presence of significant concentrations of secondary metabolites, including alkaloids, flavonoids, carotenoids, saponins, and triterpenes [7–10].

S. villosum Mill., *S. retroflexum* Dunal, *S. nigrum* L., *S. chenopodioides* Lam., and *S. americanum* Mill. were the five indigenous taxa of the *S. nigrum* complex selected for study. The pharmacologically active compounds reported in the investigated plants included α -solanine, α -solamargine, β -solamargine, solasonine, solanidine, solasodine, squalene, phytol, and glycosides of quercetin [10–13]. *S. nigrum* is a commonly occurring weed, but is also used as a food in the form of cooked vegetables or soup. The presence of polyphenols, polysaccharides, glycoproteins, and glycoalkaloids makes the plant active against various ailments, although glycoalkaloids can be toxic when taken in large amounts [14].

Studies have shown that black nightshades have in vitro anticholinesterase, anti-monoamine-oxidase, and antioxidant characteristics that enhance their importance against neurodegenerative diseases [14]. The fruit juice of ripe berries of *S. nigrum* has been reported to be active against both Gram-positive and Gram-negative bacteria, due to the presence of active phytochemicals [15]. Reports in the literature have been given on the antibacterial and antioxidant activities of *S. nigrum* L, but the medicinal value of individual morphological and chemical variants has not been explored. The authors have previously worked on the chemotaxonomy of the *Solanum nigrum* complex [12,13]. The present study includes the intensive in vitro study of five indigenous plants of the *S. nigrum* complex, focusing on the antibacterial, antioxidant, and nutritional potential of the plant.

2. Experimental Section

Five plant samples were collected from the Botanical Garden of GC University Lahore, Pakistan, and were identified by a taxonomist (Supplementary Materials, Table S1). The dried powder of leaves of the five taxa was extracted separately using methanol. The desolvated sample extracts were used to prepare 3 concentrations (1000, 2000, and 3000 $\mu\text{g}/\text{mL}$) in distilled water. Seven microorganisms (*Bacillus subtilis* PCSIR-B-248, *Bacillus licheniformis* PCSIR-B-252, *Escherichia coli* PCSIR-B-67, *Salmonella typhimurium* ATCC-14028, *Nocardia asteroides* PCSIR-B-178, *Micrococcus luteus* NRRL-B-287, and *Proteus mirabilis* ATCC-29245) were used to test the antibacterial activity of three concentrations of these five extracts via the method of Negi et al. [16]. All of the bacterial strains were cultured at 37 °C in

nutrient agar by taking the preculture from stock slants stored in the refrigerator at 4 °C. The bacterial culture (1 mL) was added to nutrient broth (9 mL) and incubated at 37 °C in a shaker to prepare the inoculum. Nutrient agar (20 mL) was allowed to settle in Petri dishes, and inoculum (0.5 mL) was added. The samples, along with three standard positive controls (benzyl penicillin, ampicillin, and streptomycin) and pure solvent as a negative control, were added to Petri dishes and incubated at 37 °C for 24 h. Readings for the zone of inhibition (mm) were taken in triplicate, and the minimum inhibitory concentrations (MICs) of the plant extracts were calculated by regression analysis. The antioxidant potential of the extracts was evaluated by applying reported protocols of the total phenolic content assay [17], lipid peroxidation assay [18], FRAP assay [19], ABTS assay [20], DPPH radical scavenging activity [21], and ferrozine metal-chelation assay [22]. Proximate analysis of the plant material—including moisture, ash, fat, nitrogen and crude protein [23], crude fiber [24], carbohydrate [25], nitrogen-free extract [26], and trace elements [27] in the five taxa—was carried out according to the standard procedures.

3. Results and Discussion

3.1. Antibacterial Activity

The antibacterial activity of the five morphologically different variants of the *S. nigrum* complex was evaluated against three Gram-negative and four Gram-positive bacteria (Table S2). Among these five taxa, *S. americanum* was found to be the most active at all concentrations, since it provided the greatest inhibition against three bacteria: *B. licheniformis* (a human pathogen causing infections and food poisoning, especially in immunocompromised patients [28]), *E. coli* (the cause most of the infections in the digestive and urinary tracts of humans and animals [29]), and *M. luteus* (reported as a cause of meningitis in humans [30]). This suggests that *S. americanum* can be used for the treatment of several human diseases and infections. All taxa, except *S. americanum*, were moderately active against *P. mirabilis*, which is usually responsible for pneumonia, septicemia, urinary tract infections, and wound infections in humans [31]. *S. nigrum* and *S. retroflexum* can be effectively used to treat typhoid fever, since they demonstrated significant activity against *S. typhimurium* [32]. *S. villosum* was most potent against *B. subtilis*, which is a food contaminant [33], and *N. asteroides*, which results in pulmonary and cutaneous nocardiosis [34]. The variation in the results shows that each taxon has its own bioactive contents, and should be used for specific infections.

The presence of alkaloids and flavonoids in *Solanum* plants plays a significant role in their use as herbal medicines [35,36]. The minimum inhibitory concentrations (MICs) of the plant extracts (Table 1) showed that *S. nigrum* and *S. retroflexum* had the lowest MIC values against *B. subtilis*, *M. luteus*, and *E. coli*. It was observed that the MIC of *S. retroflexum* against *E. coli* (30 µg/mL) was even lower than that of benzyl penicillin (32 µg/mL). Another significant result was the MIC of *S. villosum* against *P. mirabilis* (44 µg/mL), which was close to that of the standard benzyl penicillin (33 µg/mL).

Table 1. MIC values of different taxa of the *S. nigrum* complex for the analyzed bacteria.

MIC for Bacterial Strains (µg/mL)	Plant Taxa/ Drug (Code) ^a							
	SA	SC	SN	SR	SV	Amp	BP	Str
<i>B. subtilis</i>	113	560	78	74	137	8	8	4
<i>B. licheniformis</i>	34	184	49	49	84	1	4	2
<i>E. coli</i>	135	78	48	30	53	8	32	2
<i>M. luteus</i>	225	178	105	95	141	28	40	16
<i>N. asteroides</i>	85	170	195	127	116	32	18	8
<i>P. mirabilis</i>	118	382	66	53	44	2	32	8
<i>S. typhimurium</i>	77	185	96	100	111	7	16	2

^a Abbreviation of plant taxa/drug: SA: *S. americanum*; SC: *S. chenopodioides*; SN: *S. nigrum*; SR: *S. retroflexum*; SV: *S. villosum*; Amp: ampicillin; BP: benzyl penicillin; Str: streptomycin.

3.2. Antioxidant Activity

S. nigrum L. has been cited as having effective antioxidant properties [37–39]; however, the literature is silent on the antioxidant activities of the remaining four taxa under study. Hence, comprehensive research was conducted to determine and compare the antioxidant potential of these five taxa of the *S. nigrum* complex via six assays (Figure 1). Total phenolic content (TPC) is an indicator of the antioxidant potency of plants [40], and is the reason for peroxidation inhibition against the linoleic acid peroxidation system [41]. A slight variation was observed in the TPC values (in the range of 20.30–26.47 mg GAE/100 mg of dry weight) of the five taxa. The TPC of both *S. americanum* and *S. nigrum* was the highest amongst the taxa. Considerable antioxidant activity in the peroxidation system of linoleic acid was shown by all five taxa, and was comparable to that of Trolox as the positive control.

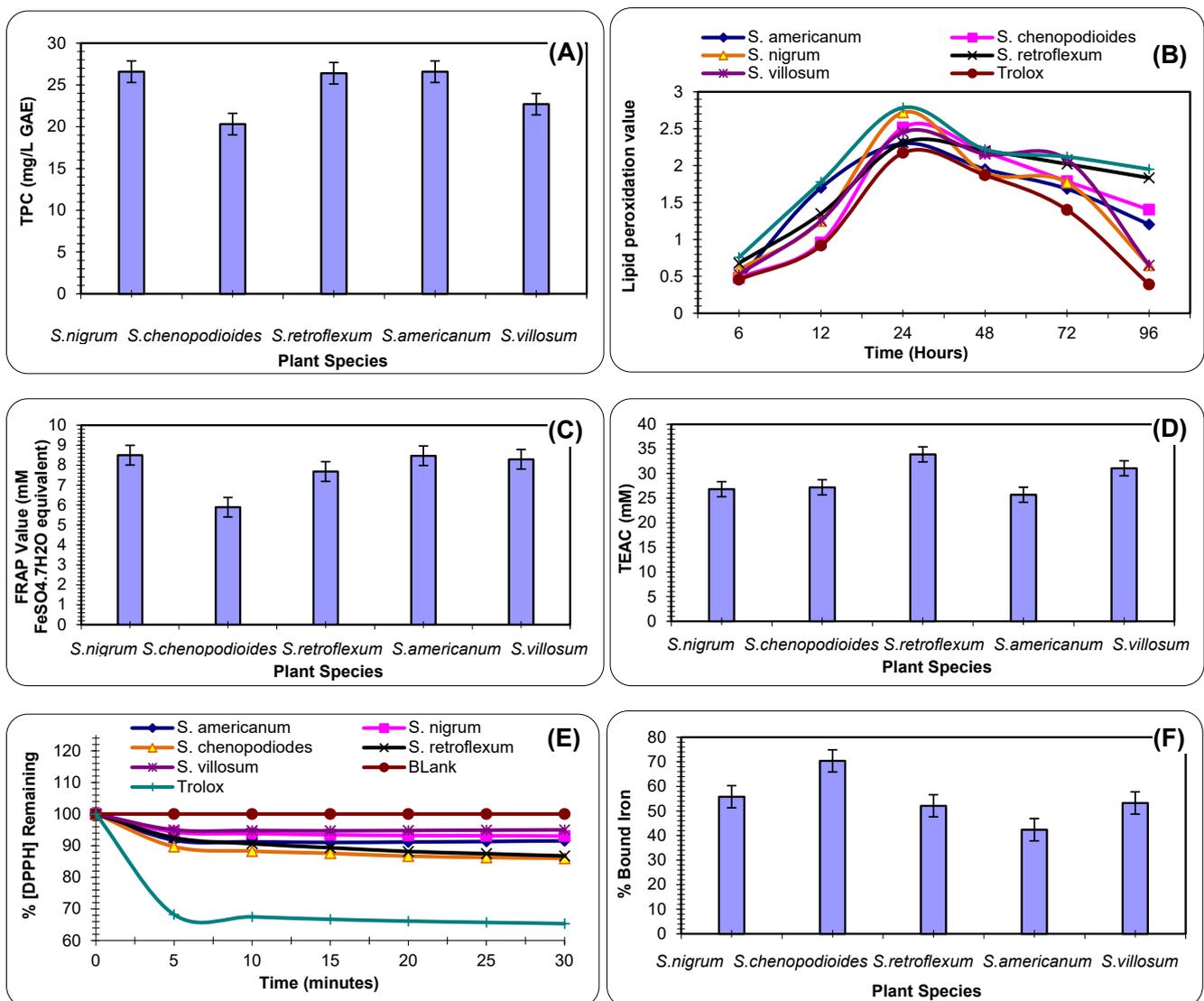


Figure 1. Antioxidant potential of five taxa of the *S. nigrum* complex: (A) total phenolic contents; (B) lipid peroxidation assay; (C) FRAP assay; (D) ABTS assay; (E) DPPH assay; (F) metal-chelating assay.

FRAP values depend on the presence of antioxidant secondary metabolites that vary in different plant species [42]. The investigated samples had FRAP values of 5.78 to 8.45 mM FeSO₄·7H₂O/mL of extract, with the observation that *S. americanum*, *S. nigrum*, and *S. villosum* were relatively more active. The Trolox equivalent antioxidant capacity (TEAC) of each of the five taxa was determined via ABTS assay. *S. retroflexum* showed the highest TEAC value (33.88 mM/100 g DW), whereas *S. americanum* had the lowest (25.69 mM/100 g DW). Significant free radical scavenging activities exhibited by plants in the ABTS assay can be used as a systematic and accurate tool for assessing their total antioxidant potential on large scale [39]. All five sample extracts showed moderate and similar DPPH radical scavenging activity. *S. villosum* had a slightly slow response, as observed from the kinetic curves. Moreover, the best EC₅₀ (48.7 µg) and T_{EC50} (7 min) results were shown by *S. chenopodioides* (Table S3).

Metal-chelating activity is an assay reported for the determination of the concentrations of transition metals. The metal-chelating activity was observed for the plant extracts, and the results showed that they possess a chelating ability for capturing the ferrous ions before they can form a complex with ferrozine. The chelating potential of *S. chenopodioides* was greater than that of the other analyzed extracts, whereas that of *S. americanum* (42.361%) was the lowest.

3.3. Proximate Analysis

Proximate analysis and elemental composition help in the evaluation of the nutritional potential of medicinal plants [43]. *Xylopiya aethiopica* is recommended as a tonic in Ivory Coast due to its high contents of carbohydrates, fat, and crude fiber [44]. The plant samples had high moisture content and produced small amounts of ash. Dietary fibers are composed of lignin, hemicellulose, and cellulose, which have certain physiological benefits for human health, and help in the absorption of trace elements in the gut while reducing the absorption of cholesterol [45,46]. Our investigations revealed that the taxa of the *S. nigrum* complex are rich in dietary fibers (Table 2). Plant proteins not only provide amino acids for the synthesis of human proteins but can also provide energy [47]. The reasonably good protein contents present in the analyzed samples supported their nutritional potential. Amino acids and proteins from *S. nigrum* have been investigated; however, the study did not specify the taxon used [47]. The investigated plant samples can serve as part of a low-calorie, high-protein diet because of their low carbohydrate and fat contents. Kareav et al. [48] evaluated the nutritional value of one taxon of this group. Generally, the investigated taxa were found to be of good nutritional quality, and to be able to provide health benefits when used in appropriate proportions with the correct classification. Comparatively, *S. nigrum* can be recommended in food due to its relative high protein and dietary fiber contents.

There are certain minerals and trace minerals (required at less than 100 mg/day) in our body that are as essential as oxygen for our existence. They exist in our food as organic and inorganic compounds. Moreover, plants also take up heavy metals from the soil. The ashes of five taxa of the *S. nigrum* complex were analyzed for 10 minerals and trace minerals (Table 3). Calcium, magnesium, and potassium were detected in significant quantities, whereas toxic metals such as cadmium, chromium, and lead were detected in the least amounts. Minerals are often found as key components of bones, teeth, muscles, blood, nerve cells, and tissues. The role of each mineral is significant in performing various fundamental physiological processes, including digestion, message transmission in the nervous system, and cell metabolism. An imbalance of certain minerals can disrupt the assimilation of vitamins. For instance, zinc is required for vitamin A, calcium for vitamin C, and magnesium for vitamins of the B complex [49]. Our results suggest that the investigated taxa have good nutritional value.

Table 2. Proximate analysis of the *S. nigrum* complex for its nutritional value.

Plant Taxa	Parameter (%)						
	Ash	Carbohydrate	Crude Fibre	Crude Protein	Fat	Moisture	Nitrogen-Free Extract
<i>S. americanum</i>	4.81	1.79	18.26	18.14	1.85	55.15	56.94
<i>S. chenopodioides</i>	1.2	2.23	14.62	20.08	2.61	59.11	61.49
<i>S. nigrum</i>	2.70	1.6	15.83	21.59	2.64	56.41	58.01
<i>S. retroflexum</i>	2.82	1.18	15.1	20.92	2.6	56.61	57.79
<i>S. villosum.</i>	1.6	1.87	14.91	20.22	2.56	58.84	60.71

Table 3. Determination of metals in the *S. nigrum* complex.

Plant Taxa	Concentration of Metal (ppm)									
	Ca	Cd	Cr	Fe	K	Mg	Mn	Na	Pb	Zn
<i>S. americanum</i>	193.21	0.044	0.091	0.018	510.04	223.61	0.003	53.11	0.041	3.38
<i>S. chenopodioides</i>	145.20	0.049	0.041	0.002	217.33	400.25	0.003	40.47	0.031	2.634
<i>S. nigrum</i>	348.15	0.048	0.075	0.051	100.23	117.14	0.004	26.02	0.039	3.018
<i>S. retroflexum</i>	340.76	0.049	0.062	0.062	370.80	143.73	0.005	29.15	0.042	3.112
<i>S. villosum.</i>	162.24	0.054	0.034	0.007	200.17	430.40	0.002	32.14	0.021	2.801

4. Conclusions

The five examined taxa showed different antibacterial activities against the seven tested bacteria. In some cases, the activities of the plant extracts were found to be comparable in effectiveness to commonly used antibiotics taken as standards. The MIC value of *S. retroflexum* was found to be lower than that of benzyl penicillin against *E. coli*. The five plant samples also showed significant antioxidant activities. The activity of *S. nigrum* was highest when the taxa were analyzed by TPC and FRAP assays, whereas *S. chenopodioides* showed the highest antioxidant potential in analysis by lipid peroxidation, DPPH, and metal-chelating assays. The overall antioxidant activities of *S. nigrum* and *S. retroflexum* were comparable. The presence of bioactive compounds resulted in significant antibacterial and antioxidant activities in the samples. These results suggest that the specific use of morphologically similar taxa can enhance the effectiveness of these traditional plants as cures for different diseases and infections. The functionality and chemical composition of individual plants can hence be determined by the systematic evaluation of their medicinal potential. These plant taxa can be used as healthy medicinal supplements when taken in the right proportions, due to their significant nutritional value.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/pr10081455/s1>, Table S1: List of plants investigated with voucher numbers; Table S2: Zones of inhibition of different concentrations of methanolic extracts of *S. nigrum* Complex and standards against different bacterial strains; Table S3: EC50 and TEC50 values of methanolic extracts of *S. nigrum* Complex.

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