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Antimicrobial assay and phyto-chemical analysis of *Solanum nigrum* complex growing in Kenya

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***Solanum nigrum* complex is a group of plants used as indigenous vegetables and also a source of traditional medicine in Kenya and other parts of the world. The main objective of this study was to determine the antimicrobial property and phytochemical composition of *S. nigrum* complex. Samples of *S. nigrum* complex were collected from Kenya. The plants were dried under the shade and then ground to a fine powder. Crude extracts were prepared from the plants using methanol and evaluated for antifungal and antibacterial activities against the fungi *Fusarium culmorum, Fusarium avenaceum, Fusarium moniliforme* and bacteria *Staphylococcus aureus, Salmonella typhi, Pseudomonas aureginosa, Proteus mirabilis, Shigella* spp., *Pseudomonas syringae, Escherichia coli* and *Basillus subtilis*. Minimum inhibition concentration (MIC) of the samples against active microbes was determined. Phytochemical composition was analysed. T-test was used to analyse the significance of the activity indexes of the extracts against the different groups of the microbes. The samples collected were found to belong to the species *S. nigrum, Solanum scabrum* and *Solanum villosum* which all belong to the *S. nigrum* complex. The samples showed a considerable antibacterial and antifungal activity against the tested microbes. The highest antibacterial activity was 29.00 mm which was shown by *S. villosum* species against *S. aureus*. The antibacterial and antifungal activities were dose dependent with low MIC values as such 0.09 g/ml given by *S. nigrum* Mill species against all eight tested bacteria. The antimicrobial activity was associated with the wide array of phytochemical compounds observed in the samples and they include tannins, saponins, flavonoids, steroids, terpenes, phenolic compounds, alkaloids and cardiac glycosides. It was concluded that the *S. nigrum* complex is rich in many phytochemical compounds like saponins, flavanoids, steroids, glycosides, terpenoids among others which are responsible for the antimicrobial activities of the plants. It can be used not only in treatment of plant fungal infections but also in the management of bacterial human diseases and further research is recommended.**

Key words: *Solanum nigrum* complex, phytochemical, antimicrobial.

INTRODUCTION

Solanum nigrum complex comprise of both native and bred *Solanum* species used as vegetables and source of fruits in Kenya and other parts of the world (Schippers, 2000). Both indigenous (wild types) and modified (hybrid)

varieties have been cultivated (Peter et al., 2009). These plants are believed to have a high nutritional value. The leaves are eaten as vegetable in most parts of the world while the ripe seeds are also edible (Edmonds and Chweya, 1997). There are several species which belong to the *S. nigrum* complex such as *S. nigrum, Solanum villosum, Solanum scabrum, Solanum americana, Solanum burkankii,* and *Solanum schenopodioides* among others.

All these species highly resemble each other and these pose a problem in the taxonomy of the species (Edmonds and Chweya, 1997). Species in the *S. nigrum* complex exhibit considerable genetic variation, both florally and vegetatively. These variations occur in different populations of the same species. Sometimes, the character may be genetically controlled in one variant, but phenoltypically plastic in another (Edmonds and Chweya, 1997).

Other than being used as vegetables, *S. nigrum* complex also forms an important part of traditional medicine in Africa. In Kenya, unripe fruits are used to soothe toothache. They are also squeezed on babies' gums to ease pain during teething. Leaves are used to treat stomach-ache and extracts from leaves and fruits are used to treat tonsillitis (Edmonds and Chweya, 1997).

In Africa, bacteria and fungi are a major challenge in both medical and agricultural fields; they cause fatal infections to humans, animals and plants. People have been using the conventional medicine made of artificial chemicals to manage and treat these infections. Some of these treatments are supposed to be used for a very long time for example treatment of tuberculosis and opportunistic infections due to human immunodeficiency virus (HIV).

Most of these synthetic drugs are also too expensive and unaffordable to most people. Given that plants in the *S. nigrum* complex are edible and universally acceptable as vegetables, there is need to establish their antimicrobial potential with a view of using them as herbal medicine for management of various diseases.

Research shows that herbal remedies have been used successfully and they pose fewer side effects as compared to the synthetic medicines. Species in the *S. nigrum* complex have been used for treatment of microbial and non-microbial diseases successfully in the traditional medicine but no research has been done in Kenya to establish its efficacy. There are very many different species and variants of *Solanum* that are grown in the country and are used traditionally both as a vegetable and as traditional medicine.

Evidently, there are no sufficient scientific studies that confirm the antimicrobial properties of the *S. nigrum* complex in Kenya. This study shows the *in vitro* antimicrobial activity of these plants against pathogenic microorganisms that cause the most common cases of infectious diseases in Kenya.

METHODOLOGY

Samples were collected from various farmers in Western Kenya. Voucher specimens of the plants were stored at the herbarium at the University of Eldoret under reference number SN23/12.

Preparation of plant sample

The samples were dried in a dark room up to a constant weight for a period of one month, turning the plants up and down daily to ensure they dried evenly without fungal growth or spoilage. The samples were then pulverized using a super mixer grinder and the powdered plants were parked in well labelled paper bags and stored at room temperature.

Preparation of crude extracts

Powdered samples (500 g) were soaked overnight in 1400 ml of distilled methanol and filtered. The residue was re-extracted three times with 500 ml of methanol and the filtrates combined. The combined filtrates were concentrated using a rotary evaporator with the water bath temperature maintained at 70°C to prevent thermal decomposition of labile compounds. The samples were placed in clean dry bottles to await further analysis.

Screening for antifungal and antibacterial activity

Preparation of the sample for antimicrobial screening

Ten (10) samples were screened. The first five samples (*S. nigrum* from Kisumu, *S. nigrum* from Bungoma, *S. cabrum* from Kakamega, *S. villosum* from Bungoma, and *S. scabrum* from Bungoma) were weighed to an approximate mass of 50 mg and dissolved in 2 ml of purified methanol to make an approximate concentration of 25 mg/ml. The last five samples (*S. villosum* from Eldoret (Ziwa), *S. scabrum* from Kisumu, *S. villosum* from Eldoret (Ziwa1) *S. nigrum* from Kakamega and *S. scabrum* from Eldoret AMP) were prepared at approximate concentration of 100 mg/ml.

Test microorganisms

The following eleven test microorganisms were used in antimicrobial sensitivity tests; Gram positive bacteria: *S. aureus, Basillus subtilis* and *Proteus* ssp.*;* Gram negative bacteria: *Escherichia coli*, *Shigella* ssp., *Salmonella typhi, Pseudomonas* ssp. and *Pseudomonas syringae*; Fungi: *Fusarium culmorum, Fusarium avenaceum* and *Fusarium moniliforme.* The strains were all obtained from Kenya Medical Research Institute in Nairobi, Kenya.

Preparation of McFarland solution

The McFarland solution was prepared by mixing water solutions of 1% (w/v) anhydrous barium chloride and 1% (v/v) sulphuric acid (H_2SO_4) in the ratio of 0.05:9.95 respectively (McFarland, 1907).

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Preparation of the inoculums

Nutrient broth and sterile distilled water were used to prepare broth cultures of bacterial and fungal test organisms respectively. The nutrient broth media was prepared according to the manufacturers' instructions. The autoclaved media and distilled water were aseptically transferred to sterile capped test tubes of about 6 ml each. Pure isolates of sub-cultured bacteria and fungi colonies were aseptically transferred to the respective broth media and the concentration of the inoculum adjusted to make an approximate cell concentration of 1.0×10^8 cells /ml (Baris et al., 2006) with turbidity approximates that of McFarland 0.5 standard.

Antimicrobial sensitivity tests

Disc diffusion method

Antimicrobial sensitivity tests of the methanol extracts was determined by disc diffusion assay method as described by Rojas et al. (2006) and Moshi et al. (2006). Muller Hilton agar and Sabouraud dextrose agar (SDA) were prepared for bacteria and fungi respectively. The media was autoclaved and allowed to cool in a 45 to 50°C water bath. The freshly prepared and cooled media was transferred to Petri dishes (90 mm in diameter) in a Laminar flow to give a uniform depth of approximately 4 mm. The agar media was allowed to cool and solidify at room temperature. About 0.2 ml of the test inoculum was evenly spread on the surface of the solidified agar media using a sterile cotton swabs. 10 µl of each sample solution was dispensed on sensitivity blank discs using a micro-dispenser and the discs allowed drying in a laminar flow hood for about 4 h after which they were placed at the centre of the labelled inoculated plates.

The treated plates were stored in a refrigerator at 4°C for 24 h to allow sufficient diffusion of the samples into the media without microbial growth and then transferred to incubator at 37°C for 24 h for bacterial and seven days for fungal. The test was carried out in triplicates. Antimicrobial activities were determined by measuring the diameters of zones of inhibition (activity index) in millimetres. Negative control was prepared by dispensing 10 µl methanol on blank discs and allowed to dry. Chloramphenical discs (30 µg/disc) and nystatin discs (25 µg/disc) were used as a positive control for bacterial cultures and fungal cultures, respectively. Some inoculated plates were prepared without any disc as a proof for viability of the inoculums. The plates were put in the same conditions as the plates with test samples and then their activity indexes were determined and recorded.

Minimum inhibition concentration (MIC) assay for antimicrobial tests

Sample preparation

A stock solution with a concentration of 25 mg/ml was prepared for each extract using methanol as the solvent. A series of dilutions with concentrations of 90, 80, 70, 60, 50 and 40% were prepared using distilled methanol. The antimicrobial activity tests were performed using these solutions as per method described earlier.

Determination of the phyto-chemical composition

Phyto-chemical tests were carried out on the methanol crude extracts of *S. nigrum* complex leaves using standard procedures to identify the constituents. The presence of alkaloids and flavonoids was determined as described by Sofowora (1993), while the methods described by Edeoga et al. (2005) was used for terpenoids, tannins, phlobatannins, saponins and cardiac glycosides. Steroids were detected using the method of Shanmugavalli et al. (2009). Presence of the compounds was identified by specific colour changes.

Statistical analysis

T-test was used to determine the level of significance between the antimicrobial results of the extracts at different sample concentrations. The efficacy of the samples against Gram negative bacteria and Gram positive bacteria was also compared, Statistical Package for Social Sciences version 16.0 (SPSS) was used for this analysis. The means and standard deviations (±SD) of the diameters of zones of growth inhibitions for the treatments were determined using Microsoft Excel software.

RESULTS AND DISCUSSION

The samples had different morphological characteristics. These morphological characteristics included the colour of flowers which was either yellow (*S. nigrum*, *S. villosum*) or purplish yellow (*S. scabrum*); colour of berries which was either black (*S. nigrum* and *S. scabrum*) or orange (*S. villosum*) when ripe; leaf size also differed among the species. Some leaves were wider (*S. cabrum*) while others were narrow (*S. nigrum* and *S. villosum*). There were also differences in the size of berries; some were smaller (about 5 mm diameter) (*S. nigrum* and *S. villosum*) while others were larger (20 mm in diameter) (*S. scabrum*). Some of the leaves had smooth edges (*S. scabrum* and *S. nigrum*) while others had serrated edges (*S. villosum*).

The samples showed variations in the percentage yields (Table 1). *S. villosum* of Bungoma had the highest yield of 15.01% while *S. scabrum* of Eldoret had the least (6.14%). The variation was attributed to the difference in the amount and type of the soluble compounds in the samples (Edeoga et al., 2005). The rest samples had the percentage yields ranging between these two values.

The different species of the *S. nigrum* complex showed a considerable amount of antibacterial activity. *S. villosum* of Bungoma was active on all the tested microbes. It gave the best activity of 29 mm against *S. aureus*. *S. scabrum* from Eldoret AMP was also active on all tested microbes followed by *S. nigrum* of Kakamega. The results showed that the three species of the *S. nigrum* complex studied had very good activity on both Gram negative and Gram positive bacteria tested. However, the results showed that differences in activity vary with concentration of the sample just like observed by other scientists in the previous studies on the same plants (Sheen, 2009; Aliero and Afolayan, 2005). The results showed that there was more antibacterial activity seen for the samples at higher concentration than for the samples at lower concentration. This was statistically proven by the T-test results where the T value of the paired means which was 24.52 was out of the expected range of 5.925 - 6.959 at 95% confidence interval

Table 1. Percentage yields of extracts from *S. nigrum* complex

Samples	K01	K02	KO3	K04	K05	K06	K07	K08	K09	K10
Plant dry mass used (g)	174.37	500	500	500	500	500	200	350	350	500
Mass of liquid phase (g)	9.76	7.12	9.27	8.26	11.13	15.76	12.05	10.08	9.43	15.24
Mass of solid phase (g)	12.40	25.30	34.40	66.80	59.50	23.66	17.39	11.79	19.38	15.45
Total mass of crude extract (g)	22.16	32.42	43.67	75.06	70.63	39.42	29.44	21.87	28.81	30.69
Percentage yield	12.70%	6.48%	8.73%	15.01%	14.12%	7.88%	14.72%	6.24%	8.23%	6.14%

K01 = *S. nigrum*; Kisumu, K02 = *S. nigrum*; Bungoma, K03 = *S. cabrum*; Kakamega, K04 = *S. villosum*; Bungoma, K05 = *S. scabrum*; Bungoma, K06 = *S. villosum*; Eldoret (Ziwa), K07 = *S. scabrum*; Kisumu, K08 = *S. villosum*; Eldoret Ziwa (A1), K09 = *S. nigrum*; Kakamega, K10 = *S. scabrum*; Eldoret AMP.

Table 2. MIC values of the liquid samples against bacteria.

Bacterial	MIC values (g/ml)									
	Samples									
	K01L	K02L	K03L	K04L	K05L	K06L	K07L	K08L	K09L	K10L
E. coli	0.49	0.60	0.40	0.70	0.42	0.09	1.20	0.22	0.30	0.60
S. aureus	0.35	0.20	0.16	0.10	0.14	0.09	0.70	0.22	0.16	0.60
P. syringae	0.21	0.70	0.56	0.60	0.35	0.09	1.20	0.22	0.14	0.30
B. subtilis	0.49	0.70	0.64	0.60	0.35	0.09	0.20	0.22	0.04	0.20
P. mirabilis	0.14	0.80	0.48	0.20	0.14	0.09	1.20	0.16	0.30	0.20
S. typhi	0.49	0.50	0.16	0.70	0.35	0.09	1.20	0.22	0.12	0.80
Shigella spp	0.75	0.70	0.56	0.60	0.49	0.09	0.60	0.22	0.14	0.30
P. aureginosa	0.75	1.01	0.82	1.02	0.73	0.09	1.20	0.22	0.12	1.32
Mean	0.46	0.65	0.47	0.57	0.37	0.09	0.94	0.21	0.17	0.54

K01 = *S. nigrum*; Kisumu, K02 = *S. nigrum*; Bungoma, K03 = *S. cabrum*; Kakamega, K04 = *S. villosum*; Bungoma, K05 = *S. scabrum*; Bungoma, K06 = *S. villosum*; Eldoret (Ziwa), K07 = *S. scabrum*; Kisumu, K08 = *S. villosum*; Eldoret Ziwa (A1), K09 = *S. nigrum*; Kakamega, K10 = *S. scabrum*; Eldoret AMP.

(Appendix 1). The antibacterial activity of the tested samples was concentration dependent (Sheeba and Thambidurai, 2009). *S. nigrum* from Kisumu, *S. nigrum* from Bungoma, *S. cabrum* from Kakamega, *S. villosum* from Bungoma and *S. scabrum* from Bungoma, were all active against *S. typhi*, *P. aureginosa* and *E. coli.* These bacteria are notorious in causing gastro-intestinal and urinary tract infections, dermatitis, bacteremia (Kenneth, 2012), traveller's diarrhea, typhoid fever, paratyphoid fever, salmonellosis and other nosocomial infections. *S. nigrum* and *S. scabrum* are capable of managing these infections. This explains the successful use of the extracts from *Solanum* species in traditional treatment of stomach infections (Edmonds and Chweya, 1997). The *S. villosum* from Eldoret (Ziwa) had no significant effect on any tested microbe. However, *S. scabrum* from Kisumu had significant effect on *B. subtilis* only. *S. nigrum* from Kakamega, and *S. scabrum* from Eldoret AMP had significant effect on all tested microbes except on *S. aureus.* Variation in activity was also associated with variation in the presence of many phytochemical compounds in the samples (Table 4) (Gugulothu et al., 2011).

The samples showed a variation in the MIC values against the different bacteria (Table 2). For instance, the lowest MIC from *S. nigrum* Mill from Kisumu, was 0.14 g/ml on *P. mirabilis*. For *S. nigrum* from Bungoma, the lowest MIC was 0.20 g/ml on *S. aureus*. For *S. scabrum* from Kakamega, the lowest MIC was 0.16 g/ml observed on *S. aureus* and *S, typhi*. For *S. villosum* from Bungoma, the lowest MIC was 0.10 g/ml on *S. aureus*. *S. scabrum* from Bungoma had the lowest MIC of 0.14 g/ml on *S. aureus* and *P. mirabilis*. *S. villosum* from Eldoret had the lowest MIC of 0.09 g/ml on all microbes tested both Gram negative and Gram positive bacteria. *S. scabrum* from Kisumu had the lowest MIC of 0.20 g/ml on *B. subtilis* and *S. villosum* from Eldoret, Ziwa had the lowest MIC of 0.16 g/ml on *P. mirabilis* and on the rest of the test microbes an MIC of 0.22 g/ml. *S. nigrum* from Kakamega had the lowest MIC of 0.04 g/ml on *B. subtilis* and *S. scabrum* from Eldoret, AMPATH had the lowest MIC of 0.20 g/ml on *B. subtilis* and *P. mirabilis*. This variation was associated with the variation in type and amount of phytochemical compounds like saponins and cardiac glycosides, which were found to be present in all the samples and have been proven to have antibacterial activity (Harbone, 1973; Foerster, 2006; Al-Bayati and Al-mola, 2008). Other compounds like tannins, sterols and

Table 3. MIC values of the liquid samples against fungi.

Fungi	MIC values (g/ml)				
	Samples				
	K01L	K02L	K03L	K04L	K05L
F. culmorum	0.35	0.20	0.16	0.70	0.28
F. moniliform	-	0.60	-	0.90	-
F. avenaceum	0.35	0.70	0.24	0.30	0.14
Mean	0.35	0.50	0.20	0.63	0.21

K01 = *S. nigrum*;Kisumu, K02 = *S. nigrum*; Bungoma, K03 = *S. cabrum*; Kakamega, K04 = *S. villosum*; Bungoma, K05 = *S. scabrum*; Bungoma.

Table 4. Phytochemical composition of liquid samples of *S. nigrum* complex.

Compound	K01L	K02L	K03L	K04L	K05L	K06L	K07L	K08L	K09L	K10L
Saponins	+	+	+	+	+	+	+	+	+	+
Tannins	+	+	+	+	+	-	-	-	-	-
Sterols	+	+	+	+	+	+	-	-	-	-
Cardiac glycosides	-	+	+	+	+	+	+	+	+	+
Flavanoids	+	+	+	+	+	+	+	+	+	+
Phlabotanins	-	-	-	+	-	+	+	+	+	+
Terpenoides	+	+	+	+	+	+	+	+	+	+
Alkaloids	+	+	+	+	+	+	+	+	+	+
Phenolic compounds	+	+	+	+	+	+	+	+	+	+

(+) Represent presence of the tested compound in the sample, (-) represent absence of the tested compound in the sample, L - liquid samples, K01 = *S. nigrum*; Kisumu, K02 = *S. nigrum*; Bungoma, K03 = *S. cabrum*; Kakamega, K04 = *S. villosum*; Bungoma, K05 = *S. scabrum*; Bungoma, K06 = *S. villosum*; Eldoret (Ziwa), K07 = *S. scabrum*; Kisumu, K08 = *S. villosum*; Eldoret Ziwa (A1), K09 = *S. nigrum*; Kakamega, K10 = *S. scabrum*; Eldoret AMP.

phlabotanins were present in some samples and absent on others in no particular order (Table 4). This qualitative variation and the possible quantitative variation in the presence of the phytochemical compounds were responsible for the variation in MIC values of the samples against the different bacterial species. Higher MIC values show that the bacteria are resistant to the test sample (Sheen, 2009).

S. nigrum from Kisumu was tested against *F. culmorum* and *F. avenaceum* giving activity indexes of 0.35 g/ml for both fungi. *S. nigrum* from Bungoma was analyzed for MIC against *F. culmorum, F. moniliform* and *F. avenaceum* with the MIC values of 0.2, 0.6 and 0.7 g/ml for the three fungi respectively. *S. scabrum* was analyzed for MIC against *F. culmorum* and *F. avenaceum.* With the two fungi having MIC values of 0.16 and 0.24 g/ml, respectively. *S. villosum* from Bungoma was tested for MIC value against the three fungal species. The MIC values were 0.7, 0.9 and 0.3 g/ml for the fungi *F. culmorum, F. moniliform* and *F. avenaceum*, respectively. *S. scabrum* from Bungoma was analyzed for MIC against the fungi *F. culmorum* and *F. avenaceum* with MIC values of 0.28 g/ml and 0.14 g/ml, respectively (Table 3).

F. culmorum and *F. moniliforme* were inhibited by *S. nigrum, S. scabrum* and *S. villosum* although the later had very low inhibition zones as compared to the standard antifungal, nystatin. *F. avenaceum* was inhibited by *S. nigrum* and *S. villosum*. These are *Fusarium* species of fungi known to cause plant infections, food poisoning and various mycoses. Therefore these samples were considered as potential treatments for the three fungi. *S. nigrum* complex species which lacked tannins had no effect on the fungi tested. According to Victor et al. (2005), tannins have considerable antifungal activity. *F. culmorum* is plant pathogenic and causes seedling blight, foot rot, ear blight, common root rot and other diseases of cereals, grasses and a wide variety of monocots and dicots (Rodriguez and Regina, 2008). It is also capable of producing mycotoxins in plants especially cereals. The most common mycotoxins of *F. culmorum* in wheat are deoxynivalenol (DON) and 3-acetyldeoxynivalelon (3-ADON) which are harmful to human and animal health (Marasas et al., 1984). This shows a potential application of these plant samples in control and treatment of plant diseases as well as preventing food poisoning by mycotoxins in cereals and grains. *F. avenaceum* is a globally distributed fungus commonly isolated from soil and a wide range of plants. Severe outbreaks of crown and stem rot of the flowering ornamental, lisianthus (*Eustoma grandiflorum*), have been

attributed to *F. avenaceum*. This fungus has been reported to be an occasional pathogen of stored cabbage (Geeson, 1983).

Extracts of the *S. nigrum* complex were found to poses a wide range of different phytochemical compounds. These compounds are tannins, saponins, terpenes, terpenoids, cardiac glycosides, phenolic compounds, anthraquinones, flavanoids and phlobatanins. All these compounds are known to be biologically active and hence the observed antimicrobial activity (Kessler et al., 2003). All the plant extracts that contained tannin can therefore be recommended for treatment of diarrhoea (Yu et al., 2000; Gertrudes, 2006), inhibit multiplication of retroviruses because of their ability to inhibit reverse transcriptase (Nonaka et al., 1990), treatment of asthma and other respiratory diseases (Burkil, 1994). These tannins can be extracted and used to prepare insecticide for spraying on plants (Buttler, 1998). Tannins have been reported to be toxic to filamentous fungi, yeasts and bacteria (Scalbert, 1991). Their antimicrobial action is made possible by their capacity for protein complexation through hydrogen and covalent bonding and inactivation of microbial adhesions, enzymes and cell envelope transport proteins (Haslam, 1996). The consumption of tannins as green teas (Gertrudes, 2006) and wines prevents different illnesses (Serafini et al., 1994) and inhibits viral reverse transcriptase. Flavonoids were present in the extracts. Their health promoting effects include anti-inflammatory, anti-viral, anti-cancer, anti-oxidant and anti-allergic effects (Balch and Balch, 2000) which make *S. nigrum* complex samples useful in all these medical applications. Saponins found in the samples are known to reduce the level of low density cholesterol and are therefore useful in human diet for controlling cholesterol levels (Assiak et al., 2001). They also control cancer by interfering with cholesterol rich membranes of cancer cells (Dong et al., 2005). They also have antioedema and immunoregulatory effects (Victor et al., 2005), antibacterial, antifungal, anti-inflammatory, antiviral (Al-bayati and Al-mola, 2008) and anti-protozoan (Cheeke, 1998) activities which add to the beneficial application of *S. nigrum* complex.

Another class of compounds which was found in all the extracts was steroids which have important application in the pharmaceutical companies in the production of sex hormones used to bring hormonal balance in expectant and lactating women. It brings libido to men and solves other fertility problems (Victor et al., 2005). This may be the reason why the leaves of *S. nigrum* are recommended as vegetable for expectant mothers or breast feeding mothers to ensure their hormonal balance. This is because steroidal structure could serve as potent starting material in synthesis of these hormones (Okwu, 2001). It has been proven that steroids also have antimicrobial effects against bacteria causing stomach infections. It has also been shown to have analgesic and anti-inflammatory effects, and also used in decreasing serum cholesterol levels (Cyberlipid, 2008). Cardiac glycosides were present in all the samples except *S. nigrum* from Kisumu. Therefore, all the studied species of *S. nigrum* complex can be used in treatment of cardiac disorders (Clifford et al., 1973; Leverin and McMatron, 1999). Phlobatanins and alkaloids were not common in most samples, but they were found to be present in *S. villosum* of Bungoma. Research shows that these compounds have antimicrobial (Ogunwenmo et al., 2007), anticancer (Sneden, 2005), anti-inflammatory (Sofowora, 1993) effects and also act as immune boosters (Jeffery and Harborne, 2000). Alkaloids have their antibacterial effect based on the fact that they help the white blood cells to dispose harmful micro-organisms, (Jeffery and Harbone, 2000).

Glycosides are responsible for the characteristic bitter taste of the black night shade. They are in high concentration in unripe berries and they prevent insects and birds from feeding on the immature fruits and seeds. They also prevent decay of damaged plant tissues (Ogunwenmo et al., 2007). Phenolic compounds, especially the hydroxylated phenols have been found to be toxic to microorganisms with their relative activity increasing with increasing level of oxidation (Scalbert, 1991), number of hydroxyl groups attached to the main structure and their specific sites (Pashin et al., 1986). Phenols and flavonoids have antibacterial activity associated with their ability to complex with nucleophilic amino acids in proteins and the bacterial cell wall leading to destruction of the protein structure and subsequent enzyme inactivation and loss of function (Mason and Wasserman, 1987).

Conclusion and recommendation

The *S. nigrum* complex of Western Kenya comprises of the *S. nigrum, S. scabrum* and *S. villosum*. These different species of the genus *Solanum* contain both antibacterial and antifungal activities against both plants and animal/human pathogens. The study also showed that the antimicrobial activity in these species is due to the presence of a wide array of phytochemical compounds which include saponins, terpenes, sterols, flavonoids, phenolic compounds, cardiac glycosides, tannins, phlabotannins and alkaloids. More work can be done in order to find out the mechanism of inhibition by these plant species which could prove to be of much importance in controlling some of the pathogens which have resistance to the existing antibiotics available in the market.

Conflict of Interests

The author(s) have not declared any conflict of interests.

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APPENDIX 1: T- test for comparing the antimicrobial activity of samples at concentrations of 100 mg/ml and 25 mg/ml

******* Two-sample T-test (paired) *******
Calculated using one-sample t-test with the null hypothesis that the mean of C1 - C2 is equal to 0

One-sample t-test

Variate: 100 mg (C1), 25mg (C2) [1].

Summary

Sample	Size	Mean	Variance	Standard deviation	Standard error of mean
C1-C2	328	6.442	22.64	4.759	0.2628

95% confidence interval for mean: (5.925, 6.959)

Test of null hypothesis that mean of C1-C2 is equal to 0

Test statistic t = 24.52 on 327 d.f.
Probability < 0.001