Antimicrobial potential and screening of phytochemical compounds of *Lantana camara Linn.*

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Abstract

Plants have a great number of chemicals that can be exploited as valuable sources of natural antibiotics and pesticides. *Lantana camara* has numerous potent phytochemicals that could be exploited in plant disease control and protection thereby reducing the indiscriminate use of synthetic pesticides. The effectiveness of extracts varies with its concentration and the kind of bacteria used in the study. Little research has documented the antimicrobial properties of *Lantana camara* plant varieties in Kenya against soil phytopathogens such as *Pseudomonas syringae* and *Phytopthora infestans*. This study was carried out to investigate on the antimicrobial potential of *Lantana camara* on the growth of *Pseudomonas syringae* and *Phytopthora infestans* and qualitative analysis of its phytochemical compounds. Roots and leaves were collected, shade dried and ground into fine powder and extracted by cold extraction method with 70% ethanol. Thereafter 10 %, 5% and 2.5% concentration of leaf extracts were constituted and distilled water used for control experiment. The experiment was arranged in a completely randomized design with three replicates. Disc diffusion method was used to assess antimicrobial activity of this plant by measuring the inhibitory growth zones formed around paper discs. Data obtained was subjected to Analysis of Variance and the means were separated and compared using the least significant difference at $p< 0.05$. The leaves extract treatments significantly reduced the growth of *Pseudomonas syringae* and *Phytopthora infestans* with increase in extract concentration, even though there were no significant differences between the two test microbes. Phytochemical analysis revealed the presence of alkaloids, flavonoids, saponins, tannins, phenols, cardiac glycosides, terpenoids and absence of steroids in the leaves and roots of the plant. Therefore it can be ascertained that certain active ingredients are present in the roots and leaves 'crude extract of *Lantana camara* and have the potential to be exploited for manufacture of pesticides for control of diseases associated with *Pseudomonas syringae* and *Phytopthora infestans*.

Keywords: Antimicrobial activity, Lantana camara, Leaf extracts, phytochemicals, Kenya, *Pseudomonas syringae*, *Phytopthora infestans.*

1.0 INTRODUCTION

Plants have a great number of chemicals that provide valuable sources of natural antibiotics. Plant parts possess antimicrobial agents (Mdee et al., 2009). Plants natural antimicrobial substances have no health environmental residual problems (Kim et al., 2004). Many pesticides sold in the markets are toxic and have adverse effects on soil ecosystems. There is development of resistant strains of pathogens due to continued use of synthetic pesticides and drugs (Goufo et al., 2008; Saravanan, 2001). Muriugi et al. (2014) indicates that the use of soil fumigants such as Methyl bromide in control of soil borne disease pathogens is not a safe method due to residual toxicity on the environment. Indiscriminate use of pesticides adversely affects soil ecosystems (Sarnaik et al., 2006; Sharma & Singh, 2011).

Lantana camara is a common weed in most agricultural lands in Kenya. This plant can grow as a compact clumps and dense thickets. The plant is an invasive species to around fifty different countries. The plant has been used to cure many health problems. The leaves of *lantana camara* have been burnt into ash and mixed with a little amount of salt to serve as a good remedy for sore throat, toothache and coughs. Leaves are chewed for toothache treatment and are also used as inhalant for headache and cold treatment (Kokwaro, 2009). Leaves have been used to treat cuts, rheumatism, malaria, cancer, chicken pox, ulcer, swelling, eczema, tumor, high blood pressure, bilious fever, sores and measles (Naeem et al., 2009). Oil from lantana plants may also be used for treatment of skin itches, wound infections, leprosy and scabies (Ashish et al., 2011).

Lantana camara have exhibited a great variation in chemical constituents according to geographical regions or climate (Singh et al., 2012). *Lantana camara* contain phenolic compounds, essential oils, flavonoids, glycosides, glycosides, quinines, saponins, steroids, triterpenes, sesquiterpenoides and tannins (Ofoegbu, 2013). Natural plant substances have been studied extensively to understand their antimicrobial activities and phytochemical compounds (Al-Rahman et al., 2013; Arora & Kaushik, 2003). Studies on human pathogenic bacteria have revealed that different parts of the plant extracts are active against *Escherichia coli*, *Bacillus subtilis* and *Pseudomonas aeruginosa* and *Vibrio cholerae* (Sanjeeb et al., 2012).

Development of resistant strains of phytopathogens and bioaccumulation of agricultural synthetic chemicals in the agricultural ecosystems are challenges that need to be addressed (Knobler et al., 2003).The emergence of microbial resistance cannot be stopped, because nature provides pathogenic organisms with too many mechanisms for survival and our challenge would be to transform this growing threat into a manageable problem (Knobler et al., 2003). Residual traces of synthetic pesticides poses international penalties on Kenya`s agricultural exports. This presents the need to venture into ecofriendly alternative methods of disease control that are able to replace the use of synthetic pesticides to suppress soil borne phytopathogens and improve crop yield.

There is need to search and come up with new antimicrobial agents that have minimal adverse effects on the environment. Many research works report on antimicrobial effects of *Lantana camara* on human pathogenic microbes but very little has been done on antimicrobial activity of the soil borne phytopathogens. This work aimed at investigating the antimicrobial activity of *Lantana camara* leaf extracts against *Pseudomonas syringae* and *Phytopthora infestans*. This investigation will lead to better understanding of the antimicrobial activities and phytochemical properties of extracts from *Lantana camara* varieties in Maseno, Kenya against *Phytopthora infestans* and *Pseudomonas syringae*.

2.0 MATERIALS and METHODS

2.1 Field collection

The leaves and roots of *Lantana camara* were collected from Maseno University veterinary farm at College campus, and were identified by taxonomists at Maseno university botany laboratory, thereafter a voucher specimen was deposited at the Department of Botany herbarium. The specimen roots were shaken to remove the adhered soil and dust and then they were placed in plastic collection bags before being ferried to the laboratory. Collected plant materials were cleaned up using tap water and spread on a sterilized laboratory bench.

2.2 Extraction

The leaves and roots of *Lantana camara* were shade dried for two weeks at room temperature conditions. They were then ground separately using an electrical grinder to obtain fine powder and finally kept in separate plastic bags at room temperature till the day of extraction.

2.2.1 Ethanol extraction

Cold extraction method was used for extraction. 100g of leaf powder was soaked in 400 ml of 70% ethanol for one week; the separated extract was then filtered through Whatman No.1 filter paper. The filtrate was then concentrated to obtain the paste using rotary evaporator. The thick extracted mass was then dried at room temperature. Thereafter, 0.25g, 0.5g and 1g respectively of the paste were weighed and each dissolved in 10 ml of tap water to make 2.5%, 5% and 10% respectively, and kept in a refrigerator until use (Rani et al., 2014).

2.3 Preparation of test microorganisms

The bacterial and fungal strains, *Pseudomonas syringae* and *Pythopthora infestans* were used for testing antibacterial and antifungal activity. The microorganisms used in this investigation were isolated from diseased tomato fruits exhibiting the pathological symptoms and identified at Maseno University Botany laboratory.

2.4 Isolation of test microorganisms

Pseudomonas syringae was isolated from a tomato fruit exhibiting its symptoms; slightly raised black specks surrounded by a green-yellow halo. 5mm by 5mm pieces were cut from the diseased parts of tomato, thereafter they were dipped into 1% Sodium hypochlorite, followed by 70% ethanol and lastly washed in distilled water three times. The sterile plant parts were then inoculated in Nutrient agar and incubated for forty eight hours at 24^{o}C. After 48 hours of incubation, bacterial colonies were observed on the medium; a loopful was scooped and streaked on the Nutrient agar plates for sub culturing and incubation. Pure culture was made by re-streaking a loopful of bacteria colony on the Nutrient agar and incubated for 48 hours (Kaiser & Ramos, 1980). The same methodology for bacteria was adopted when isolating *Phytopthora*

infestans except that PDA plates were used instead of Nutrient agar and incubated at temperature of 24ºC for two days according to Sharma & Singh (2011).

2.5 Determination of antimicrobial activities

Disc diffusion method was used and the susceptibility test determined by measuring the growth inhibitory zone on the nutrient agar and Potato dextrose agar around the conventional paper disc.

2.5.1 Antibacterial assay

Twelve Nutrient Agar plates were prepared by pouring 20 ml of molten media into sterile petri dishes. After solidification of media 100 ul of *P. syringae* suspensions were dispensed to each plate using a micropipette and spread with the aid of a sterile spreader to achieve uniform growth. The plates were then allowed to dry. Four Whatman No.1 filter paper discs each 6mm in diameter were impregnated with leaf extracts of different concentrations (2.5%, 5% and 10%) and introduced into the plates at equidistant. The sterile disc impregnated with distilled water was used as control. All the plates were incubated at 28ºC for 24 hours under static conditions. After 24 hours the zone of growth inhibition around the paper discs were measured and recorded in millimeters according to Sundaram et al. (2010).

2.5.2 Antifungal assay

Twelve PDA plates were prepared by pouring 20 ml of molten media into sterile petri dishes. After solidification of the media 100 ul of *P. infestans* suspension was dispensed to each plate using a micropipette and it was spread with the aid of a sterile spreader to achieve uniform growth, the plates were allowed to dry. Thereafter, four Whatman No.1 filter paper disc 6mm diameter, that were impregnated with different concentrations of leaf extract were introduced to each plate at equidistant. The sterile disc impregnated with distilled water was used as control. All the plates were incubated at 28ºC for 24 hours under static conditions and the zones of growth inhibition were measured and recorded in millimeters. All the assays were performed in triplicates (Sundaram et al., 2010).

2.6 Phytochemical investigations

The extract phyto-constituents analysis for the identification of bioactive chemical constituents was done according to Musyimi et al. (2008), Harborne, (1998), Mibei et al. (2012) and Akinyemi et al. (2005).

2.6.1 Test for alkaloids

Two grams of the plant material were extracted by warming it for 2 minutes with 20ml of 1% H_2SO_4 acid in a 50ml conical flask on a water bath, with intermittent shaking. It was then centrifuged and the supernatant was pipetted off into a small conical flask. One drop of Meyer's reagent was added to 0.1ml supernatant in a semi-micro tube and a cream precipitate formed indicating the presence of alkaloids (Mibei et al., 2012).

2.6.2 Test for flavonoids

Five milliliters of dilute ammonia solution was added to a portion of aqueous filtrate of the extract followed by addition of concentrated H_2SO_4. A yellow coloration was formed indicating the presence of flavonoids (Mibei et al., 2012).

2.6.3 Test for saponins

Two grams of the powdered sample was boiled in 20 ml of distilled water in a water bath and filtered. Ten milliliters of the filtrate was mixed with 5 ml of distilled water and shaken vigorously and a stable persistent froth formed. The froth was mixed with 3 drops of olive oil and shaken vigorously and an emulsion formed indicating the presence of saponins (Musyimi et al., 2008; Naz & Bano, 2013).

2.6.4 Test for steroids

One milliliter of extract was dissolved in 10 ml of chloroform and equal volume of concentrated sulphuric acid was added by side of test tube. The upper layer did not turned red and the sulphuric acid layer did not show a yellow with green fluorescence. This indicated the absence of steroids (Musyimi et al., 2008; Naz & Bano, 2013).

2.6.5 Test for tannins

Tannin presence was determined by the Folin-Denis colorimetric method described by Harborne, (1998). One gram of the dried powdered samples was boiled in 20ml of water in a test tube and then filtered through Whatman No. 42 filter paper. A few drops of 0.1% ferric chloride were added. A blue-black coloration formed indicated the presence of tannins (Akinyemi et al., 2005; Naz & Bano, 2013).

2.6.6 Test for phenols

One milliliter of extract was dissolved in 2ml of distilled water and a few drops of 2% $FeCl_3$ were added, appearance of a bluish green coloration indicated the presence of phenols (Akinyemi et al., 2005; Naz & Bano, 2013).

2.6.7 Test for Cardiac glycosides

Five milliliters of each plant extract was treated with 2ml of glacial acetic acid containing a drop of $FeCl_3$ solution. This was then underplayed with 1ml conc. H_2SO_4. A brown ring at the interface indicated a deoxy-sugar characteristic of cardenolides thereby confirming the presence of cardiac glycosides (Akinyemi et al., 2005; Naz & Bano, 2013).

2.6.8 Test for terpenoids

Five milliliters of each plant extract was mixed with 2 ml of chloroform, and concentrated sulphuric acid was carefully added by side to form a layer. A reddish brown coloration that formed at the interface indicated the presence of terpenoids (Harborne, 1998; Naz & Bano, 2013).

3.0 DATA ANALYSIS

Data on zone growth inhibition was subjected to analysis of variance and the means were separated and compared using least significant differences at $p \leq 0.05$ by use of SAS statistical package.

4.0 RESULTS

4.1. Bioassays

The results presented in table 4.1 and plates 1-4, show that different concentrations of leaf extract of *Lantana camara* inhibited the growth of *Pseudomonas syringae* and *Phytopthora infestans*; however, there was no significant difference between the two tested microorganisms ($p \leq 0.05$).

Table 4.1. Antimicrobial effects of Ethanol leaf extracts of *lantana camara* on growth of *Pseudomonas syringae* and *Phytopthora infestans.*

PHYTOPATHOGENS	Zone of inhibition (mm)
Pseudomonas syringae	8.52a
Phytopthorainfestans	8.10a
Least Significant Difference (l.s.d)	1.18

Means with same letters down the column are not significantly different.

Plates 1, 2, 3 and 4 show the inhibition of growth diameter formed on plates inoculated with *Pseudomonas syringae and Phytopthora infestans* around paper discs impregnated with different concentrations of leaf extract of 2.5%, 5% and 10% respectively. The zone of inhibition was highest at 10% leaf extract concentration

A **B**

Plate 1: Zone of inhibition of leaf ethanolic extracts on *P.*syringae at **A** 2.5% concentration and **B** 5% concentration

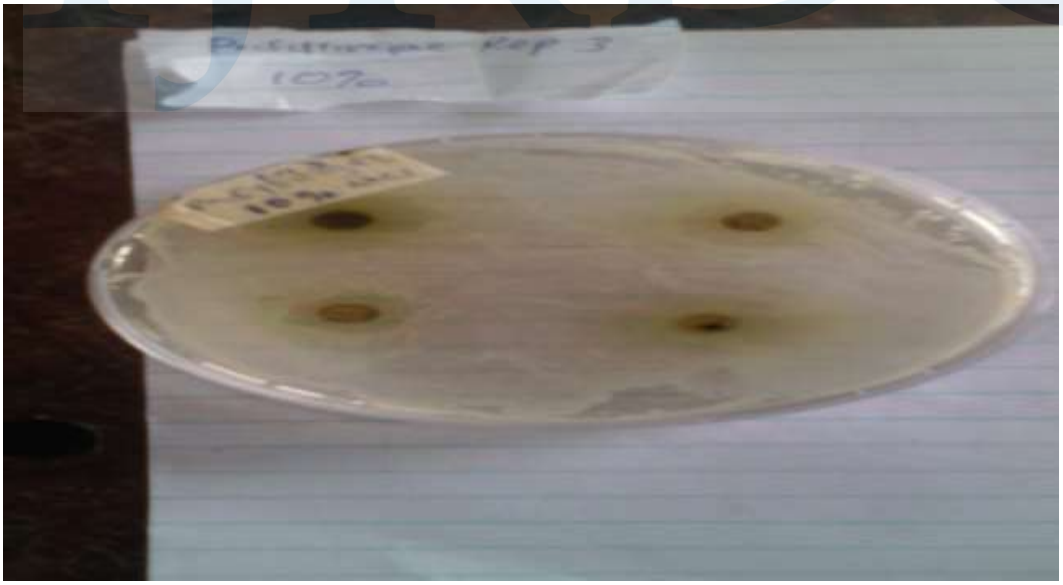
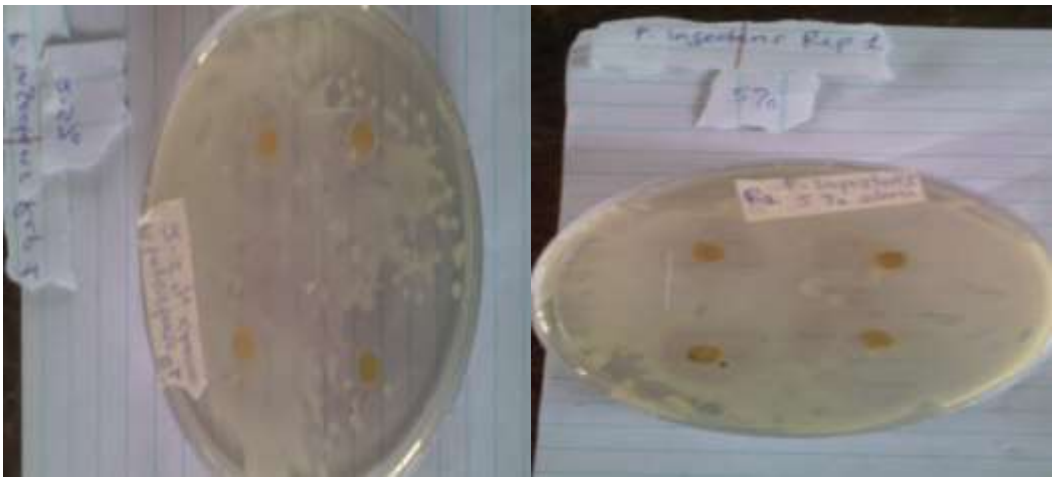


Plate 2: Zone of inhibition of leaf ethanolic extract on *P.* syringae at 10% concentration



A **B**

Plate 3: Zone of inhibition of leaf ethanolic extract on on *P. infestans* at A- 2.5% concentration and B- **5%** concentration.

Plate 4: Zone of inhibition of leaf ethanolic extract on on *P. infestans* at 10 % concentration**.**

4.2. Phytochemical compounds of ethanol extracts of *Lantana camara L.*

The results of preliminary phytochemical screening of the leaves and roots of *Lantana camara* showed the presence of alkaloids, flavonoids, saponins, tannins, cardiac glycosides and terpenoids. Steroids were absent in both leaf and root extracts (Table 4.2)

Table 4.2 Results of phytochemical screening of ethanol leaf and root extracts of *Lantana camara*.

Phytochemical name	**Leaves**	**Roots**
Alkaloids	+	+
Flavonoids	+	+
Saponins	+	+
Steroids	-	-
Tannins	+	+
Cardiac glycosides	-	+
Terpenoids	+	+
Phenols	+	+

Key: - Absent + Present

The interactions between the phytopathogens and plant extracts treatments were found to be significant at $p<0.05$ (table 4.3).

Table 4.3. Analysis of variance of effect of plant extracts on growth inhibition of phytopathogens.

SOURCE	DF	SUM OF SQUARES	MEAN OF SQUARES	F VALUE	Pr>F
Model	7	73.89	10.56	5.69	0.0019
Error	16	29.66	1.85		
Corrected total	23	103.55			

Microorganisms **(M)**	1	1.08	1.08	0.58	0.46ns
Plant extracts treatments **(T)**	3	55.31	18.44	9.95	0.006*
M x T	3	17.49	5.83	3.15	0.05*

Key: *significant ns- not significant

5.0 DISCUSSION

The systematic screening of plant extracts for antimicrobial activity is a continuous effort to find new antibacterial compounds (Mariajancyrani et al., 2014). Previous studies using extracts from *Lantana* species showed that they were able to inhibit the growth of gram positive and gram negative bacteria strains (Barreto et al. 2010). However, in this study the leaf extracts were found active against *Pseudomonas syringae and Phytopthora infestans* (Table 4.1).This could have been attributed to the presence of secondary metabolites such as alkaloids, terpenoids and phenolics in *Lantana camara* as were identified in Table 4.2. This was very pronounced with the increase in the concentration of leaf extract up to 10%. The leaf extracts significantly inhibited the growth of *Pseudomonas syringae* and *Phytopthora infestans* (Plates 1-4 and Table 4.3). In the present study *Pseudomonas syringae* growth was inhibited at the leaf concentrations of 2.5%, 5% and 10%, with10% exhibiting the highest inhibitory growth diameter of 10.6mm as evident in Plates 1, 2, 3 and 4. Similar results have been reported by Elkhalfi et al. (2013), where methanol extracts of several plant species successfully inhibited the growth of *Pseudomonas syringae*. The fungal activity was tested against *Phytopthora infestans* which showed highest growth inhibition diameters of 10.1 mm and 8.8 mm recorded at 5% and 2.5% respectively. These findings are also in agreement with the work of Yanar et al. (2011) where the fruits and root extracts from various plant species completely inhibited mycelial growth of *Phytopthora infestans* at 4% concentration.

Phytochemical compounds such as tannin, flavonoid, saponin and Cardiac glycosides were present in leaves and roots of *Lantana camara* which is a proof that lantana plants have medicinal properties (Kokwaro, 2009). The results agree with phytochemical study of aerial parts of *Lantana camara* carried out by Naeem et al. (2009). Inhibitory effect on many enzymes may

occur due to protein precipitation. The presence of these tannins could be the reason why the leaves are used locally for treatment of wound, sores and skin diseases (Kokwaro, 2009). Similar findings have been reported by Ganjewala et al. (2009). Alkaloids are synthesized in the roots of plants and then transported to the leaves for storage. Saponins, tannins and flavonoids exhibit antimicrobial activities. The use of ethanol as an organic solvent concentrates more water soluble compounds including saponins and tannins. Saponins and tannins act as antimicrobial secondary metabolite (Musyimi et al., 2008). Antimicrobial activity of Lantana species has been evaluated by several researchers (Salada et al., 2015; Barreto et al., 2010; Ganjewala et al., 2009) and it has been found that Lantana species are good antimicrobial activity. The availability of flavonoid in the leaves and roots of the plant suggests that they can be used to manufacture pesticides. This confirms the reason for the use of these plants in the treatment of microbial infections (Salada et al., 2015). The presence of cardiac glycosides in both roots and leaves shows that the plants are good for treatment of diseases associated with heart. The presence of various active ingredients revealed by the phytochemical screening (Table 4.2) supports the use of this plant traditionally for treatment of various diseases (Kokwaro, 2009).

Antimicrobial action of saponins are due to their involvement in membranolytic activities (Kensa, 2011) this may be explained by the fact that the leaf extracts exhibited pronounced inhibitory activities on *Pseudomonas syringae* than *Phytopthora infestans*. The antimicrobial activities of the plant extract may be attributed to the presence of the phytochemicals. Tannins are known to precipitate microbial proteins hence interfering with the availability of nutritional proteins (Gogoi et al., 2012). The antimicrobial activities of this plant may be attributed to the synergistic effect of the various phytochemical components of the leaf extract. The variation in the quantity of the active ingredient required to effect inhibition may not matter much since medicinal plant have been reported to have little or no side effects (Eshrat & Hussain, 2002). The present study reveals potential use of these plants for developing new antimicrobial compounds against soil phytopathogenic microorganisms.

5.1 CONCLUSIONS

The present experimental findings have revealed that the leaf extract of *Lantana camara* plant possesses inhibitory and antimicrobial activities with a more pronounced effect on the growth of

Pseudomonas syringae than *Phytopthora infestans*.The study has also revealed that this variety of *Lantana camara* plant contain phytochemical compounds such as saponin, flavonoids, tannins and cardiac glycosides and may be responsible for the growth inhibitory effects exhibited by the leaf extracts of this plant. This study provides basis for developing the plant products into useful pesticides that may be used to control soil pathogenic bacteria and fungi. To our knowledge, this is the first report of the antimicrobial activity of *Lantana camara* on *Pseudomonas syringae* than *Phytopthora infestans*.

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6.0 References

Akinyemi, K.O., Oladapo, O., Okwara, C.E., Ibe, C.C., & Fasure, K.A. (2005).Screening of crude products of six medicinal plants used in Southwest Nigerian unorthodox medicine for antimethicillin resistant *Staphylococcus aureus* activity. *BMC complementary and alternative Medicines* 5, 6-10.

Al-Rahmah, A.N., Mostaf A.A., Abdel-Megeed, A., & Hussein, S.A. (2013).Fungicidal activities of certain methanolic plant extracts against tomato phytopathogenic fungi. *African Journal of Microbiology Research*, 7(6), 518-520.

Arora, C & Kaushik, R.D. (2003). Fungicidal activity of plants extracts from Uttaranchal hills against Soybeans fungal pathogens. *Allelopathy Journal*, 112, 217-228.

Ashish, S., Sadaf, Q., Kavita S., & Noor, A.K . (2011).Antimicrobial Agrios, G. N.(1997) Plant Pathology. 4thedition. *Academic press*, Newyork, pp, 226.

Barreto, F.S, Sousa, E.O., Campos, A.R., Costa, J.G.M, & Rodrigues, F.F.G. (2010).Antibacterial Activity of *Lantana camara* Linn and *Lantana montevidensis* Brig Extracts from Cariri-Ceará, Brazil. *Journal of Young Pharmacists*, 2(1), 42-44.

Elkhalfi, B., Essari, A., Serrano, A., & Soukri, A. (2013). Antibacterial activity of plant methanolic extracts on a field isolate of *Pseudomonas syringae*pv tomato from the Casablanca region (Morocco). *Journal of Advances in Bioscience & Biotechnology*, 4, 1-10.

Eshrat H.M., & Hussain, A. (2002). Hypoglycemic, hypoipidemic and antioxidant properties of combination of *Curcuma longa Linn* and partially purified product from *Abroma augusta Linn.* In streptozotocin induced diabetes. *Indian Journal of Clinical Biochemistry*, 17(2), 33-43.

Ganjewala, D., Sam, S., & Khan, K.H. (2009). Biochemical compositions and antibacterial activities of *Lantana camara* plants with yellow, lavender, red and white flowers. *Eura Asia Journal of Biological Science*, 3, 69-77.

Gogoi, P., & Islam, M. (2012). Phytochemical screening of *Solanum nigrum* L and *Solanum myriacanthus* Dunal from Districts of upper Assam, India. *Journal of Pharmacy*, 2(3), 455-459.

Harborne, J.B. (1998). Phytochemical methods. A guide to modern techniques of plant analysis.3rd edition. Springer (India) Private Limited, New Delhi.

Kaiser, W.J., & Ramhos, H.A. (1980).Occurrence of *Pseudomonas syringae* on bean & soy beans in Kenya. *Journal of Plant Disease*, 64, 593-594.

Kensa, M.V. (2011). Studies on phytochemical screening and antibacterial activities of *Lantana camara Linn*. *Plant Sciences Feed*, 1(5), 74-79.

Kim, Y.M., Lee, C.H., Kim, H.G., & Lee, H.S.(2004). Anthraquinones isolated from *Cassia tora Leguminosae* seed show antifungal property against phytogenic fungi. *Journal of Agriculture* and *Food Chemistry*, 52, 6096-6100.

Knobler, S.L., Lemon, S.M, Najafi, M., & Burroughs, T. (2003). The Resistance Phenomenon in Microbes and Infectious Disease Vectors: Implications for Human Health and Strategies for Containment: Workshop Summary. Institute of Medicine (US) Forum on Emerging Infections. Washington (DC): National Academies Press (US).ISBN-10: 0-309-08854-2

Kokwaro J.O. (2009).Medicinal plants of East Africa.3rd edition. *East African literature bureau*, Nairobi, Kenya, pp, 289.

Mariajancyrani, J., Chandramohan, G., & Ravikumar, S. (2014). Terpenes and antimicrobial activity from *Lantana camara* leaves. *Research Journal of Recent Sciences*, 3(9), 52-55.

Mdee, L.K., Masoko, P., & Eloff, J.N. (2009). The activity of extracts of seven common invasive plant species on fungal phytopathogens. *South African Journal of Botany*, 75), 375–379.

Mibei, K.E., Ojijo, K.N., Karanja, S.M., & Kinyua, K.J. (2012). Phytochemical and antioxidant analysis of methanolic extracts of four African Indigenous Leafy Vegetables. *Annals of Food Science* and *Technology*, 13(1), 37-42.

Muriugi, S. J., Mutitu, E.W., & Muthoni, J.W. (2014). Efficacy of cultural methods in control of *Rhizoctonia solani* strains causing tomato damping off in Kenya. *African Journal of Food, Agriculture, Nutrition and Development*, 14(2), 877-878.

Musyimi, D.M., Ogur, J. A., & Muema, P.M .(2008). Phytochemical compounds and microbial activity of extracts of Aspilia plant (*Aspilia mossambicensis*) (Oliv) wild. *International Journal of Botany*, 4(1), 56 –61.

Naeem, Q., Bashir, A.C., Altaf., D., Abdul, M., & Rahan, Z.(2009). Phytochemical study of Aerial parts of *Lantana camara* for the pharmacological active compounds. *Journal of Applied Pharmaceutical Science,* 1(1):19-26

Naz, R., & Bano, A. (2013). Phytochemical screening, antioxidants and antimicrobial potential of *Lantana camara* in different solvents. *Asian Pacific Journal of Tropical Disease*, 3(6), 480-486.

Ofoegbu, O. (2013).The effects of extraction protocol on phytochemical and antimicrobial activities of *Lantana camara* leaf extract found within local environment. *Journal of Basic Applied Chemistry*, 3(1), 5-10.

Rani, J.M., Mohan, C., & Kumar, R. (2014).Terpenes and antimicrobial activity from *Lantana camara* leaves. *Research Journal of Recent Sciences*, 3(9), 52-55.

Salada, J.T., Balala, L. M., & Vasquez, E.A. (2015). Phytochemical and antibacterial studies of *Lantana camara* L. leaf fraction and essential oil. *International Journal of Scientific and Research Publications*, 5, 3. ISSN 2250-3153.

Sanjeeb, K., Guarav, K., Loganathan, K., Kokati, V., & Bhastara, R. (2012). A Review on medicinal properties of *Lantana camara*. *Research Journal of Pharmacy* and *Technology*, 5(6), 711-715.

Saravanan, T., & Velluvaparidasan, V. (2001). Fungotoxic effect of biocontrol agents and plant extracts on seed borne fungi of Sorghum, Pakistan. *Journal of Biological Sciences*, 4(6), 676-678.

Sarnaik, S.S., Kanekar , P.P., Raut, V.M., Taware, S.P., Chavan, K.S., & Bhadbhade, B.J. (2006). Effects of Application of different pesticide to Soybeans on the soil microflora. *Journal of Environmental Biology*, 27(2), 423-426.

Sharma, S.K., & Singh, P.A. (2011). Antimicrobial Investigations on rhizomes of *Cyperus rotunas Linn.* Scholars Research Library, 3(3), 428-429.

Singh, K.R., Tiwari, B., Sharma, K.D., & Satyendra, P.S. (2012).Comparative study of chemical composition of *Lantana camara* leaf, flower and fruit essential oil. *International Journal of Pharmaceutical and Chemical Sciences,* 1(3), 895-896.

Sundaram, R., Gopi, P.S., Anha, N.A.G. (2010). Antimicrobial activity of Medicinal plants along Kanyumari coast, Tamil Nadu, India. *African Journal of Basic* and *Applied Sciences,* 2(5), 6-15.

Yanar, Y., Kadioglu, I., Gocke, A ., Demirtas, G.N., Cam, H., & Whalon, M. (2011). In vitro antifungal activities of 26 plant extract on the mycelial growth of *Phytopthora infestans*. *African Journal of Biotechnology*, 10(14), 2625-2629.