ANTIBACTERIAL AND PHYTOCHEMICAL PROPERTIES OF SELECTED
POULTRY ETHNOMEDICINAL PLANTS IN MASAKA DISTRICT

BY

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DEDICATION

This work is dedicated to my Parents and my family, for always being there for me and finally to my late mother Miss. Edith Namubiru (RIP).
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ABSTRACT

Poultry farmers have problems of poor production, poultry diseases and the public is vulnerable to zoonoses. Indigenous practices such as the use of herbal medicines and concoctions have been a form of therapy for poultry among resource-poor small holder farmers. Indigenous practices are considered by scientists to be risky to both human and animal health. A few farmers use conventional veterinary drugs like antibiotics most times unnecessarily and this is making disease causing bacteria more resistant to the drugs and therefore becoming a threat to public health. Documentation and validation of indigenous medicine is therefore necessary because they are likely to be important in future especially given the trend of emerging diseases and the development of resistance of pathogens to drugs.

To solve the problems facing the poultry industry, documentation of data about indigenous knowledge data about the herbal plants as well as antibacterial and phytochemical analysis to validate this data was carried out in this research. The information, focused group discussions and key informant interviews were conducted. Ethanol, ether and water extracts of selected medicinal plants and five concoctions were screened for antibacterial activity against *Staph. aureus*, *Strep. faecalis*, *E. coli* and *S. typhimurium*, using agar well diffusion and tube dilution methods. Their Phytochemical composition of selected plants was also investigated.

The results indicated that fifty nine plants from 33 families were commonly used and family *Asteraceae* had the highest number while most frequently used plants were *Cannabis sativa* and *Nicotiana tobaccam*. Plants were mainly used for prophylaxis and the oral route was the common route of administration while leaves were the most commonly used plant parts. In general, gram-positive bacteria were more susceptible than gram-negative bacterial species. Ethanol and ether extracts had better antibacterial activity than water extracts. The water extract of *Moringa oleifera* had activity on all the four bacteria species. The water extracts of *Persea americana* had the lowest MIC on *S. typhymurium* therefore the best activity. *Leonotis nepetifolia* and *Lantana trifolia* had the lowest MIC on *Staph. aureus* therefore the best activity.

Phytochemicals such as tannins, sterols, basic alkaloids and alkaloid salts featured in most of the tested plants.
From these results, the study has found out that ethanol and ether are better solvents and can be used as alternative solvents to water by the poultry farmers to make medicinal preparations. The study has further shown that these plants have good antibacterial activity. *Moringa oleifera* leaves could be used to treat a wide number of diseases, *Persea americana* leaves could be used to treat salmonella infections while *Leonotis nepetifolia* and *Lantana trifolia* leaves could be used to treat staphylococcal infections in poultry. These plants also have phytochemicals of medicinal importance. However the study has not isolated the specific antibacterial principles, shown toxicity studies, shown activity of these plants on other organisms like other bacteria species, protozoa and helminths and carried out clinical trials. Further studies on these should therefore be carried out.
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ACRONYMS

ATCC  American Type Culture Corrections
°C  Degrees centigrade
DMSO  Dimethyl Sphoxide
E. coli  Eschericia coli
EVM  Ethno-veterinary medicine
IBD  Infectious bursal disease
MIC  Minimum Inhibitory Concentration
ND  Newcastle Disease
S. typhimurium  Salmonella typhimurium
Staph. aureus  Staphylococcus aureus
Strep. faecalis  Streptococcus faecalis
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CHAPTER ONE

INTRODUCTION

Poultry provide globally important sources of animal proteins and are amongst the most intensively reared of all live stock species (LPP, 2006). Globally, production of the primary poultry products (meat and eggs) has been rising rapidly. Over a 10 year period between 1995 and 2005, consumption, and hence production, has increased globally (percentage increase) for chicken meat (53%); turkey meat (13%), duck meat (67%), goose meat (53%), Chicken eggs (39%) and other eggs (27%) (Scanes, 2007). The total fowl population in Africa was estimated at 1,868 million in 1995 producing 1,695,620 metric tones of eggs and 2,096,000 metric tones of meat (FAO, 1996). In Uganda the poultry population was projected to be about 32.6 million birds for 2006/2007 compared to 23.5 million in 2002 (UBOS, 2002). Chickens form the main poultry types but turkeys, ducks, geese, pigeons and ostriches are also kept in some areas (UBOS, 2002).

The high growth rate of Ugandan population and increases in urban migration have increased demand for food. These factors have put pressure on land and other resources for food production and other necessities. This has led to a focus on farming systems or enterprises that maximize yield per unit area and input and poultry production has been identified as one of the enterprises that falls within this category (Byarugaba, 2007). The intensity of husbandry can only be done by controlling many infectious diseases that would otherwise inflict severe losses or even prevent intensive poultry sector. The emergency of a new pathogen or one variant of an old pathogen has the potential to spread rapidly and devastate national flocks, as has happened on several occasions with strains of highly pathogenic avian influenza (Alexander, 2007). Despite the potential of poultry to alleviate poverty and improve the quality of life of low-income earners, the rural poor and their children, little attention has been paid to it in many countries including Uganda (Sorensen, 1999).
It has been reported that the low productivity in traditional poultry systems is mainly due to high mortality which is caused by mismanagement, diseases, lack of nutritional feeding and predators. In traditional systems the mortality has been estimated to be in the range of 80-90% within the first year of hatching (Permin and Hansen, 1998). It has also been reported that in contrast to modern poultry production, village based poultry production is often characterized by a range of diseases occurring at the same time. Most often free range poultry have sub clinical infections with a high number of endoparasites and ectoparasites (Permin and Pedersen, 2002). Ojok, (1993) also reported that one of the major constraints to village poultry in Uganda is the existence of various diseases. The problem of diseases in chickens is compounded by the interaction of different entities that are of significant importance to disease epidemiology. There are uncontrolled contacts in the villages between birds from different house holds as well as frequent introduction of birds from markets as gifts. Such birds bought from markets and wild birds may be a source of infection (Byarugaba, 2007). Diseases of poultry are therefore of major concern, both locally and on international scale (LPP, 2006).

The important bacterial diseases in free range poultry include colibacillosis caused by *Escherichia coli* (*E. coli*) and Salmonellosis caused by *Salmonella species* among others (Permin and Bisgaard, 1999). Further more, several pathogens have been shown to be present in family poultry and these include salmonellosis, fowl cholera, mycoplasma, infectious laryngotracheitis virus, chicken anaemia agent, *E. coli* and fowl pox virus (Bouzoubaa et al., 1992).

In general, bacteria have the genetic ability to acquire and transmit resistance to drugs, which are utilized as therapeutic agents (Cohen, 1992). A Harvard University study showed that antibiotic-resistance genes found in bacteria infecting humans were identical to some of the bacteria infecting animals (O’Brien et al., 1982). The problem of microbial resistance is growing and the outlook for the use of antimicrobial drugs in the future is still uncertain. High level of antibiotics use, often clinically unnecessary, has led to a steady increase in drug resistance and low-income countries, home to the majority of the world's population, are believed to have an important role in this phenomenon (Aryanti and Hilbrand, 2003).
Routine, medically unnecessary use of antibiotics in livestock is making disease-causing bacteria more resistant to the drugs, diminishing the drugs’ power to treat life threatening disease in humans. The increased use of antibiotics has gone hand in hand with the development of industrial-style livestock operations. Fifty million pounds of antibiotics are produced in the United States every year; 40% of that is given to animals (American medical News, 1999). Evidence is increasing that the emergence of antibiotic resistance, caused by overuse of antibiotics, threatens public health. Drug-resistant infections, some fatal, have been increasing in people in United States. And many scientists attribute this problem to the misuse of antibiotics in humans and animals (The New York Times, 1999). In more than one-third of the Salmonella-poisoning cases in 1997, the bacteria were resistant to antibiotics used to treat the disease, according to centers for Disease Control and Prevention (The New York Times, 1999).

Staphylococcus bacteria are becoming increasingly resistant to the chief antibiotic, methicillin that has been used to treat staph infections, and to the last line of defense vancomycin (Panlilio, 1992). In the study carried out by Moniri and Dastehgoli, (2007), it was found that the prevalence rate of \textit{E. coli} resistant to ciprofloxacin and erythromycin in the samples from chickens with colibacillosis was higher than in health controls. The transfer of resistant \textit{E. coli} from chickens to humans is a common event, as has been demonstrated by several groups of researchers (Moniri and Dastehgoli, 2007). In his report, Sieradzki \textit{et al.}, (1999) suggested that there is need to search for new infection-fighting strategies to control microbial infections. Suggestions have been put forward to reduce this problem and these include controlling the use of antibiotic, developing research to better understand the genetic mechanisms of resistance, and continuing studies to develop new drugs, either synthetic or natural (Gislene \textit{et al.}, 2000). This situation has provided the impetus to the search for new antimicrobial substances from various sources like medicinal plants (Cordell, 2000).

The use of medicinal plants plays a vital role in covering the basic health needs in developing countries, particularly Africa (Munoz-Mingarro \textit{et al.}, 2003). Many of the drugs currently used to treat bacterial and other infections were first isolated from natural sources including ethno medicinal plants (Coe and Anderson, 1996). Infectious diseases are usually characterized by
clear symptoms, so it is likely that traditional healers have been able to recognize such diseases and have developed effective therapies. Medicinal plants use is widespread (Farnsworth and Suejarto, 1991). About 80% of individuals of the rural population use traditional medicine, which has compounds derived from medicinal plants (WHO Traditional Medicine Strategy, 2002). According to a study carried out by Makerere University researchers in central and eastern Uganda, about 80% of the poultry farmers surveyed know how to use medicinal plants to treat poultry diseases (Bukenya, 2007). Many farmers in Mbale, Rakai and Mbarara districts are using medicinal plants to treat coughs, diarrhoea, swollen eyes, mites, worms and lice as well as Newcastle prophylaxis and coccidiosis. Medicinal plant species such as Capsicum frutescens and Cannabis sativa were used in all three districts, while Nicotiana tabacum, Aloe sp, Vernonia amygdalina and Tagetes minuta species were used in Rakai and Mbarara (Bukenya, 2007). Such plants should be investigated to better understand their properties, and efficacy (Ellof, 1998).

Recently, much attention has been directed toward extracts and biologically active compounds isolated from popular plant species. When pure chemical entities are isolated, their structures are elucidated. These can then be developed into medicines or chemically modified for medical use (Heinrich et al., 2004). The production of medicines and the pharmacological treatment of diseases began with the use of herbs (Tyler, 1997). Plant based antimicrobials represent a vast untapped source of medicines with enormous therapeutic source potential (Cowan, 1999). They are supposedly effective in treatment of infectious diseases while simultaneously mitigating many of the side effects that are often associated with synthetic anti-microbials (Iwu et al., 1999). Since some farmers cannot afford to buy modern poultry drugs, medicinal plants would be good substitutes, sparing farmers from economic losses due to disease outbreaks (Bukenya, 2007). Any attempts to improve the lives of these people through livestock industry, must therefore begin by understanding and recognizing the evolution, application and management of ethno veterinary medicine in their cultural lifestyle. This research would therefore assess the antibacterial and phytochemical properties of selected plants from Masaka.

Ugandan farmers have problems of malnutrition, poverty, poor production, poultry diseases and people are vulnerable to zoonoses. Indigenous practices which have not been validated present the greatest threat to poultry care and human health. This study therefore sought to address this
problem by decoding the farmers’ knowledge. The information about the types, and effectiveness of ethno medicinal resources used is known by few people and it has been passed on from generation to generation but has not been described or even validated and very little literature is available. Furthermore, in the research carried out in east and central Uganda, about 80% of the poultry farmers surveyed know and are using medicinal plants to treat poultry diseases (Bukenya, 2007). However, little research has been carried out to assist these farmers. Evidence is increasing that the emergence of antibiotic resistance, caused by overuse of antibiotics, threatens public health. To address such problems, it was found necessary to do research on alternative traditional medicines to use in the poultry industry.

Emerging epidemic and zoonotic diseases are causing heavy loss of life and money. One of the main loophole in public defense against these epidemics in Uganda are the less ownership and involvement of local communities in epidemic disease prevention, and insufficient appreciation of the value and role of traditional indigenous knowledge and their integration in epidemic disease prevention by livestock service providers. These lead to lack of a primary barrier at community level against epidemics occurrence and a project was designed by TRIKOMS (Traditional Indigenous Knowledge Mystical Systems) in October 2006 as a cornerstone for Avian Influenza epidemic prevention. This current study is part of this project and is aimed at enhancing the capacity of local livestock service providers and farmers to utilize Traditional Indigenous Knowledge systems in Masaka as a remedy for fighting the existing and newly emerging poultry diseases to address the above problems. Within this aim, there were three objectives:

1) To document current ethnoveterinary knowledge among poultry farmers in Masaka District
2) To test for antibacterial activity of crude extracts and farmers’ concoctions on Salmonella typhimurium, Escherichia coli, Streptococcus faecalis and Staphylococcus aureus using agar well diffusion tests and broth dilution tests
3) To qualitatively screen for the phytochemicals of priority plants.

The current study documented the current state of knowledge and use of ethnoveterinary resources among farmers in Masaka district. Furthermore, the knowledge was decoded by
assessing antibacterial and phytochemical properties of priority medicinal resources used by poultry farmers in Masaka district.

The ultimate significance that the knowledge obtained from the study would have is to contribute to the development of intervention strategies to be used as a model for adoption by all other communities in the prevention of poultry diseases and treatment under Trikoms. The information would further contribute to reduction of threats to public health posed by some indigenous practices and routine use of antibiotics. It would avail information to the farmers after validation and decoding of their practices of using ethno veterinary materials in the management of poultry diseases. The information would contribute to reduction of the mortality and morbidity of local chicken which would in turn increase the number of local poultry, their production and productivity. The sale of chicken and the chicken products would increase and these will lead to more income and food at households. It would therefore be an opportunity for the vulnerable communities to get empowered to handle emerging disease outbreak, prevention and control of other diseases. This research related to the priorities of the Region and the country in a way that, rural poor especially women can profitably use available plant resources to treat poultry therefore alleviating poverty and improving the quality of life of the hardcore poor and their children, who account for about 45% in Uganda. Problems of diseases which are among the important constraints in the expansion of poultry industry would be solved. Local medicine would be used as an alternative instead of imported drugs which has become erratic as their importation has to compete for meager foreign exchange with other vital imports required for industrial and social development.
CHAPTER TWO
LITERATURE REVIEW

2.1 Importance of poultry
The economic and nutrient contribution of indigenous free-range poultry is estimated to be over 80% of the per capita consumption of poultry meat and eggs while the 20% is by the commercial poultry sector (UBOS, 2002). In developing countries nearly all families at the village level, even the poor and landless, are owners of poultry. The role of family poultry in poverty alleviation, food security and the promotion of gender equality in developing countries is well documented (Guèye, 2000). Poultry makes good use of locally available resources, requiring low inputs. Production is feasible at village level, where only low cost technology is needed to improve production considerably. Low investments only are required to achieve such change, land ownership is not a constraint, and village production is environmentally friendly (Upton, 2004).

Family poultry production represents an appropriate system to contribute to feeding the fast growing human populations and to provide income to poor small farmers (Kitalyi and Mayer, 1998). Though generally considered secondary to other agricultural activities by smallholder farmers, the poultry industry contributes to improved human nutrition and food security by being a crucial supplier of high quality protein in form of eggs and meat (Mukiibi-Muka et al., 2000). The poultry industry increases peoples’ incomes and improves their welfare through the sale of poultry and poultry products (Mukiibi-Muka et al., 2000). The poultry industry has the potential to generate foreign exchange earnings through export of poultry products to neighboring countries. The poultry industry acts as a key supplement to revenue from crops and other livestock enterprises, thus avoiding over-dependency on traditional commodities with inconsistent prices. Poultry products can be sold or bartered to meet essential family needs such as medicine, clothes and school fees. They are also active in pest control and provide manure to fertilise our gardens (Alders et al., 2003). In commercial poultry production, poultry contributes significantly to the incomes of both urban and suburban farmers. Urban dwellers too keep poultry from which they derive additional income in addition to other sources and on the other
hand, the big commercial producers derive most of their income from the sector as their primary business (Byarugaba, 2007).

Management of poultry has been associated with women and children for various historical and social factors. Poultry are mainly owned and managed by women and are often essential elements of female-headed households. Chicken products are among the few agricultural products directly accessible to women in rural areas and increased food production from chickens will improve household food security and is often easily combined with other household activities such as gardening because of the proximity of the chickens to homesteads (Scola, 1992; Sorensen, 1999; Byarugaba and Katunguka, 2002). Poultry are socio-culturally important with few religious taboos attached. Poultry is highly prized in many socio-cultural functions such as dowry and festivities (Alders et al., 2003; Byarugaba, 2007). It was therefore vital that research could be carried out to promote the poultry industry since it is of importance to the livelihood of the people of Uganda and the world at large.

2.2 Common bacterial diseases affecting poultry
Diseases of smallholder poultry have been identified as a major cause of mortality and a consequent loss of production. Many diseases have been documented in smallholder poultry, as diseases are easily contracted under free-range conditions due to poultry scavenging habits. Furthermore disease control is very difficult to carry out under unconfined management and is therefore rarely practiced by the owners (Ahlers, 1999, Christensen, 2000, Permin and Bisgaard, 1999). It may be roughly calculated that the annual economic impact of diseases on Small holder poultry Production amounts to at least United States $ 500 million at a market value of United States cents 50 per Kilogram (Permin and Madsen, 2002).

2.2.1 Salmonellosis
Bacteria of the genus salmonella have long presented serious challenges to the poultry industry and are responsible for significant health problems in non poultry avian species as well (FAO, 2002; WHO, 2002). In poultry, high prevalences have been reported from Thailand and Nigeria (Adesiyun et al., 1984; Aini, 1999). While prevalences of 5-10% have been reported from other
parts of Asia and Africa (Sato et al., 1997; Chrysostome et al., 1995). Salmonella infections are of three types i.e pullorum disease, fowl typhoid, and paratyphoid.

Caused by *Salmonella pullorum*, pullorum disease is an infectious, egg-transmitted disease of poultry, especially chicks and turkey poults, often characterized by white diarrhea and high mortality in young birds and by asymptomatic adult carriers (Kahn et al., 2005). Sale of exposed but apparently healthy birds to many different purchasers can result in widespread dissemination of the etiologic agent adult carriers also shed the organisms in their faeces. Sick birds appear sleepy and weak. There is anorexia, white adherent diarrhea with pasting of the vent area, huddling near heat sources and shrill chirping (Butcher, 2009). Mortality varies greatly but often is very high and can approach 100% some times mortality may be surprisingly low and the disease may go unrecognized. Insofar as chemotherapy perpetuates the carrier state, treatment of pullorum-infected birds is indefensible and should not be recommended under any circumstance (FAO, 2002).

Fowl typhoid is an infectious disease, caused by *Salmonella gallinarum* and affects primarily chickens and turkeys. The colour of the comb and wattles becomes dark red; the droppings become yellow and the birds close their eyes and keep their heads down. Usually the affected chickens die within three to six days (Butcher, 2009).

Paratyphoid infection mainly caused by 10-20 species of Salmonella including *Salmonella typhimurium* (Porter, 1998). It occurs in many kinds of birds and mammals occur frequently in poultry. In countries with intensive poultry systems, poultry meat and eggs are a major source of infection for humans. These bacterial infections are of much more importance for public health impact than for economic losses in the affected animals (FAO, 2002; WHO, 2002). Clinical signs include isomnolence, profuse diarrhea followed by dehydration, pasting or wetting of the vent area, drooping wings, shivering, and huddling near heat sources. There usually is high morbidity and mortality especially during the first 2 weeks of brooding (Porter, 1998).
2.2.2 Staphylococcosis
Staphylococcosis is a systemic disease of birds characterized most frequently by purulent arthritis and tenosynovitis. Staphylococcal infections of poultry occur worldwide and affect all classes of birds. Outbreaks are most important in turkeys and broilers. The organisms are common in the environment and are especially associated with the skin (Ondwasy et al., 2006; Butcher, 2009). Most diseases produced by staphylococcus sp are associated with a break in the skin or beak (trauma, beak trimming, toe trimming etc). Toxigenic strains capable of causing food poisoning can contaminate the skin of professed poultry. The source of these strains is usually from the processing plant environment or workers (Kahn et al., 2005).

2.2.3 Colibacillosis
Avian colibacillosis is an infectious disease in birds in which *E. coli* is the primary or secondary pathogen. *Escherichia coli* Strains cause a number of diseases in domestic poultry, leading to diseases and death, or to a decrease in egg production or condemning of carcasses (Barnes et al., 2003). Colibacillosis occurs in all types and age groups of poultry as well as in other birds and many kinds of mammals. Most reported outbreaks in poultry have been in chicken, turkeys, and ducks. It is transmitted through contamination of egg shell with pathogenic *E. coli* from poultry house or hatcher environment. It is one of the most serious threats to broiler chicken flock between 4 and 5 weeks of age with respiratory signs, omphalitis and septicemia in baby chicks causing high mortality rates (AAAPG, 2005). Due to the welfare concerns for the birds involved and the high economic loss, infections caused by *E. coli* are of great importance and disease control has been complicated over recent years by the increasing frequency of antibiotic resistance (Vandemaele et al., 2002).

2.2.4 Fowl cholera (cholera, pasteurellosis)
Fowl cholera is a contagious, widely distributed disease caused by *Pasteurella multocida* that affects domestic and wild birds. Chronically infected birds are considered to be a major source of infection. Dissemination of *P. multocida* within a flock is primarily by excretions from mouth, nose, and conjunctiva of diseased birds that contaminate their environment. Fowl cholera is a disease of many species of birds, including chickens, turkeys, geese, ducks, quail, canaries, and many wild and zoo birds (Kahn et al., 2005). It usually occurs as a septicemia of sudden onset
with high morbidity and mortality, but chronic and asymptomatic infections also occur. Fowl cholera occurs worldwide and is a relatively common disease. Prevalence records from Africa show a wide variation from less than 1% in Tanzania (Muhairwa et al., 2000) to 2% in Zimbabwe (Kelly et al., 1994).

### 2.2.5 Streptococcosis

Streptococcosis in avian species is worldwide in distribution, occurring as both acute septicemic and chronic infections. *Streptococcus* spp isolated from avian species and associated with disease include *S. gallinarum* and *S. dysgalactiae*. Infection is considered secondary, because streptococci may form part of the normal intestinal and mucosal flora of most avian species, including wild birds, and are commonly found in various poultry environments. Mortality varies, but may reach 50%. It has been reported that an outbreak of streptococcal skin infection occurred in 1978 in a factory which undertakes the slaughter, preparation and parking of chickens (Barnham et al., 1980). Streptococcus bacteria may therefore be of public health importance. Research which could help in the prevention and treatment of these diseases was therefore carried out.

### 2.3 Ethno-veterinary medicine in management of poultry diseases

Ethno-veterinary medicine (EVM) is the community-based local or indigenous knowledge and methods of caring for, healing and managing livestock. This also includes social practices and the ways in which livestock are incorporated into farming systems. Ethno-veterinary medicine consists of local peoples’ knowledge dealing with folk beliefs, skills, methods and practices pertaining to animal health care and production. This knowledge is based on close observation of animals and/or the oral transmission of experience from one generation to the next (Mathias-Mundy and McCorkle, 1989). It recognizes the cultural context of traditional practices of livestock management and marks the beginning of systematic exploration of local practices for the development of livestock population (Kamal and Anil, 2004).

Livestock owners have an excellent knowledge of ethno veterinary, which has formed the basis for screening plant materials as potential sources of medical drugs (Matekaire and Bwakura,
Despite such successes, very little of this traditional knowledge has been documented in developing countries, and ethnoveterinary knowledge has had no place in mainstream veterinary medicine (Tafara et al., 2004). In addition to this, there are problems associated with herbal medicine preparation that result into contamination of herbal drugs and can also lead to herb drug interactions. These involve the crude preparation ways, heavy microbial load resulting from the field plant contamination as well as stability and shelf life determination in terms of stabilization and preservation, particularly in liquid dosage forms that pose important fundamental challenges (Elujoba et al., 2005). There are also physical factors such as air, humidity, light and temperature can bring about deterioration directly or indirectly. These factors can lead to development of organisms such as molds, mites and bacteria. Oxidation of the constituents of a drug can be brought about by oxygen in air, causing some products, such as essential oils to resinify or to become rancid (Iqbal et al., 2006). This research therefore sought to bring out the hidden potentials that ethno-veterinary medicine has got, so as to increase its acceptance and utilization by the community. Formal research would also help to confirm the claims made by traditional healers with respect to the efficacy of their remedies and to find out if there are no possibilities of chemical, microbial and environmental contamination.

2.3.1 Economic contributions of ethno-veterinary medicine

Today, ethno-veterinary medicine remains an ethno scientific resource that is yet to be tapped and has a far-reaching implication on the economic development and enhancement of veterinary health of particularly rural poor communities, which do not have access to modern medical services. Traditional medicine is easily accessible by people of all levels, and sometimes not costing anything and can be obtained through barter trade which is very convenient for rural people (Wanzala, 2005). Traditional medicine has a lower cost for both its medicines and consultations than that of industrially produced pharmaceuticals (Rangnekar, 1997). Herbs represent one of the first pharmacological interventions attempted by healers, and even today, 25% of our conventional drugs are plant derived in a traditional format (Spore, 1992).

The industrially produced pharmaceuticals are strongly believed by a bigger proportion of rural and peri-urban people to have more damaging adverse side effects and produce more toxic residues in food than the natural drugs prepared in traditional formats while there are no harmful
effects in most cases with the use of traditional medicine (Rangnekar, 1997). With traditional medicine, there is locally available manpower, materials and equipments and good rapport in view of long association (Rangnekar, 1997). It is also postulated that reviving indigenous knowledge within communities, and its transfer between communities, can provide opportunities for sustainable and cost-effective solutions (Mathias-Mundy and Perezgrovas, 1997).

2.3.2 Poultry ethno medicine in some African countries

To a certain extent, some research has been carried out on poultry ethno medicine in Uganda and Africa at large. In Uganda farmers have traditionally used many plants to treat their chickens for example Capsicum frutescens, Cannabis sativa, Nicotiana tabacum, Vernonia amygdalina (Saimo et al., 2003; Bukenya, 2007; Olila et al., 2007) among others. Olila et al.,( 2007) reported that plants in Mount Elgon region are used to treat cough, diarrhea, worms, new castle disease, listilelessness and for prophylaxis. In Southern and Eastern Africa, it has been reported that watery extracts of Nicotiana glauca can help a chick embryo infected with influenza to survive (Watt and Breyer−Brandwijk, 1962). In Senegal, farmers have traditionally used such plants to treat their chickens against endoparasites, for example, Capsicum sp extracts and the leaves or barks of Azadirachta indica, Azadirachta. Juss are added to drinking water and given to birds. In Cameroon, good results were reported on the use of plants such as Kalanchoe crenata for coccidiosis, and pawpaw (Carica papaya) leaves for diarrhea (Agbédé et al., 1995). In Togo, farmers use various infusions (e.g. Peltophorum ferrugineum), ground pepper, and the bark of Adansonia digitata to treat diarrhea in village chickens (Lobi, 1984). Good results have been obtained after the use of the Butyrospermum parkii (or ‘karité’) oil to control various ectoparasites such as ticks, lice’s and small red ants (Lobi, 1984). According to farmers, this oil obstructs the respiratory system of the parasites. In Nigeria, poultry owners grow certain repellent plants or place sliced garlic (Allium sativum) around hen houses to keep off snakes (Ibrahim, 1996). In Nigeria the spiny fruits of Cucumis pustulatus are also placed in the drinking water of chicks to protect them against hawk attacks (Ibrahim and Abdu, 1996).

However, most of the research carried out so far in poultry ethno medicine has left gaps in the pharmacological properties and phytochemical composition of these plants. Some important
plants have also not been documented. This research therefore sought to fill some of these missing gaps.

2.3.3 Ethno veterinary medicine today
In developing countries, low-income people such as farmers, people of small villages and native communities use folk medicine for the treatment of common infections (Rojas et al., 2006). With the rise of modern medicine during the last two centuries, traditional animal health care has increasingly been superseded by modern veterinary medicine (McCorkle, 1986). This view has begun to change in light of emerging drawbacks and shortcomings of modern medicine. In developing countries modern veterinary medicine cannot deliver complete coverage in preventive and curative health care practices because of inadequate labor, logistic problems, and an erratic supply of drugs. Consequently, the majority of those raising stock in rural areas are far from the site of veterinary stations, and those who have access may not be able to afford to pay for them (Sori et al., 2004).

Training in African veterinary colleges and universities is usually based on Western models of veterinary education. However, the significance of ethnoveterinary knowledge is gaining increasing recognition even among representatives of mainstream animal science. If sufficient attention is paid to these alternative medical traditions during veterinary training, it may go a long way toward preparing students for practice as well as fostering the sustainable use of natural resources available within their local communities (Tafara et al., 2004).

2.4 Botanicals as a source of antibacterials
Botanical drugs are derived from specific plant organs of a plant species. The following plant organs are the most important: aerial parts of the herb, leaf, flower, Fruit, Bark, root, rhizome and bulb. A large majority of botanical drugs are derived from leaves or aerial parts (Heinrich et al., 2004).

It has been reported in some studies that prevalence in the use of leaves compared with other plant parts for the preparation of traditional herbal remedies is the highest (Yineger and Yewhalaw, 2007; Pradhan and Badola, 2008). Plants have served as models in drug development
for three reasons: (i) Each plant is a unique chemical factory capable of synthesizing large numbers of highly complex and unusual chemical substances. (ii) The biologically active substances derived from plants have served as templates for synthesis of pharmaceuticals. Such compounds may have poor pharmacological and toxicological profiles. (iii) Many highly active secondary plant constituents have been instrumental as pharmacological tools to evaluate physiological processes (Farnsworth et al., 1985).

Even though pharmacological industries have produced a number of new antibiotics in the last three decades, resistance to these drugs by microorganisms has increased (Santos et al., 1995). The inappropriate and indiscriminate use of antibiotics exerts a selective pressure among bacteria, encouraging the appearance of drug-resistant strains. This is an issue of major concern, especially in medical microbiology because of the increasing incidence of multiresistant bacterial infections caused by both gram positive and gram negative bacteria (Iqbal et al., 2006).

According to World Health Organization medicinal plants would be the best source to obtain a variety of drugs. About 80% of individuals from developing countries use traditional medicine, which has compounds derived from medicinal plants (Santos et al., 1995). Herbs and herbal medicine preparations have been used to treat ailments throughout the history of humanity (Iqbal et al., 2006). Therefore, such plants should be investigated to better understand their properties, safety and efficiency (Ellof, 1998). The antibacterial activity and phytochemical composition of plants used in the treatment of poultry diseases was investigated to understand some of their properties.

### 2.5 Phytochemicals under study

The use of plant extracts and phytochemicals, both with known antimicrobial properties, can be of great significance in therapeutic treatments. In the last few years, a number of studies have been conducted in different countries to test their efficiency (Artizzu, et al., 1995). Many plants have been used because of their antimicrobial traits, which are due to compounds synthesized in the secondary metabolism of the plant. These products are known by their active substances. Plants have many ways of generating antibacterial compounds to protect them against pathogens (Kuc, 1990).
External plant surfaces are often protected by biopolymers eg. waxes, fatty acid esters such as cutin and suberin. In addition, external tissues can be rich in phenolic compounds, alkaloids, diterpenoids, steroid glycoalkaloids and other compounds, which inhibit the development of fungi and bacteria (Kuc, 1985). Cell walls of at least some monocotyledons also contain antimicrobial proteins, referred to as thionins (Carr and Klessing, 1989). Plant cells contain sequestered glycosides and release them when ruptured by injury or infection. These glycosides may have antimicrobial activity against the invading pathogens or may be hydrolysed by glycosidases to yield more active aglycones in the case of phenolic compounds, these may be oxidized to highly reactive antimicrobial quinines and free radicals (Dean and Kuc, 1987). Thus damage to a few cells may rapidly create an extremely hostile environment for a developing pathogen. Phytochemical screening of bioactive plants extracts has revealed the presence of alkaloids, carbohydrates, lactones, proteins, tannins, flavanoids, sterols, terpenes, glycosides, and saponins, of these, flavonoids and tannins have been linked to antibacterial activity and antidiarrheal activity (Ahmad et al., 2006).

Different phytochemicals display various mechanisms of action such as increasing colonic water and electrolyte reabsorption and inhibiting intestinal motility, while some components have been shown to inhibit specific pathogens (Ahmad et al., 2006). Phytochemicals such as saponins, terpenoids, flavonoids, tannins, steroids and alkaloids have anti-inflammatory effects (Lui, 2003). Steroids and triterpenoids have analgesic properties (Sayyah et al., 2004 and Malairaian et al., 2006). The phytochemical composition of the plants used in the treatment of poultry diseases would be qualitatively determined to identify such compounds.

2.5.1 Higher fatty acids and glycerides
Fatty acids are very important as formulations agents and vehicles in pharmacy and as components of cosmetics and soaps. They are precursors for biosynthesis of cholesterol. Examples include: Oleic acid which is very common in nature; α-linolenic acid which is used in liniments; δ-linolenic acid which is used as a dietary supplement, a precursor to prostaglandins, involved in many biochemical pathways and aids in alleviating symptoms associated with multiple sclerosis and premenstrual tension; ricinoleic acid which is used as a purgative. They are beneficial as antioxidants (Heinrich et al., 2004).
2.5.2 Basic alkaloids and alkaloid Salts
Alkaloids have pharmacological effects and could be associated with inhibition of nucleic acid, protein, and membrane phospholipids biosynthesis (Shelton, 1991). Berberine is an important representative of the alkaloid group. The mechanism of action of highly aromatic planar quaternary alkaloids such as berberine and harmane is attributed to their ability to intercalate with DNA (Phillipson and O'Neill. 1987). Alkaloids have been found to possess antimicrobial properties (Osborn, 2003). Diterpenoid alkaloids, commonly isolated from the plants of the Ranunculaceae family (Atta-ur-Rahman, and Choudhary, 1995), are commonly found to have antimicrobial properties (Omulokoli et al., 1997).

2.5.3 Flavonosides (Flavone glycosides) and Flavone aglycones
The different classes within the group are distinguished by additional oxygen containing heterocyclic and hydroxyl groups. These include the catechins, leucoanthocyanidins, flavanones, flavanonols, flavones, anthocyanidins, flavonols, chalcones, aurones, and isoflavones (Kaufman et al., 1999). Flavonosides being phenollic compounds are water soluble antioxidants and free radical scavengers which are capable of preventing oxidative cell damage and have strong anticancer activity (Okwu, 2004). Many disease states are known to be exacerbated by the presence of free radicals such as superoxide and and hydroxyl and flavonoids have the ability to scavenge and effectively mop up these damaging oxidizing species. Alan and and Miller, (1996) also reported that the potent antioxidant activity of flavonoids, their ability to scavenge hydroxyl radicals, superoxide anions and lipid peroxide radicals may be the most important function of flavonoids. Catechins, the most reduced form of the C₃ unit in flavonoid compounds occur in oolong green teas. They therefore have important dietary significance because they are strong antioxidants (Kaufman et al., 1999).

Since they are known to be synthesized by plants in response to microbial infection, it should not be surprising that they have been found in vitro to be effective antimicrobial substances against a wide array of microorganisms. Their activity is probably due to their ability to complex with
extracellular and soluble proteins and to complex with bacterial cell walls. More lipophilic flavonoids may also disrupt microbial membranes (Tsuchiya et al., 1996). These compounds inhibited in vitro Vibrio cholerae O1 (Borris, 1996), Streptococcus mutans (Batista et al., 1994), Shigella (Vijaya et al., 1995), and other bacteria and microorganisms (Sakanaka et al., 1992). The catechins inactivated cholera toxin in Vibrio (Borris, 1996) and inhibited isolated bacterial glucosyltransferases in S. mutans (Nakahara et al., 1993) possibly due to complexing activities. This latter activity was borne out in in vivo tests of conventional rats. When the rats were fed a diet containing 0.1% tea catechins, fissure caries (caused by S. mutans) was reduced by 40% (Ooshima et al., 1993). Flavonoids have been shown to have antibacterial, anti-inflammatory, antiallergic, antimitogenic, antiviral, antineoplastic and vasodilatory activity (Alan and Miller, 1996).

2.5.4 Cyanide glycosides
Cyanide glycosides are used by plants as a chemical defense. These compounds in presence of enzymes such as β-glucosidase, lose their sugar portion to form a cyanohydrin which, in the presence of water, can undergo hydrolysis to give benzaldehyde and the highly toxic hydrogen cyanide. They are produced in plants like cassava (Heinrich et al., 2004). In medicine they are used as counter irritants.

2.5.5 Coumarins and coumarin derivatives
They are commonly found in plant families Apiaceae, Rutaceae, Asteraceae and Fabaceae. An example is the hydroxylated coumarin dimer known as dicoumarol which is used as an anticoagulant. It is also responsible for sweet clover poisoning, a form of haemorrhaging that affects animals (Heinrich et al., 2004). Hydroxycinnamic acids, related to coumarins, seem to be inhibitory to gram-positive bacteria (Fernandez et al., 1996). All in all, data about specific antibiotic properties of coumarins are scarce, although many reports give reason to believe that some utility may reside in these phytochemicals (Hamburger and Hostettmann, 1991).
2.5.6 Terpenes
They are very widespread in nature. The terpenes are a perfect example of a natural product class that is highly structurally diverse, has many members that are chiral and have extensive functional group chemistry (Heinrich et al., 2004). The general chemical structure of terpenes is $\text{C}_{10}\text{H}_{16}$, and they occur as diterpenes, triterpenes, and tetraterpenes ($\text{C}_{20}$, $\text{C}_{30}$, and $\text{C}_{40}$), as well as hemiterpenes ($\text{C}_{5}$) and sesquiterpenes ($\text{C}_{15}$) (Heinrich et al., 2004).

2.5.6.1 Triterpenes and Triterpene glycosides
There are several important groups of triterpenes, including common triterpenes, steroids in mammals, sterols in plants, sterol aglycones, saponins, and cardiac glycosides (Kaufman et al., 1999). Triterpenes are also components of resins and resinous exudates from plants. Azadirachtin, a powerful insect antifeedant from neem oil is a triterpene (Kaufman et al., 1999). Many of the terpenoid components of these resins have antimicrobial activity (Heinrich et al., 2004). Cadionotics have a profound effect on the heart rhythm. The aglycone portion is steroidal in nature and some times referred to as cardenolide, being cardioactive in nature and possessing an alkene and an olide. The aglycone portion is derived from triterpenes and these compounds may have a wide variety of sugars attached to the steroid portion. Examples are digixin and ditoxin found in foxglove used to treat congestive heart failure (Heinrich et al., 2004).

Saponins have a wide distribution in plants and are referred to as saponins as they have soap-like properties and readily form foams. Saponins have a property of precipitating and coagulating red blood cells (Okwu and Josiah, 2006). Medicinally important examples include glycyrrhizic acid used in the treatment of stomach ulcers. Studies have shown that saponins although nontoxic can generate adverse physiological responses in animals that consume them. They exhibit cytotoxic effect and growth inhibition against a variety of cells making them have anti-inflammatory and anticancer properties. Triterpene glycosides are steroid in nature and over use can lead to similar symptoms associated with steroid over use such as hypertension and thrombosis (Heinrich et al., 2004). Steroidal compounds are of importance and interest in pharmacy due to their relationship with such compounds like sex hormones (Okwu, 2001). Sterols posses antibacterial and antmicotic activity and have been shown to act as inhibitors of tumor promotion in vivo (Yasukawa et al., 1991). Sterols were found to inhibit tumor promotion in two-stage
carcinogenesis in mice (Kasahara et al., 1994). Sterols also exhibit inhibitory effect on HIV reverse transcriptase (Akihisa et al., 2001). Sterols were also shown to poses anti-inflammatory activity after topical application (Gomez et al., 1999).

2.5.6.2 Tetraterpenes (carotenoids)
Members of this class are some times referred to as carotenones or carotenoids because of their occurrence in the carrot. They are highly pigmented natual products and are responsible for very bright colours of some plants, in particular, orange of carrots due to β-carrotine and the brilliant colour of tomatoes and peppers which is due to lycopene and capsanthin, respectively. The tetraterpenes are strong antioxidants, being preferentially oxidized over biological molecules such as nucleic acids and proteins. It is thought that many disease states such as certain cancers and heart disease are excerbated by species that cause oxidation; therefore the presence of these compounds may retard the development of such diseases. The tetraterpenes are precursors of vitamin A (Heinrich et al., 2004).

2.5.6.3 Monoterpenes
Together with phenylpropenes, the monoterpenes are major constituents of the volatile oils that are common in plants and which contribute to their aroma. This group of compounds has highly characteristic odours and tastes and are used in the food and cosmetic industries as flavourings and perfumes. Volatile oils in plants are highly complex and analysis by gas chromatography can show the presence of hundreds of individual components, many of which are monoterpenoid. Examples include linalool which is used as flavouring and a carminative, α-terpineol. 1,8-cineole, which has antibacterial properties, menthol and menthone used as flavourings and carminatives, thujone which is used as an antihelmintic, α-Pinene which is used as an antiseptic, and camphor which is also used as an antiseptic (Heinrich et al., 2004).

2.5.7 Phenylpropenes
The phenylpropenes consist purely of an aromatic ring with unsaturated 3-carbon chain attached to the ring. They are biosynthesised by the oxidation of phenylalanine by the enzyme phenylalanine ammonia lyase, which through the loss of ammonia results in formation of cinnamicacid (Heinrich et al., 2004). Cinnamic and caffeic acids are common representatives of a wide group
of phenylpropane-derived compounds which are in the highest oxidation state. The common herbs tarragon and thyme both contain caffeic acid, which is effective against bacteria (Brantner, et al., 1996). Cinnamic acid may undergo a number of elaboration reactions to generate many of the phenylpropanes. Phenolic compounds possessing a C$_3$ side chain at a lower level of oxidation and containing no oxygen are classified as essential oils and often cited as antimicrobial as well.

Catechol and pyrogallol both are hydroxylated phenols, shown to be toxic to microorganisms. Catechol has two hydroxyl groups, and pyrogallol has three. The site(s) and number of hydroxyl groups on the phenol group are thought to be related to their relative toxicity to microorganisms, with evidence that increased hydroxylation results in increased toxicity. In addition, some authors have found that more highly oxidized phenols are more inhibitory (Scalbert, 1991). The mechanisms thought to be responsible for phenolic toxicity to microorganisms include enzyme inhibition by the oxidized compounds, possibly through reaction with sulfhydryl groups or through more nonspecific interactions with the proteins. The phenylpropanes are produced by steam distillation of plant material to produce an essential oil which is normally a complex mixture of phenylpropanes and other volatile natural products such as hemiterpenes, monoterpenes, and sesquiterpenes (Heinrich et al., 2004).

### 2.5.8 Polyphenols

Poly phenols occur frequently in plants mostly in the bark, leaf and fruits and they do occur in the form of tannins (Agyare et al., 2006). They are found in almost every plant part: bark, wood, leaves, fruits, and roots (Scalbert, 1991.). Tannins give rise to astringency and bitterness. "Tannin" is a general descriptive name for a group of polymeric phenolic substances capable of tanning leather or precipitating gelatin from solution, a property known as astringency. The group comprises of water soluble polyphenolic compounds. They comprise of hydrolysable tannins and non-hydrolysable tannins. A key feature of tannins is their ability to bind to proteins, which may reduce the dietary value of a plant as food. Tannins have been found to form irreversible complexes with proline-rich proteins resulting in the inhibition of the cell protein synthesis, they bind proteins and adhesins, inhibit enzymes and complex with cell wall (Iqbal et al., 2006). Tannic acid which is a mixture of gallic acid esters of glucose can be used as a topical preparation for cold sores (Heinrich et al., 2004).
Many human physiological activities, such as stimulation of phagocytic cells, host-mediated tumor activity, and a wide range of anti-infective actions, have been assigned to tannins (Haslam, 1996). One of their molecular actions is to complex with proteins through so-called nonspecific forces such as hydrogen bonding and hydrophobic effects, as well as by covalent bond formation (Haslam, 1996). The ability of tannin compounds to cause the bacterial colonies to disintegrate probably results from their interference with the bacterial cell wall; thereby inhibiting the microbial growth (Viljoen et al., 2003; Erasto et al., 2004). Thus, their mode of antimicrobial action may be related to their ability to inactivate microbial adhesins, enzymes, and cell envelope transport proteins, they also complex with polysaccharide. The antimicrobial significance of this particular activity has not been explored. There is also evidence for direct inactivation of microorganisms: low tannin concentrations modify the morphology of germ tubes of Crinipellis perniciosa (Brownlee et al., 1990).

2.5.9 Athracenocides and athracenocide aglycones (emodols)
A number of plants containing these glycosides are known for their laxative properties. Examples are aloe, senna, danthrone, rhubarb, frangula and cascara. The gel from aloe is rich in polysaccharides and these anthraquinone glycosides are incorporated into creams and ointments to treat abrasions, burns and skin irritation. Senna is widely prescribed for constipation (Heinrich et al., 2004). Cascara contains an emetic principle. Cascara is a purgative, mainly in the form of liquid extract, elixir or as tablets prepared from a dry extract. The purgative action of anthracene bearing drugs is owed to their anthracene glycosidal content rather than their content of free anthracene aglycones (i.e., glycosylation is the main requirement for activity, as the sugar moiety serve to transport the aglycone to the site of action in the large intestine).

2.4 Plant Families under study
Several studies have been carried out on plants to demonstrate there activities and phytochemical composition. In the following sections, some families which have been investigated in the study carried out are assessed.
Abiatae
*Tetradenia riparia* belongs to this family and is used to treat poultry diseases. Deacetylboronolide 3 and (+)-dideacetylboronolide 4 were obtained from *Tetradenia riparia*, a central African species widely used as a tribal medicine (Van Puyvelde *et al.*, 1979). The importance of this plant in the treatment of helminthiasis in goats in Uganda has already been reported (Saimo *et al.*, 2003). Traditional Zulu uses of the leaves of *Tetradenia riparia* are mainly for respiratory complaints, malaria, dengue fever, flu, and diarrhea, and also as an inhalant for headaches. Research has also shown that *Tetradenia riparia* has antibacterial and anti-fungal effects and some anti-malarial activity. Zulu used roots of these plants as an emetic, and an infusion of leaves has been reported to be effective against malaria (Watt *et al.*, 1962).

Acanthaceae
*Justica betonica* belongs to this family and is used to treat poultry diseases. It has been reported that *Justica betonica* is used to treat ailments in both humans and livestock in Ethiopia (Bekalo, 2009).

Agavaceae
*Agave sasalina* belongs to this family and is used to treat poultry diseases. The petroleum ether extract of *Agave sasalina* has been found to have activity against *Bacillus subtilis* while the methanol extract of *Agave sasalina* has no demonstratable antibacterial activity (Olila *et al.*, 2007). *Agave sisalana* exported by Tanzania is rich in hecogenin, employed for the partial synthesis of steroidal drugs such as corticosteroids and oral contraceptives (Elujoba *et al.*, 2005). Crude methanol extracts of *Agave sisalana*, extracts showed antibacterial activity against *Staphylococcus aureus* and *Bacillus cereus* by agar diffusion method (Kassu *et al.*, 1999).

Amaranthaceae
*Chenopodium oplifolium* belongs to this family and is used to treat poultry diseases. *Chenopodium oplifolium* is also used by the community Budiope County in Uganda to treat Malaria in human beings (Tabuti, 2006).
Apocynaceae

*Catharanthus roseus* belongs to this family and is used to treat poultry diseases. The ethanol extract of *Catharanthus roseus* has demonstrated wound healing properties and antibacterial activity against *Pseudomonas aeruginosa* and *Staphylococcus aureus* (Nayak and Pereira, 2006). Previous Studies also demonstrated the presence of tannins, triterpenoids and alkaloids (Nayak and Pereira, 2006). The plant has more than 70 types of alkaloids (mostly monoterpen indole alkaloids), and some are known to be effective in treating various types of cancers including breast and lung cancer, uterine cancer, melanomas, and Hodgkin's and non-Hodgkin's lymphoma. The anticancer drugs vincristine and vinblastine are synthesized from alkaloids of *Catharanthus roseus* (El-Sayed and Cordell, 1981; Goyal et al., 2008).

Asteraceae

*Artemesia annua, Senecio cydonii folius, Aspilia Africana, Tithonia diversifolia, Tagetes minuta, Bidens pilosa, Vernonioa auriculifera, Vernonioa amygdalina, Vernonioa cineria and symphytum asperum* belong to this family and are used to treat poultry diseases. A typical chemical trait of this family is the presence of polyfructanes as storage carbohydrates. In many taxa, some segments of the family accumulate sesquiterpene lactones which are important natural products responsible for the pharmacological effects. Polycetylenic compounds and essential oils are also widely distributed. Some taxa accumulate pyrrolizidine alkaloids. Many of these alkaloids are known for their hepatotoxic effects. Other taxa accumulate unusual diterpenoids, the diterpene glycoside stevioside is of interest because of its intensely sweet taste (Heinrich et al., 2004).

Asteraceae plant family members are antiseptic, anti-inflammatory and soothing to the skin and the digestive system (Falsetto, 2009). The methanolic extracts and aqueous extracts of the leaves of *Aspilia Africana* have exhibited differential antibacterial activities on both gram-positive and Gram-negative bacterial species (Macfoy and Cline, 1990, Adeniyi and Odufowora, 2000). *Apiria Africana* is used in Africa to stop bleeding, promote healing of wounds and sores and treatment of cardiovascular diseases (Dimo, et al., 2002). The infusion of leaves of *Aspiria Africana* is used for treatment of stomach troubles in children (Okwu and Josiah, 2006). *Apiria Africana* is effective against malaria infection (Waakoa et al., 2005; Okokon et al., 2006). It has been reported that the crude root of *Vernonia amygdalina* has antimicrobial properties against in
gingivitis and toothache (Tella, 1976). The ethanol, cold water and hot water extracts of *vernonia amygdalina* have been reported to have antibacterial activity against *Staphylococcus aureus* and *Escherichia coli* (Okigbo and Mmeka, 2008). Several of the compounds of *vernonia amygdalina* are active against gram-positive *Bacillus subtilis* and *Micrococcus lutea*, but none are active against gram negative bacteria (Kaufman *et al.*, 1999). According to Olila *et al.*, 2007, the methanol extract and petroleum ether extract of *vernonia amygdalina* has no demonstratable antibacterial activity.

In a survey of antibacterial activity of South African herbs, *Bidens* was found with high activity against gram-positive bacteria (Rabe and van Staden, 1997). Several of the flavonoids found in *Bidens pilosa* have anti-inflammatory activity as revealed in laboratory animal studies (Geissberger and Sequin, 1991). There are two main groups of active constituents of bidens; polyacetylenes which inhibit various pathogenic organisms, and flavonoids, which reduce inflammation. The polyacetylenes can also manifest an anti-inflammatory action, probably mediated by a different mechanism than the flavonoids. Bidens also has friedelane triterpenes and essential oils which may contribute to the observed therapeutic action of the herb. The ethanolic leaf extract of *Tithonia diversifolia* was reported to be active against *Staphylococcus aureus* and *E.coli* (Liasu and Ayandele, 2008).

Jah Hut people often use *Vernoni cinerea* to relief asthma. Asthma is reversible airway obstruction associated with mucosal inflammation caused by mast cells and basophils degranulation resulting in the release of inflammatory mediator (Lin, 2005). Recent studies revealed the anti-inflammatory activities of *V. cinerea* (Iwalewa *et al.*, 2003).

**Cannabacea**

*Cannabis Sativa* belongs to this family and is used to treat poultry diseases. *Cannabis Sativa* has also been documented as one of the plants used by the people of East, Central and Southern Uganda to treat poultry diseases (Bukenya, 2007).
Caricaceae

Carica papaya belongs to this family and is used to treat poultry diseases. The milky juice of Carica papaya contains proteolytic ferments, which together with papain have successfully been used as an anthelmintic agent for the treatment of Ascariasis, Trichuriasis, and ancylostomiasis (Watt and Breyer-Brandrijk, 1962). In traditional veterinary medicine, carica papaya seeds are used to deworm poultry in Indonesia and Philippines (Satyanarayanana and Krishnaiah, 1982). Extracts of pulp and seeds showed bacteriostatic properties when tested against Staphylococcus aureaus, Escherichia coli, Salmonella typhi and Bacillus subtilis (Suhaila et al, 1994).

Clusiaceae

Gacina bunchananii belongs to this family and is used to treat poultry diseases. Gacina bunchananii is used by the people around Lake Victoria in Uganda to treat a number of diseases (Katumba et al, 2004).

Cucurbitaceae

Conyza floribunda and Mordica foetida belong to this family and are used to treat poultry diseases. Phytochemical screening of the crude extract of Momordica foetida showed the presence of various secondary metabolites which included alkaloids, flavonoids, terpenoids, steroids, saponins and tannins (Olukayode and Adebola, 2008). Olukayode and Adebola, 2008 also reported that the crude extracts of Momordica foetida have good antimicrobial activity. Momordica foetida, a multipurpose plant is used as an abortifacient and ecbolic, among others. The plant has been shown to exhibit some antimalarial activity and the root extract contains foetidin as a major chemical constituent (Waako et al., 2005). According to Olila et al., 2007, the methanol extract and petroleum ether extract of Momordica foetida had no demonstratable antibacterial activity.

Euphobiaceae

Euphobia hirta, Euphorbia heterophylla, Tragia brevipes, Ricinus communis and Jatropha carcus belong to this family and are used to treat poultry diseases. Some of the reported phytoconstituents of Euphobia hirta include triterpenoids, sterols, alkaloids, glycosides, flavonoids, tannins, phenols, choline and shikimic acid, (Adedapo et al., 2005; Falodun et al.,
2006). In east, central and west Africa, a decoction of *Euphobia hirta* is used to treat asthma, oral thrush, boils, sores, skin and wound infections, in addition to its being used as an antispasmodic, antipruritic, carminative, depurative, diuretic, febrifuge, galactogogue, purgative and vermifuge (Alabashi *et al.*, 1999; Palombo and Semple, 2001; Darwish *et al.*, 2002; Ogbolie *et al.*, 2007). In Nigeria, exudates of the stem of *Euphobia hirta* are used to treat eye and ear infections (Igoli *et al.*, 2005), while a decoction of the plant is used to treat enteric infections including diarrhea and dysentery, constipations and other stomach problems, asthma, bronchitis, eczema, athletes foot and scorpion bite pains (Ogbolie *et al.*, 2007). The methanol extract of *Jatropha carcus* has been found to have activity against *Bacillus subtilis* while the petroleum ether extract has no demonstratable antibacterial activity (Olila *et al.*, 2007).

**Fabaceae**

*Tephorosia vogelli* and *Abrus precatorius* belong to this family and are used to treat poultry diseases. This Family is characterized by an impressive phytochemical diversity. Polyphenols especially flavonoids and tannins are common. From a pharmaceutical perspective, various types of alkaloids are the most relevant groups of compounds. Isoflavonoids are also important because they are known for oestrogenic activity while the Coumarins are used as anticoagulants (Heinrich *et al.*, 2004).

**Lamiaceae**

*Ocimum basilicum* belongs to this family and is used to treat poultry diseases. Essential oil in the epidermal glands is very common. Some segments of the family are known to accumulate monoterpenoid glycosides. Many species also accumulate rosmarinic acid and other derivatives of caffeic acid. Rosmarinic acid is of some pharmaceutical importance because of its non-specific complement activation and inhibition of the biosynthesis of leukotrienes leading to anti-inflammatory effects as well as its antiviral activity (Heinrich *et al.*, 2004).

**Lauraceae**

*Persia americana* belongs to this family and is used to treat poultry diseases. Lauraceae plant family members are antifungal, anti bacterial, antiviral and either tonics or stimulants (Falsetto, 2009). *Persia americana* extracts have demonstrated antimicrobial activity against Mycobacteria
strains (Gomez-Flores et al., 2008). Persia Americana is recommended for anemia, exhaustion, hypercholesteremia, hypertension, gastritis and gastrodeodenal ulcer (Pamplona-Roger, 1999). The leaves have been reported as effective, antitussive, antidiabetic and relief for arthritis and pain by traditional healers of South Africa (Adeyemi et al., 2002).

Liliaceae
Aloe ferox and Aloe belong to this family and are used to treat poultry diseases. The methanol extract and petroleum ether extract of Aloe species has been found to have activity against Bacillus subtilis (Olila et al., 2007). Aloe species are used by people in Karamoja region of Uganda to treat malaria and relief of constipation (Nanyunja, 2003). The exudates of aloe contains emodin, and anthraquinone which is a gastrointestinal irritant, hence the purgative effects. An acetylated mannan, acemannan extracted from Aloe vera has been shown to have immune modulating effects. Most researchers report an initial increase in necrosis and then more rapid healing when compared to other treatments (Kaufman et al., 1999).

Malvaceae
Sida cuneifolia belongs to this family and is used to treat poultry diseases. Sida cuneifolia has been reported to have good antibacterial activity on both gram negative and gram positive bacteria (Van-Vuurena and Viljoenb, 2006).

Meliaceae
Azadirachta indica and Melia azedarach belong to this family and are used to treat poultry diseases. The methanol extract and petroleum ether extract of Azadirachta indica has been found to have activity against Bacillus subtilis, a gram positive bacterium (Olila et al., 2007). Azadirachta indica has been extensively used for treatment of inflammations, skin infections, fever, infections and dental disorders (Subapriya and Nagini, 2005). Extracts of Azadirachta indica seeds shows activity against multidrug resistant Salmonella enterica serovar Typhi isolates (Mwatha et al., 2001). It is also effective against Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa and Candida albicans (Mwatha et al., 2001). Azadirachta indica has antibacterial activity against a variety of micro-organisms such as Staphylococcus,
Enterococcus, Pseudomonas, Escherichia, Klebsiella, Salmonella and Mycobacterium (Fabry et al., 1998).

Mimosoideae

Albizia coriaria belongs to this family and is used to treat poultry diseases. The methanol extract of Albizia coriaria have been found to have activity against Pseudomonas aeruginosa, Bacillus subtilis and E. coli, while the petroleum ether extract has no demonstratable antibacterial activity (Olila et al., 2007). Albizia coriaria is used by people in Karamoja region of Uganda to treat fever and constipation (Nanyunja, 2003). The methanol extract of Albizia coriaria have been found to have moderate antiplasmodial activity (Kigondu et al., 2009).

Moringaceae

Moringa belongs to this family and is used to treat poultry diseases. Moringa oleifera has been cited to be antibacterial (Fluglie, 1999). Moringa oleifera has shown antibacterial activity against E. coli, Enterobacter, P. aeruginosa and Staphylococcus aureus (Nepolean et al, 2009). It has been reported that antibacterial activity of the ethanolic and aqueous extracts of Moringa oleifera against Salmonella typhi. In this report, the ethanolic extracts were more active than the aqueous extracts (Doughari et al., 2007). It has also been reported that various sub species of Moringa oleifera are known to exhibit antibacterial activity (Spiliotis et al., 1998). Olila et al., 2007 reported that the petroleum ether extract of Moringa aloevera had no demonstratable antibacterial activity. Moringa is rich in compounds containing the simple sugar rhamnose and in glucosinolates and isothiocyanates (Bennet et al., 2003). Phytochemical analysis has demonstrated presence of tannins, saponins, flavonoids, glycosides and terpenoids (Nepolean et al, 2009). The presence of these constituents has been reported to account for the exertion of antimicrobial activity by plants containing them (Pretorius and Watt, 2001).

Myrsinaceae

Maesa lanceolata belongs to this family and is used to treat poultry diseases. It has been reported that Maesa lanceolata is used to treat a number of diseases in Konta, Ethiopia (Bekalo, 2009).
Myrtaceae

Syzygium belongs to this family and is used to treat poultry diseases. Ashes of the bark, mixed with water, are spread over local inflammations, or, blended with oil, applied to burns (Morton, 1987). Syzygium cumini, has been valued for its medicinal use since ages. Every part of this tree including its seeds, bark, leaves and fruit are being traditionally used for medicinal purposes. For example, seeds are widely used in diabetes; leaves in anemia & gingivitis; leaves and fruits in diarrhoea, fever and abdominal pain (Laxmi et al., 2006). It has also been reported that Syzygium cumini has been pharmacologically proved to possess hyperglycemic, antibacterial, anti HIV and antidiarrheal effects (Ravi et al., 2004).

Papilionaceae

Desmodium salicifolium and Phaseolus lunatus belong to this family and are used to treat poultry diseases. Desmodium salicifolium is also one of the plants used by the people of Bugabo in Tanzania treat malaria (Moshi et al., 2009). It has also been reported that Desmodium salicifolium is used by the Nyamwezi people in Tanzania to treat Asthma (Moshi et al., 2006).

Rubiaceae

Rubia cordifolia belongs to this family and is used to treat poultry diseases. The family is known for a large diversity of classes of natural products, including iridoids which is a group of monoterpenoids, alkaloids including indole alkaloids, methylxanthine such as caffeine, theombrromine and theophylline, and anthranoids in some taxa (Heinrich et al., 2004). Chloroform and methanol extracts Rubia cordifolia showed activity on gram-positive strains and gram-negative P. aeruginosa (Subhalakshmi et al., 2005). Rubidianin, an anthraquinone isolated from alcoholic extract of Rubia cordifolia has demonstrated significant antioxidant activity. It prevented lipid peroxidation induced by ferrous sulphate and t-butylhydroperoxide. Rubidianin depicted activity in dose-dependent manner (Malhotra and Singh, 2007).

Solanaceae

Nicotiana tobacum, Solanum mauritianum, Datura stramonium, Capsicum annua, Solanum Incanum, Solanum nigram belong to this family and are used to treat poultry diseases. Typical of this family are alkaloids, especially of the tropane, nicotine and steroidal type. Many taxa are
characterized by oxalic acid which often forms typical structures for example irregular crystals in *Datura stramonium* (Heinrich et al., 2004). *Capsicum annum* produces capsaicin and capsacin, used as spice and medicine (Elujoba et al., 2005). In that study about bovine mastitis, it was reported that the claimed effect of *Solanum mauritianum* was due to the presence of cyclooxygenase which is an anti-inflammatory (Avancini et al., 2008).

**Verbenaceae**

*Lantana trifolia* belongs to this family and is used to treat poultry diseases. *Lantana trifolia* is used by the people of Bugabo in Tanzania to treat Malaria (Moshi et al., 2009). A lot of studies have been carried out on its close relative *Lantana camara* indicating presence of phytochemicals with healing properties and antibacterial activity. Cheruiyot et al., 2009 reported that the methanol extracts of *Lantana camara* exhibited antibacterial activity against *Staphylococcus aureus* and shown no activity against *Escherichia coli* and *Pseudomonas aeruginosa*. Methanol extracts of *Lantana camara* also has activity against *Bacillus subtilis* (Olila et al., 2007). Also Ganjewala et al., 2009 reported presence of alkaloids, phenols, terpenoids, phytosterols, saponins, tannins and steroids in different varieties *Lantana camara*.

**Zingiberaceae**

*Curcuma longa* belongs to this family and is used to treat poultry diseases. This family is rich in essential oils with terpenes such as borneol, camphor and cineole which are all oxygen containing monoterpenes, camphene, pinene which are monoterpenes and zingiberene which is a sesquiterpene andphenylpropanoids which are cinnamic acid derivatives (Heinrich et al., 2004). *Curcuma longa*, is a commonly used spice and a popular remedy used for inflammatory and liver diseases and in most Asian systems used for treatment of a large variety of illnesses (Heinrich et al., 2004). Curcuminoids, a group of phenolic compounds isolated from the roots of *C. longa*, exhibited a variety of beneficial effects on health and has the ability to prevent certain diseases (Joe et al., 2004). In East Asia, the rhizomes from *C. longa*, are considered to have natural medicinal properties, including antibacterial, anti-inflammatory, antineoplastic, and analgesic activities because they contains a number of moniterpenoids, sesquiterpenoids, and curcuminoids (Fang et al., 2003). In addition, wound healing and detoxifying properties of curcumin have also received considerable attention (Joe et al., 2004). Fraction II of the oil extract from the turmeric
oleoresin containing ar-Turmerone, turmerone, and curlone showed antibacterial activity by the pour plate method against *Bacillus cereus*, *Bacillus coagulans*, *Bacillus subtilis*, *Staphylococcus aureus*, *E. coli*, and *Pseudomonas aeruginosa* (Negi et al., 1999). Methanol extract of the dried powdered turmeric rhizome and curcumin inhibited the growth of all strains of *H. pylori in vitro* with a MIC range of 6.25-50 µg/ml (Mahady et al., 2002).
CHAPTER THREE

A SURVEY OF INDIGENOUS KNOWLEDGE ON POULTRY ETHNOMEDICINAL PLANTS IN MASAKA

3.1 Summary
In this chapter, a descriptive survey was carried out in Kyanamukaka and Buwunga in Masaka district of central Uganda. This was to identify and document knowledge about the plants which are commonly used in the treatment of a variety of poultry diseases. This was done by conducting focus group discussions and key informant interviews. The chapter therefore gives a description and documents the indigenous practices and plants used in the treatment of a variety of poultry diseases. Fifty nine (59) plant species from 33 families were found to be commonly used in the treatment of a variety of poultry diseases in this area. Plants were ranked according to the frequency of use and the most frequently used plants were Cannabis sativa and Nicotiana tobaccam. Family Asteraceae had the highest number of plant species used in treatment of poultry diseases and leaves were the most commonly used parts of the plants. Most of the plants were used for prophylaxis and the oral route was the most preferred route of administration. This study serves as a basis for identifying and documenting the plants used in the treatment of poultry diseases.

3.2 Introduction
Herbs and herbal medicine preparations have been used to treat ailments throughout the history of humanity (Iqbal et al., 2006). According to a study carried out by Makerere University researchers in central and eastern Uganda, about 80% of the poultry farmers surveyed know how to use medicinal plants to treat poultry diseases (Bukenya, 2007). Livestock owners have an excellent knowledge of ethnoveterinary, which has formed the basis for screening plant materials as potential sources of medical drugs (Matekaire and Bwakura, 2004). Despite such successes, very little of this traditional knowledge has been documented in developing countries, and ethnoveterinary knowledge has had no place in mainstream veterinary medicine (Tafara et al., 2004). In developing countries modern veterinary medicine can not deliver complete coverage in preventive and curative health care practices because of inadequate labor, logistic problems, and
an erratic supply of drugs. Consequently, the majority of those raising stock in rural areas are far from the site of veterinary stations, and those who have access may not be able to afford to pay for them (Sori et al., 2004). Therefore, conservation of indigenous knowledge, skills and resources will promote the well being of farmers. The main objective of this study was to identify the plant species and indigenous knowledge used by poultry farmers in Kyanamukaka and Buwunga in Masaka district. The plant species were then ranked according to frequency of use to determine the priority plants.

3.3 Materials and methods

3.3.1 Research design and study area
This was a descriptive survey aimed at collecting information from respondents on ethnomedicine used to treat poultry diseases. The study was carried out in two subcounties of Masaka district of Uganda deemed at high risk for avian influenza. The location of Masaka district in Uganda is shown in appendix V. Masaka comprises of 21 sub counties as shown in appendix VI and these are Kyanamukaka, Buwunga, Bukakata, Masaka municipality, Mukungwe, Lukaya, Bukulula, Lwabenge, Kyamulibwa, Bigasa, Kitanda, Butenga, Kalungu, Kibinge, Kkingo, Kabonera, Kiseka, Lwengo, Ndagwe, Kyazanga and Malongo. The study was conducted in two sub counties and these were Kyanamukaaka and Buwuunga sub counties. Kyanamukaaka comprises of nine parishes and these are Kamuzinda, Kyantale, Buyinja, Kitunga, Bugere, Kyesiga, Zzimwe and Buyaga in Kyanamukaka while Buwunga comprises of six parishes and these are Mazinga, Kitengesa, Buwunga, Kamwozi Kasaka, and Kajuna. The parishes where the study was carried out were Kamuzinda, Kyantale, Buyinja, Kitunga and Bugere in Kyanamukaka Sub county while Mazinga, Kitengesa, Buwunga and Kamwozi were chosen from Buwunga Sub County.

3.3.2 Sample study population
The study population consisted of households in Kyanamukaaka and Buwuunga sub counties. Workshops were held with 180 purposively chosen farmers in focus group discussions each comprising of 20 farmers. While interviews were held with 20 key informants identified during focus group discussions. Important information was documented on the plants used and how they
are used to prevent and treat poultry diseases. The output from this phase was used in the subsequent phase of laboratory analysis.

3.3.3 Sampling Techniques
Purposive sampling was used to select the study area and the farmers. Sub counties selected using predetermined criteria of level of vulnerability to Avian influenza. Factors used for vulnerability assessment included poverty levels, swampy areas, poultry density and existence of known migratory bird landing sites within the area. Using this criterion, two subcounties were selected, Kyanamukaaka and Buwuunga. Parishes selected were those with large numbers of farmers rearing local chicken and believed to carry out traditional indigenous practices. Using this criterion, four parishes were selected from Buwunga and five from Kyanamukaaka. The people selected were those members in the community who knew and practiced traditional methods for treating poultry.

3.3.4 Research Instruments
Qualitative data collection was aided by the use of focus group discussions, key informant interviews and observations. Copies of the sample interviews used are in appendix I. Observations involved actual participation to learn how farmers process these medicinal resources and the actual resources used.

3.4 Results
3.4.1 Plants used to treat poultry diseases in Masaka
The study identified fifty nine plants which were used by poultry farmers for medicinal purposes in Masaka District (Tables 1 and 2). These were used mostly as herbs and sometimes concoctions that are personally known by farmers or by consultation with neighbors. All in all the plants belonged to thirty three different families. Family Asteraceae had 30% of plants registering the highest frequency, followed by family Solanaceae with 18% of the plants, and family Euphobiaceae with 15% of the plants. The most widely used plants were Cannabis sativa, Capsicum annum, Nicotiana tobaccam and Momordica foetida among others (Tables 1and 2). The most common concoctions used by these farmers are in appendix II.
<table>
<thead>
<tr>
<th>Family name</th>
<th>Species name</th>
<th>Local name</th>
<th>Frequency</th>
<th>Medicinal Use (s)</th>
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Table 2: Least frequently mentioned plants

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<th>Species name</th>
<th>Local name</th>
<th>Frequency</th>
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<td>Kiyondoyondo</td>
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<td>Prophylaxis</td>
</tr>
<tr>
<td>Euphorbiaceae</td>
<td>Tragia brevipes</td>
<td>Kamyu</td>
<td>8</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Spigeliaceae</td>
<td>Spilanthes Mauritania</td>
<td>Kado kanamunye</td>
<td>7</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Solanaceae</td>
<td>Solanum Incanum</td>
<td>Mukutiza ngalabi</td>
<td>6</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Convolvulaceae</td>
<td>Ipomea batatas</td>
<td>Lumonde omuganda</td>
<td>5</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Brassicaceae</td>
<td>Brassica oleracea</td>
<td>Embogga</td>
<td>4</td>
<td>Immune booster</td>
</tr>
<tr>
<td>Euphobiaceae</td>
<td>Euphoria hirta</td>
<td>Akasandasanda</td>
<td>3</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Asteraceae</td>
<td>Artemisia annua</td>
<td>Antemeziya</td>
<td>2</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Oxalidaceae</td>
<td>Oxalis corniculata</td>
<td>Mukuuta sente</td>
<td>2</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Asteraceae</td>
<td>Tithonia diversifolia</td>
<td>Kitungotungo</td>
<td>1</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Asteraceae</td>
<td>Tagetes minuta</td>
<td>Kawunyira</td>
<td>1</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Apocynaceae</td>
<td>Catharanthus roseus</td>
<td>Sipurinko</td>
<td>1</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Euphorbiaceae</td>
<td>Euphorbia heterophylla</td>
<td>Kisandasanda</td>
<td>1</td>
<td>Prophylaxis</td>
</tr>
<tr>
<td>Fabaceae</td>
<td>Abrus precantourius</td>
<td>Obuusiti</td>
<td>1</td>
<td>Deworming</td>
</tr>
<tr>
<td>Zingiberaceae</td>
<td>Curcuma longa</td>
<td>Ebinzaali</td>
<td>1</td>
<td>Fever, Diarrhoea</td>
</tr>
<tr>
<td>Lamiaceae</td>
<td>Ocimum basilicum</td>
<td>Kafumbamwanyi</td>
<td>1</td>
<td>Fever</td>
</tr>
</tbody>
</table>

The information gathered from farmers about these plants included their medicinal uses, parts used, preparation methods, and administration routes. This information has been summarized in tables 3 and 4.
<table>
<thead>
<tr>
<th>Species name</th>
<th>Medicinal Use(s)</th>
<th>Parts used</th>
<th>Preparation</th>
<th>Administration</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cannabis sativa</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Capsicum annuum</td>
<td>Diarrhoea, Respiratory</td>
<td>Seeds</td>
<td>Powder</td>
<td>Oral</td>
</tr>
<tr>
<td>Nicotiana tabacca</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Momordica foetida</td>
<td>Prophylaxis, Respiratory</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Vernonia amygdalina</td>
<td>Fever</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Chenopodium opulifolium</td>
<td>Diarrhoea, Fever</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Vernonia cinerea</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Senecio cydoniifolius</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Aspilia Africana</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Solanum mauritianum</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Albizia coriaria</td>
<td>Prophylaxis</td>
<td>Bark</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td>Conyza floribunda</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Tetradenia riparia</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Aloe vera</td>
<td>Prophylaxis, Fever</td>
<td>Leaves</td>
<td>Juice,</td>
<td>Oral</td>
</tr>
<tr>
<td>Persea americana</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Desmodium salicifolium</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Justicia betonica</td>
<td>Fever</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Jatropha curcas</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Leonotis nepetifolia</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Phaseolus lunatus</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Lantana trifolia</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Sida cuneifolia</td>
<td>Diarrhoea</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Garcinia buchananii</td>
<td>Prophylaxis, Respiratory</td>
<td>Leaves</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td>Azadirachta indica</td>
<td>Prophylaxis, Fever</td>
<td>Leaves</td>
<td>Juice,</td>
<td>Oral</td>
</tr>
<tr>
<td>Syzygium cuminii</td>
<td>Prophylaxis</td>
<td>Bark</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td>Agave sisalana</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Bidens pilosa</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Moringa oleifera</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Rubia oleifera</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Datura stramonium</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td>Carica papaya</td>
<td>Prophylaxis, Deworming</td>
<td>Leaves,</td>
<td>Powder,</td>
<td>Oral</td>
</tr>
<tr>
<td></td>
<td></td>
<td>seeds,roots</td>
<td>decoction</td>
<td></td>
</tr>
</tbody>
</table>
Table 4: Usage of least frequently mentioned plants

<table>
<thead>
<tr>
<th>Species name</th>
<th>Medicinal Use(s)</th>
<th>Parts used</th>
<th>Preparation</th>
<th>Administration</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Ricinus communis</em></td>
<td>Prophylaxis</td>
<td>Seeds</td>
<td>Powder</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Digitaria abyssinica</em></td>
<td>Prophylaxis</td>
<td>Roots</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Aloe ferox</em></td>
<td>Prophylaxis, Fever</td>
<td>Leaves</td>
<td>Juice, Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Tephrosia vogelii</em></td>
<td>Deworming, Acaricide</td>
<td>Leaves</td>
<td>Powder</td>
<td>Oral, Topical</td>
</tr>
<tr>
<td><em>Allium sativum</em></td>
<td>Prophylaxis</td>
<td>Bulbs</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Tropaeolum majus.</em></td>
<td>Immune booster</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Fleurya aestuans</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Symphytum asperum</em></td>
<td>Immune booster</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Solanum nigrum</em></td>
<td>Prophylaxis</td>
<td>Roots</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Venonia auriculifera</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Maesa lanceolata</em></td>
<td>Deworming</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Melia azedarach</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Powder</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Kalanchoe glaucescens</em> Britten.*</td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Tragia brevipes</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Spilanthes Mauritania</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Solanum incanum</em></td>
<td>Prophylaxis</td>
<td>Whole</td>
<td>Smoking</td>
<td>Inhalation</td>
</tr>
<tr>
<td><em>Ipomea batatas</em></td>
<td>Prophylaxis</td>
<td>Roots</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Brassica oleracea</em></td>
<td>Immune booster</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Euphobia hirta</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Artemesia annua</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Oxalis corniculata</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Tithonia diversifolia</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Tagetes minuta</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Catharanthus roseus</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Euphorbia heterophylla</em></td>
<td>Prophylaxis</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Abrus precantorius</em></td>
<td>Deworming</td>
<td>Seeds</td>
<td>Powder</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Curcuma longa</em></td>
<td>Fever, Diarrhoea</td>
<td>Bulbs</td>
<td>Decoction</td>
<td>Oral</td>
</tr>
<tr>
<td><em>Ocimum basilicum</em></td>
<td>Fever</td>
<td>Leaves</td>
<td>Juice</td>
<td>Oral</td>
</tr>
</tbody>
</table>

Sixty eight (68) % of the plants were used for prophylaxis. The rest of the medicinal uses of the plants that is, prophylaxis, immune boosting, treatment of fever, diarrhea, respiratory infections, deworming and as acaricides have been indicated in Table 5.
Table 5: Summary of medicinal uses of plants

<table>
<thead>
<tr>
<th>Use(s) of the plants</th>
<th>Number of plants</th>
<th>Percentage number of plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prophylaxis</td>
<td>40</td>
<td>68</td>
</tr>
<tr>
<td>Immune boosting</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Treatment of fever</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Treatment of diarrhea</td>
<td>1</td>
<td></td>
</tr>
<tr>
<td>Prophylaxis and treatment of fever</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Prophylaxis and treatment of respiratory infections</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Treatment of Fever and diarrhea</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Treatment of diarrhoea and respiratory infections</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Deworming and as an acaricide</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Deworming and prophylaxis</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

Three different administrative routes were used for different plant preparations and these were oral route, topical route and inhalation after smoking of the plant parts as shown in Table 6. Ninety six (96) % of the plants were administered orally.

Table 6: Summary of routes of administration of medicine

<table>
<thead>
<tr>
<th>Routes of administration</th>
<th>Number of plants</th>
<th>Percentage number of plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral</td>
<td>58</td>
<td>96</td>
</tr>
<tr>
<td>Tropical / oral</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Smoking</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

For 80% of the plants, leaves were used to make the drug preparations while for some either the roots or the bark or the bulbs or the whole plant or the seeds were used as indicated in Table 7.
Table 7: Summary of plant parts used by poultry farmers

<table>
<thead>
<tr>
<th>Part (s) of the plant</th>
<th>Number of plants</th>
<th>Percentage number of plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leaves</td>
<td>47</td>
<td>80</td>
</tr>
<tr>
<td>Bark</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Bulbs</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>Roots</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Seeds</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Leaves/roots/seeds</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Whole plant</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

Seventy one (71) % of the plants were prepared as juice by poultry farmers. The rest of the methods that is, making decoctions, making powders, and smoking the plant were used as indicated in Table 8.

Table 8: Summary of methods of Drug preparation

<table>
<thead>
<tr>
<th>Preparation Methods</th>
<th>Number of plants</th>
<th>Percentage number of plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decoction</td>
<td>7</td>
<td>12</td>
</tr>
<tr>
<td>Juice</td>
<td>42</td>
<td>71</td>
</tr>
<tr>
<td>Decoction / juice</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Powder</td>
<td>6</td>
<td>10</td>
</tr>
<tr>
<td>Smoked dry plants</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

3.5 Discussion

In developing countries, low-income people such as farmers, people of small villages and native communities use folk medicine for the treatment of common infections (Rojas et al., 2006). In this study, fifty nine plants were identified to be used by poultry farmers for medicinal purposes in Masaka District. Results of this study confirmed the observation of earlier studies about the use of some of these plants in treatment of poultry diseases in Uganda (Saimo et al., 2003; Olila et al., 2007; Bukenya, 2007).

The plants identified in Masaka belonged to thirty three (33) different families. The families Asteraceae, Solanaceae and Euphobiaceae registered the highest number of plants. The most frequent use of plants belonging to these families can be attributed to their being widespread and easily accessed. Previous studies have reported that plants in theses families have important
pharmacological activities. Falsetto, (2009) has reported that members of family Asteraceae have antiseptic, anti-inflammatory and soothing properties to the skin and digestive system. Some plants in this family like Vernonia cineria and Aspilia africana demonstrated presence of phytochemicals like tannins, alkaloids and Steroids. Earlier Studies have reported that these phytochemicals exhibit medicinal properties (Lui, 2003). It is therefore not surprising that plants in this family are the most frequently mentioned therefore used plants by poultry farmers. It has further been reported that alkaloids are typical of Solanaceae plant family and alkaloids are pharmacologically important (Shelton, 1991; Osborn, 2003). This also can explain why plants in this family are also among the most commonly mentioned plants by farmers. Reports also say that some plants in family Ephoriaceae have pharmacologically important phytochemicals (Adedapo et al., 2005; Falodum et al., 2006). Some of the plants in family Ephoriaceae also have good medicinal properties like antimicrobial properties (Palombo and Adebola, 2008; Darwish et al., 2002). This also may explain the relatively wide spread use of plants in this family by poultry farmers.

Cannabis sativa, Capsicum annum, Nicotiana tobacam and Momordica foetida were the most widely used plants. This study is in line with earlier studies which have reported that these same plants are used by farmers in Uganda to treat poutry diseases (Bukenya, 2007; Saimo, 2003; Olila, 2007). Some of these plants have been reported to exhibit good antimicrobial properties and other medicinal properties (Elujoba et al., 2005; Olukayode and Adebola, 2008). This may confirm the claims farmers have that these plants treat poultry diseases.

Most of the plants were used for prophylaxis purposes and according to farmers this is because very few farmers can access vaccination services in rural areas. In previous studies, many diseases have been documented in smallholder poultry, as diseases are easily contracted under free-range conditions due to poultry scavenging habits. Furthermore disease control is very difficult to carry out under unconfined management and is therefore rarely practiced by the owners of small holder poultry (Ahlers, 1999, Christensen, 2000, Permin and Bisgaard, 2000). This probably explains the reason as to why farmers commonly use these plants for prophylaxis purposes. It is also important to note that farmers use some of these plants to treat symptoms of diseases like fever, diarrhea and respiratory symptoms but not specific diseases. This is probably
because farmers are ignorant about specific diseases though with time and through experience, they have learnt that these plants are good at treating specific symptoms to these diseases.

The most frequently used route is the oral route. Farmers use the oral route most frequently because drugs are easy to administer this way and and requires less skills. Leaves were the most cited plant parts used by the healers for the preparation of traditional medicines. This finding is in line with the results of some ethnomedicinal studies (Olila et al 2007; Yineger and Yewhalaw, 2007; Pradhan and Badola, 2008) who reported that leaves were the most cited plant parts used in remedy preparations. Most plants were prepared as juice using water and probably this was because it requires less time to make juice preparations. This research also agrees with Bukenya, (2007), who reported that the most common way of preparing and administering of the medicine to poultry is simply to crush the plant material, add water to make a juice and administer the concoction orally.

In conclusion the study showed evidence that farmers in Masaka carry out various indigenous practices and use a number of medicinal plants to prevent and treat poultry diseases. This knowledge was documented and the identified plants could offer good opportunities for treatment of poultry diseases and therefore improving production amongst the rural poor and boosting the economy of the country. The documentation of this information would ensure that the knowledge is passed on from generation to generation.
CHAPTER FOUR
ANTIBACTERIAL PROPERTIES OF SELECTED POULTRY ETHNOMEDICINAL PLANTS IN MASAKA

4.1 Summary

In the previous chapter, the indigenous practices and plants used in the treatment of a variety of poultry diseases in Masaka were identified. This chapter examined the antibacterial properties of Ethanol, ether and water extracts of 30 most commonly used medicinal plants and five farmers’ concoctions. These were tested using the agar well diffusion assay for their antimicrobial activity against gram positive and gram negative bacteria: *Staphylococcus aureus, Streptococcus faecalis, Escherichia coli* and *Salmonella typhimurium*. Water extracts of eleven selected plants were tested for minimum inhibitory concentration (MIC) using the tube dilution method. To all the four extracts, *Staphylococcus aureus* was the most susceptible of the four organisms. Ethanol extracts were the most active where 100 percent of all the plants had activity to at least one bacterium, followed by ether extracts with 97% and lastly water extracts with 37%. In general, gram-positive bacteria were more susceptible than gram-negative bacterial species. The water extract of *Moringa oleifera* had activity on all the four bacteria species. The water extracts of *Persea americana* had the lowest MIC (0.25g/ml) and therefore the best activity on *Salmonella typhi*murium. *Leonotis nepetifolia* with MIC (0.25g/ml) and *Lantana trifolia* with MIC (0.15g/ml) had the lowest MICs and therefore the best activity on *Staphylococcus aureus*. Tannins could be responsible for the activity of these plants. These results suggest that *Moringa oleifera* could be used to treat a wide number of diseases, *Persea americana* could be used to treat salmonellosis while *Leonotis nepetifolia* and *Lantana trifolia* could be used to treat staphylococcal infections in poultry. However, all the 30 tested plants have antibacterial activity against at least one of the four bacteria species. The results also suggest that ethanol and ether can be used as alternatives to water as solvents. Since farmers’ concoctions showed no antibacterial activity and had contaminants, it warrants better drug preparation and preservation methods.
4.2 Introduction
The production of medicines and the pharmacological treatment of diseases began with the use of herbs (Tyler, 1997). Many of the drugs currently used to treat bacterial and other infections were first isolated from natural sources including ethno medicinal plants (Coe and Anderson, 1996). Plant based antimicrobials represent a vast untapped source of medicines with enormous therapeutic source potential (Cowan, 1999). Herbs represent one of the first pharmacological interventions attempted by healers, and even today, 25% of our conventional drugs are plant derived in a traditional format (Spore, 1992). The use of medicinal plants as traditional medicines is well known in rural areas of many developing countries (Sandhu and Heinrich, 2005 and Gupta et al., 2005). Traditional healers claim that their medicine is cheaper and more effective than modern medicine. Many plants have been used because of their antimicrobial traits, which are chiefly synthesized during secondary metabolism of the plant (Heinrich et al., 2004). The present study was aimed at screening in vitro antibacterial activity of plant used to treat poultry diseases against some major disease causing bacteria.

4.3 Methodology

4.3.1 Research design and study area
This was an experimental study aimed at decoding indigenous knowledge by seeking scientific explanations for the use of ethnomedicinal resources in the treatment of poultry diseases by farmers in Masaka. Plants were obtained from Masaka district in Kyanamukaka and Buwunga sub counties. Different ways of preparing farmers concoctions were also demonstrated by the farmers. Selection of plants for further laboratory analysis was done according to the most frequently used plants identified using participatory approaches and needs assessment as described in chapter three. Thirty most frequently used plants were considered for this study. Laboratory analysis was conducted from the Microbiology Laboratory of Makerere University Faculty of veterinary Medicine.
4.3.2 Plant collection and botanical identification
Fifty nine (59) plants used by poultry farmers were collected and taken to the Botany Herbarium of Makerere University Kampala for species identification and taxonomic identity of voucher specimen was done by comparing with those of known identity in this herbarium. Out of these, thirty plants were selected for laboratory analysis according to frequency of use. The selected plants were then collected from Masaka, air dried under a shade at room temperature i.e. 24°C for at least two weeks. The backs of the trees were further dried at 45°C in an oven over night to completely remove residual moisture before milling into fine powder. The plant material from plant parts specified by the farmers was ground in a grinding machine. The powders were then sealed in air-tight polyethylene bags and stored in a cool dry place.

4.3.3 Preparation of extracts
Extracts were prepared using the cold extraction methods. The plant samples were extracted using polar solvents which were water and ethanol, and a non polar solvent which was diethyl ether. These extraction methods isolated the active principles of these plants and crude products were obtained. Briefly, the grounded powder were weighed on Satorius balance type BA610, and soaked for three (3) days at room temperature with intermittent shaking. Filtration through cotton wool was done to remove coarse particles and then through filter paper (Whatman No.1, England) in Buchner funnel to get a pure filtrate. The crude extracts obtained were; ether extract, ethanol extract and water extract. After extraction, the extracts were concentrated by evaporation using a rotavapour (Perkins, UK), weighed and reconstituted in Dimethyl Sulfoxide (DMSO) (BDH, UK) to a concentration of 1g/ml. These samples were then stored in a refrigerator at 4°C and later used in the proceeding antibacterial and phytochemical tests.

4.3.4 Antibacterial assays
The antibacterial assay was carried out using agar well diffusion tests and broth dilution techniques. The antimicrobial activity of the plant extracts was tested on four standard bacteria species namely; *Streptococcus fecalis* (wild strain), *Staphylococcus aureus* (ATCC 25923), *Eschericia coli* (ATCC25922) and *Salmonella typhimurium* (ATCC14028) in the Microbiology Laboratory, Faculty of Veterinary Medicine of Makerere University. These were standard laboratory cultures whose susceptibility on commonly used antibiotics was already established.
*Streptococcus faecalis, Staphylococcus aureus* represented gram positive bacteria while *Eschericia coli* and *Salmonella typhimurium* represented gram negative bacteria. These bacteria species are also responsible for a cross-section of poultry diseases conditions namely; secondary bacterial infections which become opportunistic during viral infections, staphylococcosis, colibacillosis and salmonellosis. A standardized bacterial suspension was prepared by picking a colony of respective bacteria using sterile wire loop and suspending it in 5ml of Brain heart infusion liquid media (Mast diagnostics, UK). The dilutions formed the bacterial stock solutions for use in the agar well diffusion assays as outlined below.

**4.3.4.1 Agar well diffusion assay**

The agar well diffusion technique as modified by Agarry *et al.*, (2005) was the standard method used to determine the antibacterial activity of the bioactive compounds. Briefly in the method, the media of Mueller hinton agar (Becton Dicknson M.D USA) was prepared and treated according to manufacturer’s guidelines, where 35g of media was mixed with one litre of distilled water and enclosed in a container and autoclaved at 121 °C for 15 minutes. The media was later dispensed into 90mm sterile agar plates (Oxoid, UK) and left to set. The agar plates were incubated for 24 hours at 37 °C to confirm their sterility. Absence of growth after 24 hours showed that the plates were sterile. The Sterile Mueller Hinton agar plates were inoculated with the test culture by surface spreading using sterile wire loops and each bacterium evenly spread on the entire surface of the plate to obtain uniformity of the inoculum. The culture plate then had at most 4 wells of 6mm diameter and 5mm depth made into it using a sterile agar glass borer. Gentamycin was used as a positive control while normal saline was used as a negative control. Approximately 0.2ml of the bioactive test compound of concentration 1g/ml was suspended in the wells and thereafter inoculated plates/culture were incubated for 24 hours at 37°C. The plates/cultures were examined for the presence of bacterial inhibition zones around each well. Antibacterial activity was determined from the zone of inhibition around the wells. Single readings were carried out. Non-active compounds did not show any inhibition zone. The zones of inhibition were measured using a ruler and a pair of divider (Picfare) and results were reported in millimetres (mm). All zone diameters were considered important since the extracts from the plants were still crude. A zone size interpretive chart was then drawn to show the different plant extracts and their corresponding inhibition zone diameter to the nearest millimetre.
4.3.4.2 The broth dilution assay

The MIC was evaluated on plant water extracts which showed activity on any bacteria organism. The method used was the tube dilution method (Adesokan et al., 2007; Oyeleke et al., 2008; Cheruiyot et al., 2009). Dilutions of the extract were incorporated in Muller Hinton broth (Oxoid, UK) and then inoculated with 0.01ml of standardized suspension of the test organisms. The test was performed on four concentrations of each extract (0.5g/ml, 0.25g/ml, 0.125g/ml, and 0.0625g/ml) employing doubling dilutions of plant extract in brain heart infusion broth up to fourth dilution. For each extract, 1 ml of the resultant broth and 1ml of the extract were added in a test tube and serial dilution was carried out using a two fold dilution and the last 1ml was discarded. Each organism was separately suspended in 5ml of Brain heart infusion broth and incubated overnight. Thereafter, 0.1 ml was added to all the test tubes and preparation incubated at 37 °C for 18 hours. After incubation, a loop full from each tube was sub cultured on nutrient agar to see if bacteria growth was inhibited. Growth of bacteria on solid media indicated that particular concentration of the extract was unable to inhibit the bacteria. The lowest concentration of extract showing no growth indicated the amount of extract in grams per millilitre to which the organism is susceptible. This was the minimum inhibitory concentration (MIC).

The antimicrobial activity of frequently used farmers’ concoctions was also assessed using agar well diffusion tests and broth dilution techniques using the same 4 bacteria species namely; S. faecalis, S. aureus, E. coli and S. typhymurium. The same procedures for the techniques as described above were used.

4.3.5 Data analysis

Data was captured and analyzed using simple Microsoft excel.

4.4 Results

4.4.1 Antibacterial Screening tests

Out of the Fifty nine (59) plants which were collected from Masaka district, only thirty were considered for the screening tests on antibacterial activity. The antibacterial activities of the plant extracts varied according to the species of bacteria tested and the solvents used. For all the 30
plants tested at least one plant extract produced a zone of inhibition against at least one bacteria species. To all the four extracts, *Staph. aureus* was the most susceptible of the four organisms. *E.coli* was the least susceptible to both water and ethanol plant extracts while *Strep. feacalis* was the least susceptible to ether plant extracts.

4.4.1.1 Antibacterial activity of Water Extracts

Thirty six (37) % of the 30 tested plants had antibacterial activity against at least one of the 4 bacteria species as shown in Table 9.

<table>
<thead>
<tr>
<th>Table 8: Antibacterial screening on water extracts</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inhibition zone diameter (to the nearest mm)</td>
</tr>
<tr>
<td>Plant extract</td>
</tr>
<tr>
<td>1 Moringa oleifera</td>
</tr>
<tr>
<td>2 Lantana trifolia</td>
</tr>
<tr>
<td>3 Sida cuneifolia</td>
</tr>
<tr>
<td>4 Vernonia cinerea</td>
</tr>
<tr>
<td>5 Tetredenia riparia</td>
</tr>
<tr>
<td>6 Desmodium salicifolium</td>
</tr>
<tr>
<td>7 Persea Americana</td>
</tr>
<tr>
<td>8 Aspilia africana</td>
</tr>
<tr>
<td>9 Syzygium cuminii</td>
</tr>
<tr>
<td>10 Albizia coriaria</td>
</tr>
<tr>
<td>11 Leonotis nepetifolia</td>
</tr>
</tbody>
</table>

Of the eleven plants with active water extracts 9% (n=11) had activity on all the four bacteria species. Eighteen (18) % (n=11) of these plants had activity on only 3 bacteria species. Twenty seven (27) % (n=11) of these plants had activity on only 2 bacteria species. Forty five (46) % (n=11) of these plants had activity on only one (1) bacteria species (Figure 1).
Of the plants whose water extracts showed antibacterial activity 82 % (n=11) were active on gram positive bacteria. All these plants were active on *Staph. Aureus* while 44% (n=9) were active on *Strep. feacalis.*

**Table 9: Water extracts active on Gram positive bacteria**

<table>
<thead>
<tr>
<th>Plant extract</th>
<th>Activity</th>
<th>Strep. faecalis</th>
<th>Staph. Aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Moringa oleifera, Lantana trifolia, Syzygium cuminii, Albizia coriaria</em></td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>Sida cuneifolia, Vernonia cineria, Tetredenia riparia, Persea americana, Leonotis nepetifolia</em></td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
</tbody>
</table>
Of all the plants whose water extracts showed antibacterial activity 45% (n=11) plants was active on gram negative bacteria and all these were active on *S. typhymurium*. Of all the plants whose water extracts showed antibacterial activity on gram negative bacteria, 60% (n=5) were active on *E. coli*.

**Table 10: Water extracts active on Gram negative bacteria**

<table>
<thead>
<tr>
<th>Plant extract</th>
<th><em>E. Coli</em></th>
<th><em>S. typhymurium</em></th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Moringa oleifera, Persea Americana, Aspilia africana</em></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td><em>Lantana trifolia, Desmodium salicifolium</em></td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>
4.4.1.2 Antibacterial activity of Ether extracts

Ninety seven (97) % of the 30 tested plants had antibacterial activity against at least one of the 4 bacteria species as shown in Table 12.

Table 11: Antibacterial screening of ether extracts

<table>
<thead>
<tr>
<th>Plant extract</th>
<th>Strep. faecalis</th>
<th>Staph. aureus</th>
<th>E. Coli</th>
<th>S.typhymurium</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Datura stramonium</td>
<td>0.9</td>
<td>1.5</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>2. Moringa oleifera</td>
<td>1.1</td>
<td>2.8</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>3. Azadirachta indica</td>
<td>1.8</td>
<td>2.2</td>
<td>1.2</td>
<td>0.0</td>
</tr>
<tr>
<td>4. Senecio cydoniifolius</td>
<td>1.1</td>
<td>2.3</td>
<td>2.7</td>
<td>1.3</td>
</tr>
<tr>
<td>5. Carica papaya</td>
<td>1.3</td>
<td>1.9</td>
<td>0.0</td>
<td>1.8</td>
</tr>
<tr>
<td>6. Lantana trifolia</td>
<td>1.0</td>
<td>2.4</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>7. Sida cuneifolia</td>
<td>1.1</td>
<td>2.0</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>8. Vernonia cineria</td>
<td>1.3</td>
<td>3.2</td>
<td>2.1</td>
<td>0.0</td>
</tr>
<tr>
<td>9. Jatropha curcas</td>
<td>1.2</td>
<td>0.9</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>10. Nicotiana tobaccam</td>
<td>1.5</td>
<td>1.5</td>
<td>2.1</td>
<td>1.2</td>
</tr>
<tr>
<td>11. Tetredenia riparia</td>
<td>0.0</td>
<td>2.4</td>
<td>1.5</td>
<td>1.5</td>
</tr>
<tr>
<td>12. Capsicum annuum</td>
<td>0.0</td>
<td>2.9</td>
<td>2.2</td>
<td>1.4</td>
</tr>
<tr>
<td>13. Agave sislalana</td>
<td>0.0</td>
<td>2.3</td>
<td>1.1</td>
<td>2.0</td>
</tr>
<tr>
<td>14. Aloe vera</td>
<td>0.0</td>
<td>2.5</td>
<td>2.2</td>
<td>2.5</td>
</tr>
<tr>
<td>15. Justicia betonica</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>1.0</td>
</tr>
<tr>
<td>16. Chenopodium opulifolium</td>
<td>1.0</td>
<td>1.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>17. Desmodium salicifolium</td>
<td>1.6</td>
<td>1.5</td>
<td>2.3</td>
<td>2.6</td>
</tr>
<tr>
<td>18. Rubia cordifolia</td>
<td>0.0</td>
<td>1.4</td>
<td>1.6</td>
<td>1.9</td>
</tr>
<tr>
<td>19. Vernonia amygdalina</td>
<td>1.4</td>
<td>3.1</td>
<td>2.6</td>
<td>2.4</td>
</tr>
<tr>
<td>20. Persea americana</td>
<td>0.0</td>
<td>1.7</td>
<td>0.0</td>
<td>1.6</td>
</tr>
<tr>
<td>21. Aspilia africana</td>
<td>1.2</td>
<td>2.6</td>
<td>2.9</td>
<td>2.5</td>
</tr>
<tr>
<td>22. Syzygium cumini</td>
<td>0.0</td>
<td>1.9</td>
<td>2.0</td>
<td>2.5</td>
</tr>
<tr>
<td>23. Albizia cortiaria</td>
<td>1.5</td>
<td>1.7</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>24. Momordica foetida</td>
<td>0.0</td>
<td>1.1</td>
<td>1.3</td>
<td>2.3</td>
</tr>
<tr>
<td>25. Conyza floribunda</td>
<td>0.0</td>
<td>1.9</td>
<td>2.1</td>
<td>2.7</td>
</tr>
<tr>
<td>26. Phaseolus lunatus</td>
<td>0.0</td>
<td>1.9</td>
<td>1.3</td>
<td>2.0</td>
</tr>
<tr>
<td>27. Leonotis nepetifolia</td>
<td>0.0</td>
<td>1.7</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>28. Garcinia buchananii</td>
<td>0.0</td>
<td>2.4</td>
<td>1.5</td>
<td>1.7</td>
</tr>
<tr>
<td>29. Bidens pilosa</td>
<td>1.4</td>
<td>3.5</td>
<td>3.4</td>
<td>3.6</td>
</tr>
</tbody>
</table>

Of the twenty nine plants with active ether extracts, 24 % had activity on all the four bacteria species. Forty five (45) % (n=29) of these plants had activity on only 3 bacteria species.
Seventeen (17) %( n=29) of these plants had activity on only 2 bacteria species. Fourteen (14) % ( n=29) of these plants had activity on only one bacteria species (Figure 2).

![Pie chart showing percentage of plants with active ether extracts and the number of bacteria species that responded.](image)

**Figure 2: Percentage number of plants with active Ether extracts and the number of bacteria species that responded**

Of all the plants whose ether extracts showed antibacterial activity 97 % ( n=29) were active on gram positive bacteria and all these were active on *Staph. aureus*. Of all the plants whose ether extracts showed antibacterial activity on gram positive, 54% ( n=28) were active on *Strep. faecalis*. 
Table 12: Ether extracts active against Gram positive bacteria

<table>
<thead>
<tr>
<th>Plant extract</th>
<th>Activity</th>
<th>Strep. faecalis</th>
<th>Staph. aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Datura stramonium, Moringa oleifera, Azadirachta indica, Senecio cydoniifolius, Carica papaya, Sida cuneifolia, Vernonia cineria, Jatropha curcas, Nicotiana tobaccam, Chenopodium opulifolium, Desmodium salicifolium, Vernonia amygdalina, Aspilia Africana, Albizia coriaria, Bidens pilosa</em></td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>Lantana trifolia, Tetredenia riparia, Capsicum annuum, Agave sisalana, Aloe vera, Rubia cordifolia, Persea Americana, Syzygium cumminii, Momordica foetida, Conyza floribunda, Phaseolus lunatus, Leonotis nepetifolia, Garcinia buchananii</em></td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
</tbody>
</table>

Of all the plants whose ether extracts showed antibacterial activity 76% (n=29) plants were active on gram negative bacteria. Of all the plants whose ether extracts showed antibacterial activity on gram negative bacteria, 86% (n=22) were active on *E. coli*. Of all the plants whose ether extracts showed antibacterial activity 91% (n=22) plants were active on *S. typhymurium*.

Table 13: Ether extracts active against Gram negative bacteria

<table>
<thead>
<tr>
<th>Plant extract</th>
<th>Activity</th>
<th>E.Coli</th>
<th>S. typhymurium</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Senecio cydoniifolius, Sida cuneifolia, Sida cuneifolia, Nicotiana tobaccam, Tetredenia riparia, Capsicum annuum, Agave sisalana, Aloe vera, Desmodium salicifolium, Rubia cordifolia, Vernonia amygdalina, Aspilia africana, Syzygium cumminii, Momordica foetida, Conyza floribunda, Phaseolus lunatus, Garcinia buchananii, Bidens pilosa</em></td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>Azadirachta indica, Vernonia cineria</em></td>
<td>+</td>
<td></td>
<td>-</td>
</tr>
<tr>
<td><em>Carica papaya, Justicia betonica, Persea americana</em></td>
<td>-</td>
<td></td>
<td>+</td>
</tr>
</tbody>
</table>
4.4.1.3 Antibacterial activity of Ethanol extracts

All the 30 tested plants had antibacterial activity against at least one of the 4 bacteria species as shown in Table 15.

Table 14: Antibacterial screening of ethanol extracts

<table>
<thead>
<tr>
<th>Plant extract</th>
<th>Strep. faecalis</th>
<th>Staph. aureus</th>
<th>E. Coli</th>
<th>S. typhymurium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Datura stramonium</td>
<td>1.1</td>
<td>1.3</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Moringa oleifera</td>
<td>4.2</td>
<td>1.3</td>
<td>1.1</td>
<td>0.0</td>
</tr>
<tr>
<td>Azadirachta indica</td>
<td>0.8</td>
<td>1.7</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Senecio cydoniifolius</td>
<td>1.1</td>
<td>1.3</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Carica papaya</td>
<td>1.0</td>
<td>1.6</td>
<td>0.0</td>
<td>1.4</td>
</tr>
<tr>
<td>Lantana trifolia</td>
<td>1.4</td>
<td>2.1</td>
<td>1.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Sida cinefolia</td>
<td>1.2</td>
<td>2.3</td>
<td>1.5</td>
<td>1.4</td>
</tr>
<tr>
<td>Vernonia cineria</td>
<td>1.1</td>
<td>2.3</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Jatropha curcas</td>
<td>1.8</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Nicotiana tobaccam</td>
<td>1.0</td>
<td>1.4</td>
<td>1.2</td>
<td>0.0</td>
</tr>
<tr>
<td>Tetredenia riparia</td>
<td>0.0</td>
<td>2.6</td>
<td>1.2</td>
<td>0.0</td>
</tr>
<tr>
<td>Capsicum annuum</td>
<td>0.0</td>
<td>1.9</td>
<td>1.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Agave sisalana</td>
<td>0.0</td>
<td>1.2</td>
<td>0.0</td>
<td>1.6</td>
</tr>
<tr>
<td>Aloe vera</td>
<td>0.0</td>
<td>1.4</td>
<td>0.0</td>
<td>2.0</td>
</tr>
<tr>
<td>Justicia betonica</td>
<td>0.0</td>
<td>0.0</td>
<td>0.0</td>
<td>1.2</td>
</tr>
<tr>
<td>Chenopodium opulfolium</td>
<td>2.0</td>
<td>1.2</td>
<td>0.0</td>
<td>1.6</td>
</tr>
<tr>
<td>Desmodium salicifolium</td>
<td>1.0</td>
<td>1.3</td>
<td>1.3</td>
<td>2.1</td>
</tr>
<tr>
<td>Rubia cordifolia</td>
<td>0.0</td>
<td>1.2</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Vernonia amygdalina</td>
<td>1.3</td>
<td>1.4</td>
<td>0.0</td>
<td>1.3</td>
</tr>
<tr>
<td>Persea americana</td>
<td>1.2</td>
<td>1.6</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Aspilia africana</td>
<td>0.0</td>
<td>0.8</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Syzygium cuminii</td>
<td>0.0</td>
<td>1.6</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Solanum mauritianum</td>
<td>0.0</td>
<td>1.0</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Albizia coriaria</td>
<td>1.7</td>
<td>2.0</td>
<td>0.0</td>
<td>1.6</td>
</tr>
<tr>
<td>Momordica foetida</td>
<td>0.0</td>
<td>1.2</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Conyz floribunda</td>
<td>1.0</td>
<td>1.3</td>
<td>1.5</td>
<td>2.0</td>
</tr>
<tr>
<td>Phaseolus lunatus</td>
<td>0.0</td>
<td>1.3</td>
<td>0.0</td>
<td>1.3</td>
</tr>
<tr>
<td>Leonotis nepetifolia</td>
<td>1.1</td>
<td>1.3</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>Garcinia buchananii</td>
<td>1.8</td>
<td>2.3</td>
<td>0.0</td>
<td>1.5</td>
</tr>
<tr>
<td>Bidens pilosa</td>
<td>0.0</td>
<td>1.3</td>
<td>0.0</td>
<td>0.0</td>
</tr>
</tbody>
</table>

Of the thirty plants with active ethanol extracts, only (10) % plants had activity on all the four bacteria species. Twenty seven (27) % (n=30) plants had activity on only 3 bacteria species.
Thirty seven (37) % (n=30) plants had activity on only 2 bacteria species. Twenty seven (27) % (n=30) plants had activity on only one bacteria species (Figure 3).

![Pie chart showing percentage of plants with active ethanol extracts and the number of bacteria species that responded.]

**Figure 3: Percentage number of plants with active ethanol extracts and the number of bacteria species that responded**

Of all the plants whose ethanol extracts showed antibacterial activity, 97% (n=30) were active on gram positive bacteria. Of these, most extracts (93%) showed antibacterial activity against *Staph. aureus* while 60% against *Strep. faecalis*

**Table 15: Ethanol extracts active against Gram positive bacteria**

<table>
<thead>
<tr>
<th>Plants</th>
<th>Strep. faecalis</th>
<th>Staph. aureus</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Datura stramonium</em>, <em>Moringa oleifera</em>, <em>Azadirachta indica</em>,</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td><em>Senecio cydoniifolius</em>, <em>Carica papaya</em>, <em>Lantana trifolia</em>, <em>Sida</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>cuneifolia</em>, <em>Vernonia cinerea</em>, <em>Nicotiana tobacca</em>, <em>Chenopodium</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>opulifolium</em>, <em>Desmodium salicifolium</em>, <em>Vernonia amygdalina</em>,</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Persea americana</em>, <em>Albizia coriaria</em>, <em>Conyza floribunda</em>, <em>Leonotis</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>nepetifolia</em>, <em>Garcinia buchananii</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Jatropha curcas</em></td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Tetredenia riparia</em>, <em>Capsicum annuum</em>, <em>Agave sisalana</em>, <em>Aloe</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>vera</em>, <em>Rubia cordifolia</em>, <em>Rubia cordifolia</em>, <em>Aspilia africana</em>,</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Syzygium cuminii</em>, <em>Solanum mauritianum</em>, <em>Momordica foetida</em>,</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Phaseolus lunatus</em>, <em>Bidens pilosa</em></td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

Of all the plants whose ethanol extracts showed antibacterial activity on gram negative bacteria, 53% (n=17) were active on *E. coli*. Of all the plants whose ethanol extracts showed antibacterial activity on gram negative bacteria, 65% (n=17) plants were active on *S. typhymurium*. 
Table 16: Ethanol extracts active against Gram negative bacteria

<table>
<thead>
<tr>
<th>Plant extract</th>
<th><em>E. Coli</em></th>
<th><em>S. typhymurium</em></th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Sida cuneifolia, Desmodium salicifolium, Conyza floribunda</em></td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td><em>Moringa oleifera, Carica papaya, Lantana trifolia, Nicotiana tobaccam, Tetredenia riparia, Capsicum annuum</em></td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Agave sisalana, Aloe vera, Justicia betonica, Chenopodium opulifolium, Vernonia amygdalina, Albizia coriaria, Phaseolus lunatus, Garcinia buchananii</em></td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

Figure 4: Percentage of plants active on each bacteria species

4.4.1.4 Antibacterial activity of the farmers’ concoctions

Five farmers’ concoctions were analysed on all the four test bacteria. The composition of the concoctions has been indicated in appendix II. All the concoctions showed no antibacterial activity, the inhibition zone diameter was zero (0). There was growth of the common bacteria contaminants around the wells in which the drugs were put. The commonest contaminant was *Proteus vulgaris*. 
4.4.1.5 Minimum Inhibition Concentration

Water extracts from the eleven plant species showing antibacterial activity were tested further on the same bacteria species to determine the minimum inhibition concentrations. The plants which were considered for minimum inhibition concentration were those whose water extracts showed activity against at least one bacterial species. All the four plants whose water extracts were active against Strep. faecalis had a minimum inhibition concentration of 0.5g/ml and these were Moringa oleifera, Lantana trifolia, Syzygium cuminii, and Albizia coriaria. Seven out of the nine plants whose water extracts had activity on Staph. aureus had a minimum inhibition concentration of 0.5g/ml and these were Moringa oleifera, Sida cuneifolia, Vernonia cineria, Tetredenia riparia, Persea americana, Syzygium cuminii and Albizia coriaria. Leonotis nepetifolia had MIC 0.25g/ml while Lantana trifolia had the least (0.125g/ml). All the three plants whose water extracts were active on E. coli had a minimum inhibition concentration of 0.5g/ml and these were: Moringa oleifera, Persea americana and Aspilia africana. Four out of the five plants whose water extracts showed activity on S. typhymurium had a minimum inhibition concentration of 0.5g/ml and these were Moringa oleifera, Desmodium salicifolium, Lantana trifolia and Aspilia africana. While only one plant, Persea americana had the least MIC i.e. 0.25g/ml.

![Figure 5: Minimum inhibition concentration (MIC) of active water extracts on gram positive bacteria](image-url)
4.5 Discussion

Findings of this study show these plants exhibited various sizes of inhibition zones with different solvents against the bacteria they were tested on. However, this difference can’t be used as a model to draw any conclusions because the amount of active ingredients per extract is not the same. Therefore all the plant extracts which showed any activity were considered to have positive activity and those with no activity at all were considered to have negative activity. Thus in this study, Gram-positive bacteria were found to have more susceptibility as compared to Gram-negative bacterial species. This is in line with earlier studies which also owe this observed to the differences in chemical composition and structure of cell wall of both types of microorganisms (Yaghoubi et al., 2007; Nair et al., 2007).

Of the eleven plants with active water extracts 9 % had activity on all the four bacteria species. This may be indicative of presence of broad spectrum activity against both gram positive and gram negative bacteria and may explain why many farmers are using Moringa oleifera in treatment and prevention of poultry diseases. Of all the plants whose water extracts showed
antibacterial activity, 45% (n=11) plants were active on gram negative bacteria. *Moringa aloefera, Persea americana* and *Aspilia africana* were active on both *S. typhymurium* and *Escherichia coli*. The water extracts of *Persea americana* had the lowest MIC (0.25g/ml) and therefore the best activity on *S. typhymurium*.

This study therefore suggests that *Moringa aloefera, Persea americana* and *Aspilia africana* could probably be used to treat salmonelloses and colibacillosis. It has been reported by earlier studies that aqueous extracts of *Moringa oloefera* have antibacterial activity against *Salmonella typhi* (Doughari et al., 2007). *Moringa oleifera* has also shown antibacterial activity against *E.coli, Enterobacter, P. aeruginosa* and *S. aureus* (Nepolean et al, 2009). *Moringa oleifera* has been cited to be antibacterial (Spiliotis et al., 1998; Fluglie, 1999). *Persia americana* extracts have demonstrated antimicrobial activity against *Mycobacteria* strains (Gomez-Flores et al., 2008). The aqueous extracts of the leaves of *Aspilia africana* have exhibited differential antibacterial activities on both gram-positive and Gram-negative bacterial species (Macfoy and Cline, 1990, Adeniyi and Odufowora, 2000). *Apiria africana* is used in Africa to stop bleeding, promote healing of wounds and sores and treatment of cardiovascular diseases (Dimo et al., 2002). The infusion of leaves of *Aspiria africana* is used for treatment of stomach troubles in children (Okwu and Josiah, 2006).

This study has also identified two important plants whose water extracts could be used to treat diseases caused by *Staph. aureus*. These had the lowest MIC and they were *Leonotis nepetifolia* with MIC (0.25g/ml) and *Lantana trifolia* with MIC (0.15g/ml) and hence considered to be with best activity. Previous studies have shown that the methanol extract of *Lantana camara* a close relative of *Lantana trifolia* exhibited antibacterial activity against *Staph. aureus* (Cheruiyot et al., 2009). In these studies in chapter five, *Lantana trifolia* exhibited presence of tannins. The presence of tannins in *Lantana sps* has also been reported in its close relative *Lantana camara* (Ganjewala et al., 2009)).

The ether and ethanol extracts of both *Sida cuneifolia, Desmodium salicifolium* reacted on all four bacteria species and this may indicate presence of broad spectrum activity against both
gram negative and gram positive bacteria. This is in line with earlier studies carried out on *Sida cuneifolia* which reported that it has a good antibacterial activity on both gram negative and gram positive bacteria (Van-Vuurena and Viljoenb, 2006). Of all the plants whose ether extracts showed antibacterial activity, 76% (n=29) plants were active on gram negative bacteria. While of all the plants whose ethanol extracts showed antibacterial activity, 57% (n=30) was active on gram negative bacteria. Ether and ethanol extracts were more potent than water extracts to exhibit antibacterial activity. This study suggests that compared to water solvent, ether and ethanol solvents are better solvents in extraction of antibacterial substances and could also be used by farmers especially against gram negative bacteria which most times appear to cause fatal diseases like salmonelloses and colibacillosis.

All the farmers’ concoctions showed no antibacterial activity, the inhibition zone diameter was zero. There was growth of the common bacteria contaminants around the wells in which the drugs were put. The commonest contaminant was *Proteus vulgaris*. It has been reported before that the crude preparation process, heavy microbial load resulting from the field plant contamination as well as stability and shelf life determination in terms of stabilization and preservation, particularly in liquid dosage forms, would pose important fundamental challenges (Elujoba *et al.*, 2005).

In conclusion, as demonstrated by some plants in this study, there is considerable evidence that their plant extracts, have the potential to be developed into agents that can be used as preventative or treatment therapies poultry diseases. The water extracts of plants like *Moringa oleifera, Persea americana* and *Aspilia africana* could be used to treat salmonelloses and colibacillosis. The water extracts of *Leonotis nepetifolia* and *Lantana trifolia* could be used to treat diseases caused by *Staphylococcus aureus* which normally causes secondary bacterial infections in poultry. The ether and ethanol extracts of both *Sida cuneifolia*, and *Desmodium salicifolium* could be used to treat diseases caused by both gram negative and gram positive bacteria since they show broad spectum activity. The chance to find antimicrobial activity was more apparent in ethanol and ether extracts than water extracts of the same plants. It is recommended that clinical trials on poultry are carried out to establish whether these plants offer therapeutic benefits, either alone or in combination with other plants. This can help to reduce the
overall burden of poultry diseases using cheaper methods especially among rural poor people worldwide.
CHAPTER FIVE

PHYTOCHEMICAL PROPERTIES OF SELECTED POULTRY ETHNOMEDICINAL PLANTS IN MASAKA

5.1 Summary

In the previous chapters, the indigenous practices and plants used in the treatment of a variety of poultry diseases in Masaka were identified as well as the antibacterial properties of 30 selected plants. In this chapter, qualitative tests were carried out to investigate the phytochemical properties of Ethanol, ether and water extracts of eleven priority plants. Pictures of these plants were indicated in appendix IV. The phytochemicals identified in these plants were glucides, reducing compounds, anthracenosides, coumarins, steroid glycosides, polyuronides, tannins, flavone glycosides, anthracenoid aglycones, chlorophyll, sterols, flavonic aglycones, alkaloids salts, saponins, basic alkaloids, carotenoids, starch, Lipids/ fatty acids, volatile oils and anthocyanosides. These results suggest that all the 11 tested plants contain phytochemicals of potential healing properties and the most prominent phytochemicals of medicinal importance were tannins, sterols, basic alkaloids and alkaloid salts.

5.2 Introduction

Medicinal plants have been used as traditional treatments for numerous human diseases for thousands of years and in many parts of the world. In rural areas of the developing countries, they continue to be used as the primary source of medicine (Chitme et al., 2003). The search for newer sources of antibiotics is a global challenge preoccupying research institution, pharmaceutical companies and academia, since many infection agents are becoming resistance to synthetic drugs (Latha and Kannabiran, 2006). The development of medicinal chemistry, as a major route for the discovery of novel and more active therapeutic agents, further investigation into the chemical and biological activities of the plants should be carried out (Rao & Roja, 2002). As there are approximately 500 000 plant species occurring worldwide, of which only 1% has been phytochemically investigated, there is great potential for discovering novel bioactive compounds (Palambo, 2009). Therefore since there are not sufficient scientific studies on the
phytochemical composition of most plants, this study looked at the qualitative analysis of the phytochemical composition of eleven plants used to treat poultry diseases.

**5.3 Methodology**

Extracts were made from eleven plants after which phytochemicals present were identified.

**5.3.1 Preparation of extracts**

Eleven plants which are used by poultry farmers were selected for phytochemical analysis. The criterion for selecting these plants was according to the antibacterial activity of their water extracts. As indicated in chapter four, out of the thirty plants tested for antibacterial activity, the water extracts of eleven plants showed activity against at least one bacteria species. The selected plants were then collected from Masaka, air dried under a shade at room temperature i.e. 24°C for at least two weeks. The backs of the trees were further dried at 45°C in an oven over night to completely remove residual moisture before milling into fine powder. The plant material from plant parts specified by the farmers was ground in a grinding machine. The powders were then sealed in air-tight polyethylene bags and stored in a cool dry place.

Extracts were prepared using the cold extraction methods. The plant samples were extracted using polar solvents which were water and ethanol, and a non polar solvent which was diethyl ether. These extraction methods isolated the active principles of these plants and crude products were obtained. The crude extracts obtained were; ether extract, ethanol extract and water extract. After extraction, the extracts were concentrated by evaporation using a rotavapour (Perkins, UK), weighed and reconstituted in DMSO (BDH, UK) to a concentration of 1g/ml. These were then stored in a refrigerator at 4°C and latter used in the proceeding phytochemical tests.

**5.3.2 Phytochemical screening**

The extracts thus obtained were also subjected to preliminary phytochemical screening following the standard methodologies (Harborne, 1998; Houghton and Raman, 1998; Woo, 2001 and Parekh, 2006) and the laboratory procedures from the laboratory protocol for screening phytochemicals by Natural Chemotherapeutic Laboratory, Ministry of Health, Uganda.
5.3.1.1 Identification of phytochemicals in ether extract

Qualitative tests were carried out to assess for the presence of alkaloids, flavone aglycones, emodols (anthracenoside aglycones) coumarins, higher fatty acids, volatile oils, Sterols and triterpenes and carotenoids (tetraterpenes).

The identification of alkaloids was carried out on the residue obtained by evaporating 10 mls of the extract. The residue was dissolved in 1.5mls of 2% hydrochloric acid (Fisher scientific, UK). From this, a sample made up of 0.5 ml acidic aqueous solution was got and 2-3 drops of Mayer’s reagent (Fisher scientific, UK) was added to 0.5 ml of acidic aqueous solution. An opalescence, which was a cream coloured precipitate indicated presence of alkaloids.

To identify flavone aglycones, 3mls of the ether extract were evaporated until a residue was obtained. The residue was dissolved in 1-2 mls of 50% methanol (Fisher scientific, UK) in the water bath maintained at 40 °C. Metallic magnesium (Fisher scientific, UK) and 4-5 drops of concentrated hydrochloric acid (Fisher scientific, UK) were added. An orange or red colour will indicated presence of flavonic aglycones (Shibata’s reaction or cyanidin test).

The identification of emodols (anthracenoside aglycones) was done by transferring 3mls of ether extract to a test tube. One (1) ml of 25% ammonia solution (Phillip Harris, England) was added and shaking was done. A red colour indicated presence of emodols.

To determine the presence of coumarins, 3mls of ether extract were evaporated to dryness. The residue was dissolved in hot water. After cooling the solution was divided into two tubes: One tube contained the reference and, the aqueous solution of the second tube was made alkaline with 0.5mls of 10% ammonium solution (Phillip Harris, England). The occurrence of an intense fluorescence under UV light indicated presence of coumarins.

For the identification of higher fatty acids, the aqueous solution exhaustively extracted with ether was acidified with concentrated hydrochloric acid (pH = 3-4) (Fisher scientific, UK). Under these conditions the fatty acids were released from their alkaline salts. The acid aqueous solution became opalescent. The fatty acids were extracted by shaking the solution in a separating funnel.
with ethyl ether repeatedly added in small amounts. Then the other solutions were evaporated to dryness. The oily residue indicated presence of fatty acids.

To assess the presence of volatile oils, the ether extract was placed in a flask and evaporated to dryness until the residue had a pleasant odor. It was then dissolved in small amounts of alcohol, by repeated elutions. One part of the alcohol solution was evaporated to dryness. The pleasant odor indicated presence of volatile oils.

To identify sterols and triterpenes, the ether extract was evaporated to dryness. The residue was dissolved in 0.5ml of acetic acid (Fisher scientific, UK) and then in 0.5 ml of chloroform. The solution was transferred to a dry tube and 2 mls of concentrated sulphuric acid (Fisher scientific, UK) were added at the bottom of the tube (Libermann-Burchard’s reaction). If there is formation of a brownish-red or violet ring at the contact zone of the two liquids and the supernatant layer become green or violet, it indicated presence of sterols and triterpenes. If the solution where the reaction was performed became greenish, it indicated chlorophyll.

To identify carotenoids (tetraterpenes), 10mls of the ether extract was evaporated to dryness and 3 drops of saturated solution of antimony trichloride (Fisher scientific, UK) in chloroform (Sigma) were added (Carr price’s reaction). The pigments were at first blue and later become red. With concentrated sulphuric acid (Fisher scientific, UK), the carotenoids become usually deep blue or blueish-green.

5.3.1.2 Identification of phytochemicals in ethanol extract
Qualitative tests were carried out on the ethanol extracts to assess the presence of, reducing sugars, alkaloid salts, anthracenocides, coumarin derivatives, flavonosides and steroid glycosides in the plants.

The presence of reducing compounds was assessed by diluting 1ml of the alcohol extract with 2mls of water. Fehling’s solution (Sigma) was added and heated in a water bath maintained at 40 °C. A brick-red precipitate denoted the presence of reducing compounds.
Identification of alkaloid salts was done by adding 8mls of 10% hydrochloric acid (Fisher scientific, UK) and the content stirred at heat in a water bath using a glass rod. This was then cooled and 0.5g of sodium chloride (Fisher scientific, UK) was added and stirred again. The solution was filtered and the filtrate was washed with 3ml of hydrochloric acid (Fisher scientific, UK). One (1) ml of this acidic extract was then used to perform a test with Mayer’s reagent (Fisher scientific, UK). The occurrence of precipitate indicated presence of alkaloid salts.

The determination of anthracenocides, coumarins derivatives, flavonosides, steroid glycosides and anthocyanosides was done by first hydrolyising the alcohol extract. Briefly, 15 ml of 10% hydrochloric acid (Fisher scientific, UK) were added to 25 ml of alcohol extract by refluxing and heated up for 30 minutes. After cooling, the solution was extracted three times with 12ml of diethyl ether (Fisher scientific, UK) in a separating funnel. The ether extracts were placed together and dehydrated with anhydrous sodium sulphate (Fisher scientific, UK), resulting in ether and an aqueous solution.

The ether extract served to identify the anthracanosides, coumarine derivatives, flavonosides and steroid glycosides by means of a series of reactions characteristic of each group as shown in section: 5.3.1.1.

To identify anthocyanosides, if the acidic aqueous solution was red and it turned neither to violet at neutral pH, nor to green or blue in an alkaline medium, this indicated that anthocyanins were present.

5.3.1.3 Identification of phytochemicals on the equeous extract

Qualitative tests were done to assess for presence of polyuronides, glucides, tannins (polyphenols and saponins).

For the identification of polyuronides, 2ml of the aqueous extract were added drop wise in a test tube where 10ml of alcohol or acetone (Fisher scientific, UK) had already been placed. A thick precipitate was formed and it was separated off by filtration or centrifugation and washed away.
with alcohol, then stained with methylene blue (Fisher scientific, UK). The occurrence of a blue precipitate denoted presence of mucilages.

To identify glucides (oses and polyoses), 1 ml of the aqueous extract was decanted and diluted; 3 drops of lugol’s solution (Fisher scientific, UK) was added. If a blue colour developed, it indicated presence of starch.

To identify saponins, an aqueous extract was diluted to ratio of 1:1 and 2ml of the diluted solution was put in a 1.6 cm diameter and shaken for 15 minutes. The occurrence of foam column indicated presence of saponins.

The identification of tannins was carried out in 1ml of aqueous extract with a solution of ferric chloride (Sigma). If the extract contained both types of tannin, a hydrochloric formaldehyde solution (styassny’s reagent) was boiled to reflux. Under these conditions, the catechol tannins were condensed as a red precipitate which was then filtered. The solution thus obtained was neutralized with sodium acetate and some drops of ferric chloride were added. A deep blue colour indicated presence of gallic tannins.

**5.3.3 Data analysis**
Data was analyzed using simple computer packages like Microsoft excel.

**5.4 Results**

**5.4.1 Phytochemicals in the plants**
The summary of the phytochemicals present in plants as per all the different extracts are indicated in appendix IV. The phytochemicals tested in the water extracts were glucides, polyuronides, tannins and saponins (Table 19). The phytochemicals tested for in the ether extract were volatile oils, lipids/ fatty acids, sterols, carotenoids, basic alkaloids, flavonoic aglycones, nthuracenoside aglycones, coumarins and chlorophyll (Table 20).The phytochemicals tested for in the ethanol extract were reducing compounds, alkaloids salts, sterol glycosides, anthocyanosides, anthracenosides, coumarin derivatives, flavonols and flavanones (Table 21).
For water extracts, glucides and tannins were the phytochemicals that showed widespread presence in a large number of plants under study. Ninety one (91) % of the plants contained tannins and glucides. *Syzygium cumini* and *Albizia coriaria* were some of the plants that had the highest levels of a wide range of phytochemicals.

**Table 17: Phytochemicals of water extracts of selected plants**

<table>
<thead>
<tr>
<th>Phyto chemical</th>
<th>Polyuronides</th>
<th>Glucides</th>
<th>Tannins</th>
<th>Saponins</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Moringa oleifera</em></td>
<td>+</td>
<td>++</td>
<td>+(catecols)</td>
<td>-</td>
</tr>
<tr>
<td><em>Lantana trifolia</em></td>
<td>-</td>
<td>+</td>
<td>++(catecols)</td>
<td>+</td>
</tr>
<tr>
<td><em>Sida cuneifolia</em></td>
<td>+</td>
<td>++</td>
<td>++(catecols)</td>
<td>-</td>
</tr>
<tr>
<td><em>Vernonia cineria</em></td>
<td>-</td>
<td>++</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>Tetredenia riparia</em></td>
<td>-</td>
<td>+++</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td><em>Desmodium salicifolium</em></td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td><em>Persea americana</em></td>
<td>+</td>
<td>-</td>
<td>++(catecols)</td>
<td>+</td>
</tr>
<tr>
<td><em>Aspilia africana</em></td>
<td>-</td>
<td>++</td>
<td>++(catecols)</td>
<td>-</td>
</tr>
<tr>
<td><em>Syzygium cumini</em></td>
<td>-</td>
<td>+++</td>
<td>++(gallic)</td>
<td>+</td>
</tr>
<tr>
<td><em>Albizia coriaria</em></td>
<td>+</td>
<td>+++</td>
<td>++(catecols)</td>
<td>+</td>
</tr>
<tr>
<td><em>Leonotis nepetifolia</em></td>
<td>+</td>
<td>+</td>
<td>++(catecols)</td>
<td>-</td>
</tr>
</tbody>
</table>

**Key**

The extract remains clear with no change: Absence of the phytochemical  
Faint changes against dark background: Trace of the phytochemical  
Definite change noticed: Presence of phytochemical  
Heavy change noticed: Intense Presence of phytochemical
For ether extracts, basic alkaloids and sterols were the phytochemicals that showed widespread presence in a large number of plants under study. Ninety one (91) % of the plants contained, basic alkaloids and sterols. *Desmodium salicifolium* ethanol extracts had the highest levels of a wide range of phytochemicals.

### Table 18: Phytochemicals of ether extracts of selected plants

<table>
<thead>
<tr>
<th>Phytochemical</th>
<th>Sterols</th>
<th>Carotenoids</th>
<th>Basic alkaloids</th>
<th>Flavone aglycones</th>
<th>Anthracene aglycones</th>
<th>Coumarins</th>
<th>Chlorophyll</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Moringa oleifera</em></td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td><em>Lantana trifolia</em></td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td><em>Sida cuneifolia</em></td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td><em>Vernonia cineria</em></td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td><em>Tetredenia riparia</em></td>
<td>-</td>
<td>+</td>
<td>+++</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>++</td>
</tr>
<tr>
<td><em>Desmodium salicifolium</em></td>
<td>+++</td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+++</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><em>Persea americana</em></td>
<td>++</td>
<td>-</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td><em>Aspilia Africana</em></td>
<td>+</td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td><em>Syzygium cuminii</em></td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Albizia coriaria</em></td>
<td>+++</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Leonotis nepetifolia</em></td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

**Key**

- The extract remains clear with no change: Absence of the phytochemical  (-)
- Faint changes against dark background: Trace of the phytochemical  (+)
- Definite change noticed: Presence of phytochemical  (++)
- Heavy change noticed: Intense Presence of phytochemical  (+++)
For ethanol extracts, alkaloid salts were the phytochemicals that showed widespread presence in a large number of plants under study. A hundred (100) % of the plants contained, alkaloid salts. Sterol glycosides and coumarin derivatives were also present in a number of plants but were in trace amounts in most of them. *Aspilia africana* ethanol extracts had the highest levels of a wide range of phytochemicals.

**Table 19: Phytochemicals of ethanol extracts of selected plants**

<table>
<thead>
<tr>
<th>Phytochemical</th>
<th>Reducing sugars</th>
<th>Alkaloid salts</th>
<th>Sterol glycoside</th>
<th>Coumarin derivatives</th>
<th>Flavonones</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Moringa oleifera</em></td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td><em>Lantana triflora</em></td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Sida cuneifolia</em></td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td><em>Vernonia cineria</em></td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Tetredenia riparia</em></td>
<td>-</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Desmodium salicifolium</em></td>
<td>+</td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Persea Americana</em></td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Aspilia Africana</em></td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>++</td>
<td>-</td>
</tr>
<tr>
<td><em>Syzygium cuminii</em></td>
<td>++</td>
<td>++</td>
<td>+</td>
<td>++</td>
<td>-</td>
</tr>
<tr>
<td><em>Albizia coriaria</em></td>
<td>++</td>
<td>++</td>
<td>-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><em>Leonotis nepetifolia</em></td>
<td>+</td>
<td>++</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
</tbody>
</table>

**Key**
The extract remains clear with no change: Absence of the phytochemical (-)
Faint changes against dark background: Trace of the phytochemical (+)
Definite change noticed: Presence of phytochemical (++)
Heavy change noticed: Intense Presence of phytochemical (+++)
5.5 Discussion

Plant cells contain sequestered glycosides and release them when ruptured by injury or infection. These glycosides may have antimicrobial activity against the invading pathogens or may be hydrolysed by glycosidases to yield more active aglycones in the case of phenolic compounds, these may be oxidized to highly reactive antimicrobial quinines and free radicals (Dean and Kuc, 1987). Thus damage to a few cells may rapidly create an extremely hostile environment for a developing pathogen. Phytochemical screening of bioactive plants extracts has revealed the presence of alkaloids, carbohydrates, lactones, proteins, tannins, flavanoids, sterols, terpenes, glycosides, and saponins, of these, flavonoids and tannins have been linked to antibacterial activity and antidiarrheal activity (Ahmad et al., 2006). Different phytochemicals display various mechanisms of action such as increasing colonic water and electrolyte re absorption and inhibiting intestinal motility, while some components have been shown to inhibit specific pathogens (Ahmad et al., 2006). Phytochemicals such as saponins, terpenoids, flavonoids, tannins, steroids and alkaloids have anti-inflammatory effects (Lui, 2003). Steroids and triterpenoids have analgesic properties (Sayyah et al., 2004 and Maldirajan et al., 2006). It is therefore probable that these phytochemicals are responsible for the healing properties of the plants which have been claimed by the farmers in this study.

In this study, tannins were present in 91% of the plants tested and may be responsible for the good antibacterial activity demonstrated by these plant extracts. Previous studies have shown that tannins have been found to form irreversible complexes with proline-rich proteins resulting in the inhibition of the cell protein synthesis, they bind proteins and adhesins, inhibit enzymes and complex with cell wall (Iqbal et al., 2006). Tannic acid which is a mixture of gallic acid esters of glucose can be used as a topical preparation for cold sores (Heinrich et al., 2004). Many human physiological activities, such as stimulation of phagocytic cells, host-mediated tumor activity, and a wide range of anti-infective actions, have been assigned to tannins (Haslam, 1996). One of their molecular actions is to complex with proteins through so-called nonspecific forces such as hydrogen bonding and hydrophobic effects, as well as by covalent bond formation (Haslam, 1996). Thus, their mode of antimicrobial action may be related to their ability to inactivate microbial adhesins, enzymes, and cell envelope transport proteins, they also complex
with polysaccharide (Brownlee et al., 1990). This probably explains the reason as to why the plants containing these tannins showed good antibacterial activity. It is also not surprising that over 22% of the farmers used these plants in the management of poultry diseases. Basic alkaloids and alkaloid salts were also present in most of the plants tested and may be responsible for the good antibacterial activity demonstrated by these plant extracts. Alkaloids have been reported to be responsible for the antibacterial activity in some plants (Doughari, 2006). Earlier studies have also shown that alkaloids possess antimicrobial properties (Osbow, 2003). Studies have demonstrated that alkaloids have pharmacological effects and could be associated with inhibition of nucleic acid, protein, and membrane phospholipids biosynthesis (Shelton, 1991). The mechanism of action of highly aromatic planar quaternary alkaloids such as berberine, an important representative of the alkaloid group and harmine is attributed to their ability to intercalate with DNA (Phillipson and O'Neill, 1987). This probably explains the reason as to why the plants containing these basic alkaloids and alkaloid salts showed good antibacterial activity.

Furthermore, sterols and sterol glycosides were present in most of the plants tested and may be responsible for the good antibacterial activity demonstrated by these plant extracts. Earlier studies have shown that sterols possess antibacterial and antimicotic activity and have been shown to act as inhibitors of tumor promotion in vivo (Yasukawa et al., 1991). Sterols were found to inhibit tumor promotion in two-stage carcinogenesis in mice (Kasahara et al., 1994). Sterols also exhibit inhibitory effect on HIV reverse transcriptase (Akihisa et al., 2001). Sterols were also shown to possess anti-inflammatory activity after topical application (Gomez et al., 1999). The presence of these sterols has been reported to account for the exertion of antimicrobial activity by plants containing them (Pretorius and Watt, 2001). The presence of these sterols may contribute to the good antibacterial activity exhibited by these plants.

A few plants contained traces of flavonoids in this study and presence of these may be partly responsible for the medicinal properties of these plants. Previous studies have reported that flavonoids being phenolic compounds are water soluble antioxidants and free radical scavengers which are capable of preventing oxidative cell damage and have strong anticancer activity (Okwu, 2004). Reports also say that many disease states are known to be exacerbated by the
presence of free radicals such as superoxide and and hydroxyl and flavonoids have the ability to scavenge and effectively mop up these damaging oxidizing species (Okwu, 2004). Alan and Miller, (1996) reported that the potent antioxidant activity of flavonoids, their ability to scavange hydroxyl radicals, superoxide anions and lipid peroxide radicals may be the most important function of flavonoids. Catechins, the most reduced form of the C₃ unit in flavonoid compounds occur in oolong green teas the reason why they have important dietary significance is because they are strong antioxidants (Kaufman et al., 1999). It is therefore probable that the reason as to why these plants were used by over 22% of the farmers in the management of poultry diseases is because of such important phytochemicals with medicinal properties.

A few plants also contained traces of carotenoids and presence of these may be partly responsible for the medicinal properties of these plants. The carotenoids are strong antioxidants, being preferentially oxidized over biological molecules such as nucleic acids and proteins. It is thought that many disease states such as certain cancers and heart disease are exacerbated by species that cause oxidation; therefore the presence of these compounds may retard the development of such diseases (Heinrich et al., 2004). Probably the reason as to why these plants were used by some of the farmers in the management of poultry diseases was partly because of these important phytochemicals with medicinal properties.

In conclusion therefore, the study showed that these plants contained compounds of medicinal importance. It is probable that these constituents were responsible for the antibacterial activity that they exhibited against the four bacterial species; Streptococcus feacalis, Staphylococcus aureus, Eschericia coli and Salmonella typhymurium. These active compounds in these plants could find place in treatment of various bacterial infections in poultry where they can be used as alternatives to conventional antibacterial drugs. This could reduce the unnecessary use of antibiotics in poultry which is making disease-causing bacteria more resistant to the drugs and diminishing the drugs’ power to treat life threatening disease in humans and animals.
CHAPTER SIX
GENERAL CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

- The study has been able to establish and document the important medicinal plants used in management of poultry diseases in Masaka and how they are used.
- The study has also demonstrated significant antibacterial activity in some of these plants especially in their ether and ethanol extracts. The water extracts of some plants have also demonstrated good antibacterial activity.
- This study has further confirmed that these plants contain the phytochemicals which have healing properties according to previous reports.
- The study will solve some of the problems in the poultry industry farmers face. Farmers could also use plants like *Moringa oleifera* to treat a number of bacterial diseases while *Persia americana* could be used to treat salmonellosis. The other important plants are *Leonotis nepetifolia* and *Lantana trifolia* and these could be used to treat staphylococcal infections in poultry. The water extracts of these plants demonstrated good antibacterial activity.
- Positive assays of tannins, sterols, basic alkaloids and alkaloid salts in these plants clearly demonstrate that these plant species may provide bioactive compounds.

However in this study I did not isolate the active compounds responsible for antibacterial activity while clinical trials and toxicity studies were also not carried out. Therefore further research should be carried out on these and once it is done these will serve as lead compounds in the manufacture of novel drugs which farmers can use as one of the approaches in prevention and control of diseases. This could also be an important health and economic resource.

6.2 Recommendations

- The extracts of these plants should be further analyzed to isolate the specific antibacterial principles in them.
Toxicity studies of the effective plants should also be done to determine the safety indices of the extracts. Studies to determine the mechanisms of the action, compatibility with other drugs, side effects and other important parameters should be done.

These plants should be studied more extensively to explore their activity on other organisms like other bacteria species, protozoa and helminths among others.

Clinical trials should be carried out to explore the potential of these plant extracts in the treatment of these infectious diseases.


Map of Masaka District showing subcounties as it appeared on 9/7/2010. [http://www.pakistan-karachi.info/Masaka](http://www.pakistan-karachi.info/Masaka). 

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Map of Uganda showing Masaka District as it appeared on 9/7/2010. [http://www.icoduganda.org](http://www.icoduganda.org)


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APPENDIX
Appendix I

Sample interview I

Checklist questions for focus group discussions

1. List the main poultry disease/conditions in the community and carry out pair wise ranking on the major five
2. Discuss those ones that can affect human beings
3. Discuss the main methods of controlling poultry diseases in the community
4. Discuss whether there is any specific person knowledgeable in the control of poultry diseases in the community
5. What indigenous knowledge do you know in the control and treatment of poultry diseases in the community?
6. For each plant you give, list down the number of people in your group who use them in the treatment of poultry diseases and conditions.
7. How do you prepare these remedies before you administer them to your birds?
8. Discuss the advantages and disadvantages of indigenous technical knowledge in the control and treatment of poultry diseases.

Sample interview II

Checklist questions for key informants

1. Give the main methods you use to control and treat poultry diseases
2. Do you use any conventional methods to treat and control poultry diseases, if not why?
3. What indigenous knowledge do you know in the control and treatment of poultry diseases?
4. How did you get to know this knowledge?
5. How do you prepare these remedies before you administer them to your birds?
6. Which difficulties do you encounter in this practice?
7. Do your birds really cure?
8. Discuss the advantages and disadvantages of indigenous technical knowledge in the control and treatment of poultry diseases.
Appendix II

Five most frequently used concoctions selected and prepared under field conditions.

Concoction 1
In a big container, five leaves of aloe vera plus a hand full of Vernonia amygdalina leaves plus a hand full of Vernonia cineria leaves plus ten leaves of Jatropha curcas were put in seven liters of water. These were then squeezed to produce juice. The juice was decanted and packed and the leaf particles left behind for disposal. The juice was stored in a cool dry place. This concoction was used by farmers for prevention of all poultry diseases.

Concoction 2
In a big container, a hand full of Sida cuneifolia leaves plus a hand full of Momordica foetida leaves plus ten tea spoons of ash were squeezed in five liters of water. The juice was decanted off and stored in a cool dry place and the leaf particles left behind for disposal. This concoction was used by farmers for treatment of fever in poultry.

Concoction 3
In a big sauce pan, five leaves of Aloe vera, a hand full of Bidens pilosa leaves and a hand full of Vernonia amygdalina leaves were put in fifteen cups of water and left to boil until at least five cups of water evaporated off. It was then left to cool to room temperature, sieved and the juice kept in a cool dry place. The residues were disposed of. This concoction was used for treatment of fever in poultry.

Concoction 4
In a big sauce pan, five leaves of Aloe vera plus a half a hand full of Capsicum annum plus ten leaves of Jatropha curcas were put in fifteen cups of water and left to boil until at least five cups of water evaporated off. It was then left to cool to room temperature, sieved and the juice kept in a cool dry place. The residues were disposed of. This concoction was used for treatment of fever in poultry.

Concoction 5
In a big sauce pan, five leaves of Aloe vera plus a hand full of Vernonia amygdalina leaves plus a hand full of Vernonia cineria leaves plus five roots of the male plant of Carica papaya were put in fifteen cups of water and left to boil until at least five cups of water evaporated off. It was then left to cool to room temperature, sieved and the juice kept in a cool dry place. The residues were disposed of. This concoction was used for treatment of fever in poultry.
### Appendix III

**Table 20: The phytochemicals in each of the eleven plants screened**

<table>
<thead>
<tr>
<th>Plant Name</th>
<th>Phytochemicals</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lantana trifolia</em></td>
<td>Tannins, Steroid glycosides Glucides, Alkaloid salts, Saponins, Sterols, Basic alkaloids, Chlorophyll, Carotenoids, Anthracenoside aglycones, Polyphenols, Alkaloids salts, Reducing compounds, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Sida cuneifolia</em></td>
<td>Glucides, Tannins, Steroid glycosides, Alkaloid salts, Polyuronides, Reducing compounds, Starch, Coumarins, Basic alkaloids, Flavone aglycones, Chlorophyll, Sterols, Alkaloids salts, Coumarin derivatives, Polyphenols, and Flavanones</td>
</tr>
<tr>
<td><em>Vernonia cineria</em></td>
<td>Glucides, Reducing compounds, Steroid glycosides, Alkaloid salts, Tannins, Basic alkaloids, Lipids/ fatty acids, Sterols, Carotenoids, Chlorophyll, Alkaloids salts and Coumarins, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Tetredenia riparia</em></td>
<td>Glucides, Reducing compounds, Tannins, coumarins, Steroid glycosides, Alkaloid salts, Basic alkaloids, Chlorophyll, Coumarin derivatives, Volatile oils and Carotenoids.</td>
</tr>
<tr>
<td><em>Desmodium salicifolium</em></td>
<td>Steroid glycosides, Glucides, Alkaloid salts, Saponins, Anthracenosides, Anthocyanosides, Anthracenoside aglycones Sterols, Carotenoids, Basic alkaloids, Flavonoic aglycones, Reducing compounds, Alkaloids salts and Coumarins derivatives</td>
</tr>
<tr>
<td><em>Persea Americana</em></td>
<td>Reducing compounds, Tannins, Anthracenosides, coumarins, Steroid glycosides, Anthocyanosides, Alkaloid salts, Polyuronides, Saponins, Sterols, Basic alkaloids, Chlorophyll, Volatile oils, Alkaloids salts, Polyphenols, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Aspilia Africana</em></td>
<td>Glucides, Reducing compounds, Tannins, coumarins, Steroid glycosides, Starch, Chlorophyll, Basic alkaloids, Flavonoic aglycones, Sterols, Polyphenols, Alkaloids salts, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Syzygium cuminii</em></td>
<td>Glucides, Anthracenosides, Reducing compounds, Tannins, coumarins, Flavone glycosides, Starch, Alkaloid salts, Saponins, Sterols, Basic alkaloids, Polyphenols, Alkaloids salts, Sterol glycosides, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Albizia coriaria</em></td>
<td>Glucides, Reducing compounds, Tannins, Anthracenosides, coumarins, Flavone glycosides, Sterol glycosides, Alkaloid salts, Polyuronides, Saponins, Sterols, Volatile oils, Basic alkaloids, Alkaloids salts, and Polyphenols, Coumarin derivatives</td>
</tr>
<tr>
<td><em>Leonotis nepetifolia</em></td>
<td>Tannins, coumarins, Flavone glycosides, Sterol glycosides, Glucides, Alkaloid salts, Polyuronides, Basic alkaloids, Sterols, Coumarin derivatives, Carotenoids, Chlorophyll, Alkaloids salts, Reducing compounds.</td>
</tr>
</tbody>
</table>
Appendix IV

The pictures of selected plants

Moringa oleifera

Lantana trifolia

Sida cuneifolia

Tetredenia riparia
Aspilia Africana

Persea americana

Syzygium cuminii

Leonotis nepetifolia
Desmodium salicifolium

Albizia coriaria

Vernonia cinerea
Appendix V
Map of Uganda showing the location of Masaka

Key
• Masaka District
Appendix VI

Map of Masaka showing sub counties under study

Sub counties under study