

بسم الله الرحمن الرحيم

Sudan University of Science and Technology

College of Graduates Studies



**Effect of Mixture of Three Herbal Essential Oils on
Performance, Carcass Yield and Blood Serum
Constituents of Broiler Chicks**

أثر إضافة خليط ثلاث من الزيوت الأساسية العشبية على الاداء
الانتاجى وصفات الذبيح وخصائص الدم للدجاج اللحم

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الآية

بسم الله الرحمن الرحيم

(يَا أَيُّهَا الَّذِينَ آمَنُوا إِذَا قِيلَ لَكُمْ تَفَسَّحُوا فِي
الْمَجَالِسِ فَافْسَحُوا يَفْسَحِ اللَّهُ لَكُمْ وَإِذَا قِيلَ
انشُرُوا فَاَنْشُرُوا يَرْفَعِ اللَّهُ الَّذِينَ آمَنُوا مِنْكُمْ وَالَّذِينَ
أُوتُوا الْعِلْمَ دَرَجَاتٍ وَاللَّهُ بِمَا تَعْمَلُونَ خَبِيرٌ).

صدق الله العظيم

سورة المجادلة الآية (١١)

Dedication

الى من احمل اسمه ولم اره الى روجه

ابي

الى من بها اكبر وعلها اعتمد بعد الله ... الى شمعة متقدده تنير ظلمة حياتى ... الى من بوجودها اكتسب قوة ومحبه لا حدود لها ... الى من كافحت لتتنير لى دربي وتعلمنى.

امى الحبيبة

الى سندی وقوتى

الى من اثرونى على انفسهم

الى من علمونى علم الحياة

الى من اظهروا لى ماهو اجمل من الحياة

إخوانى

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Abstract

The experiment was conducted to evaluate the response of broiler chicks diets supplemented with graded level of mixed essential oils (Anise , Clove , Caraway) (1:1:1) on the Performance, Carcass Yield and Blood Serum Constituents of Broiler Chicks .

A total of 160 seven days old unsexed (Ross 308) broiler chicks were subjected to 42 days experimental period. Chicks were randomly divided into five groups of 21 chicks per each pen with four replicate for each group. Five experimental diets were formulated (A ,B ,C ,D , and E) as follows , diet A as negative control , diet B was control diet supplemented with antibiotic (Neomycin 16mg/kg) diets C , D , and E were control diet supplemented with mixed essential oils (MEO) at 200 , 400 , and 600g/kg respectively .

Result obtained showed no significant difference in the performance parameters (body weight, body weight gain, feed intake and FCR). However group fed a 200g/kg MEO recorded the highest values. Result also showed no significant difference in dressing percentage, giblet, commercial cuts, subjective and objective of meat quality parameters. The addition of 200g/kg MEO recorded economic benefits, also the study showed that the addition of MEO as growth promoter in broiler diets have a similar effect as that with antibiotic without any adverse effects.

ملخص التجربة

تم اجراء هذه التجربة لدراسه اثر تغذيه الدجاج اللاحم على علائق تحتوي على تراكيز مختلفه من خليط مكون من زيت القرنفل واليانسون والكرابوية كاضافة علفية طبيعية على الاداء الانتاجي وصفات الذبيح للدجاج اللاحم

تم استخدام 160 كتكوت لاحم عمر يوم واحد غير مجنس , ووزعت عشوائيا الى خمس معاملات بواقع اربع مكررات لكل معاملة و8 كتاكيت بكل مكرر , تمت تغذيه المجموعه الاولى على عليقة اساسية بدون اضافع اى محفز للنمو والمجموعه الثانية عذيت على عليقة اساسية مضاف اليها المضاد الحيوي الكولاسين 20جم/كجم عليقة كمحفز للنمو اما بقية المجموعات الاخرى فقد غذيت على العليقة الاساسية مضاف اليها خليط من زيت القرنفل واليانسون والكرابوية كمحفز للنمو بتركيز 600/400/200 جرام /طن تم تكوين العليقة الاساسية وفقا للاحتياجات الغذائية لدجاج اللحم الصادر من (NCR 1994) .

تمت مراقبه القطيع وتسجيل قياسات الاداء الانتاجي كما تم حساب نسبه التصافي بالاضافه الى حساب التقييم الاقتصادي

دللت النتائج المتحصل عليها من التجربة بانه لا توجد اى فروقات معنوية ($P < 0.05$) في الوزن النهائي ووزن الجسم المكتسب ومعدل التحويل الغذائي بين المعاملات المحتوية على خليط الزيوت بمستوي 200, 400, 600 جم /طن مقارنة بالمجموعه القياسية الضابطة والمجموعه الضابطة المضاف اليها المضاد الحيوي .

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Chapter One

Introduction

Chapter one

Introduction

In the Sudan the poultry industry has shown the most rapid increase in five last years. In name the broiler are the most efficient in converting raw feed stuffs by products into high protein food which is urgently needed to improve the nutritional standards of human.

The practice of feeding live stock with sub therapeutic levels of antibiotics has been in use for over fifty years antibiotic, usage is possibly the most important factor that promotes the emergence , selection and dissemination of antibiotic resistant microorganism in both veterinary and human medicine, At slaughtering, resistant strains from the gut may contaminate poultry carcasses and as a result poultry meats are often associated with multi resistant microorganism therefore, the use of antibiotic growth promoters has been banned in many countries, especially in European union (Cantonal 2007) as a result new commercial additives of plant origin, considered to be natural products that consumers would accept, have been proposed to live stock producers. Herbs, spices and various plant extracts have received increased attention as possible antibiotic growth promoters replacement, in this view , aromatic plants and essential oils extracted from these plants become interesting due to their antimicrobial (Sing etal , 2002) antioxidant (Dargland etal., 2003, Botsogou .,etal 2004) anti Bacterial (Dorman and Deans, 2000) antifungal (Jantan etal ,2003) activates and as hypo cholestoleemics and stimulate effect of animal digestive enzyme and improve utilization of digestive products through enhanced liver functions.

As aromatic plants caraway (Caram carvi) is annual herbs its oil has carvone and limonene as the essential components (Sedlakova etal, 2001) has the property of antibacterial fungicidal improve test , gas reliving or to relive gassy indigestion

, it has effect calming and soothing the nerves and digesting system . Similar as a medicinal plant, Anise has been used as stimulating effect of digestion and anti parasitic (Cabuck et al., 2003) anti bacterial antifungal and antipyretic Eugenol is a major component of Clove extract and exhibits wide , range of anti microbial activity in vitro in addition , antiseptic , appetite and digestion stimulant strong anti microbial and anti fungal, antiphrostatic (Kim et al., 2004) and anti oxidant activities of clove and its ingredients have been reported .

The objective of the present study was to examine the effect of dietary supplementation of an herbal essential oil mixture (EOM) at different level on growth performance carcass yield and blood serum constituents of broiler chicks.

Chapter Two

Literature Review

Chapter Two

Literature Review

2-1: Feed additives:

Feed for broilers and laying hens is formulated to contain an optimum nutrient concentration obtainable at reasonable cost for desirable growth production and efficiency of feed utilization. To insure that dietary nutrition are ingested, digested, protect from destruction, absorbed and transported to the cells of body, certain non-nutritive feed additives are sometimes used in addition on the optimum concentration and balance nutrients .Other feed additives have been used to alter the metabolism of the chicken in an effort to produce better growth or more desirable finished products (Lessons and Summers, 2001)

Additives are usually included in the feed mixture in very small quantities and require very careful Modern intensive poultry production has achieved phenomenal gains in the efficient and economical production of high quality and safe chicken meat, eggs and poultry bio products. At the same time as making gains in production and efficiency, the industry has had to maximize the health and well-being of the birds and minimize the impact of the industry on the environment.

Feed additives have been an important part of achieving this success feed additives.

2-2: Antibiotic

Antibiotic feed additives have been used for more than 50 years to enhance growth performance .However, the currents endistotives because of public concern about antibiotic residues in animal products and the potential evolving of antibiotic resistant bacteria. As a consequence, new commercial additives of plant origin, considered to be natural products that the consumer would accept, have been proposed to animal producers. The chemical components of most essential oils from plants are generally recognized as safe, and are used

commonly in the food industry (Varel, 2002). Essential oils derived from herbs have been shown to have antimicrobial effects. Their antimicrobial mode of action consists of interactions with the cell membranes of microorganisms by changing permeability for nutrients (Ultee et al., 1999). Moreover, there is evidence to suggest that herbs, spices and various plant extracts have appetizing and digestion-stimulating properties and antimicrobial effects (Langhout, 2000 Madrid et al., 2003, Alçiçek et al., 2004) and (Zhang et al., 2005). The improvement in feed efficiency achieved with essential oil mixtures could be attributed to their positive effects on nutrient digestibility. Breeder age was found to influence broiler performance differently during the various phases of the growing period (Peebles et al., 1999a; b). It was shown that body weight of broilers from 51-wk old breeders was significantly higher than that of broilers from 63-wk old breeders. Information in the literature regarding the effects of added dietary essential oils and breeder age throughout production on performance and slaughter yield is lacking. Antibiotics represent a group of chemical compounds produced biologically by certain plants or microorganisms, usually a fungus, which possess bacteriostatic or bactericidal properties. Some antibiotics are particularly effective against negative bacteria, other antibiotics are most effective against positive bacteria. While some, termed broad spectrum antibiotics are effective against a wide range of gram negative bacteria the use of antibiotics as growth promoters in poultry diet was started around years ago, when the first indication of beneficial effects on production efficiency in poultry was reported by (Moore et al, 1946) antibiotics had been approved for growth promotion in experimental, and many different groups of antibacterials have subsequently been approved for on farm use as growth promoters and the antibiotic as growth promoter may produce one or more of the following effects and they may favor the growth nutrients synthesizing microbes or inhibit that of nutrient destroying microorganisms and antibiotics may inhibit the growth of organisms that produced excessive amount of ammonia and other toxic nitrogenous waste

products in the intestines and they may improve availability or absorption of certain nutrients and they may improve feed or water consumption or both and antibiotic may instances prevent or cure actual pathological disease . Which occur either in the intestinal tract or systemically; and they may reduce that maintenance cost associated with turnover of the intestinal epithelium (Kahn et al, 2005).

Antibiotics have been used in poultry nutrition for improving growth performance, preventing some specific pathogenic microorganism and increasing some useful microorganism in intestinal micro flora over the years. However, antibiotics used as growth promoters in animal feeds have been banned recently due to potential development of anti-biotic resistant human pathogenic bacteria. Nowadays, the possibility of using new natural alternative additives instead of antibiotics in animal diets is being researched (Weber et al., 2012). Probiotics as alternatives of antibiotics are live microbial feed supplements which beneficially affect the host by improving its intestinal microbial balance (Fuller. 2001). There are viable single or mixed cultures of microorganisms that when given to animals or humans, beneficially affect the host by improving the properties of the indigenous micro flora (Kabir, 2004).). Other alternatives are plant ex-tracts. Plant extracts contain many active components, including essential oils, which have a wide range of pharmacological activities. The essential oil extracted from aromatic plants have been shown antibacterial, anticoccidial (Giannenas et al, 2003), antifungal antioxidant (Bostoglou et al, 2004) activities. Some results showed that aromatic plants and their extracts have a positive effect on growth performance in chickens (Guler et al. 2005) or have not any significant effect (Bostoglou et al. 2002). When the effect was positive, weight gain and feed intake were increased where as the feed gain ratio was lowered when compared to control. Dietary essential oils can also improve digestion. A number of studies have reported the positive effect of spices or their active components on digestion process. They

have been shown to stimulate bile salt secretion and digestive enzyme activities of intestinal mucosa and of pancreas (Hernandez et al. 2004).

2-2: Essential oils in poultry nutrition:

2-3-1: Definition of essential oils:

There are no doubts that dietary antibiotics have played a fundamental role in animal production as growth and health promoter. However, it is the current trend to look for alternatives to antibiotics due to public concerns as to their in-feed use and the resulting residues, and subsequent occurrence of antibiotic-resistant bacteria. Dietary antibiotics presumably act on intestinal microflora, leading to improved animal performance. Most supplements claimed to be alternatives to antibiotics have effects on the microflora either directly or indirectly (Taylor, 2001). Thus, the microflora in chickens should not be ignored in relation to birds' performance. However, unlike ruminant and non-ruminant herbivores, chickens have little nutritional benefit from the microflora (Moran, 1982). Microflora, on the other hand, can adversely affect the host if it is not properly controlled. It is well-known that non-starch polysaccharides present in cereals stimulate growth of microflora (Smits, 1998), leading to lower growth performance. Gut microflora can hydrolyze conjugated bile salts (Feighner, 1987) which limits fat digestion (Krogdahl, 1985). It is thus clear that controlling the microflora could positively influence birds' performance and that feed supplements with antimicrobial activity are potential alternatives to antibiotics.

Various authors have extensively reviewed and compared various compounds regarded as alternatives to antibiotics in animal production (Mellor, 2000a,b; Wenk, 2000; Taylor, 2001). Essential oils are already marketed for use in animal production and are claimed to be "digestive enhancers" (Williams 2001). This urged us to collect published information on essential oils and assess the possible application in poultry nutrition.

Essential oils are complex compounds, and their chemical composition and concentrations of various compounds are variable (Lee et al., 2004). Essential Oils basically consist of two classes of compounds, the terpenes and phenylpropenes, depending on the number of carbon building blocks. The exact anti-microbial mechanism of essential oils is poorly understood. However, it has been suggested that their lipophilic property (Cornner, 1993) and chemical structure (Farag et al., 1989a; 1989b) can play a role. It was suggested that terpenoids and Phenyl ropanoids can penetrate the membranes of the bacteria and reach the inner part of the cell because of their lipophilicity (Helander et al., 1998). Moreover, structural properties, such as the presence of the functional groups (Farag et al., 1989c) and aromaticity (Bowles and Miller, 1993) are also responsible for the antibacterial activity of essential oils. Specific effects of the essential oils on chicken performance have not received much attention because poultry may not acutely respond to flavor when compared to pigs, although there is evidence (Deyoe et al., 1962) that flavors could affect feed intake negligibly in chicken. Various plant extracts, especially essential oils, have been studied for their antimicrobial abilities. Most of researches done in this area have been performed in vitro, but there have been few studies with live poultry flocks. Recent study involving live birds showed that blends of the primary components of the essential oils could be used to control *Clostridium perfringens*, the bacterium that causes necrotic enteritis in broilers (Mitsch et al., 2004). Ground thyme has been shown to inhibit the growth of *S. typhimurium* when added to media (Aktug, 1986). The essential oil of the thyme has been shown to inhibit the growth of the *E. coli* in media al., Aromatic plants and essential oil extracted from these plants has been used as alternatives to antibiotics. For this reason, these plants are becoming more important due to their antimicrobial effects and the stimulating effect on animal digestive system (Osman et al., 2005).

Essential oil is a mixture of a fragrant , volatile compounds ,named after the aromatic characteristics of plant materials from which they can be isolated (Oyen and Dung,1999) the term essential was adapted from the theory of “quanta essential “ proposed by Paracelsus who believed that this quintessence was the effective element in medical preparation. Because the term, essential oil, is poorly defined concept from medieval pharmacy, the term ‘volatile oil ‘ has been proposed (Hay and Waterman 1993). However the name of essential oil will be used preferentially in this review.

Essential oils are very complex mixtures of compounds and their chemical compositions and concentration of individual compounds are variable for example the concentration of two predominant components of thyme essential oils. (Lawrence , 1984).

2-4-2: Classification of essential oils:

Essential oils have long been recognized because of their antimicrobial activity (Deans and Ritchie, 1987 Paster et al 1990 Reddy et al 1991S and Hammer et al 1999). Due to this property, essential oils have gained much attention in investigations on their potential as alternatives to antibiotics.

2-5-3: Effect of essential oils on digestion process:

There are suggestions that dietary essential oils can improve digestion it might be reasoned that spices and herbs, from which essential oils are derived ,will positively affect food digestion (Pradeep et al ,1991Pradeep and Geervain,1994). A number of studies have reported the effect of spices or their active components on bile salt secretion (Bhat et al 1984, 1985 Bhat., , 1987.,Sambaiah, 1991) in addition , the dietary pungent principles I.e curcumin capsaicin , and piperine , have been shown to stimulate digestive enzyme activates .

2-6-3: In-vitro antimicrobial activities of essential oil

Essential oils have long been recognized because of their antimicrobial activity (Lis-Blachin et al.,). Due to this property, essential oils have gained much attention in investigations on their potential as alternatives to antibiotics for therapeutic purposes and applications in the cosmetics and food industry.,(Lee and Ahn 1998) found that cinnamaldehyde, derived from the cinnamon essential oil strongly inhibits and moderately inhibits *Lactobacillus acidophilus* isolated from human feces. The selective inhibition by cinnamaldehyde of pathogenic, intestinal bacteria may have a pharmacological role in balancing the intestinal micro biota. The wide range of antimicrobial activities of essential oils derived from cinnamon, thyme and oregano have been published, which supports their possible use as antimicrobial agents. It is reasonable to expect that the main components of essential oils displaying in vitro antimicrobial activity are responsible for this activity.

It is considered beneficial to keep the effective antimicrobial concentration of essential oils as low as possible due to their characteristic flavors. This problem can be overcome, as suggested by (Moleyar 1992), by using synergistic properties of different oils, thus improving the antimicrobial activity in spite of low dosages. This synergism was highlighted in studies of (Didry et al. 1994 and Montes:, 1998).

2-7: Antimicrobial mode of action of essential oils

The exact antimicrobial mechanism of essential oils is poorly understood. However, it has been suggested that their lipophilic property (Conner, 1993) and chemical structure could play a role. investigated how two isomeric phenols, carvacrol and thymol, and the phenylpropanoid, cinnamaldehyde, exerts their antibacterial effects on *Escherichia coli* and *Salmonella typhimurium*. Carvacrol and thymol both, in a similar fashion, disintegrated the membrane of bacteria, leading to the release of membrane-associated material from the cells to the external medium. On the other hand, cinnamaldehyde failed to affect the

membrane but did exhibit antibacterial activity, indicating that two molecules have different mechanisms underlying antibacterial activity. The authors, however, suggested that terpenoids and phenylpropanoids can penetrate the membrane of the bacteria and reach the inner part of the cell because of their lipophilicity, but it has also been ascribed to structural properties such as the presence of the functional groups and aromaticity. It is thought that membrane perforation or binding is the principle mode of action (Shapiro and Guggenheim, 1995; Stiles et al., 1995), leading to an increase of permeability and leakage of vital intracellular constituents (Juven et al., 1994), resulting in impairment of bacterial enzyme systems. The mechanism of antifungal action of cinnamaldehyde has been investigated (Kurita et al., 1979) and it was proposed that it takes place through the reaction with sulfhydryl groups, which are indispensable for the fungal growth, and that the formation of charge transfer complexes with electron donors in the fungus cell could lead to inhibition of cell division and thus interferes with cell metabolism. It was also reported that cinnamaldehyde inhibits fungal cell wall synthesizing enzymes (Bang et al., 2000).

2-8: In vitro antioxidant effects of essential oils

The anti-oxidative properties of the extracts of oregano, dittany, thyme, marjoram, spearmint, lavender and basil have been evaluated when added to lard kept at 75 °C (Economou et al., 1991). Oregano extract was found to be most effective in stabilizing lard, followed by thyme, dittany, marjoram and lavender. (Schwarz et al., 1996) and thymol and carvacrol (Aeschbach et al 1994, Aruoma 1997 and Baratta et al 1998) which are found in thyme showed strong antioxidant properties.

2-9: Effects of essential oils on lipid metabolism

Craig (1999) reviewed the role of herbs and their essential oils as to their cholesterol lowering properties and in the protection against cancer. (Elson et al 1989) reported the hypocholesterolemic effect of lemongrass oil, which is rich in

geraniol and citral, in human subjects. On the contrary, hardly any effects on plasma lipids other than cholesterol were observed (Cooke et al 1998).

2-10: Effect of essential oils on growth performance in chickens

The observed effects of essential oils on growth performance in chickens are either positive (Anonymous, 1997, Bassett 2000, Langhout, 2000, Kamel, 2001, Botsoglou et al., 2002). The authors explained the lack of effect by pointing out that the birds' performance was already superior, leaving no room for growth enhancing effects of the additives. Caution is required when interpreting the results of this may indicate that when experimental conditions and diets are marginal for the birds, growth enhancing effects of essential oils will be seen.

2-11: Micro flora fat digestibility and essential oils:

Schaedler (1973) stated that an ideal flora would allow optimum growth performance. Any alteration of the indigenous flora by diet or environment can be deleterious to the host. Extensive reviews (Jukes, 1955; Visek, 1978; March, 1979; Fuller, 1989; Vanbelle et al., 1990; Ewing 1994; Stavric, 1995) on the role of microflora on animal performance support the view of .It was thus assumed that dietary essential oils could have growth enhancing effects due to their actions on the intestinal microflora. This implies that the efficacy of essential oils on animal performance could be affected by the microbial status. In other words, their effects on germ-free chicks are expected to be negligible at best. This statement is backed up by a study of (Coates et al., 1963) who demonstrated that dietary penicillin had no apparent effect on growth performance by germ-free chicks. There are also indications showing that dietary antibiotics may not play a significant role in growth performance when birds are kept in a clean environment and fed well-balanced diets. On the other hand, when birds are challenged by gut microbial loads, then dietary essential oils can positively affect growth performance. This concept has been tested as to the efficacy of dietary antibiotics in counteracting the negative effects of rye in chickens (MacAuliffe, 1971; Wagner 1978; Patel et al., 1980) and Antonoiu

and, 1982; Honeyfield et al., 1983). It is well known that feeding rye to chickens increases number of bacteria in the intestine so that growth-enhancing property of dietary antibiotics can be seen. The microbial over-population by rye feeding is attributed to its pentosan contents which raise intestinal viscosity. It is suggested that intestinal viscosity, caused by ingestion of soluble fiber, impairs the normal digestion process so that more undigested materials travel to distal parts where they can be used as substrates by the microflora. Increased microbial populations are also apparent in the upper part of the small intestine where digestion occurs.

There is evidence (Kussaibati et al., 1982) that the intestinal microflora has a pronounced impact on fat digestion in chickens because of lowered bile acid availability. Bile salts are known to be a limiting factor for efficient fat digestion (Redinger et al. (1973) found that 87% of total bile acid pool is present in the intestine. Birds are well equipped with the efficient re-absorption of bile acids, the absorption be as high as 93% (Hurwitz et al., 1973). This reduces the need of hepatic synthesis of bile acids in order to maintain the bile salts pool in the chicken (Freeman, 1984), and can be an important factor especially in young chicks with limited secretion of bile acids . reported that when the bile concentration is lower than 50 to 60 mol/g of fat-free matter, the digestibility of tallow will be impaired.

It has been reported that the intestinal microflora can hydrolyse bile salts (Freigher 1987). This flora includes Clostridium, Lactobacillus, Pepto streptococcus, bifid bacterium, Fusobacterium, Eubacterium, Streptococcus, and Bacterioids reported that chickens fed on a diet rich in rye exhibited an 18-fold increase in bacterial cholytaurin hydrolase activity in ileal homogenates when compared their counterparts fed on corn diet. Unconjugated bile acids are less effective in forming micelles .It is thus expected that feeding soluble fiber to chicken can severely affect fat digestion.suggested that carboxy methylcellulose (CMC), which is a non-fermentable soluble fiber, can impair re-absorption of

bile acids, leading to lower bile acid availability. The extra fecal loss of bile acids can occur by either direct binding to the soluble fiber or by less efficient utilization of unconjugated bile acids (Angelin et al., 1982) .

2-12: Anise oils:

For centuries aromatic plants have been used worldwide as food and for medicinal purposes. Various biological activities, such as antioxidative (Botsoglou et al., 2002;; Florou et al., 2006), anticoccidial (Christaki et al., 2004; Florou-Paneri et al., 2006) or antimicrobial (Govaris et al., 2007;Botsoglou et al., 2010) properties have been identified in these plants. Consequently, an increasing interest in the use of these products in poultry nutrition has been experienced, especially since the complete ban by the European Union countries in 2006 (EU, 2005) on the use of antibiotics as growth promoters in animals.

Anise (*Pimpinella anisum.*), a member of the Apiaceae family, is an annual aromatic plant, native to the eastern Mediterranean and southwestern regions of Asia. The part of the plant used, is the fruit, in particular the seed and its essential oil., (Franz et al., 2005; Al-Beitawi et al., 2009).

Anise has been examined for its ant parasitic and digestion stimulating properties (Cabuk et al., 2003), as well as its antibacterial ,antifungal antipyretic (Afifi et al., 1994), antioxidant (Gulcin et al., 2003), antimicrobial (Al-Kassie et al., 2008), anthelmintic (Bhatti et al., 1996) and hypo cholesterolemic activities. Additionally, anise is reported to possess anticonvulsant (Pourgholam et al., 1999), antiepileptic (Janahmadi *et al.*, 2008) and muscle relaxant (Albuquerque et al., 1995) properties. Some studies have been conducted to evaluate the use of anise seed oroil in poultry nutrition, especially to replace antibiotics as growth promoters (Ciftci et al., 2005; Soltan et al., 2008).

It is well documented that dietary vitamin E, in the form of α -tocopheryl acetate, is a strong antioxidant in the body; though limited research has been conducted on its potential to improve performance and egg quality traits of laying Japanese

quail (Sahin et al., 2006). Antioxidants in poultry diets perform an important role in the good health and performance of poultry and the oxidative stability of their products, and poultry diets are routinely supplemented with antioxidants. Nevertheless, it remains unclear whether the antioxidant properties of aromatic plants are comparable to that of tocopheryl acetate, which is usually included in commercial poultry diets.

2-13: Clove oils

Clove oil is a dark-brown liquid, a distillate of flowers, stalks and leaves of the clove tree *Eugenia aromatic* (Soto 1995).

At present, effects on clove oil on commercially produced fish are studied in a project regarding the application of principles of pharmacovigilancy in aquaculture in the Czech Republic. In the first stage of the project, effects of clove oil on rainbow trout were studied (Velisek et al., 2005).

Anesthetics are used with increasing frequency in aquaculture, mainly to reduce the stress and to prevent mechanical damage to fish during handling.. At present, clove oil is used in the Czech Republic for short-term immobilization of fish before artificial spawning. The recommended concentration for anesthetic purposes is 30 mg/L water bath (Svoboda, 1999; Hamackova et al., 2001).

Clove oil has been used in food products perfumery in addition, antiseptic and digestion stimulant (Kamel, 2001) strong antimicrobial and anti fungal (Erhrch et al.,1995) ., an angelic and anti inflammatory (Feng et al . , 1987) an esthetic and anti carcinogenic, anti parasitic and anti oxidant (Dargland etal ., 2005) activities clove Eugenol is a substance found in clove oil that anti microbial properties .

Mukhtar (2010) studied that effect of dietary clove oil as a natural growth promoter on broiler performance , He found that addition of clove oil to broiler diets improve body weight and feed intake and the lowest rate mortality compound to antibiotic groups .

2-13: Caraway oils

Caraway (*Carum carvi*) is one of the most important medicinal plant cultivated in Poland on the area (Selder et al 2012). Caraway fruit (*Carvi Fructus*) are used as a component of herbal mixtures. Recommended as a digestive, carminative and lactogenic. Caraway essential oil obtained from fruit by hydro-distillation is transparent liquid, colorless with pleasant smell and spicy taste (Sadowska, 1998). The main constituents of oil are monoterpenes: carvone and limonene which usually make 95% of all oil. According to carvone content determines the quality of reports that most of essential oils performs strong antimicrobial activity specially oils (thyme, savory, clove) contained such phenols as: thymol, carvacrol or eugenol. Similar results were obtained by who indicated high activity of cinnamon, clove, oregano, savory and thyme oils, while testing 20 different oils Caraway oil was not tested.

Diabetes mellitus is a metabolic disorder of the endocrine system that is found in all parts of the world and is rapidly increasing worldwide. In spite of all the advances in therapeutics, diabetes still remains a major cause of morbidity and mortality. Currently available therapies for diabetes include insulin and various oral antidiabetic agents such as sulfonylureas, biguanides, α -glucosidase inhibitors, and glinides, which are used as monotherapy or in combination to achieve better glycemic regulation. Many of these oral antidiabetic agents have number of serious adverse effects; thus, managing diabetes without any side effects is still a challenge (Jung *et al.* 2006). The search for a cure for diabetes mellitus continues along traditional and alternative medicine. Many herbal supplements have been used for the treatment of diabetes, but not all of them have scientific bases to support their effectiveness. Caraway is a member of the group of aromatic Apiaceae. It is naturally found in North Africa, Turkey, Iran, India, Siberia, and Northern and Central Europe. The fruit of caraway is a schizocarp, which at harvest splits into two halves called seeds. Caraway has been used since ancient times, especially for the treatment of digestive disorders. For ancient

Egyptians, caraway soothes flatulence, digestive, breath freshener and anti helminthic (Abou El-Soud, 2010). Caraway is known to have anti hyperglycemic effect. showed that aqueous extract of caraway has antihyperglycemic effect in induced diabetic rats. The plant extract and volatile oils from *Carum carvi* have also been used as antiulcerogenic agents and anti-flatulent colic in infants ,Caraway has been commonly used in phytomedicines as antibacterial and laxative agent Caraway seed oil is promising oil with high levels of bioactive compounds. The major constituents of its seeds are carvone, flavonoids and limonene. In addition, myrcene, betacaryophyllene, thujone, anethole, and pinene are present as minor components The flavonoid constituents of caraway have been separated by means of chromatography on cellulose columns and constituents such as quercetin-3-glucuronides, isoquercitrin, quercetin 3-Ocaffeylglucoside, and kaempferol 3-glucoside were obtained However, little information is available on the effect of administration of caraway oil on kidney of diabetic experimental animals.

Mixed oil

Khattak et al., (2013) studied the effect of a novel commercial preparation of natural blend of essential oils from basil , caraway, lemon , orange , sage , tea and thyme (Tecnaroma herbal mix pl) on growth performance , blood biochemistry , cecal morphology and carcass quality of broilers . They found that birds fed diets supplemented with Tecnaroma herbal mix pl had significant heavier, And higher ($p < 0.05$) weight gain and had improved feed to gain ratio compared with the control group . the blood biochemistry results showed no difference between treatment , the carcass weight , breast weight , and relative percentage of breast meat increased when diets were supplemented with Tecnaroma herbal mix pl compared to control group .

Alcicek et al (2003) reported significant improvement in body weight , feed conversion ratio and carcass yield of broiler , fed on 48mg/kg of essential oil combination derived from selected herbs growing wild in turkey , of different

levels of studied the effect of an essential oil mix derived from oregano , clove and anise , they concluded that 200ppm essential oil significantly improves the daily live weight gain and feed conversion , ratio during a growing period of five week .

Cabuk eta l., (2014) fed 7 week old laying quail diets supplemented with essential oil mixture (24mg/kg feed) derived from many herbs : oregano laurel feed , sage feed , my rile feed , fennel seeds and citrus peel in comparison with the control diet or antibiotics . the result indicate that the essential oil mixture has beneficial effect as a dietary supplemental on egg production and feed conversation ratio.

Chapter Three

Material and Method

Chapter Three

Material and Method

3: Response of broiler chicks to different dietary levels of mixture essential oils:

This experiment was conducted at Poultry Production Farm College of Agricultural Studies Sudan University of Science and Technology during the period from 6 December 2013 – 10 January 2014, in which the ambient temperature ranged between 18-30C°

3-1: Experimental chicks:

A total of 160 one day old unsexed broiler chicks were purchased from commercial chicks Production Company in Khartoum (Mico poultry production farm) for the first week chicks were fed on a commercial prestart. After a week of adaptation period chicks were randomly distributed to five treatment group with four replicates of eight chicks per each replicate. Feed and water were provided adlibitum. Chicks were vaccinated against Gumboro disease at 11 day age and against Newcastle disease at 22 days of age through the drinking water. Soluble multivitamins compound given to chicks before and after three days of vaccinations in order to guard against stress.

3-2: Housing:

An open wire mesh side house was used ,the house was constructed on a concrete floor with local materials , with corrugated metal sheet roof and a solid brick western – eastern , wall up to 3 meters the eaves and 4-5 meters for apex ,One meters square pens , inside the house were prepared using wire mesh partitioning and each pen was equipped with one cleaned and disinfected feeder and drinker to allow adlibitum consumption of feed and water ,light was provided 24 hours in a form of natural light during the day and artificial light during the night . The house was cleaned and disinfected before commencement

of the experiment. About 3 cm wood shaving layer was laid and on pen floor as a litter material.

3-3: Experimental rations:

Essential oils (caraway, anise and clove) used in this experimental were purchased from Khartoum market, they were mixed at (1:1:1) ratio to be used as a natural growth promoter. Five experimental diets were formulated to meet or exceed the requirements of broilers chicks according to (NRC 1994) the diets were iso nitrogenous and isocaloric the chicks were fed on 5 dietary treatments. The first group A fed on based diet (negative control) without growth promoters. The second group B fed on basal diet containing an antibiotic as chemical growth promoter, Neomycin 20g/t(positive control). The other groups C,D and E were fed on the basal diet supplemented with mixture essential oils (anise ,clove , caraway) as natural growth promoter at levels of 200 , 400 , 600 mg/kg respectively .

The ingredients percent composition and the calculated chemical analysis of the experimental diet were presented in table 3 and 4 experimental diets were fed for 6 weeks.

Table (1) The ingredients percent composition and the calculated chemical analysis of the experimental diets:

Item	Control	antibiotic	Mix 200	Mix400	Mix600
Sorghum fetarita	65.5	65.5	65.5	65.5	65.5
Ground nut cake	13	13	13	13	13
Sesame cake	15	15	15	15	15
Oyster shell	1	1	1	1	1
Concentrate	5	5	5	5	5
Salt	0.25	0.25	0.25	0.25	0.25
Lysine %	0.1	0.1	0.1	0.1	0.1
Methionine	0.15	0.15	0.15	0.15	0.15
Total	100	100	100	100	100

Antibiotic (Neomycin)	-	20	-	-	-
Oil mg/kg	-	-	200	400	600

Table No (2): Calculated chemical analysis of experimental Diets:

Item	control	Antibiotic	Mix 200gm/kg	Mix400gm/kg	Mix600gm/kg
Energy	3176	3176	3176	3176	3176
Protein	23.13	20.03	20.03	20.03	20.03
Laycin	1.137	1.17	1.17	1.17	1.17
Methuonin	0.44	0.54	0.54	0.54	0.54
Calcium	1.1	0.99	0.99	0.99	0.99
Available Phosphorus	0.73	0.47	0.47	0.47	0.47

Data collected:

3-4: Performance data:

Average live body weight and feed consumption (g) for each group were taken weekly intervals throughout the experimental period with the help of standard balance. Health of the experimental stock and mortality data were closely observed and recorded daily, weight gain and feed conversion ratio were calculated weekly minimum and maximum temperature were recorded daily .

3-5: Blood serum profile:

Sera sample obtained from each group following collection of blood for concentrations of metabolism total protein, cholesterol, urea, glucose, triglyceride, calcium, and enzyme activates ALP, AST, ALT, and minerals.

3-6: Slaughter procedure and carcass data:

At the end of the 6th week birds were fasted overnight with only water allowed. Four chicks that their body weight close to group average a were selected from each treatment, they were weighed individually there slaughtered weighed individually slaughtered by severing the right and left carotid and jugular vessels, trachea and esophagus. After bleeding they were scalded in hot water, hand plucked and washed .The head was removed closed to skull, feet shanks were removed at the hock joint and eviscerated characteristics for carcass was accomplished . Hot carcass and each evisceration have the liver, heart and gizzard was separately weighed the carcass were stored for 24 hours for carcass characteristic and meet yield.

Carcass data:

The hot carcass was prepared for analysis by removal of the skin and neck near to the body and each was weighed separately .The carcass was divided into right and left side by mid sawing along the vertebral column and each side was weighed. The right side was divided into three commercial cuts, breast, drum, stick and thigh, each cut was weighed separately, then they were deboned, the

meat and bone were weighed separately, the meat was frozen and stored for chemical analysis.

3-8: Chemical analysis:

Stored meat samples were cut into small pieces, minced twice and three samples were from each samples were taken to determine, moisture, protein, fat and ash content according to AOAC (1995). They separated serum from the collected blood samples were analysis to determine total plasma cholesterol concentration, total protein, uric acid and enzyme activists (AST and ALT) calorimetrically using enzymatic colorimetric test .

3-8: Panel taste:

The stored carcasses meat were slightly seasoned wrapped individually in aluminum foil and roasted at 19 C for 70 minutes with average internal temperature of 88 c and served warm to the panel test to determine flavor, tenderness, juiciness and color of meat (Cross et al 1978) on scale of 1-8 (appendix) water was provided for the panelists to rinse their mouth after tasting each sample.

3-9: Calculation:

The hot and cold carcass weights were expressed as a percentage of live weight, the commercial cuts were expressed as a percentage of hot carcass. Non- carcass compounds (heart, liver, gizzard and leg) were expressed as a percentage of cold carcass meat commercial cuts were expressed as % g, of their cuts.

3-10: Statistical analysis:

Completely randomized design used in this experiment. Data collected were supplied to analysis of variance (ANOVA) means were further separated by Dun cans multiple range test(1955),the level of significant difference set up at ($p < 0.05$).

Chapter Four

Result

Chapter four

Results

The effect of feeding different levels of mixture essential oils (MEOs) compared with antibiotic on the performance of broiler chicks was summarized in table (3). Result obtained showed no significant differences ($P>0.05$) in the performance parameters (Body weight gain, feed intake and body weight).

The body weight gain dialer between tested groups, the highest live body weight gain recorded in 200g MEOs, group followed by antibiotic group, while group fed on 600 g MEOs recorded the lowest body weight gain

Feed intake also was similar between groups, but a numerical improvement was observed in group fed on 200g MEOs.

Result revealed no significant ($P>0.05$) difference in FCR among all tested groups

The mortality rate did not influenced by the dietary treatment

Result revealed no significant difference ($P>0.05$) among all treatment groups in the percentages of carcass dressing , giblets, commercial cuts and their separable tissue percentages , meat chemical composition and subjective meat quality parameters.

Result of this study showed that addition of 200g /kg MEOs recorded the best values for all performance parameters.

Economically the addition of MEO as a natural growth promoter alternative the antibiotic resulted in economical benefits. This study revealed that addition of MEO at 200g/kg as growth promoter in broiler diet recorded the highest revenue compared to chicks fed a control or supplemented with antibiotic diets.

Table No (3) The effect of adding a mixture of clove oil, Anise and caraway into the feed on the predictive performance of broiler chicks during period (6) weeks:

Item	Control	antibiotic	Mix 200 gm/kg	Mix400gm/kg	Mix600gm/kg
Initial weight (g/bird)	40	40	40	40	40
Final weight (g/bird)	2417.1	2381.9	2335.6	2261.7	2200.4
Weight gain (g/bird)	2377.1	2341.9	2295.6	2221.7	2160.04
Feed intake (g/bird)	4012.5	4417.2	3943.8	3796.3	3709.8
F.C.R	1.80	1.88	1.86	1.88	1.87
Mortality	0	0	0.32	0	0.32

Figure No (1) the effect of adding a mixture of clove oil, Anise and caraway into the feed on the predictive performance of broiler chicks during period (6) weeks:

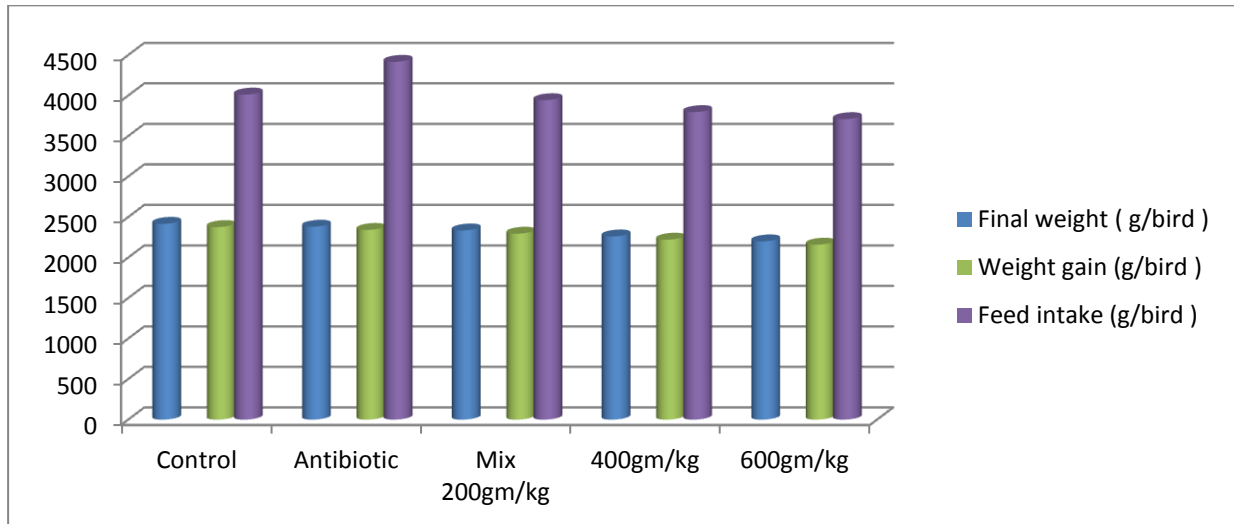


Table No (4) The effect of different dietary amounts of mixture essential oils (Anise, clove caraway) on the meat subjective value of broiler chicks for 6 weeks:

Item	control	antibiotic	Mix200gm/kg	Mix400gm/kg	Mix600gm/kg
Tnderness	6.5	6.3	6.5	6.7	6.8
Flavor	6	6.4	6.1	6.5	6.8
Color	6.4	6.3	6.1	6.4	6.5
Juiciness	6.2	6.2	6.5	6.3	6.6

Figure No (2) The effect of different dietary amounts of mixture essential oils (Anise, clove caraway) on the meat subjective value of broiler chicks for 6 weeks:

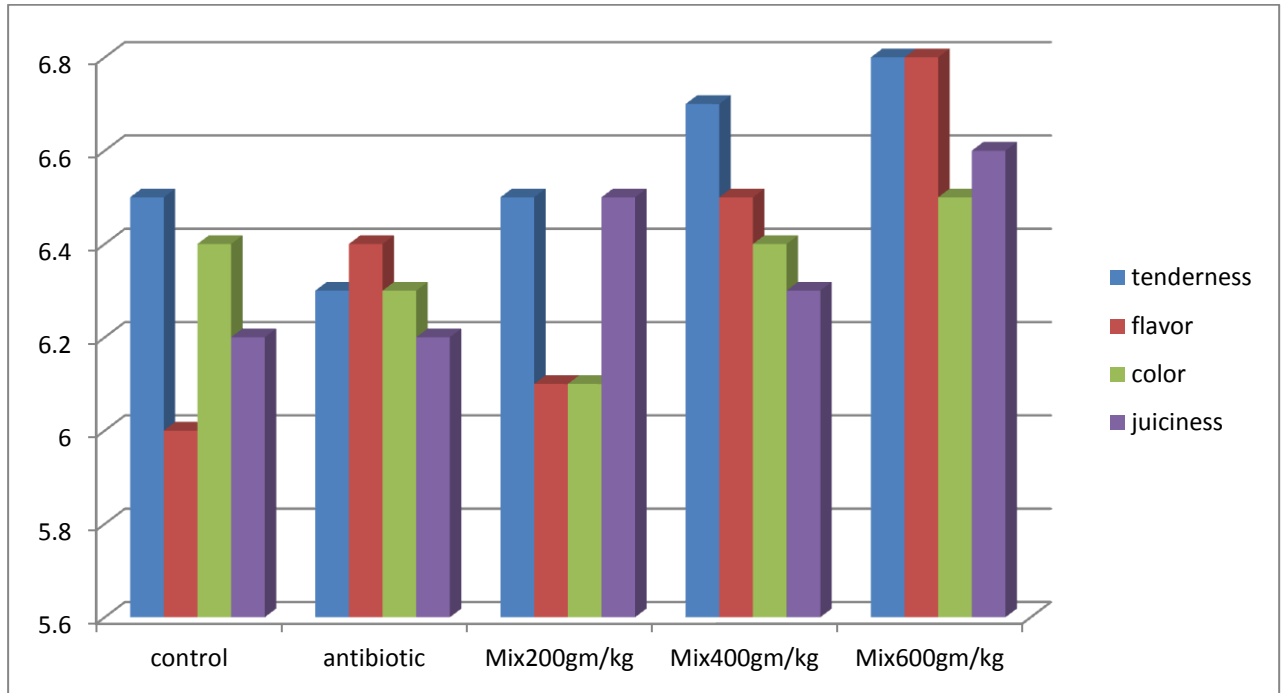


Table No (5) The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (Lipid Profile):

item	A	B	C	D	E
Cholesterol mg/dl	95	86	82	91	92
Glucose	220	177	172	193	211
Tri Glycride	43	43	57	35	29

A: control

B: antibiotic

C: mixture 200gm/kg (Clove, Anise and caraway)

D: mixture 400gm/kg (Clove, Anise and caraway)

E: mixture 600gm/kg (Clove, Anise and caraway)

Figure No (3)The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (Lipid Profile):

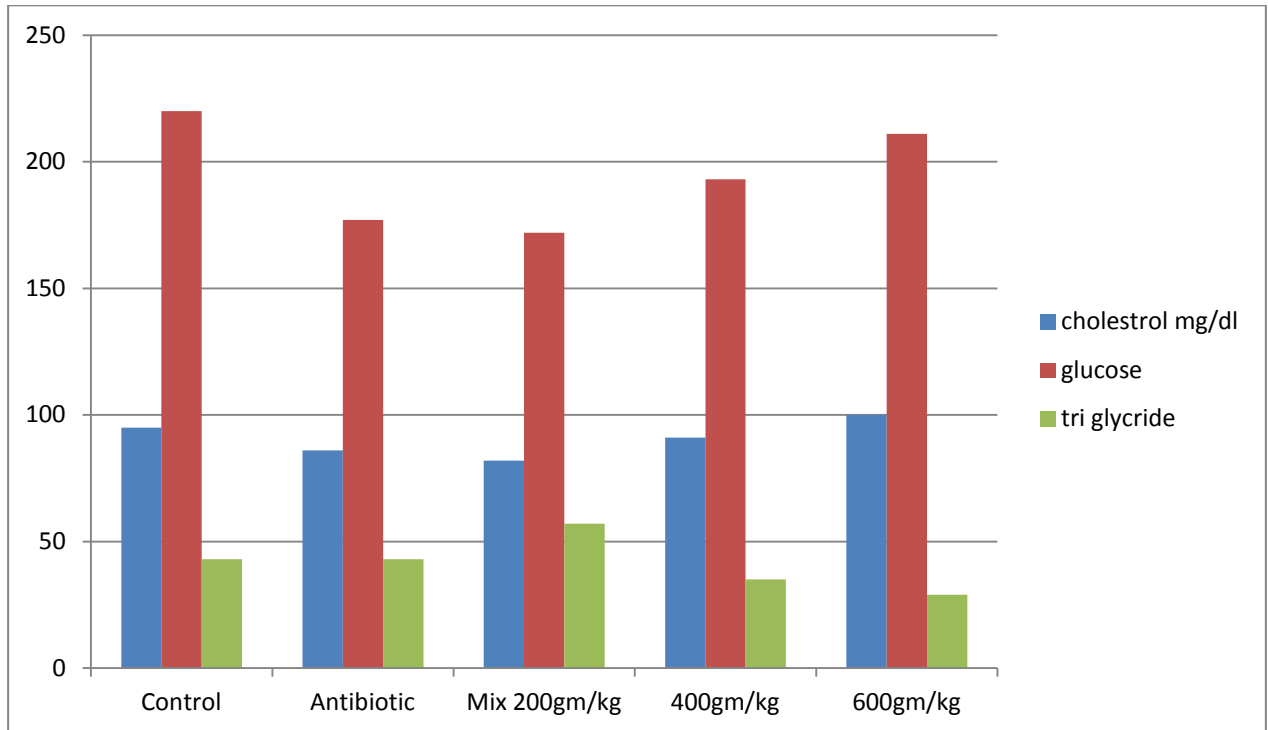


Table No (6)The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (Liver Function):

ITEM	A	B	C	D	E
Hb. g/dl.	11.5	10.8	10.3	11.3	11.3
ALP	33	30	41	38	25
ALT	17	19	16	17	23
AST	16	16	14	10	14
Total protein g/dl	4	3.7	4.4	4	3.9

A: control

B: antibiotic

C: mixture 200gm/kg (Clove, Anise and caraway)

D: mixture 400gm/kg (Clove, Anise and caraway)

E: mixture 600gm/kg (Clove, Anise and caraway)

figure No (4) The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (liver function) :

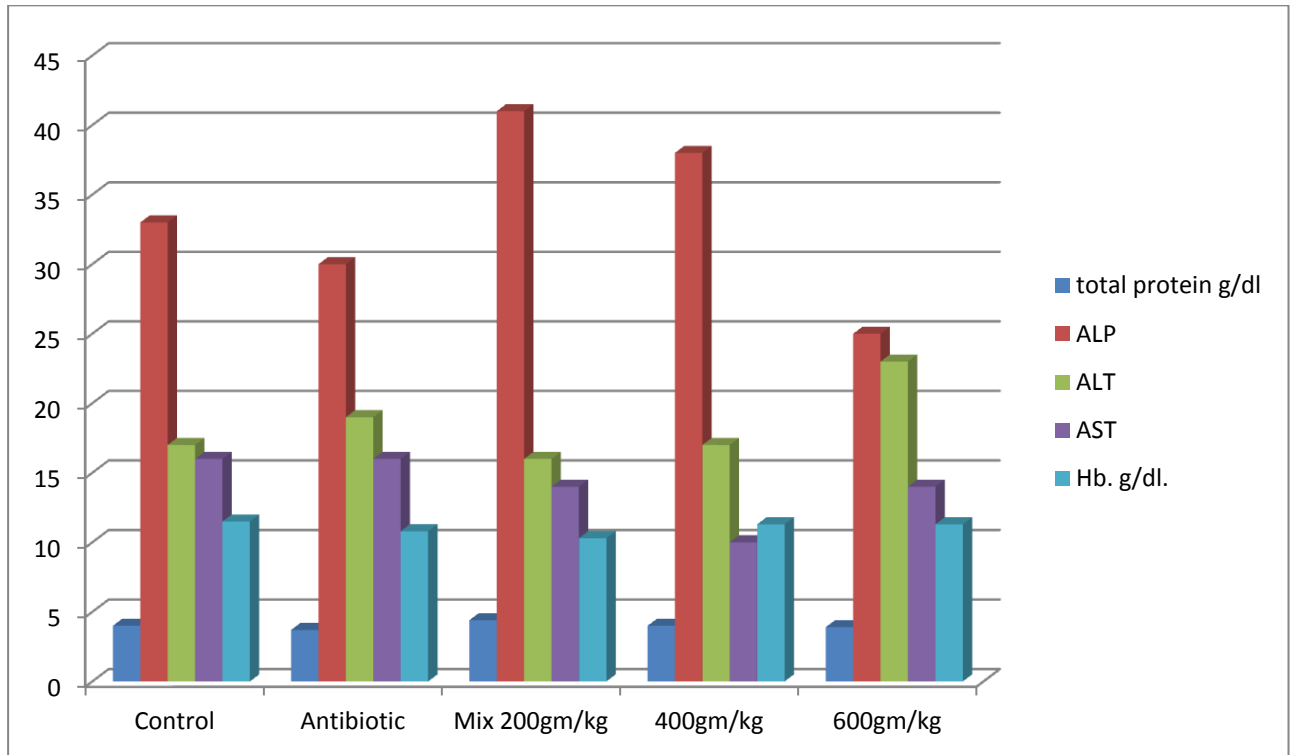


Table No (7)The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (Renal Function):

ITEM	A	B	C	D	E
Uric acid	3.7	4.4	1.9	1.9	2.5
Creatinine	0.37	0.76	0.3	0.34	0.24
Phosphorus	5	2.5	3.8	5	6.3
Urea	12	32	8	13	13
Calcium	9.6	9.3	8.2	9.3	8.6

A: control

B: antibiotic

C: mixture 200gm/kg (Clove, Anise and caraway)

D: mixture 400gm/kg (Clove, Anise and caraway)

E: mixture 600gm/kg (Clove, Anise and caraway)

Figure No (5) The Effect of feeding broiler chicks on diets supplemental with mix essential oil of Blood and Serum analysis (Renal Function):

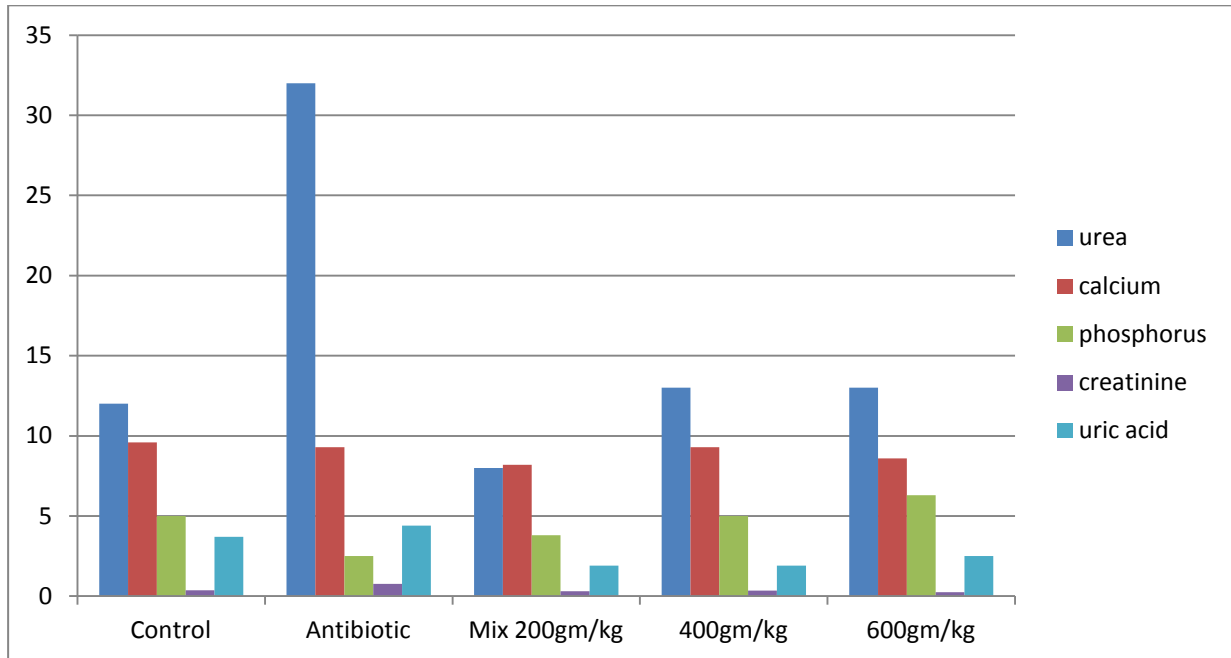


Table No (8): The effect of mixture essential oils (anise, clove caraway) on the commercial cuts percentage of broiler chicks for 6 weeks :

ITEM	Breast	thigh	Drum Stick
Control %	34.25	14.30	11.30
Antibiotic %	28.45	12.90	8.6
Mix 200 %	31.65	13.75	9.8
Mix 400 %	33.50	14.04	10.4
Mix 600 %	29.95	14.70	10.70

Figure No (6): The effect of mixture essential oils (anise, clove caraway) on the commercial cuts percentage of broiler chicks for 6 weeks:

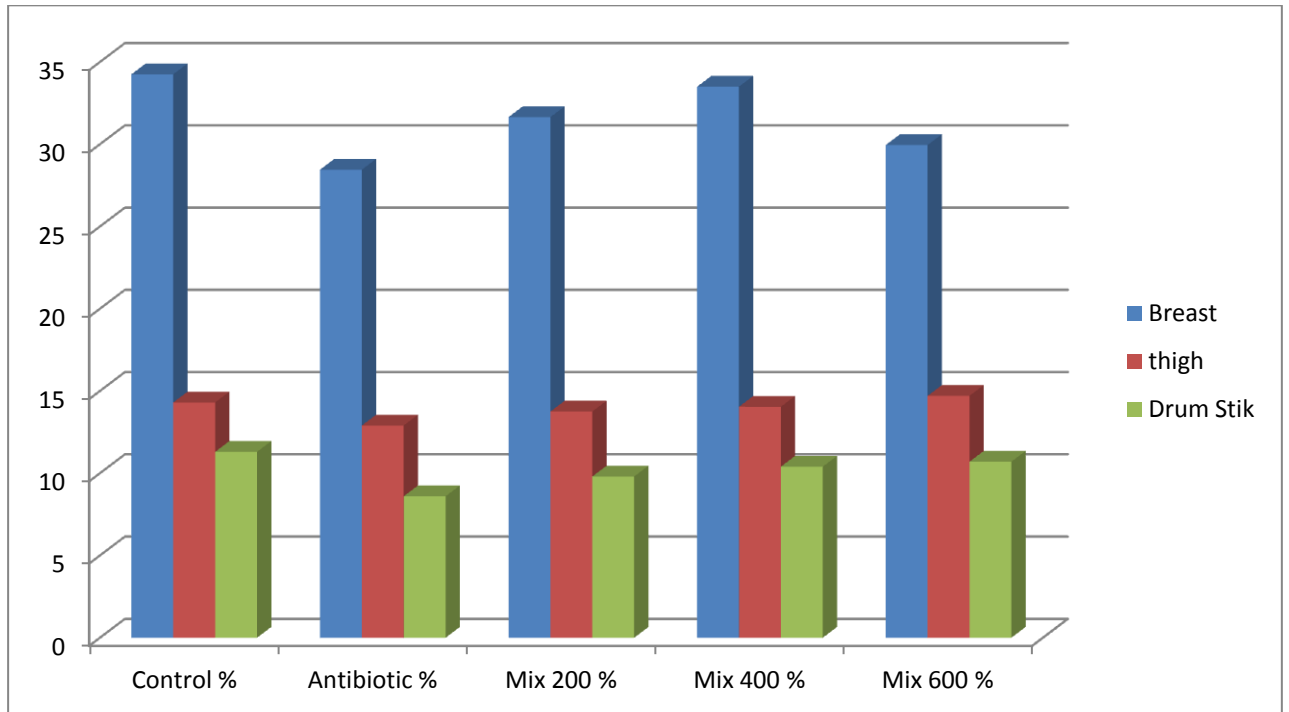


Table No (9): The effect of mixture essential oils (anise, clove. caraway) on percent of carcass and giblets (gizzard, liver, heart) of broiler chicks for 6 weeks:

ITEM	control	antibiotic	Mix 200	Mix 400	Mix 600
Gizzard %	3.55	3.05	3.30	4.10	4.25
Liver %	5.85	5.20	5.40	5.70	5.90
Heart %	1.6	1.3	1.1	1.2	1.3

Figure No (7): The effect of mixture essential oils (anise, clove, caraway) on percent of carcass and giblets (gizzard, liver, heart) of broiler chicks for 6 weeks:

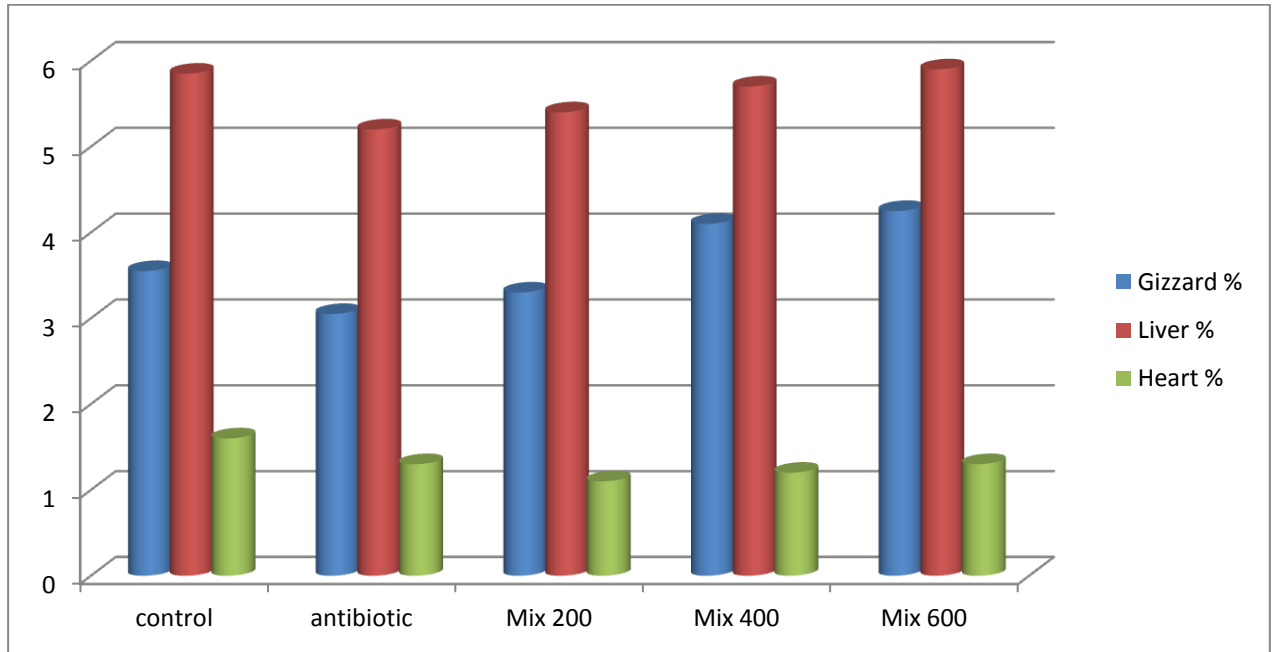


Table No (10): The effect of mixture essential oils (anise, clove. caraway) on meat chemical value of broiler chicks for 6 weeks:

ITEM	DM	Fat	CP	Ash
Control %	26.98	1.37	23.4	4.28
Antibiotic %	26.79	2.55	21.3	4.51
Mix 200 %	25.11	5.1	22.75	4.40
Mix 400 %	24.16	8.17	25.55	4.79
Mix 600 %	32.35	7.28	24.85	3.84

Figure No (8): The effect of mixture essential oils (anise, clove. caraway) on meat chemical value of broiler chicks for 6 weeks :

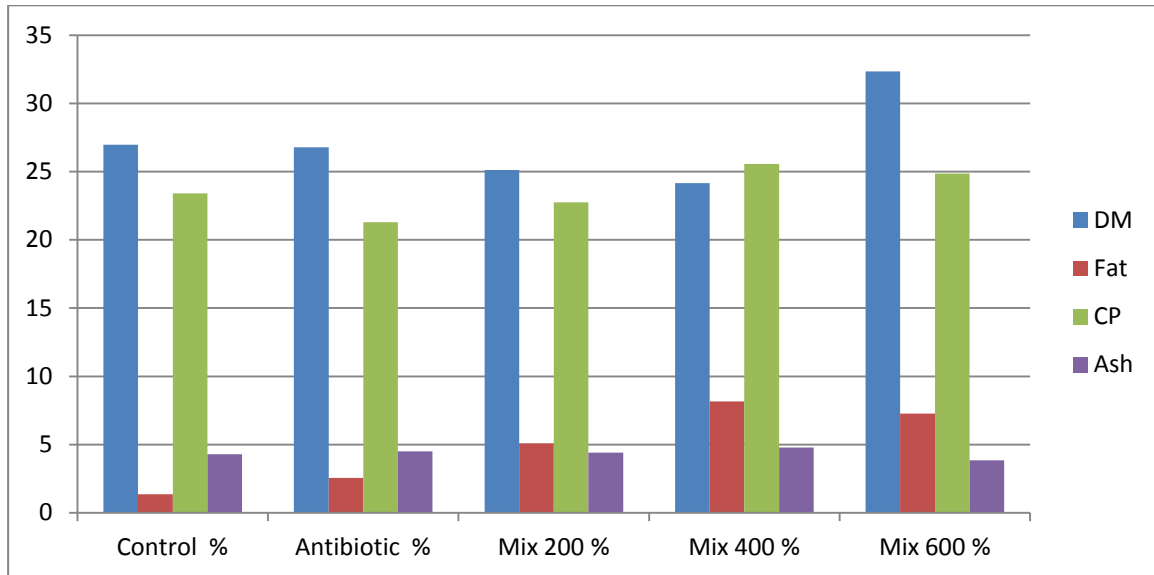


Table No. (11) the economic evaluation of the experience:

Cost per bird

Items	Control	antibiotic	Mix200gm/kg	Mix400gm/kg	Mix600gm/kg
Chickens	5.0	5.0	5.0	5.0	5.0
Nutrition	14.8	13.2	12.4	12.4	12.2
Management	3.0	3.0	3.0	3.0	3.0
Total	22.2	21.2	20.4	20.4	20.2

Revenues

Dressing %	70.52	69.62	69.63	69.64	69.64
Average carcass weight	1704.5	1658.2	1627.9	1574.5	1523.3
Price /kg	21	21	21	21	21
Total revenue pound	35.7	34.8	34.1	33.0	32.1

Profits

Total revenues	35.7	34.8	34.1	33.0	32.1
The total cost	22.2	21.2	20.4	20.4	20.2
Net profit in pounds	13.5	13.6	13.6	12.6	11.9

Chapter Five

Discussion

Chapter five

Discussion

Result obtained for broiler chicks taken different level of Eos compound of negative and positive controls showed no effect on the mortality rate of experimental chicks due to treatments, the rate of mortality was lower than commercial situation, this might be due to the good hygiene conditions , and it could be suggested that the actives substances contained in the MEOs product, might have had a positive effect on gut micro flora by reduce the amount of pathogenic bacteria. This result was in line with the finding of Osman et al., 2005, and Mukhtar 2010 and Mukhtar et al 2011).

Result showed that feed intake was similar between groups, but a linear improved in group fed on 200g/kg MEO, This improvement may be due to the appetizing effect of active ingredient in MEO (Cabuk et al ., 2003), essentials oils exhibit antioxidants activities (Botoglou et al .,2002) and can stimulate animal digestive system (Jamroz and Kamel 2002 ,).

Result revealed no significant difference in body weight gain and FCR, although group fed on 200 g/kg MEO recorded the best values, this might be due to more feed intake compared to control and antibiotic groups.

This statement could be in line with founding's of, (Osman et al 1991, Case et al., 1995, , kamel , 2001and Mukhater et al , 2013). Who observed no effects on growth performance with essential oils supplementation?

Result observed no significant effects on the broiler non carcass components, commercial cuts and their separal tissue percentages, meat chemical composition which was confirmed with the panel taste values.

The results were in line with finding's of Mukhtar 2011 Amal et al 2013 , but not agreed with the report of Alci cek et al ., 2004) who observed improvement in dressing percentage by the dietary essential oil.

The results of this study contradict with (Wang et al 2005, who observed that multination and enzyme supplement significant ($p>0.01$) increased body weight.

Chapter Six

Conclusion and Recommendations

Chapter six

Conclusion and Recommendations

From the result obtained it can be concluded that mixed essential oils (Anise, Clove, Caraway) can be a good natural growth promoter substance alternative of antibiotic without any adverse effect.

Antibiotic of mix Eos at level of 200g/kg improved the broiler chicks performance and recorded the significant profitability ratio compound in to the negative and positive controls.

Most essential oils consist of mixture of compounds such as phenolics of poly phenols, terpenoids , sapcnines , quinme , esters , flavcne , flavonoids , tannins , alkaloids and non volatile residues and their chemical composition of consent ration of compounds is variable.

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Appendix

Appendix No (1): Temperature during experimental period

Week	maximum	minimum	Average
1 st week	31	11	21
2 nd week	30	12	21
3 rd week	33.5	16	25.8
4 th week	29	17	23
5 th week	26.5	16.5	21

Appendix No (2):

Card use for judge of subjective meat quality attributes-Sensory Evaluation Card

Evaluate this sample for color – Flavor – Juiciness and Tenderness, for each sample, use appropriate scale to show your attribute by checking the point that best describe your feeling about the sample, please ask. Thanks for your cooperation

Name:.....

Date:

Tenderness	Flavor	Color	Juiciness
8-Extremely tender	8-Extremely intense	8-Extremely desirable	8-Extremely juicy
7-Very tender	7-Very intense	7-Very desirable	7-Very juicy
6-Moderately tender	6-Moderately intense	6- Moderately desirable	6-Moderately juicy
5-Slightly tender	5-Slightly intense	5-Slightly desirable	5-Slightly juicy
4-Slightly tough	4-Slightly bland	4-Slightly undesirable	4-Slightly dry
3-Moderately tough	3-Moderately bland	3-Moderately undesirable	3-Moderately dry
2- Very tough	2-Very bland	2-Very undesirable	2-Very dry
1-Extremely tough	1-Extremely bland	1-Extremely bland	1-Extremely dry

Appendix No (3):

Serial sample code:

1					
2					
3					
4					
5					

Appendix No (4):

Anise Oil



Appendix No (5):



Appendix No (6):



Appendix No(7):



Appendix No (8):



Appendix No (9):



Appendix No (10):



Appendix No (11):



Appendix No (12):



Figure 2: Benefits of essential oils