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Impact of the Incorporation of Essential oils from some medicinal plants on the zootechnical performance of broiler chickens

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I pray to Almighty to make this research and our knowledge beneficial to our people and all mankind. Ameen.

Dedication

This work is dedicated to our loving parents whose support and encouragement have always been with us.

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Abstract

This study aimed to evaluate the impact of Thymus vulgaris and Lavandula angustifolia essential oils (TEO and LEO) on growth performance, carcass traits, and biochemical and hematological blood parameters of broiler chickens. A total of 160 day-old broiler chicks (Cobb 500) of both sexes were used in the form of a completely randomized design with 5 treatment batches. Broiler chickens in batches 1, 2 and 3 received drinking water supplemented with TEO 250µL/L, LEO 250µL /L, and TEO 125µL /L + LEO 125µL /L, respectively. Those in batch 4 (negative control) were received drinking water with no EO addition. Broiler chickens in batch 5 (positive control) were treated with several antimicrobial and antiparasitic agents. Over 42 days, growth metrics, feed intake, feed conversion ratio (FCR), mortality rates, and carcass characteristics were measured. The results of this study showed that TEO, T+LEO and LEO supplementation increased body weight by 10.37%, 5.42% and 4.33% at 42 day of age, respectively (P < 0.05). The feed intake was reduced in the group treated with T+LEOs by 2.27% when compared to the control group (P < 0.05) and it was increased by 12.16% in the positive control group (P < 0.05). Also, the feed conversion ratio (FCR) decreased and the average daily gain (ADG) increased in chickens that had EOs added to drinking water when compared to the negative control group (P < 0.05). The batch 3 which received a mixture had a significantly higher mortality rate (18.75) when compared to the control. The T+LEO group had the highest carcass percentage (74.6) while the control group had the lowest (69.7) (P < 0.05). The TEO group had the highest thigh percentage (18.9%)(P < 0.05). The negative control group had the highest liver weight percentage (3.07%), while the TEO and T+L EO groups had lower liver weight percentages (2.4 and 2.48% respectively)(P < 0.05). In addition, the results of this study reported that TEO and LEO did not affect the biochemical and hematological blood parameters. In conclusion, TEO and LEO alone or combined can improve growth performance and carcass characteristics in broiler chickens, providing a viable alternative to antibiotics in poultry farming.

Keywords: Thyme, lavender, essential oil, antibiotics, growth performance.

Résumé

Cette étude visait à évaluer l'impact des huiles essentielles de Thymus vulgaris et de Lavandula angustifolia (HET et HEL) sur les performances de croissance, les caractéristiques des carcasses et les paramètres sanguins biochimiques et hématologiques des poulets de chair. Au total, 160 poussins de chair d'un jour (Cobb 500) des deux sexes ont été utilisés sous la forme d'un plan complètement randomisé avec 5 lots de traitement. Les poulets de chair des lots 1, 2 et 3 ont reçu de l'eau potable additionnée de TEO 250 µL/L, LEO 250 µL/L et TEO 125 μL/L + LEO 125 μL/L, respectivement. Ceux du lot 4 (témoin négatif) ont reçu de l'eau potable sans ajout d'HE. Les poulets de chair du lot 5 (témoin positif) ont été traités avec plusieurs agents antimicrobiens et antiparasitaires. Pendant 42 jours, les paramètres de croissance, la consommation alimentaire, le taux de conversion alimentaire (FCR), les taux de mortalité et les caractéristiques des carcasses ont été mesurés. Les résultats de cette étude ont montré que les suppléments de HET, HET+L et HEL augmentaient le poids corporel de 10,37 %, 5,42 % et 4,33 % à l'âge de 42 jours, respectivement (P < 0,05). La consommation alimentaire a été réduite de 2,27 % dans le groupe traité avec HET+L par rapport au groupe témoin (P < 0,05) et augmentée de 12,16 % dans le groupe témoin positif (P < 0,05). En outre, le taux de conversion alimentaire (TCA) a diminué et le gain moyen quotidien (GMQ) a augmenté chez les poulets auxquels on avait ajouté des HE à l'eau potable par rapport au groupe témoin négatif (P<0,05). Le lot 3 qui a reçu un mélange d'HE présentait un taux de mortalité significativement plus élevé (18,75) par rapport au témoin négatif. Le groupe HET+L avait le pourcentage de carcasses le plus élevé (74,6ù) tandis que le groupe témoin négatif avait le plus faible pourcentage (69,7%) (P<0,05). Le groupe HET présentait le pourcentage de cuisses le plus élevé (18,9 %) (P < 0.05). En outre, le groupe témoin négatif avait le pourcentage de poids du foie le plus élevé (3,07 %), tandis que les groupes HET et HET+L avaient des pourcentages de poids du foie plus faibles (respectivement 2,4 et 2,48 %) (P < 0,05). De plus, les résultats de cette étude ont montré que la HET et la HEL n'affectaient pas les paramètres sanguins biochimiques et hématologiques. En conclusion, le HET et le HEL seuls ou en association pourraient améliorer les performances de croissance et les caractéristiques des carcasses des poulets de chair, offrant ainsi une alternative prometteuse aux antibiotiques dans l'élevage de volailles.

Mots clés : Thym, lavande, huile essentielle, antibiotiques, performance de croissance.

ملخص

هدفت هذه الدراسة إلى تقييم تأثير الزيوت العطرية Thymus vulgaris و Lavandula angustifolia (LEO وLEO) على أداء النمو، وصفات الذبيحة، ومؤشرات الدم البيوكيميائية والدموية للدجاج اللاحم. تم استخدام 160 فرخاً من فراخ اللحم بعمر يوم واحد (Cobb 500) من كلا الجنسين على شكل تصميم عشوائي كامل بخمس مجموعات معالجة. تلقى الدجاج اللاحم في المجموعات 1 و2 و3 مياه شرب مكملة ب TEO 250 ميكرولتر /لتر، LEO 250 ميكرولتر /لتر، وTEO 125 ميكرولتر /لتر + LEO 125 ميكرولتر/لتر، على التوالي. تم تزويد الفراخ الموجودة في المجموعة 4 (التحكم السلبي) بمياه الشرب بدون إضافة EO. تم علاج الدجاج اللاحم في الدفعة 5 (التحكم الإيجابي) بالعديد من مضادات الميكروبات والطفيليات. على مدار 42 يومًا، تم قياس مقاييس النمو وتناول العلف ونسبة تحويل العلف (FCR) ومعدلات الوفيات وخصائص الذبيحة. أظهرت نتائج هذه الدراسة أن مكملات TEO وT+LEO وLEO زادت من وزن الجسم بنسبة 10.37% و5.42% و4.33% عند عمر 42 يومًا، على التوالي (P <0.05). انخفض تناول العلف في المجموعة المعالجة بـ T+LEOs بنسبة 2.27% بالمقارنة مع مجموعة التحكم السلبي (P <0.05) وزاد بنسبة 12.16% في مجموعة التحكم الإيجابي (P 0.05>). كما انخفضت نسبة التحويل الغذائي (FCR) وزاد متوسط الكسب اليومي (ADG) في الدجاج الذي تمت إضافة EOs إلى مياه الشرب بالمقارنة مع مجموعة التحكم السلبي (P <0.05). كان للدفعة 3 التي تلقت خليطًا معدل وفيات أعلى بكثير (18.75) مقارنةً بمجموعة التحكم السلبي. كانت لمجموعة T+LEO أعلى نسبة للذبيحة (74.6) بينما كانت لمجموعة التحكم السلبي أقل نسبة (69.7) (P <0.05). حصلت مجموعة TEO على أعلى نسبة فخذ (18.9%) (P<0.05). كان لدى مجموعة التحكم السلبي أعلى نسبة لوزن الكبد (٪3.07)، في حين كان لدى مجموعتي TEO و T + L EO نسب أقل لوزن الكبد (2.4 و 2.48٪ على التوالي) (P <0.05). بالإضافة إلى ذلك، أشارت نتائج هذه الدراسة إلى أن TEO وLEO لم يؤثرا على مؤشرات الدم البيوكيميائية والدموية. في الختام، يمكن لـ TEO وLEO تحسين أداء النمو وخصائص الذبيحة في الدجاج اللاحم، مما يوفر بديلاً قابلاً للتطبيق للمضادات الحيوية في تربية الدواجن.

الكلمات المفتاحية: Lavandula angustifolia · Thymus vulgaris ، الزيت العطري، المضادات الحيوية، أداء النمو.

Introduction

Introduction

The poultry sector contributes significantly to the global food supply, with broiler chickens providing a key source of protein for millions of people globally. However, the sector has various challenges, such as improving bird welfare, reducing environmental impact, and increasing output while maintaining food safety and quality. One option for resolving these issues is to use essential oils from medicinal herbs in broiler chicken production (Samad, 2022; Valco, 2024).

Essential oils are natural, volatile molecules derived from plants that have been utilized for ages in traditional medicine and agriculture. A recent study has focused on their ability to increase animal health and productivity, especially in chicken production (Arif et al., 2024; Dnyaneswarn et al., 2024). Medicinal plants like thyme, oregano, and rosemary have essential oils that have antibacterial, antioxidant, and anti-inflammatory qualities, making them excellent for broiler chicken production.

The use of essential oils from medicinal herbs in broiler chicken production has various potential advantages. For starters, essential oils can improve bird health and welfare by lowering stress, increasing immunological function, and managing infections. Second, essential oils can boost feed efficiency and growth performance, resulting in improved productivity and lower expenses. Finally, essential oils can reduce the demand for antibiotics and other chemicals in chicken production, resulting in a more sustainable and eco-friendly sector (Raseem et al., 2016).

Despite these potential benefits, the effect of essential oils on broiler chicken performance is poorly understood. Previous research has looked into the effects of individual essential oils or blends on various areas of broiler chicken production, but there is still a need for a more comprehensive understanding of their influence on zoological performance. The purpose of this study is to evaluate the effect of the incorporation of *Thymus vulgaris* and *Lavandula angustifolia* essential oils in drinking water on the zootechnical performance of broiler chickens. It will specifically look into the impact of essential oils on growth performance, carcass traits, as well as their effect on biochemical and hematological blood parameters in broiler chickens.

This study's findings will help to improve understanding of the potential benefits and limitations of employing essential oils in broiler chicken production, as well as provide vital insights for developing sustainable and efficient production systems, which aligns with the ideals of sustainable agriculture and food security.

Chapter I : Antibiotics in poultry and

alternatives to antibiotics

ANTIBIOTICS IN POULTRY AND ALTERNATIVES TO ANTIBIOTICS

1. Antibiotics in poultry

1.1. Definition

An antibiotic is a chemical that either kills infectious bacteria or inhibits their growth. The term antibiotic is also used for chemicals that kill other microorganisms, such as single celled organisms (also known as protozoans). Antibiotics were initially produced by microorganisms but are now synthesized chemically. Antibiotics do not affect viruses (Scanes et al., 2018).

1.2. Use of antibiotics in poultry

In the wild, chickens live in small flocks often with only a handful of members, traversing a range of habitats as they forage for insects, seeds, and other food. Disease can lurk around every corner and beneath every leaf. Despite innumerable hidden threats, wild chickens can enjoy a lifespan of a decade or more, all without any intervention from human beings. Life in the wild does not require the constant application of drugs: a fact that renders the woeful living conditions on factory farms all the more stark. Factory farms are the antithesis of natural living conditions for chickens. Without antibiotics, raising chickens on factory farms would be extremely expensive because the conditions on these farms are otherwise unlivable. Drugs are regularly administered via daily water and food rations and are generally used throughout the chicken's lifetime, starting when chicks are only a few days old. On factory farms, antibiotics are used for two reasons: to promote growth and to prevent or treat infection. They're administered regularly in the chickens' feed, and they're so effective at encouraging rapid growth that today's chickens are twice as large as chickens were 60 years ago. This is a problem because broiler chickens' bodies can't support this much weight. This unnatural growth leads to skeletal and joint issues, and it often causes so much stress on the chickens' legs that they become painfully lame.

1.3. Production of antibiotics

Antibiotics can be produced using three three methods:

- Fermentation;
- Semi-synthetic;
- Synthetic.

1.3.1. Fermentation method

Fermentation is used to produce antibiotics when the source microorganism is grown in large containers (100000 to 150000 liters or more) containing a liquid growth medium. Oxygen concentration, temperature, pH, and nutrients are closely controlled. The population size of the bacteria must be controlled carefully in order to ensure that the maximum yield is achieved before the cells die.

1.3.2. Semi-synthetic method

This method of production is used in modern times. It involves a combination of natural fermentation and laboratory work to maximize the antibiotic. Maximization can occur through the efficacy of the drug itself, the amount of antibiotics produced, and the potency of the antibiotic being produced. Depending on the drug being produced and the ultimate usage of said antibiotic determines what one is attempting to produce. An example of semi-synthetic production involves the drug ampicillin. A beta-lactam antibiotic just like penicillin, ampicillin was developed by adding an addition of amino group (NH₂) to the R group of penicillin. This additional amino group gives ampicillin a broader spectrum of use than penicillin.

1.3.3. Synthetic method

Not all antibiotics are produced by bacteria; some are made completely synthetically in the laboratory. These include the quinolone class, of which nalidixic acidis often credited as the first to be discovered. Like other antibiotics before it the discovery of nalidixic acid has been chalked up to an accident, discovered when George Lesher was attempting to synthesize chloroquine. However, a recent investigation into the origin of quinolones has discovered that a description for quinolones happened in 1949 and that patents were filed concerning quinolones some 5 years before Lesher's discovery.

Chapter I :

1.4. Major antibiotics used in poultry production

There are many different kinds of antibiotics, and they destroy bacteria in different ways. The antibiotics within a class generally have similar effectiveness and mechanisms of action and resistance and they tend to attack the same types of bacteria. Some antibiotics, referred to as broad-spectrum antibiotics, treat a wide range of infections. Others, called narrow-spectrum antibiotics, are effective against only a few types of bacteria. Although antibiotics are sometimes used in conventional animal feeds, some of the antibiotics discussed below can be used only under the supervision of a veterinarian (Jacob, 2024).

Popular antibiotics used in poultry production include:

1.4.1. Aminoglycosides

Aminoglycosides are antibiotics derived from various species of *Streptomyces* bacteria. They act by blocking the synthesis of proteins vital to bacterial growth. The aminoglycosides remain in the digestive tract and are effective in the treatment of enteric infections. Examples include gentamycin, neomycin, spectinomycin, and streptomycin.

1.4.2. Bambermycins

Bambermycins are antibiotics that act by inhibiting the synthesis of the bacterial cell wall. Examples include bambermycin and flavophospholipol, which are effective against grampositive pathogenic bacteria and do not affect *Lactobacillus*, *Bifidobacterium*, and other protective bacteria. They are derived from *Streptomyces bambergiensis*.

1.4.3. Beta-Lactams

These are antibiotics that contain a beta-lactam ring in their chemical structure. The principal beta-lactam antibiotics include.

- Penicillin
- Cephalosporins

1.4.4. Penicillin

Penicillin was the first antibiotic to be discovered and they were produced by a mold called *Penicillium notatum*. They kill bacteria by inhibiting the formation of the bacteria cell wall. Penicillins are effective in the treatment of sinusitis and chronic respiratory disease in poultry. Examples include Amoxicillin and Ampicillin.

1.4.5. Cephalosporins

Cephalosporins also interfere with the formation of bacterial cell walls. This class of antibiotics is further divided into the first, second, and third generations. Each generation has a broader spectrum of activity than the one before. Like penicillin, cephalosporins interfere with the formation of bacterial cell walls. The Food and Drug Administration (FDA) issued an order on January 4, 2012, that prohibits certain extra-label uses of the cephalosporin class of antimicrobial drugs in cattle, swine, chickens, and turkeys. The order became effective on April 5, 2012. (The term extra-label refers to the use of a drug in a way that is not listed as an approved use on the label) (Jacob, 2024).

1.4.6. Glycopeptides

Glycopeptide antibiotics are a type of antibiotic that inhibits bacterial cell wall formation by inhibiting peptidoglycan synthesis. They are used to treat infections caused by Gram-positive bacteria.

1.4.7. Ionophores

Ionophores are feed additives used in the control of coccidiosis, primarily when raising broilers, broiler breeders, and replacement pullets. Coccidiosis is a disease in poultry worldwide and is caused by a protozoan parasite (*Eimeria*) that invades the cells of poultry intestines. Ionophores are used primarily as an anti-microbial but can control some bacteria so is often grouped with antibiotics. Ionophores are not used in human medicine.

1.4.8. Lincosamides

Lincosamides are antibiotics produced by Streptomyces lincolnensis. Lincomycin is a lincosamide that penetrates most tissues and bones too. It is effective against bone and joint infections, as well as necrotic enteritis caused by *Clostridium perfringens*.

1.4.9. Macrolides

Macrolides are antibiotics derived from the Streptomyces bacteria. They are bacteriostatic and act by interfering in protein production (the bacteria is not able to complete proteins that are essential for life). Macrolides can also be used to treat necrotic enteritis.

1.4.10. Polypeptides

The polypeptides are bactericidal antibiotics with activity against gram-negative aerobic bacilli including Pseudomonas aeruginosa. They are not active against Proteus sp and have no activity against Gram-positive organisms

1.4.11. Quinolones

The fluoroquinolones are synthetic, broad-spectrum antibacterial agents with bactericidal activity. They exert their effects by binding to and inhibiting bacterial DNA-gyrase. This enzyme produces supercoiling of cellular DNA which is needed for bacterial DNA synthesis. Fluoroquinolones are not derived from bacteria or fungi; they are synthetic antibiotics.

1.4.12. Streptogramins

Streptogramins are antibiotics produced by Streptomyces species and consist of two structurally unrelated molecules. One inhibits protein synthesis, and the other plays a role in cell wall formation. Individually, the molecules are bacteriostatic but the combination of the molecules is bacteriocidal.

1.4.13. Sulfonamides

Sulfonamides (SAs), synthetic antibiotics, are commonly used by veterinarians in chicken for therapeutic, prophylactic, or as growth promoters and halt the growth of bacteria in animal production. They interfere with RNA and DNA, which are necessary for cell growth and replication.

1.4.14. Tetracyclines

Tetracyclines are broad-spectrum bacteriostatic agents derived from the bacteria Streptomyces. They prevent bacteria from multiplying while the host animal's immune system deals with the original infection.

1.4.15. Coccidiostats

Coccidiosis is a protozoal disease causing diarrhea, weight loss, and decreased production in poultry by infestation of the intestines. It can be fatal. Diagnosis is by fecal flotation to detect oocysts, often in combination with characteristic necropsy findings.

Coccidiostats like:

- Amprolium ; •
- Decoquinate ;
- Halofuginone hydrobromide ;
- Lasalocid; •
- Monensin, are used as anticoccidials to prevent or treat Coccidiosis.

1.5. Antimicrobial resistance

The main cause of antibiotic resistance is antibiotic use. When we use antibiotics, some bacteria die but resistant bacteria can survive and even multiply. The overuse and misuse of antibiotics make resistant bacteria more common. The more we use antibiotics, the more chances bacteria have to become resistant to them. The worldwide increase in the use of antibiotics as an integral part of the poultry and livestock production industry to treat and prevent infectious bacterial diseases and as growth promoters at sub-therapeutic levels in feeds has led to the problem of the development of bacterial antibiotic resistance during the past years. Recent scientific evidence has shown that resistance to antibiotics is not only due to the natural ability of a tiny fraction of the bacteria with unusual traits to survive antibiotic's attack, enabling resistant strains to multiply, but also stems from the transmissibility of

acquired resistance to their progeny and across to other unrelated bacteria species through extrachromosomal DNA fragment called the plasmid which provide a slew of different resistances. The emergence and spread of resistant bacterial strains like Campylobacter sp, Escherichia coli and Enterococcus sp. from poultry products to consumers put humans at risk to new strains of bacteria that resist antibiotic treatment. Resistant bacteria thwart antibiotics by interfering with their mode of action via a range of effectors' mechanisms, including synthesis of inactivating enzymes, alteration in the configuration of cell wall or ribosome and modification of membrane carrier systems. These mechanisms are specific to the type of resistance developed (Apata, 2009). Acquired bacterial resistance is caused by four general mechanisms including inactivation, target alteration, decreased permeability, and increased efflux. First, target site changes typically occur from spontaneous mutation of a bacterial gene with selection pressure of antibiotics. Two examples consist of mutations in RNA polymerase and DNA gyrase which facilitate resistance in rifamycins and quinolones, respectively. Second, target alteration uses a strategy to make the antibiotic ineffective through enzymatic degradation. commonly occurring among aminoglycosides. chloramphenicol, and beta-lactams. Third, Gram-negative bacteria can decrease permeability to selectively filter antibiotics from entering the cell membrane. Fourth, efflux pumps function mainly to release toxic substances from the bacterium and many of these pumps can transport an extensive variety of compounds (Hedman et al., 2020).

Other causes of antimicrobial resistance include:

- Poor infection control;
- Poor hygiene and sanitation;
- Absence of new antibiotics being discovered.

2. Alternatives to antibiotics in poultry

The poultry industry rapidly decades. has risen over the last three Antibiotics are increasingly being used for both medical and growth-promoting reasons. Antibiotic resistance in bacterial populations, antibiotic residues in poultry products, and consumer demand for antibiotic-free products have led to a search for alternatives that can replace antibiotics without compromising productivity or quality. Alternatives to antibiotics in poultry include probiotic microorganisms, prebiotic substrates that encourage beneficial bacterial populations, and synbiotics (prebiotic and probiotic), combinations that improve production and poultry health. Phytobiotics, antimicrobial peptides (AMPs), plant extracts, organic acids, herbal drugs, vitamins, and minerals, are some examples (Samad, 2022).

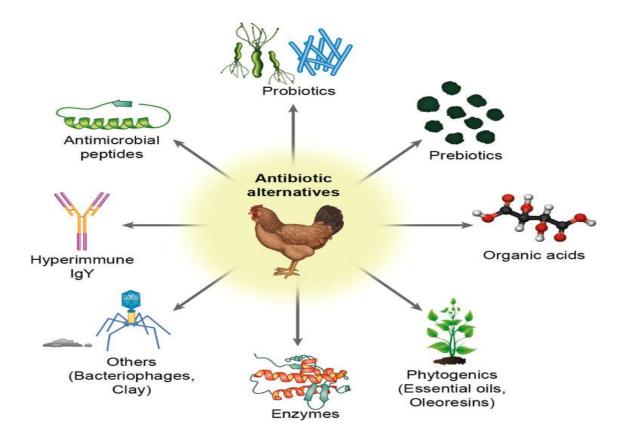


Figure 1. Various classes of antibiotic alternatives are available for use in poultry production (Gadde et al., 2017).

2.1. Probiotics

The term "probiotic" is derived from the Latin words "pro" and "bios". Probiotics are microbial cultures that can be isolated or mixed to improve the host's health. Probiotic bacteria's mode of action includes competition with intestinal receptor sites, formation of antimicrobial metabolites (short organic fatty acids, hydrogen peroxide), and immune stimulation. Probiotic microorganisms include *Lactobacillus*, *Streptococcus*, *Enterococcus*, *Bacillus*, *Clostridium*, *Bifidobacterium*, and *E. coli*. Probiotics made from yeast and fungi include *Saccharomyces cerevisiae* and *Aspergillus oryzae*. Yeast and bacteria have been inserted as living organisms or spores. *Saccharomyces cerevisiae* and *Bacillus* spp. (spores) are non-colonizing probiotics, while *Lactobacillus* and *Enterococcus* spp, are colonizing

species. *Saccharomyces* is rich in B complex vitamins and high-quality proteins. Yeast extract, a non-antibiotic functional substance, can reduce pathogenic bacteria in turkey production because of its immunomodulatory capabilities (Samad, 2022). As zootechnical feed additives, yeast cell derivatives are becoming increasingly essential. Similarly, feeding *Aspergillus awamori* (0.05%) promoted meat quality by increasing the content of unsaturated fatty acids in grilled chicken breast meat while also improving growth performance by releasing growth boosters (Eden, 2003). Probiotics are primarily used to build healthy intestinal flora to avoid or reduce disturbances caused by enteric infections. Probiotics do not replace medications in birds suffering from major illnesses, but they do assist restore the natural bacterial population (Samad, 2022).

The effectiveness of a probiotic depends on several factors, including the bird's physiological condition, the type and concentration of the strain, its ability to survive feed processing and the gastrointestinal tract, and compatibility with the intestine's natural microbiota. The ideal probiotic strain should be resistant to bile salts, acid, and digestive enzymes. To achieve the intended impact, the microbial population should be able to multiply rapidly. Furthermore, the chosen strain should not alter the gut microflora's susceptibility to antibiotics (Samad, 2022).

2.1.1. Benefits of Probiotics

- Improves gut health by preserving the desired bacteria population balance and reducing diarrhea outbreaks;
- It slows the spread of diseases and reduces mortality;
- Increases the effectiveness of feed conversations;
- Improves body weight gain and growth rate;
- Improves digestive enzymes, allowing the body to absorb nutrients more efficiently;
- Regulates lipid metabolism, lowering circulating cholesterol levels;
- Increases the efficiency of vaccinations;
- Contributes to the rapid removal of mycotoxins

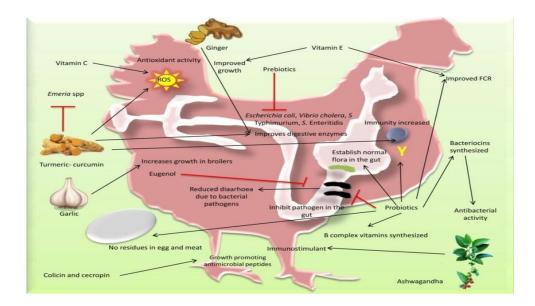


Figure 2. Illustration of benefits of probiotics in the poultry industry.

This illustration depicts various natural supplements and their beneficial effects on chicken health and growth. Key elements include; Vitamin E: which increases immunity and improves feed conversion ratio (FCR), Ginger: Which provides antioxidant activity and improves growth, Turmeric (Curcumin): which enhances broiler growth, Garlic (Eugenol): which reduces diarrhea from bacterial pathogens, etc. (Ajit Singh Yadav et al., 2016).

2.2. Prebiotics

According to Gibson and Roberfroid (1995), prebiotics are non-digestible food ingredients that have a positive effect on the host by selectively promoting the growth and/or activity of a single or small group of bacteria in the colon. These provide food for the stomach's healthy microbes. Prebiotics have multiple functions, including altering gut microflora, stimulating the immune system, preventing colon cancer and pathogen invasion, reducing cholesterol and odor compounds, promoting enzyme reactions, reducing ammonia and phenol products, and lowering production costs (Ghiyasiet al., 2007; Khksar et al., 2008; Peric et al., 2009; Samad., 2022).

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2.2.1. Some commonly used prebiotics

Oligosaccharides, such as fructo-oligosaccharides (FOS), mannon-oligosaccharides (MOS), lactulose, and inulin, are common prebiotic ingredients. Prebiotics can come from both natural and synthetic sources. Natural sources include legumes (oligosaccharides) such as field peas, black gram, and chickpea, which contain raffinose, staychose, and verbascose. Synthetic oligosaccharides are produced through direct polymerization of disaccharides, fractionation of microbial cells to extract material from the cell wall, and polysaccharide fermentation (Apata, 2009).

2.2.2. Advantages of prebiotics supplementation

In poultry production, prebiotic supplementation has grown in popularity as a natural way to improve bird health, productivity, and overall well-being. Prebiotic supplementation in chickens has several advantages, including:

- Prebiotics can help reduce the colonization of pathogens in chicken diets;
- Promotion of enzyme reaction;
- Improve gut health and microbial balance;
- Improve performance;
- Improve nutritional utilization (including amino acids and proteins);
- Reduce environmental pollution;
- Reduce production costs;
- Reducing stress;
- Reducing antibiotic use.

2.3. Antimicrobials of plant origin or phytobiotics

Phytobiotics are substances from plants that have various effects on plants, animals, and humans. They are secondary metabolites that give plants their particular flavor and taste and are often used to protect against pests and animal grazing. Over the years, more than 80000 compounds have been identified, including phenols, flavonoids, tannins, saponins, and essential oils. These molecules, which were once deemed waste products, are now recognized

as antioxidants, digestive aids, and health-promoting components. They exhibit antimicrobial activity in a variety of organisms and are utilized as an alternative antibiotic growth promoter in monogastric animal production. They have been shown to improve gut microflora in less hazardous organisms, with the most plausible mechanism being a change in membrane permeability to hydrogen ions (H+) (Samad, 2022). Their antibacterial, antiviral, antifungal, and anti-protozoan characteristics are becoming increasingly important as they are used in ecologically friendly and cost-effective fungicide preparations and fly-repelling solutions.

Tannin-containing plants such as chestnut, Acacia karoo, and grape pomace influence rumen fermentation and reduce disease-causing bacteria in animals. They lower methane emissions while enhancing energy for growth and production (Samad, 2022). Tannins can increase production, intestinal health, and immunological function, but saponins destroy cells and harm microorganisms (El-Ghany, 2020). Essential oils derived from plants are used as feed additives in animal diets to eradicate pathogenic microorganisms in the rumen. Examples include rosemary, lavender, mints, thyme, jamrosa, and jasmine oil. Essential oils degrade Gram-positive and Gram-negative bacteria's cell membranes, rendering them useless in the animal's body. Supplementing broiler meals with essential oils, oregano, or saponins improves growth and immunity by eliminating pathogens (Ikusikaet al., 2022).

Herbal medicine has gained popularity due to its advantages over chemical medications, such as lower toxicity, natural availability, and appropriate feed additive properties. Plant components like herbs and spices have antibacterial properties, with essential oils being a key component. These volatile chemicals have a wide antibacterial range and are effective against a variety of bacterial and fungal pathogens. The antibacterial properties of essential oils are regulated by biological factors, industrial operations, and storage environments. The study of their impact on feed efficiency and health status in chicken birds is still ongoing, with a focus on herbs that inhibit bacterial quorum sensing (Samad, 2022).

Plant extracts, also known as phytobiotics, have been utilized as an alternative to antibiotics because of their antibacterial, anti-inflammatory, antioxidant, and antiparasitic effects. They contain minor metabolites such as terpenoids, phenolics, glycosides, and alkaloids, which can improve poultry development and health. However, excessive use of these secondary metabolites may impair digestive efficiency. Plant extracts are both safe and efficient against specific germs. Plant extracts from aromatic spices, pungent spices, and herbs have surpassed antibiotics in popularity in African countries because of their low cost and availability. Plant extracts have been shown in trials to boost feed intake, feed conversion ratio, and body weight gain in chickens, improving both productivity and health. Plant extracts enhance the gut microbial ecosystem by Plant extracts also improve the gastrointestinal microbiota ecosystem by controlling pathogen volume in birds' small intestines. Herbs like black pepper are growth promoters without adversely affecting performance (M.M. Andrew et al., 2020).

2.3.1. Advantages of phytobiotics

- Favorably alters the microbial community to promote gut health;
- Increases feed efficiency and body weight growth;
- Reduces cholesterol levels by inhibiting hepatic enzyme activity;
- Increases antioxidant defenses against oxidative stress;
- Promotes the release of digestive enzymes and nutritional absorption.

2.4. Antimicrobial peptides (AMPs)

Antimicrobial peptides (AMPs) are small proteins found in almost every living organism, with broad-spectrum antimicrobial activity against bacteria, fungi, protozoa, and viruses. They have various advantages, including easy degradation, increased host immunity, neutralization of many pathogens, and a low resistance frequency. AMPs can disrupt the bacterial membrane in a variety of ways, including electroporation, non-lytic membrane depolarization, membrane destabilization, hole formation, membrane thinning or thickening, and oxidized lipid targeting. They can also interact with intracellular targets, preventing cell wall, protein, and acid nucleic acid synthesis, as well as interfering with bacterial metabolism. AMPs can be used in the prophylactic control or treatment of bacterial infections while promoting broiler growth. They can reduce pathogenic bacterial burdens in the avian gut, boost the number of beneficial bacteria, and alter the intestinal microbiota. Synthetic and recombinant AMPs can enhance the antibacterial activity of traditional antimicrobials used in chicken production. Examples of AMPs used in poultry: are Cathelicidins, Defensins, Cecropins, and Magainins (Abreu et al., 2023).

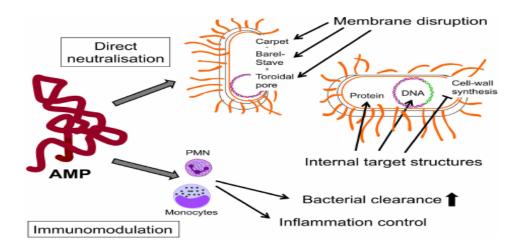


Figure 3. Different Mode of Action AMPs.

2.4.1. Benefits of Antimicrobial peptides (AMPs)

AMPs used as growth promoters: Studies have shown that AMPs, such as Microcin J25 and Nisin, can promote growth and improve intestinal health in poultry. Microcin J25, a bacteriocin produced by *E. coli*, has a high level of inhibition against other bacteria, while Nisin, a bacteriocin produced by *Lactococcus lactis*, is used in the food sector due to its ability to suppress bacterial infections. However, the EFSA has not approved its usage in poultry, rendering it prohibited in animal diets. Nisin alters the animal's gut microbiota, reducing possible infections in the ileum, decreasing nutritional competition, and boosting energy utilization. AS7, a bacteriocin produced by *Carnobacterium divergens*, has been proven to stimulate growth and have antibacterial properties in grill chickens. Cecropin A-D-Asn, a recombinant AMP, reduces gut bacterial growth, increases nutrition utilization, and stimulates growth. Synthetic AMPs A3 and P5 can boost growth, improve nutrient absorption, and minimize intestinal damage (Abreu et al., 2023).

AMPs use as immunomodulators: Studies have shown that several AMPs, derived from various sources, can reduce bacterial infections in broiler flocks through immune modulation. AMPs can control the expression of proinflammatory and anti-inflammatory cytokines in the digestive tract, hence increasing intestinal morphometric characteristics and productivity. They can also imitate the physiological release of endogenous AMPs, making them viable alternatives to antibiotics. In addition, studies have indicated that bovine lactoferrin (bLf) can improve broiler chicken body weight, feed conversion, and intestinal morphology. Oral

administration of rabbit Sacculus rotundus antimicrobial peptides (RSRP) has been shown to increase intestinal mucosal immune responses in broilers, hence improving growth performance and intestinal health. Overall, AMPs have demonstrated potential as antibiotic alternatives in preserving intestinal balance in avian species (Abreu et al., 2023).

AMPs can be used in the prophylactic control or treatment of bacterial infections while promoting broiler growth. They can reduce pathogenic bacterial loads within the avian gut, increase the population of beneficial bacteria, and modulate intestinal microbiota. Synthetic and recombinant AMPs can improve the antimicrobial activity of conventional antimicrobials in poultry production (Abreu et al., 2023).

1.5. Organic acids

Organic acids are an effective alternative to antibiotics in poultry due to their role in reducing pH in the gut, improving nutrient utilization, and fighting against pathogenic bacteria. They also reduce the colonization of gut wall infections such as *Salmonella* and *Escherichia coli*, which harm epithelial cells. Acetic, formic, butyric, propionic, malic, lactic, and tartaric acids are among the most widely utilized organic acids. These acids are essential for digestion in poultry, particularly in diets with low protein quality. High protein fermentation can produce volatile fatty acids, ammonia, and other undesirable gases, all of which can be uncomfortable and detrimental to growth. However, adding organic acids improves the digestion of proteins and carbs. Organic acids have been found in studies to improve broiler development, feed conversion rate, and nutrient utilization while also providing protection against Campylobacter infection, and improve cell proliferation in the gastrointestinal tract. Supplementing with butyric acid in broiler diets has also been shown to improve chicken growth performance.

Chapter II : Essential oil from medicinal and aromatic plants

ESSENTIAL OILS

Essential oils are volatile, natural compounds formed by aromatic plants as secondary metabolites. They have antiseptic, therapeutic, and aroma characteristics and are used in embalming, food preservation, antibacterial, analgesic, sedative, anti-inflammatory, spasmolytic, and locally anesthetic medicines. They are important in plant protection as antibacterials, antivirals, antifungals, insecticides, and herbivore repellents. They are derived from various aromatic plants found predominantly in moderate to warm climates such as the Mediterranean and tropical regions. Volatile aromatic compounds from a scientific perspective, essential oils are often referred to as volatile aromatic compounds. Volatile aromatic compounds are small organic molecules that are known to change quickly from a solid or liquid state to a gas when put at room temperature. The word "volatile" refers to the quick rate at which these molecules change their state. Because essential oils change so quickly from a liquid state to a gas state, they are potent and easy to smell, even from a distance. This is because of the volatile nature of essential oils. The physical and chemical makeup of volatile aromatic compounds allows them to move quickly through the air and interact with special sensors in the nose. The type of volatile aromatic compounds found in an essential oil will determine what kind of aroma and benefits the oil has (Bakkali et al., 2008).

Approximately 3000 essential oils are known, with 300 being commercially important for the pharmaceutical, agronomic, food, sanitary, cosmetic, and perfume industries. They are utilized in perfumes, cosmetics, sanitary products, dentistry, agriculture, food preservation, additives, and natural cures. Some essential oils are claimed to develop a better understanding of essential oils' biological action is important for new applications in human health, agriculture, and the environment to treat organ failure or systemic illnesses (Bakkali et al., 2008).

1. Quality and authenticity of essential oils

The quality and authenticity of essential oils are critical considerations when choosing and using them. Here are some important factors to ensure quality and authenticity:

1.1. Source and origin

Essential oils are found in plants and plant parts, which play a crucial role in animal and human life by producing oxygen, providing food, regulating water, and creating habitats. Several plant types, species, and families have distinct features that make them useful for certain purposes. Essential oils are present in the seeds, bark, stems, flowers, roots, wood, needles, and fruit of various plants. Essential oils have been extracted from plant components for thousands of years, with ancient civilizations recognizing their utility for everyday chores like health care, cosmetic treatments, religious ceremonies, and burials. Technology and research have helped us learn the benefits and applications of plant components and essential oils, but ancient civilizations had it right. Because of the diverse range of plant types and species available, there are several applications for plants. Essential oils are created in small specialized glands on plants and are either expelled or saved for later use.

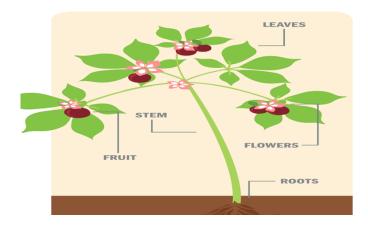


Figure 4: The schematic image showing the source of essential oils (https://media.doterra.com/us/en/ebooks/essential-oils-origins.pdf)

1.2. Botanical names

There are over 100 different species of plants that are used to produce essential oils, each with its unique chemical composition and therapeutic benefits. These plants can be classified into several categories, including:

- Herbs (e.g. basil, rosemary, thyme);
- Spices (e.g. cinnamon, ginger, cloves);
- Flowers (e.g. rose, lavender, jasmine);
- Trees (e.g. eucalyptus, tea tree, cedar wood);
- Roots (e.g. ginger, turmeric, valerian);
- Resins (e.g. frankincense, myrrh, dragon's blood).

The number of botanical species is estimated between 700000 and 1700000 (many of which remain to be discovered), and only a small group can be described as aromatic plants, 17500 according to Lawrence. These plants fall into a small number of groups based on their ability to produce the components of EOs (Laguerre and Virginie, 2015). In this chapter, we will explore 6 of the most popular essential oil-producing plants, including their common names, botanical names, and plant families. We will also examine the therapeutic benefits and uses of some oils (Thyme and Lavender), as well as their cultural and historical significance (Jill Taylor, 2004)

Table 1: List of some essential oil-producing plants (Adaszyńska-Skwirzyńska etSzczerbińska, 2016).

Common Name	Botanical Name	Family
Ylang Ylang	Cananga odorata	Annonaceae
Thyme	Thymus vulgaris	Lamiaceae
Rose	Rosa damascena	Lamiaceae
Lavender	Lavandula angustifolia	Lamiaceae
Ginger	Zingiber officinale	Zingiberaceae
Basil	Ocimum basilicum	Lamiaceae

1.3. The therapeutic benefit of Thyme and Lavender for chickens

Poultry, like all living things, can benefit from natural remedies to improve their health and well-being. Thyme and lavender are two herbs that have shown tremendous promise in helping chickens stay healthy. In this chapter we will investigate the medicinal effects of thyme and lavender EO on chickens, including their antibacterial, anti-inflammatory, and soothing characteristics.

1.3.1. Thyme (*Thymus vulgaris*)

Thyme is a perennial herb rich in antioxidants, vitamins, and minerals, as well as antibacterial, antifungal, and antiviral qualities, making it a natural cure for poultry.

Thyme can be used fresh or dried, as a full sprig (a single stem clipped from the plant), or as an essential oil extracted from the plant parts (Jill Taylor., 2004). Thyme volatile oils are one of the most commonly utilized essential oils as preservatives and antioxidants in the food and cosmetic industries. Specific applications investigated in poultry include:

Boosting the immune system ;

- Thyme contains antioxidants and essential oils that support the immune system;
- These natural compounds contribute to overall health and resilience in chickens;
- Antimicrobial properties: Thyme has been shown to have antibacterial and antifungal properties, effective against *E. coli*, *Salmonella*, and other pathogens that can harm poultry.

Respiratory health :

- Thyme has antimicrobial and anti-inflammatory properties;
- Thyme can help alleviate respiratory issues such as coughs, colds, and bronchitis in chickens

> Natural pest control :

- The strong scent of thyme is believed to repel pests such as mites, lice, and fleas, promoting a healthier environment for chickens;
- Placing thyme near the chicken coop can help keep unwanted critters away.

Enhancing egg flavor :

- Chickens that consume thyme tend to produce eggs with a more flavorful and aromatic taste;
- So not only does it benefit the hens, but it also adds a little extra yumminess to your breakfast table (Jill Taylor, 2004).

1.3.2. Lavender (Lavandula angustifolia)

Lavender (*Lavandula angustifolia*) is an annual or short-lived perennial herb and one of 39 species in the *Lavandula* genus of flowering plants in the mint family (Lamiaceae). Lavender is mostly renowned for its usage in aromatherapy and the calming, soothing, and sedative qualities of its fragrance. Specific applications studied in poultry include:

- ➢ Relaxation:
- Hanging dried lavender in chicken coops has a natural relaxing effect on the chickens.
 - Respiratory system:

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- Lavender oil is commonly used to treat various respiratory issues. The stimulating properties of lavender essential oil can also release phlegm and reduce congestion associated with respiratory problems, hastening the recovery process and assisting the body in naturally eliminating phlegm and other undesirable material.
 - Pain relief
- Lavender has analgesic properties, alleviating pain and discomfort in chicken

Ways to use lavender in poultry

- Place dried lavender buds in their bedding or nesting boxes;
- Use lavender essential oil in a diffuser or blend with water to spray in the coop;
- Mix lavender into their feed or water;
- Make a lavender-infused spray for use on wounds and injuries (Jill Taylor., 2004)



Figure 5: The illustration Image of lavender plants. (<u>https://stock.adobe.com/dz/search?k=lavender</u>)

1.4. Extraction methods of essential oils

Several components of various aromatic plants can be extracted to produce essential oils, which then have several applications in cosmetics, pharmaceuticals, poultry, and food safety. The manufacturing method and technique for extracting essential oils are determined by the botanical extract's features and components. The key aspect determining the quality of essential oils is the extraction process utilized, as improper extraction procedures can destroy and alter the action of phytochemicals found in aromatic oils. The resulting impacts can be, for example, the loss of pharmacological ingredients, stain effect, off-flavor/odor, and

physical change in essential oils. Such extraction techniques can be divided into two categories: classical methods and innovative methods. The use of innovative techniques, such as ultrasonic and microwave-enhanced processes, has improved the efficiency of the extraction process in terms of time required for essential oil isolation and energy dissipation, as well as improvement in production yield high-quality of EOs (Rassem et al., 2016).

1.4.1. Classical methods

The methods applied to extract essential plant oils are based on water distillation by the heating process.

1.4.1.1. Hydrodistillation

Hydrodistillation is the oldest and simplest oils extraction method which was discovered by Avicenna, the first to develop extraction through the alembic. Rose was the first plant extract refined using this technology. The operations begin with immersing the plant components straight into water inside the alembic (vessel), and the entire mixture is cooked. The devices include a heating source, a vessel (Alembic), a condenser to convert vapor from the vessel into liquid, and a decanter to collect the condensate and separate essential oils from water (Rassem et al., 2016)

This extraction technique is regarded as a unique approach for extracting plant materials such as wood or flowers, and it is frequently used for extractions involving hydrophobic natural plant material with a high boiling point. Because the oils are surrounded by water, this process allows essential oils to be extracted at a controlled temperature without being overheated. The primary advantage of this extraction approach is its ability to isolate plant components below 100°C (Aziz et al., 2018). Hydrodistillation can be done through water immersion, direct vapor injection, or a combination of the two. This multilateral procedure is suitable for both large and small enterprises. The time required for distillation varies according to the plant material being treated. Prolonged distillation yields minimal essential oil but introduces undesirable high boiling point chemicals and oxidation byproducts.

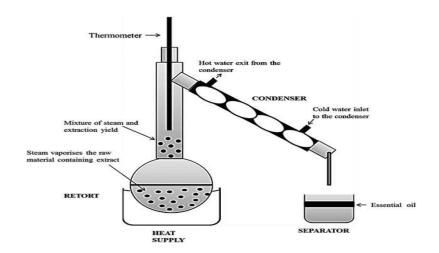


Figure 6: The schematic apparatus for hydro distillation (Oreopoulou et al., 2019).

1.4.1.2. Steam distillation

Steam distillation is a popular method for extracting essential oils from plant sources, accounting for 93% of all extractions. This technique involves heating plant material with steam from a steam generator, which is essential for breaking down and releasing aromatic components. Masango created an innovative steam distillation technology to increase isolated essential oil yields while reducing wastewater generation.

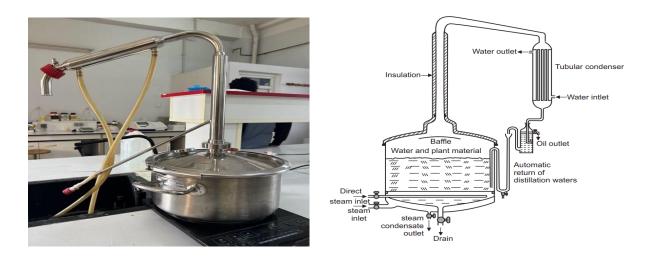


Figure 7: Schematic apparatus for Steam distillation (https://www.solnpharma.com/2022/03/steam-distillation-construction-working.html)

The technique employs a densely packed bed of plant samples above the steam source, allowing only steam to flow through the plants, and minimizing the amount of water in the distillate. Steam distillation is an ancient and officially permitted process of extracting essential oils from plant sources. It was previously a prominent laboratory method for purifying organic molecules, but it is now outdated due to vacuum distillation. However, it is still vital in certain industrial sectors. Steam distillation creates a mixture of vapor and the desired essential oil, which is subsequently condensed to collect the essential oil. This approach works on the idea that the total vapor pressure matches the ambient pressure at around 100°C, allowing volatile components to evaporate at temperatures similar to those of water (Aziz et al., 2018).

1.4.2. Innovative methods

Classical extraction techniques have disadvantages due to the thermolability of essential oils, which can undergo chemical changes at high temperatures. This can damage the quality of extracted oils, particularly if the extraction duration is prolonged. Modern industrial production prioritizes economics, competitiveness, environmental friendliness, sustainability, high efficiency, and quality. With technological advancement, new techniques have been developed that may not necessarily be widely used for the commercial production of essential oils but are considered valuable in certain situations, such as the production of costly essential oils in a natural state without any alteration of their thermosensitive components or the extraction of essential oils for micro-analysis

1.4.2.1. Solvent Free Microwave Extraction

Solvent-free microwave extraction (SFME) is a method of extracting essential oils from plant material without the use of solvents. It was invented by Cheat and colleagues, and it involves microwave dry-distillation at atmospheric pressure, which eliminates the need for water or chemical solvents. The plant material is moistened with water for 1 to 2 hours before being placed in a microwave oven cavity and the extracted oils are collected using a condenser. The instrument panel controls irradiation power, temperature, and extraction time. The extracted oil is dried with sodium sulfate and kept at 4°C in the dark.

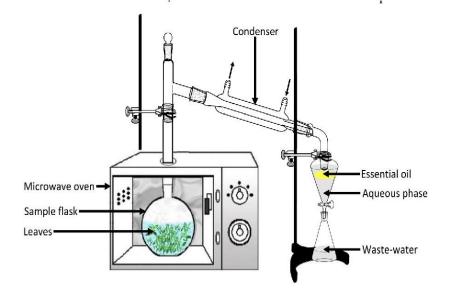


Figure 8. Schematic apparatus of the solvent-free microwave extraction (www.researchgate.net/figure/Schematic-representation-of-the-solvent-free-microwaveextraction-apparatus)

1.5. Chemical composition of EOs

As their definition specifies, EOs is a complex mixture of various constituents. In total, more than 3,000 constituents were isolated from the EOs. As the therapeutic activity of an EO is linked to its biochemical structure, only the main biochemical families will be presented (Virginie, 2015).

1.5.1. Hydrocarbons

The vast majority of essential oils belong to this group; they solely include molecules of hydrogen and carbon and are categorized as terpenes. These hydrocarbons can be acyclic, alicyclic (mono, bi, or tricyclic), or aromatic. This product family includes limonene, myrcene, p-menthane, α -pinene, β -pinene, α -sabinene, p-cymene, myrcene, α -phellandrene, thujone, fenchane, farnesene, azulene, cadinene, and sabinene, among others. These chemicals have been linked with a variety of therapeutic benefits.

1.5.2. Esters

Esters are sweet-smelling and add a nice aroma to oils; they are typically present in a wide variety of essential oils. Linalool acetate, geraniol acetate, eugenol acetate, and bornyl acetate

are a few examples. Esters possess anti-inflammatory, spasmolytic, soothing, and antifungal properties.

1.5.3. Oxides

Oxides or cyclic ethers are the strongest odorants, and by far the most known oxide is 1,8cineole, as it is the most omnipresent one in essential oils. Other oxides include bisabolone, linalool, sclareol, and ascaridole. Their therapeutic benefits include nervous system stimulants and expectorants.

1.5.4. Lactones

Lactones are quite high in molecular weight and are commonly found in pressed oils. Lactones include nepetalactone, bergaptene, costuslactone, dihydronepetalactone, alantrolactone, epinepetalactone, auscultating, citropene, and psoralen. They can be used for antipyretic, sedative, and hypotensive reasons, although allergies, particularly those involving the skin, are contraindicated.

1.5.5. Alcohols

In addition to their attractive scent, alcohols are the most therapeutically effective of essential oil components, with no recorded side effects. They are antibacterial, antiseptic, tonifying, balanced, and spasmolytic. Essential oil alcohols include linalool, menthol, borneol, santalol, nerol, citronellol, and geraniol.

1.5.6. Phenols

These aromatic components are among the most reactive, potentially toxic, and irritant, especially for the skin and the mucous membranes. They have qualities comparable to alcohols, but more pronounced. They have antibacterial and rubefacient effects, which stimulate the immunological and neurological systems and may lower cholesterol. Thymol, eugenol, carvacrol, and chavicol are some of the most common phenols that crystallize at ambient temperature.

1.5.7. Aldehydes

Aldehydes are common essential oil components that are unstable and oxidize quickly. Many aldehydes cause mucous membrane irritation and skin sensitization. They have a sweet, fruity odor and are found in some of our most well-known culinary herbs, including cumin and cinnamon. Certain aldehydes have been identified as antiviral, antimicrobial, tonic, vasodilators, hypotensive, relaxing, antipyretic, and spasmolytic. Common examples of aldehydes in essential oils are citral (geranial and neral), myrtenal, cuminaldehyde, citronellal, cinnamaldehyde, and benzaldehyde.

1.5.8. Ketones

Ketones are uncommon in most essential oils; they are relatively stable compounds that play little role as perfumes or flavorings. In some situations, ketones are neurotoxic and abortifacients, such as camphor and thujone (Gali-Muhtassib et al., 2000), although they also have some medicinal properties. They can be mucolytic, cell-regenerating, sedative, antiviral, analgesic, or digestive. Ketones are difficult for the liver to metabolize because they are so stable. Carvone, menthone, pulegone, fenchone, camphor, thujone, and verbenone are some common ketones found in essential oils (De Groot et al., 2016).

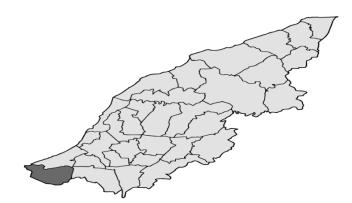
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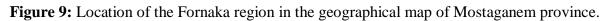
Material & Methods

Material and methods

1. Study area

Fornaka is a coastal commune in the Mostaganem province (figure 9). It is located 27 km from Mostaganem and 60 km from Oran. It covers an area of 4456.26 km²; it is a municipality with high agricultural and industrial potential and is characterized by a dry and hot semi-arid climate.





The farm "Mostaganem Food Poultry" (MFP) is located in a place called Douar Mkadid class 09 Fornaka. This place is characterized by the presence of numerous poultry farms as well as several slaughterhouses and hatcheries.

2. Presentation of the farm "Mostaganem Food Poultry"

The MFP broiler farm is made up of four hard, dark-type houses running parallel to a northsouth axis. Each house covers an area of 1034 m² (94m-11m) with a height of 4m in the middle of the building and 2.5m at the end. The capacity of each building is estimated at 18000 birds. The walls are made up of two walls, internal and external, made of brick. The roof is of the sandwich panel or one-piece double-skin cladding type, consisting of a layer of insulating material between two sheets of profiled material. The floor is flat and concrete, easy to clean and disinfect. Each house consists of two compartments separated by a hard wall. The space between the houses is 14 m (figure 10).



Figure 10: Broiler farm "Mostaganem Food Poultry"

3. Environmental factors

In all the houses, ventilation is dynamic, of a bilateral type. It is provided by two types of extractors, 8 larges and 2 smalls, which are located on both sides of the house. The temperature is ensured by radiant heaters that work with propane gas with a power of 1200 watts. Temperature and humidity are recorded using the sensors that operate automatically from the control cabinet. The cooling system is provided by pad cooling at a rate of two per house. Each house has a feed storage silo with a capacity of 15 tons. Lighting is provided by lamps suspended at a height of 2.5 m all along the house in 2 parallel lines to distribute the light evenly. It is controlled automatically with a light dimmer in order to guarantee optimal light intensity (20 lux minimum on 80% of the floor area) and to ensure an identical light program for the 4 houses (the choice of light program varies according to the high stump, the density of the animals, the average daily gain and the desired slaughter weight) (Table 2).

Age (days)	Duration of darkness (hours)
0	0
1	1
2-21	Progressive increase in darkness up to 8 hours
22	8
23	7
24	6
5 days before slaughter	5
4 days before slaughter	4
3 days before slaughter	3
2 days before slaughter	2
1 day before slaughter	1

Table 2: The standard light program was applied in the broiler houses.

A standard feed ration adapted to each phase of rearing (start-up, growth, and finishing) of broiler chickens and strictly identical was distributed to all the 4 houses. The feed is based on corn, soybean meal, and other feed additives such as VMS (vitamin-mineral supplement) (Table 3). It is distributed on round linear plates throughout the barn, which is distributed automatically but is regulated by the farmer every 4 hours and as needed.

	Start-up 0-13 days	Growth 13-28 days	Finishing 29 days- withdrawal
Type of feed	Flour	Flour / Pellets	Pellets
Corn	55.50	57.90	59.10
Soy	35.40	31.75	27.50
Calcium	1.35	1.10	1.20
Oil	0.50	0.50	0.60
Low Flour	5.00	7.00	10.00
Phosphate	1.25	0.75	0.60
VMS 260 (phytase + Rovabio + Cox)	1.00	1.00	1.00
Polycalcium	-	-	-
TOTAL	100	100	100

Table 3: Composition of feed used in the three phases of rearing (start-up, growth, finishing) in the four houses.

Watering was *ad libitum* throughout the breeding period. It is provided by 2 types of drinking troughs: plastic siphoid drinking troughs, for the start-up period (1st to 7th day), and linear drinking troughs, used during all growth and finishing phases.

4. Animals and treatments

Plant material and extraction procedure

The aerial parts of the two plants were collected from Algiers area, respectively, during their flowering period in 2023. Aerial parts were separated and dried at room temperature from 2 to 3 weeks in darkness and then stored in sealed paper bags. The essential oils were extracted using the steam distillation method for 2 h (for both *Thymus vulgaris* and *Lavandula angustifolia*) and stored in sealed glass vials at 4°C prior to use.

Animals

This study was carried out from March 17, 2024 to April 28, 2024, on a total of 160 day-old broiler chicks (Cobb 500) of both sexes in a completely randomized design with 5 treatment groups, consisting of four replications with 8 birds in each replicate. One day old chicks

were randomly assigned to experimental treatments after weighing. All chicks were vaccinated against several diseases (Table 4).

Age	Disease name	Type of Vaccine	Mode of
			administration
0	- Gumboro	- CEVAC	Transcutaneous
(at hatchery	- Newcastle	TRANSMUNE	Injection for
level)	- Infective	IBD	Transmune
	Bronchite/	- VECTORMUNE	/Vectormune
	Newcastle		and
	- Infective	- VITABRON	nebulization for
	Bronchite		bronchitis and
	(virion)		Newcastle
10	- Newcastle	Hipraviar B1	Nebulization
18	- Infective	Volvac IB Mass	Nebulization
	Bronchite		

Table 4: Vaccination program carried out during the breeding period.

Broiler chickens in Batches 1, 2 and 3 received drinking water supplemented with TEO 250μ L/L, LEO 250μ L/L, and TEO 125μ L/L + LEO 125μ L/L, respectively. The broiler chickens were provided with water in reversible waterers on a 5 L manual reservoir tank (in each pen). Once all the LEO-infused water was consumed, the reservoir tanks were replaced with new ones containing clean water without the addition of LEO. Those of batch 4 (negative control) were received drinking water with no EO addition throughout the 42-day rearing period. Broiler chickens in batch 5 (positive control) were treated with various antimicrobial (table 5) and antiparasitic agents (table 6) throughout the production cycle.

AGE (days)	ANTIBIOTIC	QUANTITY
0	ENROBROXINE 1L	3
0	COLISTINE 500ML	2
10	FOSFOMYCINE AAC 500G	8
26	TYLVALOSINE	8
26	COLISTINE 500ML	7

Table 5: Antibiotics used during the experiment

Table 6: Antiparasitic used during the experiment

AGE (days)	ANTIPARASITE	QUANTITY
15	TECHNOZURIL	20
	250ML	
28	ALGICOX	7

5. Zootechnical and health performance

At the end of the rearing period (42 days), the chickens were weighed individually. To determine the feed intake over the 42 days, the amount of feed remaining at the end of the rearing period was subtracted from the total feed given during the period. The feed conversion ratio was calculated by dividing the average feed intake by the average weight gain. The average daily gain was obtained by dividing the average weight gain by the number of days in the rearing period. Finally, the mortality rate was determined by dividing the number of deaths in each batch by the initial number of individuals and then multiplying it by 100.

6. Carcass characteristics

To determine the impact of the essential oils on the carcass components, three (3) birds were randomly selected from each batch with treatments replicated 3 times. They were

slaughtered and their weights before and after slaughter were recorded. After weighing the weight of the whole carcass, other components were also measured such as the breast, thighs, wings, heart, liver, pancreas, gizzard, and abdominal fat.

7. Blood biochemical & hematological parameters

To measure the blood biochemical and hematological parameters, 5 ml of blood samples from birds in each replicate were collected through the wing vein and then 2.5 ml was transferred to test tubes without anticoagulant to extract the serum and another 2.5 ml to tubes containing anticoagulant to prepare plasma. The blood samples were transported to the laboratory and processed immediately for biochemical (cholesterol, triglycerides, glycemia, high-density lipoprotein, low-density lipoprotein, etc.) and hematological parameters (leukocytes, erythrocytes, hematocrit, hemoglobin, mean corpuscular volume, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration).

8. Statistical analysis

All data collected were statistically analyzed in a completely randomized design using the GLM (General Linear Model) option in SPSS 2024 to conduct an analysis of variance (ANOVA) test. A one-way ANOVA test was used to compare the effect of the different treatments (essential oils) on the carcass components and blood parameters. The treatments were considered as the independent variable, while the carcass components and blood parameters and blood parameters were considered as the dependent variables. The statistical model applied was: $Yij = \mu + Ai + eij$, where Yij is the value of each observation, μ is the overall mean of the trait, Ai is the effect of essential oils, and eij is the standard error. Duncan's multiple range test at a 5% probability level was used to separate the means.

Results & Discussion

Results and discussion

1. Results

1.1. Zootechnical and health performance

The first part of this study aimed to study the impact of the EO preparations on the growth performance of industrial broiler chickens, hatched and reared under commercial conditions. The effect of T+LEOs on broiler chickens performance is presented in the table 7. The results of this study showed that the weight at day 42 of the broilers in batches 1, 2, 3 and 5 was higher than that of batch 4 (the negative control). It shows that TEO, T+LEO and LEO supplementation increased body weight by 10.37%, 5.42% and 4.33% at 42 day of age, respectively (P < 0.05). The feed intake was reduced in the group treated with T+LEOs by 2.27% when compared to the control group (P < 0.05) and it was increased by 12.16% in the ATB group (P < 0.05). Also, the feed conversion ratio (FCR) decreased and the average daily gain (ADG) increased in chickens that had EOs added to drinking water when compared to the control group (P < 0.05).

	Batch 1	Batch 2	Batch 3	Batch 4	Batch 5	P value
	(TEO)	(LEO)	(T+LEO)	(Control)	(ATB)	
Average live weight on D 0	42.8	42	40.5	41.2	43	0.385
Average live weight on D 42	2172.7	2053.8	2075.3	1968.5	2081.2	0.004
Total feed intake (FI) in (g)	4125	4125	4031	4125	4627	0.02
Feed conversion ratio (FCR)	1.89	2.00	1.94	2.09	2.2	0.014
Average daily gain (ADG)	51.73	48.9	49.4	46.8	49.5	0.048

Table 7. Effect of essential oils on the broiler chicken performance.

Figure 11 shows the weekly growth curves of the animals according to the type of EO they received. It can be seen that a regular growth curve is observed in batches 2 and 3, batch 4 started the same way but the growth declined towards the end of the breeding period. Batches 1 and 2 showed better results, starting similarly to the other batches. The batch 1 showed a significant increase in growth from day 12 up until day 30 when it slightly declined but picked up the pace and ended with the highest average weight. The batch 5 on the other hand,

showed a consistent high growth curve from the start of the breeding period finishing slightly below the batch 1.

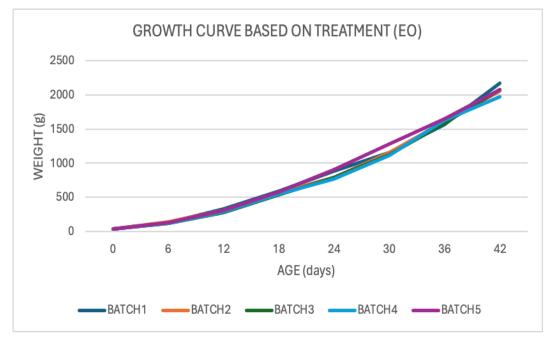


Figure 11: Growth curve of broiler chickens complemented with EO

The batch 3 which received a mixture had a significantly higher mortality rate (18.75) when compared to the control (P > 0.05) (figure 12). Four chickens died within the last two weeks of the rearing period. The TEO group had a significantly lower mortality rate. The batch 1 and 5 had two (2) deaths each, batch 2 and 4 had four (4) deaths each while batch 3 had a total of 6 deaths.

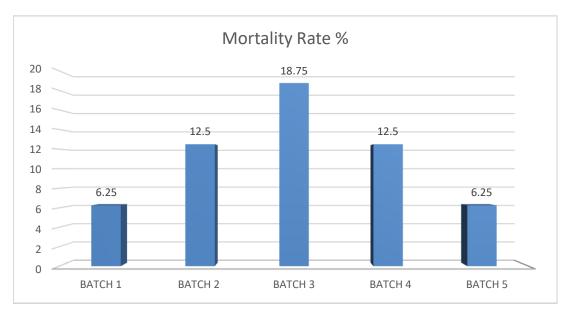


Figure 12: Mortality rate of broiler chickens at the end of the rearing period.

1.2. Carcass characteristics

The effect of EO supplementation on carcass characteristics of broiler chickens is presented in the table 8. The breast meat yield shows variations across treatments, with the highest mean value observed in the batch 5, the antibiotic group (ATB) (19.7), and the lowest in the control group (15.7). However, the differences were not statistically significant (P > 0.05). Liver percentages differ significantly between the different batches (P < 0.05). The control group had the highest liver weight (3.07), while the TEO and T+L EO groups had lower liver weights (2.4 and 2.48 respectively). The gizzard percentages do not show statistically significant differences among the treatments (P > 0.05) although the control group had the highest mean value (3.34) and the T+L EO group had the lowest (2.72). Heart percentages show statistically significant differences (P < 0.05). The LEO group had the highest heart percentage (0.68), while the T+L EO group had the lowest (0.53). Pancreas weights also exhibited significant differences (P < 0.05) when compared to the control. Pancreas percentage increased in birds treated with EO. Fat percentage showed significant differences among treatments (P < 0.05). The fat percentage decreased in birds treated with antibiotics. The LEO group had the highest fat percentage (2.1), while the ATB group had the lowest (1.0). The carcass percentage increased in birds treated with EO when compared to the control (P < 0.05). The T+L EO group had the highest carcass percentage (74.6) while the control group had the lowest (69.7). The thigh percentage also increased in birds treated with EO when compared to the control (P < 0.05). The TEO group had the highest thigh percentage (18.9) implying that TEO had a positive effect on thigh muscle development. Wing percentages also did not show statistically significant differences when compared to the control (P > 0.05). However, birds treated with antibiotics had the highest wing percentage (8.8) and the control group had the lowest. This indicates that neither antibiotics nor EO significantly influenced the wing development of broilers used in this study.

Items		Treatments					
	Control	ATB	LEO	T+L EO	ΤΕΟ	F value	P value
Carcass	69,7±2,20 ^b	74,3±2,90 ^a	72,5±0,40 ^{ab}	74,6±1,50ª	74,2±0,50 ^a	3.89	0.037
Breast	15,7±1,6	19,7±2,40	16,8±0,9	18,4±1,9	18,9±2,2	2,14	0.15
Thigh	15.73°±0.01	19.66 ^a ±0.02	$16.84^b \pm 0.03$	18.42 ^{ab} ±0.09	18.91 ^{ab} ±0.06	2.13	0.013
Wings	8,6±0,50	8,8±0,30	8,6±0,50	8,9±0,10	7,9±0,60	2.11	0.154
Liver	3,07±0,05 ^a	2,73±0,30 ^{ab}	2,85±0,28 ^{ab}	2,48±0,14 ^b	2,47±0,13 ^b	4.66	0.02
Pancreas	0,2200±0,02 ^c	0,25±0,02 ^c	0,32±0,03 ^{ab}	0,27±0,02 ^{bc}	0,34±0,04 ^a	8.18	0.003
Heart	0,67±0,03 ^a	0,59±0,09 ^{ab}	0,68±0,01ª	0,53±0,02 ^b	0,57±0,03 ^b	5.43	0.014
Gizzard	3,34±0,19	2,98±0,48	3,02±0,11	2,72±0,21	2,92±0,28	1.86	0.19
Abdominal Fat	$1,7\pm0,40^{ab}$	1,0±0,20 ^b	2,1±0,40 ^a	1,4±0,50 ^{ab}	$1,8\pm0,10^{a}$	3.61	0.045

Table 8 : Effect of EO treatments on carcass characteristics of broiler chickens.

^{a,b,c}Values with different letters significantly differ (P < 0.05).

1.3. Biochemical and hematological blood parameters

The table 9 presents a comprehensive analysis of the biochemical blood parameters of broiler chickens subjected to EO and antibiotic treatments.

Table 9 : Effect of experimental treatments on biochemical blood parameters of broiler chickens.

	Group						
Trait	ATB	CONT	LAV	T+L	TH	F value	P value
Cholesterol	1,03±0,25	0,87±0,10	0,91±0,20	0,94±0,11	0,94±0,15	0,37	0,82
Triglycerides	1,02±0,33	0,80±0,09	0,67±0,29	0,69±0,26	0,71±0,27	0,93	0,48
CHOL/HDL	1,52±0,11	1,39±0,06	1,42±0,25	1,36±0,03	1,32±0,13	0,55	0,70
Glycemia	2,49±0,24	1,84±0,50	2,53±0,32	2,82±0,31	2,54±0,51	0,37	0,82
HDL	0,67±0,12	0,63±0,07	0,64±0,07	0,69±0,08	0,71±0,05	0,95	0,47
LDL	0,18±0,10	0,10±0,03	0,15±0,17	0,13±0,03	0,10±0,06	2,59	0,10
TGO	482,00±281,73	336,67±184,72	270,00±87,58	214,00±15,87	248,33±42,25	1,37	0,31

HDL high-density lipoprotein; LDL low-density lipoprotein; GOT; glutamate oxaloacetate transaminase CHOL/HDL cholesterol/ HDL ratio.

According to the table, the cholesterol levels across different treatment groups ranged from 0.87, in the control group, to 1.03, in the ATB group, indicating no statistically significant differences among the groups (P > 0.05). This suggests that none of the treatments had a notable impact on the cholesterol levels. Triglyceride levels varied, with the control group exhibiting 0.80 and the ATB group showing 1.02, signifying no significant differences among them (P > 0.05) even though the levels were slightly decreased in the birds treated with EO. This means that the experimental treatments including the use of EO did not significantly alter the triglyceride levels, suggesting stable lipid metabolism under these conditions. The ratio between cholesterol and HDL ranged from 1.32 in the TEO group to 1.52 in the ATB group, indicating no significant differences compared to the control (P > 0.05). The absence of substantial differences could mean that the EO did not negatively impact the cardiovascular risk profile of the broiler chickens. Glycemia levels ranged from 1.84 in the control group to 2.82 in the T+L EO group indicating no statistically significant differences among the groups (P > 0.05). This parameter is critical for assessing glucose metabolism, and the results suggest that the treatments did not adversely affect blood glucose levels. HDL levels were fairly consistent across the groups, ranging from 0.63 in the control group to 0.71 in the TEO group, which also indicates that there were no significant differences (P > 0.05). HDL is considered a good cholesterol and its stable levels suggest that the EO treatments did not detrimentally affect the lipid profiles of the broiler chickens. LDL levels ranged from 0.10 in the TEO group, to 0.18 in the ATB group which indicates no significant differences recorded (P >0.05). GOT levels varied widely from 214.00 in the T+L EO group to 482.00 in the ATB group which also indicated no statistically significant differences when compared to the control (P > 0.05).

The effect of experimental treatments on hematological blood parameters of broiler chickens is presented in the table 10. The results of this study showed that EO supplementation did not significantly affect the hematological blood parameters of broiler chickens.

	Group					F	Р
Trait	ATB	CONT	LAV	T+L	TH	value	value
Leukocytes	44956,7±9220,6	44966,7±15695,6	33593,3±7952,4	30250,0±2790,9	32323,3±22510,3	0,85	0,52
Erythrocytes	3,2±1,0	2,4±0,1	3,6±1,1	$2,4\pm0,1$	2,6±0,2	1,78	0,21
Hematocrit	43,9±15,3	31,3±1,6	48,0±15,6	31,4±1,8	34,0±2,1	1,83	0,19
Hemoglobin	15,4±5,3	11,2±0,5	16,9±5,4	11,5±0,3	12,1±0,9	1,72	0,22
MCV	135,1±3,9	130,1±1,4	132,6±1,4	128,8±3,1	129,9±3,0	2,56	0,10
MCH	47,5±1,2	46,7±0,6	46,8±0,9	47,1±0,6	46,1±0,6	1,22	0,36
MCHC	35,2±0,2	35,9±0,6	35,3±0,4	36,6±1,0	35,5±0,4	2,72	0,09

Table 10: Effect of experimental treatments on hematological blood parameters of broiler chickens.

MCV mean corpuscular volume; MCH mean corpuscular hemoglobin; MCHC mean corpuscular hemoglobin concentration

Leukocyte levels decreased in broiler chickens treated with essential oils. However, they were not statistically different compared to the control (P > 0.05). Chickens treated with EO and antibiotics had slightly higher hemoglobin, hematocrit, and erythrocyte levels compared to the control but the differences were not also statistically significant (P > 0.05). MCHC values ranged from 35.2 (ATB) to 36.6 (T+L EO), MCH values from 46.1 (TEO) to 47.5 (ATB), and MCV values from 128.8 (T+L EO) to 135.1 (ATB). These values were, however not significantly different compared to the control (P > 0.05).

2. Discussion

The current study was carried out to explore the effects of EO on growth performance, carcass characteristics and biochemical blood parameters in broilers. Our findings showed that TEO, T+LEO and LEO supplementation increased body weight by 10.37%, 5.42% and 4.33% at 42 day of age, respectively. Weighing broilers is a crucial aspect of poultry farming, providing valuable insights into the health, growth, and overall performance of the birds. Weighing around 42g at hatch, broilers can achieve a weight of 2,800g and an average daily growth of 66g under normal conditions (Valco, 2024). In addition to enhancing body weight, the results of this study showed that the use EOs lowered feed intake in broilers, decreased feed conversion ratio (FCR) and increased the average daily gain (ADG). These results are in agreement with those reported in several studies (Alçiçek et al., 2003; Arif et al., 2024; Dnyaneswarn et al., 2024). There is strong evidence that EOs have a positive effect on the production performance of broiler chickens, which is reflected in reduced feed intake,

increased body weight gains, and better immunity and health as previously reported (Michalina and Danuta, 2016). Feed Conversion Ratio (FCR) is a critical problem in livestock production, particularly on commercial animal farms. Feed accounts for almost 70% of the entire cost of production in poultry production, hence the FCR in livestock farming is quite important to a livestock farmer, it's used to assess an animal's production and efficiency. A standard broiler chicken should have an FCR of 1.7-1.9 (Poultry care, 2022). Several studies have also recorded an increase in mortality rates of broilers as they approached the end of the rearing period when they added two blends of EO components into the feed (Alexander et al., 2015; Mitsch et al., 2004; Shamma et al., 2019).

Concerning carcass characteristics, the results of this study showed that the carcass percentage increased in broilers who had essential oils added to drinking water. This result corresponds to the results obtained by Hoseinyan Bilandi et al. (2018) who reported that the active compounds of herbal medicines could increase the growth rate of the carcass of broiler chickens. Thanks to the antimicrobial properties of medicinal plants, bacterial population is reduced, which could lead to better absorption and utilization of nutrients and thus, improve carcass percentage. The results of the current study showed also that the liver percentage in chickens treated with thyme essential oil and a mixture of thyme and lavender essential oil was lower. This significant difference indicates that essential oil treatments might reduce liver weight compared to the control and antibiotic treatments. This result is similar to that of Babak et al. (2024), Bölükubaşi et al (2006), and Tilhonen et al (2010). The liver is an important organ that plays a role in the detoxification and filtration of toxins produced by harmful microbes (Tilhonen et al, 2010). The decrease in liver weight signifies a reduction in the detoxification activity (Babak et al., 2024). Therefore, this may be taken as one of the positive effects of adding Thyme EO and a mixture of both Thyme and Lavender EO to the drinking water of broilers. Thigh muscle percentage also increased. This could be a result of the anti-oxidative properties of plants. Lipid oxidation causes deterioration in the quality of meat and causes undesirable effects on nutritive value (Shakeri et al., 2019). Acute thermal stress also increases the oxidative damage to chicken skeletal muscles (Mujahid et al., 2007).

The lack of statistical significance in the gizzard percentage suggests that EO treatments did not have a strong impact on gizzard weight compared to the negative control and positive control, which is similar to the results obtained by Fawaz et al. (2021). The increase in the pancreas percentage of chickens treated with essential oils in this study suggests that they positively influence pancreas development or function by affecting digestive enzyme activity or nutrient absorption. The pancreas is one of the central organs of the digestive system which secretes enzymes that break down proteins, fats, and carbohydrates into monomers capable of absorption into the blood (Vladimir et al., 2023). From the current results, the percentage of abdominal fat increased in chickens with essential oils added to their diet. This result opposes the result obtained by Al-Kassie (2009) who added different levels of thyme and cinnamon oil to the diet of broiler chickens. The effect of these essential oils lowered the fat percentage in the chickens.

The results of this study reported that thyme and lavender essential oils did not affect the biochemical and hematological blood parameters. Therefore, TEO and LEO did not negatively affect these parameters. The evaluation of the effect of EO on biochemical and hematological blood parameters is very important, serving as a vital tool to help detect any deviation from normal state of well-being in animals (Gbolabo et al., 2015). According to the results obtained by Christensen et al. (2010), S. officinalis (sage) lowered plasma cholesterol and triglyceride but increased HDL levels in lipidemic rats. Z. Mohammadi et al. (2013) also obtained similar results when they added clove essential oil to the diet of broilers. The absence or presence of cholesterolaemic effects of essential oils in an animal depends on the breed, gender, age, and composition of the feed (Lee et al., 2003). Z. Mohammadi et al. (2003) reported higher values in Mean corpuscular volume (MCV) and mean corpuscular hemoglobin (MCH) in their study. The parameters that influence the hematology and serum biochemistry of various livestock animals are typically under two broad categories; genetic and non-genetic parameters. The breed and genotype of the animal are under the genetic parameters, while the non-genetic parameters include age, sex, management system, medication, health status, and environmental factors such as nutrition, hormones, and climate. Hematological values of farm animals could also be influenced by geographical location, season, climate, length, and time of day (Etim et al., 2014).

Conclusion

Conclusion

This study has demonstrated that the use of TEO and LEO, either individually or in combination, can significantly enhance growth performance and carcass characteristics in broiler chickens. The findings showed notable improvements in body weight, feed conversion ratio, and average daily gain when essential oils were added to the drinking water of the chickens. Additionally, the study observed that the essential oil treatments did not adversely affect the biochemical and hematological parameters of the chickens.

The implications of these results are substantial for poultry farming, offering a promising alternative to antibiotics, which are commonly used to promote growth and prevent disease. By reducing the reliance on antibiotics, the use of essential oils like TEO and LEO could contribute to more sustainable and health-conscious poultry production practices.

Overall, the study supports the potential of essential oils as viable substitutes for antibiotics in poultry farming, paving the way for innovative and sustainable practices in the industry

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