

ADDIS ABEBA UNIVERSITY
COLLEGE OF VETERINARY MEDICINE AND AGRICULTURE
DEPARTMENT OF ANIMAL PRODUCTION STUDIES



**Effects of Dietary Garlic (*Allium sativum*), Thyme (*Thymus vulgaris*) and Their
Combination on Growth, Carcass Yield and Gut Microbial Population of Broiler Chicken**

MSc Thesis

By

Israel Yakob

June, 2023
Bishoftu, Ethiopia

**Effects of Dietary Garlic (*Allium sativum*), Thyme (*Thymus vulgaris*) and Their
Combination on Growth, Carcass Yield and Gut Microbial Population of Broiler Chicken**

**A Thesis submitted to College of Veterinary Medicine and Agriculture of Addis Ababa
University In Partial Fulfillment of the Requirements for the Degree of Master of Science
in Animal Production**

By
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June, 2023
Bishoftu, Ethiopia

ADDIS ABEBA UNIVERSITY
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Dedication

I dedicate this thesis manuscript to my two sons. My children Amenen and Dagim for motivating me to work towards self-improvement in aspects of life.

STATEMENT OF THE AUTHOR

First I declare that this thesis is my *bonafide* work and that all sources of material used for this thesis have been duly acknowledged. This thesis has been submitted in partial fulfillment of the requirement for MSc degree at Addis Abeba University. College of veterinary medicine and agriculture and is deposited at the university/college library to be made available to borrowers under rule of library. I solemnly declare that this thesis is not submitted to any other institution anywhere for the award of academic degree, diploma or certificate.

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BIOGRAPHICAL SKETCH

The author Israel Yakob was born in September 1994 in Addis Abeba capital city of Ethiopia. She attended her elementary and junior education in Yenegewsew private school after that she attended her senior last year in Dandii Boro school. Then joined Mekelle university and was awarded Doctorate in veterinary medicine in June 20 soon after graduation she started working at her own animal production farm and marketing business until she joined Addis Abeba university to pursue master of science degree in animal production.

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LIST OF ABBREVIATIONS

ADG	Average Daily Gain
ALP	Alkaline Phosphatase
AOAC	Associate Of Official Analytical Chemistry
BW	Body weight
CF	crude fiber
Cfu	Colony Counting Unit
CP	Crude protein
CSA	Central statistical agency
DADS	Diallyl disulfide
DAS	Diallyl sulfur
DATS	Diallyl trisulfide
DM	Dry matter
EE	Ether extract
EFSA	European Feed Standard Agency
EMA	European medicines agency
FAO	Food and Agriculture Organization
FCR	Feed Conversion Rate
FSA	Foreign Agricultural service
GC-MS	Gas Chromatography-mass Spectroscopy
GIT	Gasterointestinal tract
GRAS	Generally Regarded as Safe
HPLC	High performance liquid chromatography
LDL	low density lipoprotien
ME	metabolizable energy
NSP	Non- starch Polysaccharide
SAC	G-glutamyl S-allylcysteine
SAMC	S-allyllmercaptocysteine
Spss	statistical package for social science
WHO	World Health Organization

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Effects of Dietary Garlic (*Allium sativum*), Thyme (*Thymus vulgaris*) and Their Combination on Growth, Carcass Yield and Gut Microbial Population of Broiler Chicken

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ABSTRACT

*The study targeted evaluation of garlic (*Allium sativum* L.), thyme (*thymus vulgaris*) powders and their mixtures in broiler diets on growth performance, carcass yield and gut microbial population. The experimental trial was conducted using 156 unsexed day-old broiler chicks of Cobb500 breed which were divided into four treatment groups. Each treatment had a total of 39 chicks with three replicates and each replicate containing 13 chicks in completely randomized design. The treatments groups were diet containing 1% garlic powder (T1), 1% thyme powder (T2), 0.5% garlic + 0.5% thyme mixture powder (T3) and control group only basal diet (T4). Daily feed refusal, weekly body weight gain (BWG), carcass yield and fecal microbial counts were measured. The study showed that inclusion of 1% of garlic in the diet chicks had lower feed intake (1060.4g) during the grower phase followed by the control group (1063.05g), while higher feed intake was from 1% of thyme (1072.8g) followed by combination group (1070.9g). The results also showed that there was significantly lower feed intake (996.1g) for finisher phase and the entire period (2332.7g) for 1% thyme group. There was a higher feed intake (1716.1g) for 1% garlic and combined group (3055.0g) in the finisher phase and overall period, respectively. However, average final body weight, weight gain, feed conversion ratio, and carcass yield were not significantly improved by inclusion of dietary treatments compared to the control. Also, *E. coli* and *lactobacillus* count in the gut of broilers did not show any significant difference among treatment groups. The economic analysis revealed, highest profitability was acquired from herbal treatments than the control with sole garlic and thyme treatments showing a higher positive impact on the profitability of broiler production. The study also showed that addition of phytobiotic additives had a favorable impact on the quality of broiler meat, as demonstrated in the sensory taste. Therefore, it can be concluded that incorporating garlic and thyme into the diet of broiler chickens can yield a higher economic advantage and increase the quality of broiler meat.*

Key words: Broiler chicken; Feed additives; Garlic; Thyme; Growth performance; Microbial load; Carcass traits

1. INTRODUCTION

Medicinal plants have significantly contributed to the wellness of humans globally (Lucy, 1999). The use of botanical additives has been observed in human civilization since ancient times whereby herbs were employed for therapeutic purposes as well as preventive measures in both humans and animals. The use of these herbs has been successfully demonstrated by great civilization such as the Egyptians, Chinese, Greek, and the Romans (Jaric *et al.*, 2007). In Ethiopia, approximately 80% of people and 90% of livestock animals rely upon the use of medicinal plants to cure illnesses traditionally (Abebe *et al.*, 2001). In addition to essential nutrients, minerals and vitamins, the integration of feed additives has been shown to promote effective feed utilization. Herbal plants have been used as dietary supplements in the feed of various avian species including broilers, layers, local chickens, quails, ducks and pet birds (Haniarti *et al.*, 2019).

According to the EFSA (2003), feed additives are described as products used in animal nutrition with the aim of improving quality of feed and food of the animal origin while simultaneously enhance the performance and overall health of the animal. The use of natural plant based additives has become more popular in chicken production and authorization been granted based on the result of scientific evaluation on the safety of these additives on humans, animals as well as the environment (Pirgozliev *et al.*, 2019). In broiler production, chickens are confronted with various obstacles including climatic conditions, microbial load, changes in feed and stress during rearing period which can interfere with the proper functioning of their organs. Mainly the gastrointestinal tract (GIT) can be affected due to impaired absorption of nutrients resulting in reduced performance and increased mortality. The main characteristics of functional GIT include optimal digestion and absorption, regulating fluid and electrolyte balance and elimination of waste products (Celi *et al.*, 2017).

Phytobiotics are becoming important due to their natural composition, easy accessibility, absence of toxicity and residue free properties (Alagawany *et al.*, 2019). Several researches have demonstrated that the inclusion of phytobiotics in animal feed has the potential to promote growth, enhance gut functionality, antioxidant action, nutrient absorption and boosting of immunity along with reducing diarrhea occurrence (Zeng *et al.*, 2015; Gong *et al.*, 2013,

Zdunczyk *et al.*, 2010). Phytogetic additives in poultry have been found to improve microbiological status of the intestine thereby reducing the presence of pathogenic bacteria while increasing the number of beneficial microorganisms ultimately leading to improvement in immunity and resistance to GIT diseases as well as an increase in production performance. These plants based additives exert their immuno-modulatory effects by stimulating antioxidants and anti-inflammatory responses with GIT leading to improved nutrients absorption (Liu *et al.*, 2014; Mueller *et al.*, 2012).

It has been indicated that round 21,000 plant species can potentially be used as medicinal plants (Lucy, 1999). Ethiopia is believed to have rich biodiversity harboring approximately 6500 to 7000 species of which nearly 12% are identified as endemic (Abebe *et al.*, 2001). Phytobiotics feed additives are commonly used in terms of enhancing feed consumption, promoting body weight gain, improving feed conversion ratio (FCR) and enhancing meat quality. Among these additives are Thyme (*thymus vulgaris*), Garlic (*allium sativum*) that are administered either singly or in combination (Grashhorn, 2010; Puvaca *et al.*, 2014). The botanical species *allium sativum* commonly referred to as garlic is one of the most traditionally used plant additive that played a significant role on seasonings of foods. Furthermore, Garlic has been known for its medicinal value with a potential to prevent and treat various illness including infections and cardio vascular ailments (Javandel, 2008).

It has been proven scientifically of its antibiotic, anticancer, antimicrobial, antioxidant, immune modulatory, anti-inflammatory, hypoglycemic and cardiovascular-protecting effects (Reuter *et al.*, 1996). According to CSA (2018), Garlic is recognized as the second most extensively cultivated allium species in Ethiopia, following onion and the estimated production of garlic surpasses 1665.28 tons. Garlic is composed of numerous active compounds, such ajoene, s-allyl cysteine, diallyl sulphide and the most active compound allicine (Rahmatnejad and Roshanfekar, 2009). According to Alder and Houlub (1997), it is possible that allicin has the ability to reduce low density lipoprotein (LDL), triglycerides and cholesterol present in serum. The use of garlic as a natural feed additive in broiler feed has shown a significant enhancement in weight gain while reducing mortality rate (Stanacev *et al.*, 2012). Additionally, a separate study has reported that inclusion of garlic in broiler feed improved their productive performance (Demir *et al.*,

2003). A study showed that incorporating garlic in broiler chickens feed resulted in improved weight gain and FCR (Kirubakan *et al.*, 2016).

Thyme (*Thymus vulgaris*) is a botanical species known as plant with various medicinal properties. Since ancient times thyme has been used to achieve healing and relieve pain. *Thymus vulgaris* is used as antiseptic, antiviral and antimicrobial agent (Shyamapada and Manisha, 2016). Thyme leaves are used as flavors in variety of food products (Damite and Mekonnen, 2015). The use of thyme in poultry diet has been practiced to increase weight gain and health of the chickens (Khan *et al.*, 2012). It was reported by Basilica and Basilco (1999) that thyme has many secondary metabolites which are active compounds. The main phenolic compounds in thyme are thymol (5-methyl-1-1-isopropyl phenol) and carvacrol (5-isopropyl-2-methyl-phenol) which act as potent antioxidants (Hoffman and Wu, 2010). According to Cross *et al.* (2007), thyme can improve the general health of broilers with its anti-bacterial, anticoccidial and antifungal activities. Adel *et al.* (2013) reported that mixing thyme in broiler feed would promote weight gain, increase feed consumption and decrease mortality. Also Abdel-Wareth *et al.* (2012) reported that adding thyme at different level in feed improved weight gain and FCR in broiler chickens.

According to Khaligh *et al.* (2011), the administration of combined phytobiotic additives may exhibit greater efficacy when compared to administration of a single additive. Furthermore a number of studies have demonstrated that the use of different herbal mixtures have yielded positive outcome in the performance of broiler chickens (Ademola *et al.*, 2007, Oleforuh-okoleh *et al.*, 2015). Although several studies have been conducted on the effect of garlic in broiler chickens, no report have been made regarding the potential impact of thymus or its combination with garlic on the performance of broiler chickens in Ethiopia. As mentioned above, the production of broilers is faced with many challenges which motivate researches to explore the potential of phytogenic feed additives in Ethiopia as an option for tackling these challenges which can offer substantial amount of benefits to broiler chickens. Therefore, the aim of this study was to investigate the outcome of supplementing garlic, thyme and their combined mixture with broiler feed and evaluated the production performance, gut microbial population and economic efficacy with the following specific objectives.

1. To determine the feed intake, growth performance and carcass characteristics of broilers supplemented with garlic, thymus and their mixture,
2. To determine the effect of dietary garlic, thyme and their mixture on gut microbial population,
3. To evaluate the effect of dietary garlic, thyme and their mixture on quality of broiler meat, and
4. To analyze the economic benefits of inclusion of garlic, thyme their combination in the diet of broiler chicks.

2. LITERATURE REVIEW

2.1. Broiler Production and Feed Resources in Ethiopia

Poultry meat is the most common type of meat that has been widely produced in recent years. Poultry meat accounts for 40.6 percent of the total meat production which amounts to 337.3 million tons in the year 2020 (FAO, 2020). The production of broilers holds a significant role in ensuring food security for the rapidly increasing human population in Ethiopia. The Ethiopian production system is strongly drawn toward broiler chicken production due to its short production cycle, high feed efficiency and high biomass per unit of agricultural land (Smith, 2001). Broilers are capable of achieving a substantial weight within five to six weeks however the ability to achieve favorable level of production is contingent upon the accessibility of high quality feed and effective preventive measures against diseases (Elijah and Ruth, 2012).

Alema poultry farm, recognized as the second largest commercial chicken farm in Ethiopia facilitates the production of more than half a million broilers annually for local markets. Ethiopia has thirty-five large scale commercial chicken farms operating with a capacity ranging from 10000 to 10,000 birds. According to FAO (2019), the farms are predominantly situated in Adama, Modjo, and Debrezite. A significant number of these establishments have been intentionally located within a 100 kilometer zone situated to the south of Addis Abeba to facilitate convenient access to feed veterinary services and market channels to Addis Abeba (FAS, 2017).

Feed resources in Ethiopia can be divided into two primary classifications conventional and non-conventional. Conventional feed resources refer to those feeds that have been traditionally used in numerous countries these conventional feed ingredients are maize, wheat, soybean, barley and byproducts such as wheat-bran, animal and vegetable protein sources like fish-meal, meat-meal, soybean-oil-meal, groundnut-cake and more according to their availability (Younas and Yaqoob, 2005). However non-conventional feed is not commonly used in commercial production of feed the primary origin of these feeds are agricultural and forest-by product. The use of unconventional feeds is increasing due to shortage of conventional ingredients (Rajesh, 2021).

2.2. Phytobiotics as Feed Additives in Poultry

Feed additives are generally divided into organic and non-organic additives. The organic feed additives are products derived from plants which are used in feeding animals to improve their performance while inorganic feed additives are agrochemicals such as antibiotics (Nakatani, 2000). Furthermore, the EFSA recognizes five categories of feed additives as Technological (preservatives antioxidants), Sensory(colorings), Nutritional (Vitamins, trace elements,) Zoo-technical (gut flora stabilizers, probiotics, phytobiotics etc) and Coccidiostats and Histomonostats (inhibit protozoa, Ionophores). The application and advancement of enzymes, probiotic and phytobiotics in poultry nutrition has gained significant attention (Pirgozliev *et al.*, 2019).

Herbs, spices and plant extracts are known as botanicals, Phytobiotics. They are also referred to as plant secondary metabolites or phytochemicals, Phytobiotics, are plant-based products like herbs, spices, essential oils and oleoresins, having the potential to advance growth and serve as therapeutic agents (Windisch *et al.*, 2008). The use of phytogenic additives as a substitute for antibiotics in poultry production has gained attention worldwide as means to prevent the development of antibiotic resistant pathogens and meet consumer demand for drug free food chain (Dibner and Richards, 2005). The ability of phytogenics to contribute to the health of the chickens have been reported by Windisch *et al.* (2008). Phytobiotics can also be mixed with feed of commercial poultry to enhance feed utilization, improved animal production performance and improving the quality of the animal product (Windisch *et al.*, 2008).

2.2.1 Mechanism of action

Medicinal plants have been known to exhibit growth promoting effects in poultry. However the understanding of their mode of action and aspect of application is still limited and their effect remains speculative (Pirgozliev *et al.*, 2015). These may involve change in intestinal microbiota, by increasing digestibility and nutrient absorption (Prajapat, 2016). Numerous bioactive compounds derived from plants including many antimicrobial agent, exert their effect by the means of modulating the cellular membrane of microorganisms (Kamel, 2000). A variety of essential oil blends that feature natural polyphenolic compound or flavonoids as their primary

active constituents have been recognized as potential antimicrobial and antioxidants (Friedman *et al.*, 2004).

The improved nutrient digestibility leads to enhanced performance and health status in broilers. Additionally, it has been found that phytochemicals obtained from medicinal plants have the ability to relieve the host animal from stress due to immune defense at harsh conditions which in turn increases the intestinal absorption rate of vital nutrients and facilitates better growth of animals in accordance to their genetic potential (Hashemi and Davoodi, 2010). The integration of essential oil mixtures as a dietary supplement for broiler has been shown to promote the development of beneficial gut microflora leading to optimum digestion and improved performance (Cruickshank, 2001).

2.2.2. Preparation of phytobiotics

Herbal plant preparation is processed through techniques such as extraction, distillation, expression, fractionation, purification, concentration, dehydration and fermentation which are employed to convert to powdered herbs, tinctures, essential oils and expressed juices and processed exudates (EMA, 2021). Drying of medicinal plants is a process of reducing the moisture content of the herb in order to prolong its shelf life. It is a widely-adopted technique used for post-harvest preservation of medicinal plants primarily due to its easy and quick conservation of the medicinal properties in a simple way (Heeger, 1989).

Post-harvest medicinal plants are vulnerable to fungus infection because of their high moisture content. Hence it is crucial to reduce the level of moisture in the drying process by about 10% to 12% (Azizi *et al.*, 2010). In general, the effectiveness of phytochemical supplements can be affected by four factors: the plant's physical characteristics, the part of the plant used, when and where it was harvested, and how well it can blend with the animal feed (Wang *et al.*, 1998). These lead to a 50% difference in body weight gain and 63% variation in feed conversion rate among different studies (Gong *et al.*, 2013).

2.3. Garlic (*Allium sativum* L, *Nich-shinkurt*)

The exact origin of Garlic remains unknown however it is believed to have originated indigenously in central Asia and northern Iran. Garlic is diverse, including approximately 750 species but only seven of these species are commonly cultivated worldwide. Among these species onion (*Allium cepa* L), shallot (*A. cepa* var. *ascalonicum* L), garlic (*A. sativum* L.) and leek (*A. A. ampeloprasum* L.) are significant in Ethiopia. *A. sativum* was domesticated in ancient times and its historical documentation is found in the literatures of Egypt, Greece, India and China. In recent years, the annual global production of garlic amounts to be 10 million tons with China, Korea, India, The United States, Spain, Egypt and Turkey emerging as the world's largest producer (Acosta-Rodríguez *et al.*, 2008).

2.3.1 Botanical description of garlic

Garlic is a bulbous perennial plant that belongs to the family Alliaceae and genus *Allium* (Rekowska and Skupien, 2007). *Allium Sativum* known as garlic is a shallow rooted vegetable crop that can reach a height of 1.2m. It possesses a distinct aroma and pungent taste which is the hallmark of its properties. Garlic is widely cultivated and used in different regions depending on the geographic area its given different local names including rocambole, stinking, rose, rustic treacle, nectar of gods, camphor of the poor, poor man's treacle and clove of garlic. Garlic is widely consumed second to onion, it is known for its easy growth in temperate and tropical regions across the globe. Various subcategories or subspecies of garlic exist, where the commonly known ones are hard-neck garlic and soft neck (Acosta-Rodríguez *et al.*, 2008). Garlic has been employed as a flavor enhancer and common seasoning in the culinary practice (Adaki *et al.*, 2014).

2.3.2 Chemical Composition

Fresh garlic typically comprises of a variety of chemical constituents which include water, fiber, lipids, proteins and carbohydrates including fructose. The vitamin content of garlic is mainly comprised of vitamin C and vitamin A. In addition, it contains different minerals such as

potassium, phosphorous, magnesium, sodium, iron, calcium and also seventeen amino acids as well as phytosterols and phenolic compounds. However the medicinal effect comes from bioactive chemicals that are organic sulfur compounds which can be divided in two categories first group lipid –soluble compounds containing allylulfur, including diallyl sulfur (das, diallyl disulfide(dads and diallyl trisulfide (dats) and second group into water soluble compounds such as g-glutamyl S-allylcysteine (sac and s-allylmercaptocysteine(SAMC) (Bayan *et al.*, 2014).

Garlic has an acidic nature with a pH of 3.91. It possesses various components such as alkaloids, tanins carotenoids, saponins, flavonoids, steroids and cardenolides. purified alkaloids and its synthetic derivatives has bactericidal properties (Evans *et al.*, 2002). Also, a report by Yusuf *et al.*(2018) indicated that garlic powder contains 4.55 moisture, 15.33 crude protein, 2.10 crude fiber and 4.08 of ash content.

2.3.3 Antimicrobial effect of garlic in broilers chickens

The medicinal properties of garlic have been studied, indicating the effectiveness as a therapeutic agent. The effect of garlic was observed to decrease the pathogenic microorganism in the digestive system, leading to accelerated performance, improved digestion and enhanced immunity and health of broilers (Kumar *et al.*, 2010). According to Stanačev *et al.*(2010) garlic is known to possess beneficial properties such as antimicrobial and antioxidant effect which has shown a reduced mortality rate and increased gastric secretion in broilers. Moreover, due to its antimicrobial characteristics, garlic is considered medicinal plant for the growth promotion of broiler chickens (Darc, 2006). Purified alkaloids and its synthetic derivatives has bactericidal properties (Evans *et al.*, 2002). Plants that produce saponins have a defense mechanism that protect against external pathogens which make them a natural antibiotic. The significant concentration of saponin present in garlic may be responsible for its antimicrobial effect (Okwu and Emenike, 2006; Okwu and Nnamdi,2008). Abidullah *et al.* (2021) showed in their research that garlic had the highest range of antibacterial effect against both gram negative (*E. coli*) and gram positive bacteria(staphylococcus).

2.3.4 Effect on growth and carcass characteristics of broiler chicks

According to Rekowska and Skupien (2007) the inclusion on garlic in animal feed can enhance its flavor and make it more appealing, leading to increased voluntary consumption of the feed. Also, Rekowska and Skupien (2007) reported that even a relatively small amount of garlic extract added to the feed can stimulate broilers appetite and significantly increase their feed intake, ultimately resulting in greater weight gain. Kirubakaran *et al.*(2016) reported that the inclusion of garlic in the diet of broiler chickens may contribute to the weight gain also speculated that garlic has the potential to enhance the digestive process by amplifying the production of saliva and gastric fluid, ultimately leading to better nutrient absorption and increased body weight .

Table 1. Summary of response of broilers to dietary inclusion of different levels of garlic as phytobiotics

Dietary dose of garlic	Treatment effects	Reference
0.1 %	increased body weight gain a and FCR	Mansoub (2011)
1% & 3%	Improve FCR	Raeesi <i>et al</i> (2010)
0.25%	Improved Body weight gain & FCR	Onu (2010)
0.5 % &1%	improved bodyweight gain and feed conversion ratio	Nikola et al (2016)
0.1%,1.4%,1%,2% &3%	Improved feed intake and body weight gain and Reduced abdominal fat percentage	Oleforuh-okoleg et al 2014

2.4. Thyme (*Thymus vulgaris*, *tosign*)

Thyme (*T. vulgaris*) belongs to the family *Lamiaceae* known for its diverse species. *Thymus vulgaris* originated from southern Europe and has recently been introduced and cultivated in Wondogenet by the essential oil research center (Ermias *et al.*, 1998). According to Destaw *et al.* (2017). *T serrulatus* and *T schimperi* are indigenous to Ethiopia representatives to this genus are locally known as tosign in Amharic. Furthermore, this same report pointed out that *T. serrulatus* was found mainly in West Gojjam (Yilmana Densa) while *T. schimperi* was found from Tarma Ber, Buta Jira, and Bale.

2.4.1 Botanical description of thyme

Thymus vulgaris also known as garden thyme or common thyme is a perennial plant that has a woody base and a generally upright growth. It is largely cultivated as a culinary herb in gardens. A cluster of woody-based stems ascends to create a foliage mound that measures 6 -12 inches tall. The stems of the plant are clothed with leaves that are tiny, opposite, greyish green colored, shape that ranges from oblong-lanceolate to linear and are gland-dotted. Their size ranges from 5-10 in length and 0.8-2.5mm in width with their margins curved backwards. The flowers show a pale violet featuring two lip and having a glandular calyx covered in hair (Stahl and Venskutonis, 2012).

The fragrant leaves emit their stronger scent right before flowering and commonly used as fresh or dried for flavoring in various culinary practices, such as soups, stews, sauces, and meals featuring meat or fish. Cluster of mixture, cylindrical lavender blossoms emerge at the tips of the stems during latter part of the spring through the beginning of the summer. Bees are drawn to allure of the flowers. The environmental condition can cause variations in the morphological features of *Thymus vulgaris* it flourishes effectively in dry moderate and open areas. one can reproduce the plant through seeds, cuttings or by separating rooted portions Thyme has the ability to survive in frigid conditions and can be observed growing in elevated regions (Kuete, 2017).

2.4.2 Chemical Composition of Thyme

Thyme is mainly made up of wide range of chemical substances including phenolic compounds terpenoids, flavonoids, steroids, alkaloids, tannins and sponins. Majority of them are volatile compounds that have been acquired from plant oils using extraction. The composition of phytochemicals has been evaluated primarily by the use of gas chromatography-mass spectroscopy GCMS and High-performance liquid chromatography/thin layer chromatography HPLC/HPTLC. The findings reveal that essential oil components are more widespread compared to other metabolites Al-Asmari *et al.*, 2017).

Additionally, only a limited number of studies were able to identify the existence of steroids, alkaloids, tannins and saponins. however, there is a lack of knowledge regarding the specific chemical constitutes within these classifications (Fayad *et al.*, 2013). Besides analyzing the qualities of plant oil, it is crucial to conduct detailed examination of ethanolic and aqueous extracts to detect the existence of various significant elements. Significant pharmacological benefits can be derived from various phytochemicals that have been isolated from this plant. Although *thymus vulgaris* contains phytochemicals the process of isolating and identifying them has not yet been undertaken (Kuate, 2017). *Thymus vulgaris* is reported to have dry matter of 40.89%, EE 5.90%, CP 9.00%, Ash 12.10% (Jiang *et al.*,2020) while in a different report by Gerencsér *et al.* (2014) that crude fiber to be 18.10% in content.

2.4.3 Antimicrobial effect of thyme in broiler chickens

Thymus vulgaris is commonly known as thyme is a well-known as a medicinal plant which received significant interest due to its powerful antioxidant and antibacterial properties. According to Vincent (2002) thyme is reported to have antibacterial properties against various pathogenic microorganism. Friendman *et al.* (2002) also reported that thymol and carvacrol have antibacterial effects against *E.coli*, *S.enterica*, *C.jejuni*, and *L.monocytogenes in vitro*. It was also reported that *carvacrol*, *cinnamaldehyde* and *thymol* are particularly effective against *E.coli* (Tian *et al.*, 2019). The essential oil extracted from *thymus vulgaris* contains carvacrol and thymol which possess microbial and fungal properties. Such findings indicate that thyme plant has the potential to be used as an alternative natural enhancer of broilers. (Basilico and Basilico, 1999).

2.4.4 Effect on growth and carcass characteristics of broiler chicks

The inclusion of thyme oil in broiler feed led to an improvement in body weight gain, feed intake and feed conversion rate. This may be due to the presence of digestion stimulating compounds like thymol and carvacrol as well as their ability to fight bacteria in the intestine (Oliviero *et al.* , 2016). Thymol impacts harmful bacteria by altering the permeability of their cell wall, which leads to the pore formation, osmotic shock and leakage of cytoplasm, ultimately leading to the

bacteria cell death. Thymol's antimicrobial effects are brought by vital membrane ions such as potassium and hydrogen on the equilibrium pumps (Vouillamoz and Christ, 2020).

Table 2. Summary of response of broilers to dietary inclusion of different levels of thyme as phytobiotics

Dietary dose of thyme	Treatment effects	Reference
0.5,1,1.5 %2%	increased body weight gain, decreased FCR and mortality rate	El-Ghousein and Al-Beitawi, (2009)
1g/kg	increased body weight gain	Cross et al. (2007)
5g/kg	increased feed intake	Fallah and Mirzaet 2016
10mg/kg	improved bodyweight gain and feed conversion ratio	Wade et al., (2018)
150 & 200mg/kg	improved immune system and negative reduce effect of heat stress	Olfati and Mojtahedin, 2018

2.5 Gastro-intestinal Tract Microbes

The role of gut microorganisms is crucial in regulating energy metabolism and immunity in commercial poultry. The GIT is remarkably populated with divers range of bacteria, viruses fungi and protozoan, with bacteria mostly dominating (Sommer and Backhed, 2013; Wei *et al.*, 2013). The microbiome interacts with its hosts in ways that can be either damaging (pathogenic) or beneficial(symbiotic), and it can play important roles in human and animal health (Aruwa *et al.*,2021). Several studies in the past have highlighted the beneficial effect of gut bacteria in hosts, including epithelial barrier maintenance, inhibition of pathogenic bacterial adhesion to the intestinal surface, enhancement and maturation of hosts immunity, degradation of non digestible plant polysacchrdes and production of metabolites suach as hshort chain fatty acids (SCFAs) and vitamins (Al-Asmakh and Zadali, 2015; Sanchez *et al.*, 2017).

The crop, proventriculus, gizzard duodenum, jejunum ileum, cecual, and cloaca are all parts of chicken's GI tract (Carl *et al.*, 2012). Lu et al. (2003) examined 16S rRNA gene sequences to determine the ileal bacterial population and discovered lactobacillus as the dominant group

(70%), followed by the Clostridiaceae family (11%), Streptococcus (6.5%), Enterococcus (6.5%). Although most E.coli strains are harmless commensals, some strains have acquired the ability to cause intestinal or extra intestinal diseases (Mullata *et al.*, 2013). Lactobacillus species inhibit pathogenic growth and regulate gut flora (Fuller, 1989). The main goal of broiler chicken production is to increase body weight in a short period of time and gut microbiota are important factors that can benefit interaction with the host (Turnbaugh *et al.*, 2009; Dahiya *et al.*, 2017)

2.6. Meat Eating Quality

Meat quality is determined by sensory characteristics as well as the chemical composition of the meat. Tenderness, juiciness, and flavor/odor of cooked meat are all key sensory characteristics. A taste panel can evaluate meat flavor by testing and scoring meat samples subjectively by a group of individuals under more or less controlled conditions (Warriss, 2000). Sensory analysis has been utilized as a powerful technique in determining how alternative diets, processing, packaging, storage duration, and so on affect poultry meat quality.

Traditionally, concerns about chicken quality have centered on the appearance of both the carcass and the tenderness of the meat. Meat softness was traditionally associated with live bird quality criteria such as breed, sex, or age (Fletcher, 2002). However, juiciness refers to the amount of visible fluid that comes from meat when chewing. Juiciness and softness are strongly associated, and it is frequently observed that the more tender the flesh, the more easily juices are exhaled, and the juicier the meat is thought to be. Flavor is the result of the interaction of aromatic chemicals within the nasal cavity and chemical compounds that come into contact with the tongue during eating (Sativisal, 2012).

Another quality factor that consumers consider when determining the desirability of poultry meat is flavor. While eating, it is difficult to discriminate between flavor and odor. They both enhance the flavor of poultry. Sugar and amino acid interactions, lipid and heat oxidation, and thiamin breakdown all contribute to flavor production in poultry flesh during cooking. These chemical changes are not specific to chicken, but the lipids and fats in poultry are distinct and interact with odor to account for the 'poultry' flavor (Mir *et al.*, 2017). Fatty acid compositions in muscle are

important in the flavor development of poultry meat (Ismail and Joo, 2017), and glutamic acid has been demonstrated to have a detectable effect on the taste of chicken meat which may be due to the different flavor between the meats (Wattanachant, 2008; Ismail and Joo, 2017).

Before making a purchasing choice, consumers place a high value on the color, appearance, and texture of poultry meat (Liu *et al.*, 2004). Lyon *et al.*(2001) states that scientific assessment of chicken meat quality, which includes evaluating sensory, flavor, and texture profiles, has been widely utilized to justify pre-processed treatment and post-harvest processing for broilers.

3. MATERIALS AND METHODS

3.1. Description of the Study Area

The research was conducted from February to April 2023 at the poultry farm at college of veterinary medicine and agriculture, located at latitude: 08^o44' N; longitude: 38^o 57'E in East Shewa Zone of Oromia Regional state. Bishoftu is 47km far away from the capital city of Addis Ababa to the East at an altitude of 1900 meter above seas level and experiences A bimodal rainfall pattern with long rainy season from June to October and short rainy season from March to May. The average annual rainfall and averages maximum and minimum temperature of the center are 851mm, 28.3 and 8.9 °C, respectively (EIAR,2016).

3.2. Source and Preparation of Selected Phytobiotics

Fresh bulb of garlic was acquired from Debrezite Agricultural Research center found in Bishoftu. To prepare fresh garlic, bulbs were peeled of their outer and inner cover, using a knife, and then were air-dried. The specific species (*thymus vulgaris*) was brought from Wondogenet Research Center found at about 267 km South of Addis Ababa. The leaves of the thymus were taken apart from the stem carefully. Both additives were placed in different plates spread evenly allowed to air dry. After it is dried, it was ground to fine powder to pass through 1mm sieve screen. Dried garlic and thyme powders were stored at room temperature in a clean plastic bags and until added to the rations, which were carefully hand-mixed to ensure homogenous distribution of the powder given with the morning feed.

3.3. Preparation of Experimental House and Equipment

Initially the poultry shed was cleaned of the waste deposited by chickens from previous research. The waste was eliminated from the entire space after that the whole house was thoroughly rinsed with water and allowed to dry. Further two disinfectants were used for treating different parts of the shed. The floor was disinfected with sodium hydroxide known us caustic sodas while the mesh wire, the roof and the wall was disinfected with bio-safe disinfectant which was safer for

metallic compounds. Throughout the duration of the experiment drinking and feeding equipment's were washed with cleaning liquid soaps on a daily basis every morning. In addition, strict biosecurity measures were followed throughout the end of experiment.

3.4. Management of Experimental Animals

A total of 156 day-old unsexed Cobb500 broiler chicks were purchased from Daira farm and randomly distributed to four groups each with 3 replications making up 13 chicks per replicate and 39 chicks per group or treatment. Broiler chickens were kept for up to 42 days in a tin-roofed and tin walled poultry shed with half opened wall enclosed with wire-mesh and concrete flooring. The area was divided in 12 pens there were 4 treatments each with 4 replications. Each pen was partitioned with wire-mesh and wood at the bottom. The experimental pen enclosures were crafted from tin material and in addition it had an external storage room. The chicks were brooded under 200-watt bulb hanging at just the optimum height to ensure that every chick received adequate heat and light coverage. Despite the frequent electricity problems encountered, lightning program was for duration of 23 hours. *Teff* straw served as the bedding material for the use of deep liter. Replacing of wet litter was done by changing it with clean dry litter every 2 weeks. Temperature was reduced gradually every week, which was achieved by removing some bulbs from pens in the middle area. The chicks were vaccinated via eye-drop HB1, Gumboro, Thermo stable, Gumboro booster dose and Newcastle Thermo stable at day 2,7,12,17and 22 respectively. The vaccines were purchased from National Veterinary Institute in Bishoftu, Ethiopia.

3.5. Experimental Diet and Feeding Management

Experimental diets namely starter, grower and finisher diets were purchased from Alema koudijs feed plant for the respective feeding phases (Table 1). These diets did basically make-up the control diets. The rest of the treatments were formed by adding of the respective feed additives and their combination. The daily amount of feed given was minimum 20 gram/bird/day up to 160 gram/bird/day feed was offered twice a day at 8:30 AM and 5:00 PM. The chickens had free

access to clean and fresh water. Starter feed was given during week 1 and 2, grower during week 3 and 4 and finisher during week 5 and 6.

Table 3 : Ration composition of broiler chickens as given by the manufacturer

	Starter	Grower	Finisher
Dry Matter	91.76	91.69	91.41
%DM			
Crude Protein	20.54	19.00	17.36
Crude Fiber	5.50	5.00	4.94
Ash	5.86	5.62	4.88
Moisture	8.24	8.31	8.59
ME, kcal/kg	3000	3150	3250

3.6. Experimental Design and Treatments

The experiment was employed using a completely randomized design (CRD) consisting of four treatments replicated three times. Chicks were equally and randomly distributed to 4 treatments each with three replicates making up 13 per replicate and 39 chicks per treatment. Treatments were T1, T2, T3 and T4, where T1 and T2 were fed control diets plus 1% garlic and 1% of thyme respectively, while T3 was given the combination of 0.5% Garlic and 0.5% thymus in addition to control feed, and T4 was control diet without any additive (Table 2).

Table 4: The arrangement of treatment groups

Treatments	Types of Phytobiotics	Level of inclusion	No of birds per replication			Total no of birds
			R1	R2	R3	
T 1	Garlic	Basal diet + 1%	13	13	13	39
T 2	Thymus	Basal diet + 1%	13	13	13	39
T 3	Combination	Basal diet + 0.5% (G) + 0.5% (T)	13	13	13	39
T 4	Control	Basal diet	13	13	13	39
Overall			52	52	52	156

3.7. Measurements and Observations

3.7.1 Chemical analysis

Representative samples of garlic and thyme were taken and analyzed for Dry matter (DM), crude protein (CP), ether extract (EE), crude fiber (CF) and ash content in Debre-Birhan agricultural research center nutrition laboratory based on proximate method of analysis (Table 3). Samples were grounded by using Wiley mill (Thomas® Wily Cutting Mill) to pass through 1mm for chemical analysis. Nitrogen (N) was analyzed using Kjeldahl (AOAC, 1998) procedure and CP was calculated as $N \times 6.25$. Metabolized energy value (ME) of diets was calculated using the equation proposed by Wiseman (1987): $ME \text{ (kcal/kg DM)} = 3951 + 54.4EE - 88.7CF - 40.80ash$

Table 5: Chemical composition of feed additives

Chemical composition	Garlic	Thyme
DM,	96	88
%DM		
Ash	2.08	18.86
CF	4.78	31.45
CP	20.06	9.07
EE	1.4	1.9
ME, kcal/kg	3518.31	495.26

3.7.2 Feed intake

Throughout the experiment the measured amount of feed was offered twice daily at 8:00AM and 4:00PM hours. The feed refusal from each pen was collected every day in the morning before giving fresh new feed. The feed offered and the refused feed was recorded for each pen. The amount of feed intake is calculated by the difference between feed offered and feed refusal on a DM-basis.

Feed intake = Daily Feed Offered – Daily feed Refusal

$$\text{Average daily feed intake(g)} = \frac{\text{Feed intake}}{\text{Experimental days}}$$

3.7.3 Body weight gain

On the first day after being randomly assigned to their pens, the chicks were weighed in groups using sensitive balance. The total weight in every pen was divided by the number of chicks per pen to identify the mean weight of a chick per pen and recorded as initial body weight. Final body weight was taken at 42 day of the experiment and recorded. Body weight gain per pen per chick was calculated as the difference between the final and initial body weight. Average daily body weight gain per chick for each pen was measured by dividing total body weight gain per pen to the number of experimental days.

Body weight gain = Final body weight – Initial body weight

$$\text{Average Daily weight gain (ADG)} = \frac{\text{Body weight gain}}{\text{Number of experimental days}}$$

3.7.4 Feed conversion ratio

The feed conversion ratio (FCR) was determined by dividing average daily feed intake with average daily body weight gain.

$$\text{Feed conversion ratio(FCR)} = \frac{\text{Average daily feed intake (g)}}{\text{Average daily body weight gain(g)}}$$

3.7.5 Mortality rate

The Birds were monitored carefully during the day and night time . Throughout the experiment, mortality was recorded and the mortality rate percentage were calculated

$$\text{Mortality rate} = \frac{\text{Number of dead chicken}}{\text{Total number of chicken at the beginning of the experiment}} \times 100$$

3.7.6 Carcass measurements

At the end of the experiment 2, randomly selected birds per pen were put in a different pen and starved overnight, early in the morning they were weighed first and then slaughtered by cutting the jugular veins complete blood loss then become unconscious after a certain amount of blood loss. After de feathered by hand picking and removal of feet, head and the viscera dressed carcass was weighed. The dressed carcass, breast, thighs, drumsticks, wing and bone were weighed. Dressed and eviscerated weights were calculated following method of FAO (2001) and determined as

$$\text{Dressing percentage (\%)} = \frac{\text{Carcass weight}}{\text{Live body weight}} \times 100$$

$$\text{Carcass yield (\%)} = \frac{\text{Weight of dressed carcass (g)}}{\text{carcass weight (g)}} \times 100$$

3.7.7 Cecal bacterial load determination

At the end of the experiment (42 days) 24 birds (two bird from each replicate that have been selected for slaughtered for carcass evaluation and intestinal samples. Microbial load, was determined after the collection of intestinal content. Starting from the Meckel's diverticulum to 4 cm above the ileo-cecal junction was quickly dissected and the Cecal contents of each bird was squeezed out onto sterilized tube and transport to CVMA microbiology laboratory in icebox for enumeration of microbial, A 10-fold serial dilution method was used to determine colony forming unit (cfu) in each gram of intestinal content sample by means of pour plate method. E. coli was cultured on Eosin Methylene Blue (EMB) agar at 37°C for 48 hours. Lactobacilli were enumerated on lactobacilli MRS agar at 37°C for 72 hours. The counted coliform forming bacteria values were transformed to log₁₀ cfu/ml.

3.7.8 Sensory evaluation

Skinless breast meat was sliced into pieces of meat and was frozen until frying. For frying, the pieces of meat were thawed at room temperature, minced and cut into 2.5 cm cubes. The breast meat was fried for 15 min on a pan with vegetable oil, but without salt. After frying, the pieces

were cooled to room temperature. The panelist were given preliminary instruction before the actual testing session and finally asked to evaluate the appearance, Taste/Flavor, Texture, Aroma/odor, and overall acceptability of the breast meat cuts. Panelists were instructed to chew and taste meat and drink water and rinse their mouth with bottled drinking water kept at room temperature between each sample and pause for 20 seconds before tasting the next sample. Panelists were instructed to sensory evaluation using 5-point hedonic scale test procedure (1 = dislike extremely, 2 = dislike slightly, 3 = neither like nor dislike, 4 = like slightly, 5 = like extremely) of the American Meat Science Association (AMSA, 2016). Sensory properties were determined by 10 panelists (5 females and 5-males) from CVMA staff and students selected randomly to judge the portion of broiler chicken meat served for their evaluation. The whole sensory analysis was repeated twice.

3.7.9 Partial budget analysis

Partial budget analysis was conducted in order to evaluate the feasibility of adding phytobiotic additives to broiler diets at a certain level. In conducting the analysis, the variable cost of feed consumed by the chicks, the purchased price of the chicks and the selling price of the broiler meat were taken into account. Expenses, such as labor and vaccination cost were assumed to be the same for all treatment.

The partial budget analysis considerations were the cost of feeds and supply costs at the time of experiment used. The analysis involved calculation of the variable cost and profits of the experimental diet. Net return (NR) of the experimental group was calculated as the amount of money left when total variable costs (TVC) were subtracted from the total returns (TR) as follows: $NR = TR - TVC$ The change in net income (ΔNR) or Net return (ΔNR) was calculated as the difference between change in total return (ΔTR) and the change in total variable costs (ΔTVC) as follows $\Delta NR (= \Delta TR - \Delta TVC)$, where: NR: net return, TR: total return and TVC: total variable costs

$$NR = \text{Total return} - \text{Total variable cost}$$

The marginal rate of return (MRR) measures increases in net income (ΔNI) associated with each additional unit of expenditure (ΔTVC) and it will be calculated as $MRR = (\Delta NI / \Delta TVC)$

$$\text{Marginal rate of return} = \frac{\text{Net return}}{\text{Total Variable cost}}$$

3.8. Statistical Analysis

The data collected were analyzed using one way ANOVA procedures of Statistical Analysis Systems software (SPSS, 2015). Duncan's multiple range test was used to detect the differences among the treatment means and significance was considered at $p < 0.05$ (Duncan, 1955).

The model used was:

$$Y_{ij} = \mu + \alpha_i + \epsilon_{ij}$$

Where: Y_{ij} = the j th observation with treatment i

μ = overall mean

α_i = the i th treatment effect

ϵ_{ij} = the random error term normally and independently distributed $(0, \delta^2)$

4. RESULTS

4.1. Growth Performance

4.1.1 Feed intake

The effect of phytobiotic feed additives on the growth performance of chickens is summarized in Table 4. The result showed that there was no significant difference ($P>0.05$) in feed intake among all treatment groups during the starter phase. However, significant differences ($p<0.05$) for feed intake of broiler chickens were observed among treatments during grower, finisher and entire experiment. During the grower period, although T1 and T4 had lowest feed intake as compared to T2 and T3. Birds fed in T2 had the significantly ($p<0.05$) lower feed intake during the finisher and throughout the entire period. In this study, birds that were fed in T1 and T3 had the highest ($p<0.05$) feed consumption in the finisher phase and overall period respectively.

Table 6. The effect of phytobiotic additives in feed intake of broilers during the starter, grower, finisher and the entire period of the experiment

Feed Intake(g)/bird	T1	T2	T3	T4	P-value	Sign
Starter	261.5 ^{abc} ±7.67	263.8 ^{abc} ±4.04	269.6 ^{ab} ±2.53	271.8 ^a ±1.24	0.402	ns
Grower	1060.4 ^d ±4.27	1072.8 ^a ±1.58	1070.9 ^b ±0.37	1063.0 ^c ±1.54	0.18	**
Finisher	1716.1 ^a ±0.31	996.1 ^d ±1.47	1714.6 ^b ±0.54	1714.4 ^c ±1.04	0.000	**
Entire	3038.0 ^c ±6.78	2332.7 ^d ±5.04	3055.0 ^a ±2.79	3049.2 ^b ±1.97	0.000	**

a-d Means with different superscripts within the same column are significantly different

(P<0.05); T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant.

4.1.2 Body weight gain

Table 5 shows body weight gain, final body weight and average daily body weight gain during the starter, grower, finisher and the entire period. There was no significant difference ($P>0.05$) in the body weight between dietary treatments. Even though, there was no statistical difference there was numerical difference during the final body weight where T1 had a high finishing weight than the other treatments and T2 had the lowest final weight.

Table 7. The effects of phytobiotics additives on body weight or broiler chickens during the starter, grower, finisher and the entire period of the experiment.

BW(g/bird)	T1	T2	T3	T4	P-Value	Sig
IBW(g)	38.7±0.36	39.4±0.40	39.2±0.78	39.1410±0.40753	0.808	ns
BW gain(g)						ns
Starter	257.6±2.89	263.6±5.078	259±2.71	253.1±0.92	0.517	
Grower	976.7±37.24	961.7±41.06	972.5±11.58	977.0±21.81	0.981	ns
Finisher	1928.3±51.99	1912.5±30.41	1781.5±48.28	1887.5±29.19	0.149	ns
FBW(g)	2671.67±108.06	2556.7±57.76	2563.3±86.67	2563.3±54.34	0.704	ns
ADG(g)					0.569	ns
Starter	19.3±0.20	19.6±0.44	19.5±0.23	18.9±0.49		
Grower	37.7±1.58	37.3±1.58	37.6±0.50	37.5±0.80	0.996	ns
Finisher	58.3±1.35	56.7±0.66	55.0±1.70	56.5±1.05	0.379	ns
Overall	2632.9±107.79	2517.2±58.06	2524.1±87.38	2524.2±53.94	0.702	ns

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant. BW=body weight, ADG= average daily gain.

4.1.3 Feed conversion ratio

The results of feed conversion ratio is shown in Table 6 and no significant difference ($p>0.05$) was observed in FCR among the treatment groups during starter grower and finisher phase.

Table 8. The effect of phytobiotic additives on performance of broilers during the starter, grower, finisher and the entire period of the experiment

FCR	T1	T2	T3	T4	p-value	Sig
Starter	1.6±0.06	1.5±0.05	1.6±0.04	1.7±0.71	0.356	ns
Grower	2.0±0.08	2.0±0.08	2.0±0.03	2.0±0.44	0.962	ns

Finisher	2.1±0.45	2.1±0.02	2.2±0.07	2.2±0.00	0.352	ns
Overall	1.9±0.02	1.9±0.51	2.0±0.03	1.9±0.44	0.574	ns

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant

4.2. Mortality Rate

Table 7 indicates the mortality rate of broiler chickens during the experimental periods. Highly significant difference ($p<0.05$) was observed during the grower phase, where T1 had the lowest mortality rate while the highest mortality rate was determined for T4. However, no significant difference ($p>0.05$) was observed in the mortality during the starter, finisher and entire period.

Table 9. The mortality rate of broilers fed with phytobiotic additives during the starter, grower, finisher and the entire period of the experiment

	T1	T2	T3	T4	p-value	sig
Starter	2.7±2.56	2.6±2.56	5.1±2.56	7.7±4.44	0.627	ns
Grower	2.6±2.56	10.3±2.56	17.9±2.56	20.5±2.56	0.004	**
Finisher	25.6±18.49	10.3±2.56	15.4±4.44	15.4±4.44	0.738	ns
Overall	30.8±19.36	23.0±4.44	38.5±4.44	43.6±6.78	0.578	ns

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant

4.3 Carcass Characteristics

Carcass evaluation result is shown below in Table 8 Broiler's carcass weight or body cuts were not significantly different ($p>0.05$) among treatment groups fed with different phytobiotic additives.

Table 10. The effects of phytobiotics additives on Carcass yield of broiler chickens at the end of the experiment

Carcass weight	T1	T2	T3	T4	P-value	sign
Live weight(g)	2671.7±70.97	2556.7±70.70	2563.3±57.37	2563.3±84.09	0.622	ns
Carcass weigh(g)	1851.7±70.97	1736.7±70.70	1743.3±57.37	1748.3±85.14	0.634	ns
Carcass w %	69.2±0.80	67.8±0.84	67.87±0.71	67.97±1.14	0.658	ns
Wing(g)	188.5±5.43	180.0±6.42	169.0±7.64	180.3±8.69	0.314	ns
Wing%	10.2±0.16	10.3±0.13	9.7±0.38	10.2±0.04	0.149	ns
Breast(g)	571.5±22.06	534.8±23.74	530.0±18.18	549.0±28.40	0.602	ns
Breast%	30.8±0.09	30.7±0.32	30.4±0.50	31.4±0.49	0.364	ns
Drumstick(g)	210.5±8.40	199.3±9.56	209.8±12.91	196.5±8.61	0.679	ns
Drumstick %	11.3±0.025	11.4±0.14	12.0±0.39	11.2±0.32	0.231	ns
Thigh(g)	296.2±13.8	262.8±26.86	289.5±11.80	283.0±17.05	0.609	ns
Thigh%	15.9±0.24	15.97±0.29	16.6±0.30	16.1±0.28	0.402	ns
Bone(g)	586.7±23.22	547.0±16.47	549.8±15.73	541.3±27.44	0.441	ns
Bone%	31.7±0.11	31.5±0.44	31.5±0.34	31.0±0.55538	0.636	ns

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant

4.4 Cecal Microbial Count

Table 9 represents the effect of dietary garlic, thyme and their mixture on E.coli and lactobacillus count in the gut of broilers. The results showed that there was no significant ($p>0.05$) effect on the E.coli and lactobacilli count due to dietary supplementation of the feed additives.

Table 11. The effects of phytobiotic additives on gut microbial population of broilers at the end of the experiment

	T1	T2	T3	T4	p-value	Sig
E. coli count	7.3± 0.11	7.4± 0.34	7.3±0.17	7.4±0.13	0.977	ns
Lactobacilli count	7.7 ±0.19	7.6±0.93	7.7±0.09	7.6±0.17	0.903	ns

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet= SE standard error of mean, P= Probability, ns= not-significant.

4.5. Sensory Evaluation

The results of sensory evaluation of broiler meat is presented in Table 10. Among the treatments, T2 exhibited the highest favorability rating for its aroma, texture and overall appeal, surpassing the other groups by slight margin. While T1 ranked second in terms of acceptance due to its taste and flavor, T3 received the highest score for visual appeal.

Table 12. The effects of phytobiotic additives on meat Quality of broiler chickens

Treatment	T1	T2	T3
Appearance	30	50	60
Taste	60	40	40
Texture	30	60	50
Aroma	10	60	20
Acceptance	80	90	80

T1= Garlic, T2 = Thyme, T3 = Garlic and Thyme

4.6 Partial Budget Analysis

Table 11 provides a summary of the profitability of broilers that had been supplemented with two phytobiotic additives and their mixture. The T3 diet had the most expensive total variable cost, with T4 and T1 following close behind. The T2 diet had the lowest price in total cost which yielded the greatest net profit, with the T1, T4, and T3 following close behind. The diet containing a combination of garlic and thyme (T3) resulted in the lowest net profit from bird

consumption. Nevertheless, the marginal rate of T3 showed similarity to that of T4, control group which had the lowest marginal rate.

Table 13. Economic efficiency of inclusion of phytobiotic feed additives in broiler chickens

Description	T 1	T 2	T3	T 4
Cost				
A day-old chick cost(ETB)	80	80	80	80
Total feed cost(birr/bird)				
Starter	10.5	10.6	10.8	10.9
Grower	41.7	42.2	42.1	41.8
Finisher	66.9	38.8	66.8	66.8
Additive cost (ETB)	6.38	4.68	7.31	0
Fixed cost (ETB)				
<i>labor</i>	32.0	32.0	32.0	32.0
<i>vaccine</i>	5.4	5.4	5.4	5.4
Total variable cost (ETB)	242.9	213.8	244.6	237.1
Revenue				
Average carcass weight in (kg)	1.85	1.74	1.74	1.75
Price of carcass/kg (ETB)	210	210	210	210
Total return (ETB)	388.8	364.7	366.1	367.1
Net return	145.9	150.9	121.5	130.1
Marginal rate of return	0.60	0.70	0.50	0.55

T1=1% garlic, T2 = 1% thyme, T3 = 0.5%+0.5% of garlic and thyme, T4= control basal diet

5. DISCUSSION

The supplementation of herbal feed additives garlic and thyme didn't affect feed intake of birds during the starter phase, where as an opposite effect was observed in feed intake during the grower and the finisher phase, highly significant difference occurred, the control diet and 1% garlic supplementation had the highest feed intake during the grower and finisher phase respectively. overall feed intake was higher in combined treatment of garlic and thyme. While the lowest FI recorded was at 1% thyme supplementation. Birds had higher feed consumption at a combined treatment level of 0.5%+0.5%. In general, over the entire period lower feed consumption was from thyme treatments whereas higher feed consumption was from. This report agrees with Isa (2011) who states that the inclusion of mixture containing garlic and thyme leave powder resulted in a significant increase in broilers feed intake when compared to separately supplementing these additives. This result might be due to increased palatability of diets when mixture of garlic and thyme leaves are added to the diet. On the contrary Schutte *et al.* (1993) and Jansman *et al.* (1999) reported that feed intake of birds was not significant ($p>0.05$) affected by any of the additives garlic, thyme or their mixture among the supplemental groups.

Despite the anticipation that adding garlic and thyme leave powder to the broiler diet whether independently or combined would enhanced their growth performance, there was no statistically significant ($p>0.05$) impact observed on the broiler body weight gain, average daily BW gain or FCR between the dietary treatments. These findings from the current study contradicts several studies according to Ademola (2004) when birds were given garlic in their feed at different level of 5g/kg, 10g/kg and 15g/kg, their average weight and final live weight improved in comparison to the birds that were only fed basal diet. Ramiah *et al.* (2014) also reported similar result that birds given diets containing garlic at the level of 0.5% exhibited higher body weight compared to the control group. Another studies found that the addition of just 1% garlic powder to animal feed led to a reduction in FCR Raesi *et al.* (2010). Toghyami *et al.* (2010) reported that administering 5g/kg of thyme had a notable impact on the body weight and feed conversion rate of broiler. Similarly scholars Najafi and Toriki (2010) found that broilers fed with diet containing thyme showed significant improvement in body weight and feed conversion rate.

However a report by Tekeli *et al.* (2006) and Demir *et al.* (2003) found contradictory results, indicating thyme had no effect on broilers performance which is consistent with the present study but according to a report by Abid (2013), the use of both garlic and thyme in treatment groups resulted in the increase in body weight. However, our current research contradicts these findings and aligns with Demir *et al.* (2008) which was determined that the addition of garlic or thyme did not have any impact on the body weight or FCR.

During the experimental period, there was no significant difference in mortality rate during the starter period among groups however there was significant difference during the grower period there was a sign of morbidity, and mortality occurrence due to ascites and the criteria for diagnosis of ascites syndrome were water belly during the opening of dead birds . This finding aligns with Tona *et al.* (2005) that the peak of ascites related mortality occurs at the end of growing phase of birds. Highest mortality occurred in the control group then followed by combination group and 1 % thyme group while the lowest mortality rate was from the group of 1% garlic this finding is in alignment with Saifali *et al.* (2015) who concluded that garlic bulb with the inclusion level of 5g/kg decrease the incidence of ascites thereby reduce mortality without impairing broiler chicken performance. Furthermore, there was no significant difference ($p>0.05$) in mortality rate of the birds during starter finisher and the entire period however there was numerically higher dead birds in 1% garlic during the finisher phase the cause was due to the attack of a ferocious predator from the garlic group only, managed to kill majority of birds in a single night. A study by Meskerem (2017) and Reta (2009) concluded that the major constrain in urban areas of Bishoftu city was the losses of birds due to disease and predators which agrees with the present study.

The present study revealed that there was no difference in the carcass cuts among the various treatment groups when they were fed diet including garlic powder or dried thyme leaves independently or combined. These findings contradict with Raeesi *et al.* 2010, the study observed that adding 1% garlic powder to the diet resulted in higher thigh yield. Conversely the group supplemented with 3% garlic powder had the lowest thigh yield. In another study the breast yield of groups who were given 1% garlic powder was notably higher compared to the other groups Raeesi *et al.* 2010. The present research aligns with Hosseini and Mohammed

(2011) report they stated that adding 1.5% garlic to the diet of broilers did not impact other characteristics of the carcass, and there was no significant variance in the carcass yield of the broilers between the treatment groups fed combined or independently. Likewise, Habiban *et al.* (2010) also reported that the incorporation of 20 g/kg of garlic in broiler diets resulted in a reduction in abdominal fat, but did not impact on the characteristics of carcass.

The result of the current study investigated revealed no statistically significant ($p>0.005$) difference in the count of *E.coli* bacteria present in the cecum of broiler chickens with respect to the dietary treatment. which agrees with a report from Phyto *et al.* 2017 that supplementing garlic, thyme or combined didn't not affect the *E.coli* population in broiler chickens. These reports are in contrast to Ramiah *et al.* (2014) who observed that adding dietary garlic powder (0.5%) resulted in a significant decrease in the presence of *E.coli* bacteria and significantly elevated lactobacillus when compared to the control group. Also, there was no statistically significant difference in the colony forming unit of lactobacilli count among treatment groups these findings are in contradiction with Phyto *et al.* (2017) who determined that thyme powder exhibited a favorable impact on the population of lactobacillus bacteria. These findings correspond with the previous work of Rahimib *et al.* 2011, who confirmed that the count of lactobacilli in the thyme group demonstrated significant increase when compared to the other groups. The findings of the current investigation are consistent with the report from Tschirich (2000) who suggested that cavacrol, in thyme has a stimulating effect on lactobacillus proliferations. The variance in these reports could be attributed to the procedures involved from the collection of samples to the microbial count.

The study found that the addition of thyme powder at 1% level resulted in reduced feed cost from broiler chickens in the finisher phase, based on the mean feed cost per bird. As a result, the thyme treatment exhibited the greatest net return. While the combination of garlic and thyme resulted in the lowest NR due to high feed intake and lower carcass weight at the last period. On the other hand findings by Hassan and Awad (2017) suggest that feeding birds with 8g/kg of thyme resulted in the highest feed cost and lowest net revenue as opposed to the other group. According to the present study the garlic treatment group resulted in a greater total return, primarily due to increase carcass weight. Pourali *et al.* (2010) suggested that broiler growth may be enhanced by garlic allicin compound which could support intestinal flora activity. The present

study also revealed that garlic is followed by control group, combination then thyme group for higher total return respectively. Nonetheless there was no significant difference in total return a month these three groups. The thyme group recorded the highest marginal rate of 0.60 closely followed by garlic at 0.70 while the control group had a rate of 0.55 then combination group had 0.50 marginal return. the current study found that the combined treatment had a lower marginal return rate compared to the control group. The thyme group reduced feed intake but had similar body weight gain to the other groups, leading to a higher net return. meanwhile there was no significant difference in body weight among the treatment groups, garlic group achieved the highest body weight resulting in the thyme and garlic in broiler diets may be profitable than using their combination or solely feeding basal diet.

The study conducted on evaluating the sensory aspect of broiler breast meat from the three different treatments, found that the treatment group that were supplemented thyme had the highest level of acceptance as well as preference for aroma and texture. This study supports a previous result by Mohamed *et al.* (2022) who found that adding thyme essential oil at level of 0.2ml/kg improved sensory and quality parameters of broiler meat and had no negative impact on either the birds or human safety. Also, the present study revealed that the taste for garlic supplementation was favored the most amongst all the other groups In comparison to the other groups. These findings were in agreement with previous study which concluded that chickens fed with a garlic supplemented diet had superior texture and flavor in their thigh meat (Kim, 2009). The treatment groups that received both garlic and thyme had the highest score for appearance. Hence the broiler diet incorporated 1%thyme demonstrated the most notable outcome in terms of sensory evaluation.

6. CONCLUSION AND RECOMMENDATIONS

6.1. Conclusion

Despite significant variation in feed consumption among the treatments, the study demonstrated that the overall performance of broiler remained mostly unaffected by the feeding of either garlic powder or dried thyme leaves alone or in combination. One can draw a conclusion that neither of the mixtures enhanced the birds productivity regarding their weight gain and feed conversion ratio and there was no impact on the carcass characteristics, also the introduction of these additives independently or combined did not have any impact on the microbial population of E.coli and lactobacillus bacteria in GIT of broilers. However broilers fed with thyme and garlic yielded a higher economic benefit compared to the control group or their combination. These variations observed could be explained by factors including the plant species, harvesting time, storage time and place as well as preparation method of the additives. It can be concluded that thyme and garlic can have a positive impact on the profitability in raising broiler chickens. Furthermore, the research showed that addition of thyme leaves powder in broiler feed had the higher appeal in terms of aroma, texture and overall acceptance while garlic had the highest for its flavor and taste which seems to have a positive effect for the consumption of broiler meat. Therefore, it could be concluded that thyme and garlic can be used as feed additive in broiler diet and can offer economic benefits and enhance the quality of broiler meat as a result individuals may become more inclined to consume chicken meat.

6.2. Recommendations

- Inclusion of thyme and garlic at 1% in the diet of broiler chickens could possibly enhance their growth making it a viable option as a natural growth promoting additive which in turn can lead to improved economic appraisal
- Detailed study is necessary to investigate the impact of thyme and garlic on microbial population of broiler chickens during all phases

- Further studies are required to fully understand the mechanism of action of phytobiotic compounds, determine the optimal level of inclusion and preparation method to enhance gut health and attain higher growth performance in broiler chickens.

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8. APPENDICES

Appendix I

Daily Feed Offered and Refused Record Sheet

Breed: Cobb500

Day of Entrance:

Experiment: Phytobiotic feed additive Garlic, thyme and combination

Treatment group: _____ Replication no: _____

Date	Day	Week	No of Birds Alive	Type of Additive Given	Feed Offered(g)		Feed Refused(g)	Mortality	Remark
					Morning	Afternoon			

Appendix II

Weekly body weight measurement record

Breed: Cobb500

Day of Entrance:

Experiment: Phytobiotic feed additive Garlic, thyme and combination

Treatment group: _____ Replication no: _____

Date	Week	No of weighed Birds	Body weight(g)	Total weight(g)	Average weight(g)	Remark

Appendix III

Sensory evaluation form

Meat type: Breast meat

Name: _____ Occupation: _____

Directions: Check one rating for each of the following: Appearance, Taste/flavor, Texture/Consistency, Aroma/smell and Over all acceptability

Group _____

Rating scale	Appearance	Taste/flavor	Texture/Consistency	Aroma/smell	Over all acceptability

Appendix table 1. The effect of phytobiotic additives in feed intake of broilers during the starter, grower, finisher and the entire period of the experiment

Feed intake Parameter	Source of variation	Sum of Squares	Df	Mean Square	F	Sig.
Starter	Between Groups	206.941	3	68.980	1.106	.402
	Within Groups	498.974	8	62.372		
	Total	705.915	11			
Grower	Between Groups	323.709	3	107.903	6.190	.018
	Within Groups	139.463	8	17.433		
	Total	463.172	11			
Finisher	Between Groups	1162866.907	3	387622.302	142793.546	.000
	Within Groups	21.717	8	2.715		
	Total	1162888.624	11			
Overall	Between Groups	1149770.050	3	383256.683	6154.967	.000
	Within Groups	498.143	8	2.268		
	Total	1150268.193	11			

Appendix Table 2. The effects of phytobiotics additives on body weight, ADG and FCR on broiler chickens during the starter, grower, finisher and the entire period of the experiment

BW parameter	Source of variation	Sum of Squares	df	Mean Square	F	Sig.
Starter	Between Groups	165.692	3	55.231	.823	.517
	Within Groups	536.740	8	67.092		
	Total	702.431	11			
Grower	Between Groups	463.932	3	154.644	.056	.981
	Within Groups	22096.094	8	2762.012		
	Total	22560.026	11			
Finisher	Between Groups	36001.562	3	12000.521	2.349	.149
	Within Groups	40866.667	8	5108.333		
	Total	76868.229	11			
Intial Bw	Between Groups	.779	3	.260	.324	.808
	Within Groups	6.416	8	.802		
	Total	7.195	11			
Final Bw	Between Groups	27589.583	3	9196.528	.481	.704
	Within Groups	152866.667	8	19108.333		
	Total	180456.250	11			

ABWG	SOURCE OF VARIANCE	Sum of Squares	df	Mean Square	F	Sig.
Starter	Between Groups	.864	3	.288	.716	.569
	Within Groups	3.216	8	.402		
	Total	4.079	11			
grower	Between Groups	.240	3	.080	.018	.996
	Within Groups	35.272	8	4.409		
	Total	35.512	11			
finisher	Between Groups	16.608	3	5.536	1.172	.379
	Within Groups	37.776	8	4.722		
	Total	54.384	11			
overall	Between Groups	27859.542	3	9286.514	.485	.702
	Within Groups	153209.442	8	19151.180		
	Total	181068.984	11			

FCR	Source of variation	Sum of Squares	df	Mean Square	F	Sig.
starter	Between Groups	.034	3	.011	1.244	.356
	Within Groups	.072	8	.009		
	Total	.106	11			
grower	Between Groups	.003	3	.001	.093	.962
	Within Groups	.093	8	.012		
	Total	.096	11			
finisher	Between Groups	.025	3	.008	1.257	.352
	Within Groups	.054	8	.007		
	Total	.079	11			
overall	Between Groups	.009	3	.003	.707	.574
	Within Groups	.035	8	.004		
	Total	.044	11			

Appendix table 3. The mortality rate of broilers fed with phytobiotic additives during the starter, grower, finisher and the entire period of the experiment

Mortality	Source of variation	Sum of Squares	df	Mean Square	F	Sig.
GROWER	Between Groups	591.716	3	197.239	10.000	.004
	Within Groups	157.791	8	19.724		
	Total	749.507	11			
STARTER	Between Groups	54.241	3	18.080	.611	.627
	Within Groups	236.686	8	29.586		
	Total	290.927	11			
FINISHER	Between Groups	374.753	3	124.918	.429	.738
	Within Groups	2327.416	8	290.927		
	Total	2702.170	11			
Entire	Between Groups	724.852	3	241.617	.700	.578
	Within Groups	2761.341	8	345.168		
	Total	3486.193	11			

Appendix table 4. The effects of phytobiotics additives on Carcass yield of broiler chickens at the end of the experiment

Carcass parameters	Source of variation	Sum of Squares	df	Mean Square	F	Sig.
Live weight (g)	Between Groups	55179.167	3	18393.056	.601	.622
	Within Groups	611883.333	20	30594.167		
	Total	667062.500	23			
Cacrcass weight(g)	Between Groups	53766.667	3	17922.222	.581	.634
	Within Groups	617233.333	20	30861.667		
	Total	671000.000	23			
Wing (g)	Between Groups	1153.125	3	384.375	1.252	.317
	Within Groups	6138.833	20	306.942		
	Total	7291.958	23			
Breast (g)	Between Groups	6237.000	3	2079.000	.634	.602
	Within Groups	65614.333	20	3280.717		
	Total	71851.333	23			
Drumstick(g)	Between Groups	925.792	3	308.597	.511	.679
	Within Groups	12085.167	20	604.258		
	Total	13010.958	23			
Thigh(g)	Between Groups	3733.458	3	1244.486	.623	.609
	Within Groups	39971.167	20	1998.558		
	Total	43704.625	23			
Bone (g)	Between Groups	7646.458	3	2548.819	.937	.441
	Within Groups	54381.500	20	2719.075		
	Total	62027.958	23			
% Cacrcass weight(g)	Between Groups	7.683	3	2.561	.543	.658
	Within Groups	94.279	20	4.714		
	Total	101.962	23			
% Wing (g)	Between Groups	1.649	3	.550	1.982	.149
	Within Groups	5.548	20	.277		
	Total	7.197	23			
%Breast (g)	Between Groups	3.055	3	1.018	1.122	.364
	Within Groups	18.152	20	.908		
	Total	21.206	23			
% Drumstick(g)	Between Groups	1.936	3	.645	1.557	.231
	Within Groups	8.287	20	.414		
	Total	10.223	23			
%Thigh(g)	Between Groups	1.455	3	.485	1.027	.402
	Within Groups	9.442	20	.472		

	Total	10.896	23			
% Bone (g)	Between Groups	1.661	3	.554	.579	.636
	Within Groups	19.125	20	.956		
	Total	20.786	23			

Appendix table 5. The effects of phytobiotic additives on gut microbial population of broilers at the end of the experiment

Microbial parameter	Source of variance	Sum of Squares	Df	Mean Square	F	Sig.
E.coli count	Between Groups	.042	3	.014	.066	.977
	Within Groups	3.863	18	.215		
	Total	3.905	21			
Lactobacillus count	Between Groups	.082	3	.027	.187	.903
	Within Groups	2.194	15	.146		
	Total	2.276	18			

