T.R

ERCIYES UNIVERSITY

GRADUATE SCHOOL OF NATURAL AND APPLIED SCIENCES

DEPARTMENT OF AGRICULTURAL SCIENCE AND TECHNOLOGIES

COMPARISON OF ZINC NANOPARTICLES OBTAINED FROM PLANT EXTRACT WITH DIFFERENT ZINC SOURCES AND THEIR EFFECTS ON EGG PRODUCTION AND QUALITY FEATURES IN LAYER QUAIL

Prepared by

Saif Shakir ALJASSAN

Supervisor

Assoc. Prof. Dr. İsmail ÜLGER

Master's Thesis

June 2022

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COMPLIANCE WITH SCIENTIFIC ETHICS

I confirm that all data used in this work was obtained in compliance with academic and ethical guidelines. At the same time, as required by these rules and norms, I declare that all materials and results not central to this work have been fully attributed and referenced.

Saif Shakir ALJASSAN

İmza

“Comparison of Zinc Nanoparticles Obtained from Plant Extract with Different Zinc Sources and Their Effects on Egg Yield and Quality Traits in Layer Quail” The master's thesis, named Erciyes University, was prepared in accordance with the Graduate Thesis Proposal and Thesis Writing Directive.

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Saif Shakir ALJASSAN

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BİTKİ ÖZÜTÜ İLE ELDE EDİLEN ÇİNKO NANOPARTİKÜLLERİN FARKLI ÇİNKO KAYNAKLARI İLE KARŞILAŞTIRILMASI VE YUMURTACI BILDIRCINLARDA YUMURTA VERİMİ VE KALİTE ÖZELLİKLERİ ÜZERİNE ETKİLERİ

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Yüksek Lisans Tezi, Haziran 2022

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ÖZET

Bu araştırmada, yumurtacı bıldırcın karma yemlerine 3 ve 6 mg/kg düzeylerinde inorganik, organik ve bitki özütlü nano çinko ilave edilmiş ve 56 gün süreyle beslenmiştir. Rasyonlarına ilave edilen farklı Çinko kaynaklarının canlı ağırlık, canlı ağırlık değişimi, yem tüketimi, yemden yararlanma oranı, yumurta ağırlığı, yumurta kabuk ağırlığı, yumurta eni, yumurta boyu, şekil indeksi, sarı ağırlığı, yumurta sarısı rengi (L*a*b*) gibi özellikleri üzerine etkileri incelenmiştir. Çalışmada 175 adet dişi bıldırcın kullanılmıştır. Yapılan çalışmada gruplar arasında bitiş canlı ağırlık farkları istatistiksel olarak önem arz etmemiştir ($P>0.05$). Yapılan çalışmada rasyonlarına farklı oranda farklı Zn kaynaklarının ilavesi deneme süresince ortalama yem tüketimlerini yükselttiği tespit edilmiştir. Farklı Zn kaynaklarının rasyona ilavesi ile yapılan çalışmada yumurta boyu en yüksek rasyona 3 mg/kg organik Zn ilave edilen grupta tespit edilmiştir. Deneme gruplarının rasyonlarına farklı Zn kaynaklarının ilavesi deneme süresince yumurta ağırlığı ve yumurta eni 6 mg/kg inorganik Zn katkılı yem tüketen grup ta en yüksek tespit edilmiştir ($P<0.05$). Yapılan çalışma gruplarının rasyonlarına farklı Zn kaynaklarının ilavesi deneme süresince yumurta sarı ağırlığı üzerine etkisi istatistiksel olarak tespit edilmemiştir.

Sonuç olarak bitki özütü ile elde edilen nano çinkonun rasyona ilavesinin yumurtacı bıldırcınlarda kullanılabileceği belirlenmiştir.

Anahtar Kelimeler: Bıldırcın, Çinko, Organik, İnorganik, Nano, Performans, Yumurta verimi

COMPARISON OF ZINC NANOPARTICLES OBTAINED FROM PLANT EXTRACT WITH DIFFERENT ZINC SOURCES AND THEIR EFFECTS ON EGG PRODUCTION AND QUALITY FEATURES IN LAYER QUAIL

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Erciyes University, Graduate School of Natural and Applied Sciences

Master's Thesis, June 2022

Supervisor: Assoc. Prof. Dr. İsmail ÜLGER

ABSTRACT

In this thesis, 3 and 6 mg/kg inorganic, organic and plant extract nano zinc were added to layer quail compound feed and animals were fed with these diets for 56 days. Live weight, live weight gain, feed consumption, feed conversion ratio, egg weight, eggshell weight, egg width, egg length, shape index, yolk weight and egg yolk color (L*, a* and b*) parameters were investigated. Totally, 175 female quail were used in present experiments. Effects of experimental treatments on final live weights were not found to be significant ($P>0.05$). It was determined that the addition of different Zn sources at different rates to quail rations increased the average feed consumption throughout the experiments. In present experiments conducted with the addition of different Zn sources to the ration, the greatest egg length was determined in 3 mg/kg organic Zn-supplemented group. The greatest egg weight and egg widths were determined in 6 mg/kg inorganic Zn- supplemented groups ($P<0.05$). The effects of adding different Zn sources to the diets on egg yolk weight of treatment groups were not found to be significant.

It was concluded that nano-zinc particles obtained from plant extracts could safely be used in layer quail diets.

Keywords: Quail, Zinc, Organic, Inorganic, Nano, Performance, Egg Production

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RESUME ..... **Hata! Yer işareti tanımlanmamış.**

ABBREVIATIONS

Zn	: Zinc
ZnO	: Zinc Oxide
Nano	: Nanoparticle
L*	: Egg yolk brightness value
a*	: Egg yolk redness value
b*	: Egg yolk yellowness value
Ag	: Silver
Cu	: Copper
Fe	: Iron
Mn	: Manganese
A	: Carbon
CaCO3	: calcium carbonate
P	: Phosphorus
MgO	:Magnesium Oxide
KCl	:Potassium chloride
ZnSO4	:Zinc Sulphate
CuSO4	:Copper Sulphate
Co	:Cobalt
CO3	:Carbon monoxide
Se	: Selenium
Nm	: Nanometer
mg	: Milligram
kg	: Kilogram
DNA	: Deoxyribo nucleic acid
ZnSO4	: Zinc sulfate

FC	: Feed consumption
LW	: Live weight
ME	: Metabolic Energy
FCV	: Feed coefficient value
SEM	: Mean standard error
P	: Level of significance
LWG	: Body weight gains
FCR	: Feed conversion ratio

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INTRODUCTION

Animal products are a source of critical nutrients in human nutrition around the community and help to maintain a healthy diet. In order to boost the production of animal protein, which is essential for human nutrition, both existing sources and the hunt for protein sources obtained from new animal products are being pursued. Alternative poultry farming has recently been exploded worldwide. Alternative poultry consumption represents 22% of global poultry consumption (Roenigk, 1999).

Quail is an alternative avian breed that is produced more quickly and efficiently than the other poultry species (Şeker, 2003).

In recent years, there has been an increase in the production of quail meat and eggs as an additional source of protein, as well as its use for experimentation and research purposes. Importing and breeding quail from the other countries for scientific and industrial purposes in Turkey occurred 50 years after Japan began extensive quail breeding. Quail meat and eggs, which were once utilized in animal experiments in colleges, drew consumer interest that led to the formation of quail businesses. Few researchers were initially interested in quail, both as animal experiments and as production animals, because of their low feed utilization, short generation intervals and the other factors.

In this sense, the need to minimize vitamin inadequacies in poultry mixed feeds has prompted numerous studies. Minerals and chemicals are at the center of these deficits. Minerals are essential in poultry for both their vital activities and to increase output. Mineral shortages or antagonistic interactions of minerals can cause major health concerns (Ecevit and Olgun, 2019; Ülger et al., 2020; Ülger and Fidan., 2021). Mineral compounds must therefore be present in sufficient quantities in the feed. The propensity to use organic minerals rather than inorganic elements is biologically more beneficial and efficient in poultry nutrition. Minerals can be used at reduced levels in poultry feeding without affecting animal performance, which ultimately result in savings in

production costs. Furthermore, by lowering the level of minerals emitted in poultry feces, environmental pollution is also reduced. Amino acid complexes, proteinates, zinc, copper and manganese chelates are the most commonly used organic forms in poultry nutrition. In majority of previous research, organic minerals had a favorable influence on inorganic minerals in poultry. Because of their higher absorption and reduced excretion with feces, they require less addition into the meal. (Zafar and Fatima, 2018). In a study, researchers compared the effects of inorganic and organically bonded essential minerals on mineral buildup in tissues and fecal excretion in broiler breeders. It was found that replacing feed inorganic essential minerals with organic essential minerals increased mineral deposit in tissue and reduced feces mineral excretion in broilers (Wang et al., 2019a). As is well known, using organic mineral resources rather than inorganic minerals has been popular in recent years in order to maximize the value of minerals added to the poultry feeds. Nano-minerals, which offer an alternative both to organic and inorganic natural minerals and have a high usability, are currently being studied as feed additives (Tatlı et al., 2019). It was indicated that nano-minerals promoted mineral deposit in tissues and decreased fecal extraction of minerals in broiler breeders (Wang et al., 2019a). In order to actively profit from of the mineral added to poultry feeds, the utilization of organic natural resources rather than inorganic minerals has gained popularity in recent years.

Nano-minerals have a particle size range of 1-100 nm. They can withstand extreme temperatures and pressures (Stoimenov et al., 2002). They are more effective at lower dosages than bigger sized ones because they are easily absorbed by gastrointestinal tract and utilized in the animal system (Feng et al., 2009). Because of larger surface area, they react more with the other organic and inorganic compounds (Zaboli et al., 2009). Nano-sized minerals also have minimal adverse effects on human cells (Reddy et al., 2007), which is an added advantage. There may be greater dispersal of nano-minerals across the small intestine and into the blood, brain, lung, heart, kidney, spleen, liver, intestine and stomach (Hillyer et al., 2001). The size, shape and composition of nano-metals are primarily dictated by their crystal structure and morphology (Dickson and Lyon, 2000). The particle size of nano-minerals has a significant impact on their functional activities, such as chemical, catalytic and biological impacts (Lewis and Klibanov, 2005; Rosi and Mirkin, 2005).

In this thesis, effects of zinc nano-particles obtained from plant extracts with different zinc sources on live weight, live weight change, feed consumption, feed conversion ratio, egg weight, eggshell weight, egg width, egg length, shape index, yolk weight, egg yolk color parameters (L*, a* and b*) of laying quail were investigated.

CHAPTER 1

1.1.GENERAL INFORMATION AND LITERATURE STUDY

1.1.1.Mınerals

Minerals are required for all living things to function properly. Vitamins and minerals are essential elements that animals require to survive. According to the demand levels of animals, minerals are split into macro and micro (trace) minerals, thus trace minerals such as zinc, copper, iron, iodine and manganese are minerals which are required less than 100 mg/kg in the diet and are represented as mg/kg or ppm in the ration. In the body, trace minerals provide structural, enzyme activator, antioxidant, and immune system support activities. Anemia, reduced egg production, poor and aberrant shell formation, weakened immunity and weak feathering are all the symptoms of a lack of essential minerals in the ration. Minerals are components required for optimal muscle and nerve activities, as well as optimum body growth. They're also necessary components of cells, hormones and bodily enzymes. To fully fulfill the mineral requirement, care must be given. In general, there are issues with production and animal health. Although the effects are not immediate, when animals are malnourished for a long time, their mineral requirements grow. Mineral deficiencies can quickly affect the performance if the clinical indications of inadequacy arise. Most minerals can be hazardous to animals at high concentrations (Taurus and Filik, 2011). Copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) are important trace elements in poultry nutrition because they are involved in various physiological and metabolic processes. In enzyme systems, trace minerals mostly serve as a catalyst or as a component of the enzyme (Yazgan, 1991; Bao et al., 2007). Trace minerals in inorganic form provided to chicken feeds first must dissolve into ionic form before being absorbed by the digestive system. Mineral elements in the ionic state, on the other hand, can mix with feed components and form non-absorbable compounds. To guarantee that the animal needs are met, some trace minerals are added into poultry feeds in excess of the estimated demand. Excess

trace minerals in the ration not only raise the cost of the ration, but also cause environmental issues by increasing the amount of minerals expelled inside the feces (Close, 1998; Cao et al., 2002; Rutz et al., 2004; Yenice et al., 2015; Singh et al., 2015). In fact, poultry manure has 660 percent more Zn and 560 percent more Cu than plant requirements (Dozier et al., 2003). Inorganic compounds make up all of the minerals utilized in nutrition. Minerals are found in solids and crystalline forms, and although they are less significant than proteins and energy, they play an important role in living organisms. Mineral compounds must present in sufficient quantities in the feed in order for animal species to be healthy and active. These are compounds that can't be made by ordinary chemical processes. About 96 percent of the living organism is made up of organic components including oxygen, hydrogen, nitrogen and carbon. Anions and cations account for 3.5 percent of the total, with the remaining minerals accounting for the remainder. Mineral compounds that cannot be generated in the animal body, unlike many nutrients, play a significant role in the organism (Ülger and Küçük, 2011; Ülger and Fidan, 2021).

1.1.2.Organic Minerals

Organic minerals are metal ions that create a complex system with amino acids, peptides, proteins or polysaccharides (Çoşkun, 2006). Organic minerals include a carbon (C) atom in their structure. Carbon (C) atoms do not exist in inorganic mineral sources. Organic minerals could be produced through reaction of metal salts with organic molecules or by biologically supplying soluble metal salts into yeast culture medium (Çoşkun, 2006). Mineral supplements have been created with a number of methods over the years, but the majority have come from inorganic sources. Inorganic materials are either natural (found in the form of minerals on the ground or refined) or chemically created. $CaCO_3$ (calcium carbonate), P (phosphorus), MgO (magnesium oxide), KCl (potassium chloride), $ZnSO_4$ (zinc sulphate), $CuSO_4$ (copper sulphate), Co (cobalt), CO_3 (carbon monoxide) and the other elements are among them. All of these substances are either salts or inorganic metal complexes (Miles and Henry, 2000; Ahola et al., 2004). Organic trace minerals are said to be given to rations less frequently than inorganic essential minerals due to their better absorption or bioavailability, resulting in less contamination. It has been found that when organic minerals account for 20% of inorganic minerals, a comparable degree of performance is attained (Nocek, 2006).

Instead of digesting typical mineral ions in the small intestine, organic minerals are utilized inside the organism through the digestion of peptides and amino acids. These mineral forms are significantly more readily transported and absorbed in the colon, in addition to having a high bioavailability. They are biochemically protected against undesirable interactions which may occur with particular ration components, which might lower their absorption rate, because they are more stable (Close, 1998). Organic minerals are more stable than simple ions and do not react as readily. They have no interactions with vitamins or the other ions, are active in small levels and do not react with vitamins in the digestive system or premixes. Inorganic minerals in feed and the digestive system interact with ligands to generate low-solubility salts or a variety of non-absorbable inorganic complexes. It's unclear if chelate or complex organic mineral supplements, like inorganic free ions, breakdown into free ions at low pH and reach the intestinal mucosa's glycocalyx for absorption. Except for large molecule proteinates, which are used in metabolism or stored in various tissues with the same organic forms, organic forms can be absorbed by amino acids or peptide transport pathways, depending on the conditions and durability of the digestive tract. Proteinases are hydrolyzed and trace elements in their structures are converted into amino acids without becoming free ions or because of peptides (Ahola et al., 2004; Spears, 2003; Miles, 2000; Boland, 2003).

1.1.3.Inorganic Minerals

When comparing the productivity between organic and inorganic minerals in poultry nutrition, it is more physiologically friendly and efficient to employ organic minerals rather than inorganic minerals. Organic minerals also lower the expense of feeding by lowering the dosage rate without compromising their performance. Furthermore, due to the poor elimination of mineral elements in poultry feces, environmental contamination is decreased. Amino acid complexes, proteinates, zinc, copper and manganese chelates are the most often used and assessed organic forms in poultry. Both broilers and layers showed favorable impacts of organic minerals upon inorganic minerals in the majority of investigations (Zafar and Fatima, 2018; Ülger and Fidan 2021). When the effects of various levels of ration trace elements acquired from organic or inorganic sources on broiler growth parameters, carcass characteristics, meat quality and fecal mineral excretion were investigated, it was observed that reduced trace element supplementation

had little negative effect on growth performance, carcass yield and meat quality. Although it had a negative impact, it was shown to lower fecal mineral excretion. It's also been demonstrated to play a little effect on trace metal supply, performance and excretion, with just a minor boost to animal productivity if trace element availability is improved (Zhu et al., 2019). Wang et al. (2019b) examining the effects of replacing inorganic trace minerals with organic trace minerals on the production performance, blood profiles and antioxidant status of broiler breeders and indicated that substituting the commercial inorganic mineral level in broiler breeder diets with LOTM and OTM diets had a negative effect on production and procreative performance, tissue antioxidant enzyme activity and blood profile. In a different research, it was reported that in majority of the tissues examined, substituting diet inorganic essential minerals with low-dose complex glycinate minerals boosted apparent bioavailability of Fe, Mn and Zn without decreasing mineral retention rates (Sun et al., 2020). It was discovered that substituting LOTM and OTM diets with commercial inorganic minerals in broiler breeder diets had no significant impact on productivity and reproduction rate, tissue antioxidant enzyme activity or blood profile (Wang et al., 2019b).

1.1.4.Nano Minerals

The merger of biological sciences, materials science and computer technology is referred to as nanotechnology. This technology is made to work on an atomic level. Although nanotechnology has aided the growth of the electronics, energy, environment and health sectors, livestock development remains limited (Rajendran, 2013).

Nanotechnology is a new growing science that employs materials with unique characteristics and applications in sizes ranging from 1 to 100 nanometers. Stoimenov et al. (2002) found that nanoparticles are stable at high temperatures and pressures. Nano-sized ZnO particles are more rapidly absorbed by the gastrointestinal system and utilized in vivo than larger-sized ZnO, making it more effective at lower dosages [Fenget al., 2002]. It reacts efficiently with the other organic and inorganic chemicals in vivo because of its larger surface area (Zaboli et al., 2013). An extra benefit is that nanosized ZnO has negligible deleterious effect on human cells (Reddy et al., 2007). Because nano minerals enter via the small intestine, absorption will be improved, and blood, brain, lung, heart, kidney, spleen, liver and other organs will benefit.

The characteristics of nanoscale material differ from those of base material and solitary atom (Albert et al., 2006). Richard Feynman, an American physicist, proposed the concept of nanotechnology in 1956 (Rajendran et al., 2013). Nanotechnology is used in a wide range of fields, including physics, chemistry, engineering, medicine and more lately, livestock and agriculture. Nanometer-sized materials are nothing new. Living systems rely on many nanoscale things such as proteins, DNA and enzymes, hence nano molecules are widespread in nature. Nanoparticles can also be found in the natural environment (Albrecht et al., 2006). It is one of the most cutting-edge technologies for creating materials and components with enhanced structure, texture and performance at the molecular level (Usama, 2012).

Nano-sized mineral particles can be generated either by the "bottom-up technique" based on wet chemical reduction (growth and nucleation of mineral atoms) of mineral salts, or by the "top-down method" requiring mechanical grinding of the heap (Rajendran et al., 2013).

Inorganic, organic, emulsion, dispersion and clay nanoparticles are classified as inorganic, organic, emulsion, dispersion and nano clay (Gopi et al., 2017). In poultry, silver (Ahmadi & Kurdestani, 2010; Felehgari et al., 2013; Vadalasetty et al., 2018), zinc oxide (Zhao et al., 2014; Fathi, 2016), copper (Wang et al., 2011; Joshua et al., 2016; Ognik et al., 2016), iron (Zhou and Wang, 2011) and selenium (Saki and Abbasinezhad, 2014) were employed. The potential of nanotechnology in poultry industry has yet to be completely realized due to a lack of relevant understanding (Usama, 2012).

Minerals, especially trace minerals, have a poor bioavailability in mammalian bodies. Mineral antagonism in the stomach can be reduced by trace element nanoparticles, resulting in enhanced absorption and less excretion to environment. They may also improve avian immunological responses and digestion efficiency, leading to increased feed conversion efficiency (Gopi et al., 2017). For laying and breeding flocks, metals in the diet also lower the frequency of early embryonic mortality and cystic ovaries (Rajendran et al., 2013). The poultry industry's nanotechnology potential has yet to be completely realized (Usama 2012).

1.2.PREVIOUS STUDIES

1.2.1.Studies On The Use Of Nano Minerals In Animal Nutrition

Nanotechnology can help researchers pinpoint the exact position of a trace mineral or nutritional elements in a tissue, cell or cellular compartment. Hypersensitive nutrient and metabolite detection can help to expand understanding of nutrition and their biomolecular interactions in many tissues (Thulasi et al., 2013). Such an amazing technology has the potential to improve bioavailability and the other nutritional factors.

In recent years, there has been a tremendous interest in investigating the possible use and usefulness of nanoparticles in animal production. Major uses of these nanomaterials in animal nutrition as supplements, medicines and probiotics are thoroughly investigated. Because of their improved bioavailability and less antagonism behavior in the intestine, nanotechnology is mostly used in poultry nutrition in the form of nano-minerals (Gopi et al., 2017). Broiler growth was boosted with the use of nano-Zn (Mohammadi et al., 2015b). Similarly, broiler diets containing silver (nano-Ag) and selenium (nano-Se) nanoparticles have been found to minimize oxidative stress due to their significant antioxidant properties (Ahmadi and Kurdestani, 2010; Aparna and Karunakaran, 2016). Furthermore, copper nanoparticles (nano-Cu) increased not only the growth performance but also immunological response of poultry (Wang et al., 2011). The inclusion of nano-Fe has also been linked to improved growth and hatchability of poultry species (Saki and Abbasinezhad, 2014; Sizova et al., 2015).

Zhao et al. (2014) conducted a research in which they added nano ZnO to broiler diets. The reference group's ration had 60 mg/kg of inorganic ZnO, whereas the other three experimental groups' rations contained 20, 60 and 100 mg/kg of nano ZnO, respectively. The researchers found that the group receiving 100 mg/kg nano ZnO gained less live weight than the others. The feed conversion ratios were not altered in any way. According to the researchers, the amount of 100 mg/kg nano Zn was greater than it should have been, therefore the group fed with the ration containing 20 mg/kg nano ZnO had the best performance and this level would be adequate to offer antioxidant capacity, which was another criterion studied.

In a research using broilers, Sahoo et al. (2014a) found that there were substantial variations in biological availability and immune system impacts among inorganic ($ZnSO_4$), organic (Zn-methionine) and nano (ZnO) Zn sources. According to the study's

findings, both inorganic minerals were employed in the same ratios (15 ppm), the organic source yielded better results for investigated criteria and micro ZnO was as efficient as organic fertilizers even when just 0.06 ppm was utilized. The same team conducted another investigation (Sahoo et al., 2014b) and discovered that equal feeds in broiler chicks produced equivalent blood values.

In laying hens, Tsai et al. (2016) looked at the effects of inorganic, organic and nano Zn sources on YV and eggshell quality, immunological and serum parameters. The first of two efforts at this goal were to estimate the quantity of Zn excreted in the feces in a research that involved 20 white layer chickens aged 68 weeks that lasted for five days. The animals were given a 40 mg/kg Zn-base feed as well as four alternative meals including inorganic ZnO to achieve a Zn concentration of 60 mg/kg, organic Zn methionine and nano Zn. The quantity of Zn maintained in the body was greater in the groups fed with nano and organic Zn sources as compared to the inorganic Zn and control groups. In the second study, 80 laying hens of the same line and age were utilized and the eggshell thickness (CC) was found to be larger in the groups fed with nano and organic Zn sources than in the control and inorganic Zn-containing groups. In terms of immune system characteristics, there was no difference between the groups, although serum growth hormone and alkaline phosphatase enzyme levels were greater in the nano and organic Zn-containing groups than in the other groups. In comparison to inorganic Zn sources, researchers discovered that nano ZnO enhanced small intestine absorption. In comparison to other groups, serum growth hormone and alkaline phosphatase enzyme levels were greater in nano and organic Zn-containing groups.

Olgun and Yıldz (2017) performed a 42-week experiment on 144 laying hens. A total of 12 distinct meals were employed in the experiment, each with three extra Zn levels (0, 75, and 100 mg/kg) and 4 different Zn sources ($ZnSO_4$, ZnO, Zn-glycine and micro ZnO). Egg weight, egg mass and feed conversion ratio were all lower in the Zn-glycine group than in the ZnSO4 group. The eggshell thickness of the group given the nano ZnO ration was found to be lower than the other groups. As a consequence, the researchers concluded that nano Zn had a detrimental effect on KK, that nano Zn sources should not be used in laying hen feeds and that inorganic and organic Zn sources may safely be employed.

Misra et al. (2014) investigated the effects of Nano Zn on layer chick development and serum biochemical parameters. The animals were separated into groups with baseline diet (without extra Zn, comprising 30 mg/kg Zn), ZnO at the needed level and Nano ZnO at the level of 1/100, 1/500, and 1/1000 of the basal ration in the research, in which 300 chicks of daily age were used. Researchers employed five different rations in all. While the effects of a treatment in diets containing (YT) and YDK were minor, the group fed with a ration containing micro ZnO at a rate of 1/500 of the basal diet had a higher growth rate of chicks.

Mohammadi et al. (2015) investigated the effects of different Zn sources on performance of broilers. A total of 6 different rations including 80 mg/kg of $ZnSO_4$, ZnMet, nano $ZnSO_4$, nano ZnMet and Nano Zn max (Nano chelate form) sources were used . It was revealed that the group using nano $ZnSO_4$ had a lower weight (CA) and PT at the end of the trial (42 days). It was also found that in the nano $ZnSO_4$ and control groups, carcass production was lower and abdominal fat was higher. The therapies used did not appear to make a meaningful impact on the SAI. The use of nano ZnMet and Zn max sources at a level of 80 mg/kg level in broilers had a beneficial influence on animal performance.

Bahakaim et al. (2014) studied the effects of organic and inorganic Zn sources, as well as their various levels (0, 50, 100, and 150 mg/kg), on laying hen performance, egg quality, and serum parameters. Feed conversion ratio and egg quality traits were better in the group fed with the ration containing 100 mg/kg organic Zn, while serum Zn levels were greater in the groups supplemented with organic Zn.

CHAPTER 2

2.MATERIAL AND METHOD

2.1.Animal Material

This research was carried out in quail breeding unit of Erciyes University, Faculty of Agriculture, Department of Animal Science. The animals to be used in the experiment were obtained from the quail unit of the faculty. In the study, 175 8-week-old quail were used as animal material. Before starting the study, the animals were subjected to a 14-day pre-adaptation feeding.

2.2.Creation of treatment groups

At the beginning of the experiment, the quail were weighed and assigned to the treatment groups such that live weight variations were small. Each cage held 5 quail and each treatment group was made up of 5 replications, resulting in a total of 5 cages and 25 layer quail (Table 1). In terms of sensible animal nutrition, it's critical to understand the illnesses that might result from mineral and vitamin deficiencies or excesses. Several illnesses can develop as a result of mineral excess or particularly deficiency and these can result in severe economic losses in quail breeding. Both excess and deficiencies of the dosages administered to the mixed diet were determined in this study and the toxic impact was explored. The nutritional contents of the diets were within the feeding guidelines. Zinc was also supplemented into the mixed meals of the experimental groups. The amount of zinc in the concentrate feed was determined. All of the feeds were made isonitrogenously and calorically. The treatment groups were given 3 and 6 mg/kg of inorganic, organic and plant-derived nano-zinc in their mixed meal for 56 days. Nano-zinc was created by combining inorganic and organic zinc sources with extracts from commercial firms.

Table 2.1. Creation of treatment groups

Groups	Feed Content	Number	Total
1	The control group consumed the feed without Zn supplementation.	5x5	25
2	The group consuming 3 mg/kg inorganic Zn-supplemented feed	5x5	25
3	The group consuming 6 mg/kg of inorganic Zn-supplemented feed	5x5	25
4	The group consuming 3 mg/kg organic Zn-supplemented feed	5x5	25
5	The group consuming 6 mg/kg organic Zn-supplemented feed	5x5	25
6	The group consuming 3 mg/kg plant extract nano Zn-supplemented feed	5x5	25
7	The group consuming 6 mg/kg plant extract nano Zn-supplemented feed	5x5	25
Total			175

2.3.Supplementation into animal feeds

Mineral nanoparticles, as an alternative to organic and chemical minerals now utilized in animal nutrition, will be less poisonous, organic and more useable by animals, resulting in more cost-effective, healthy and productive animals, as well as animal products for human consumption. Products that are healthful and organic are available. Plant extract-derived zinc nanoparticles were manufactured in powder form and supplemented into animal diets.

2.4.Feed analysis and preparation of mixed feeds

Dry matter, crude ash, crude oil, crude protein and crude cellulose of the compound feeds were analyzed. Sugar analysis was conducted according to Zoll's rule. Starch analysis was conducted with the use of polarization method in Feeds and Animal Nutrition laboratories of Erciyes University, Faculty of Agriculture, Department of Animal Science (Akyıldız, 1984).

Metabolic Energy (ME) values were calculated according to the WPSA method as fallows (Vogt 1984).

ME (kcal/kg) = (3.69 x % crude protein + 8.18 % crude fat + 3.99 x % starch + 3.17 x % Sugar) x 10

To establish the treatment combinations in Table 1, a basal ration (Table 2) was prepared and zinc levels to be supplemented were calculated. Then the remaining zinc quantities supplied from different zinc sources were supplemented into animal diets. The ration was prepared in the ERU Scientific and Application Farm's feed facility. Feed minerals were measured and ground in appropriate amounts before being fed into the mixer. Anticoccidial, DL-methionine, L-lysine, limestone, dicalcium phosphate and vitamin E were delivered to the mixer (Table 3) as a premix and then zinc supplies were added at appropriate proportions. Feed intake and egg quality were monitored daily after 14-day adaption period to determine feed consumption, egg production and feed efficiency, as well as zinc levels.

Table 1.2. The composition of the concentrated feed to be used in the experiment

Feed Ingredients,	**% Composition**
Corn	52.21
Soybean Meal	40.79
Corn Gluten	3.09
Vegetable oil	one
Marble dust	1.20
Dicalcium Phosphate	0.78
Lysine	0.05
Methionine	0.13
Vitamin-Mineral Premix*	0.50
Salt	**0.25**

*Vitamin A 12 000 000 IU, Vitamin D3 2 400 000 IU, Vitamin E 20 000 mg, Vitamin K3 4000 mg, Vitamin B1 3 000 mg, Vitamin B2 7 000 mg, niacin 25 in every 2.5 kilograms of vitamin-mineral mix 000 mg, calcium D pantothenate 10 000 mg, vitamin B6 5 000 mg, vitamin B12 15 mg, D biotin 75 mg, folic acid 1 000 mg, choline chloride

125 000 mg, manganese 100 000 mg, iron 60 000 mg, zinc 60 000 mg, copper 5 000 mg, cobalt 200 mg, iodine 1 000 mg.

Table 2.3. Raw nutrient content of the concentrated feed to be used in the experiment

Calculated Nutrients**	**Contents**
Metabolic Energy, Kcal/kg	2910
Dry Matter, %	89.90
Crude Protein, %	24.00
Calcium, %	0.80
Phosphorus, %	0.32
Sodium, %	0.12
Lysine, %	1.30
Methionine + Cysteine, %	0.89

*: Vitamin-Mineral mixture amounts per 2.5 kg: vitamin A, 12,500,000 IU; vitamin D3, 3,000,000 IU; vitamin E, 20,000mg; vitamin K3, 3,000 mg; vitamin B1, 2,500 mg; vitamin B2, 7,000 mg; vitamin B6, 5,000mg; vitamin B12, 20mg; niacin, 20,000mg; Cal-D-Pan, 15,000mg; folic acid, 1,000 mg; biotin, 20mg; vitamin C, 50,000 mg; cholincholoride, 300,000 mg; manganese, 80,000 mg; iron, 70,000 mg; zinc, 50,000 mg; copper, 6,250 mg; iodine, 1,250mg; cobalt, 200mg; selenium, 150mg; canthaxanthin, 0mg; apo-carotenoicacidest., 0 mg; lasolosidsodium, 90,000 mg. **: Values are calculated from NRC (1994) tables, excluding dry matter.

2.5. Determination Of Live Weight And Weight Gains

Animal live weights were determined by weighing the animals individually at the beginning, 1st, 2nd, 3rd and 4th week of the experiments. A precise balance (±0.01 g) was used to weigh the animals. The difference between consecutive weighing was taken as live weight gain.

2.6. Determination Of Feed Consumption And Feed Utilization Rate

The quantity of feed left in the feeders in the first, second, third, fourth, fifth and sixth weeks of the research was deducted from the amount of feed supplied for each subgroup at the beginning of that week, yielding total amount of feed used for each subgroup in a week. Each subgroup's weekly feed consumption was divided by the number of available animals in its own subgroup and weekly feed consumption was determined

using the averages of the subgroups and groups. The feed efficiency ratios were obtained through multiplying that averaged amounts of food used by animal in two subsequent weighing by the average live weight gain in these two subsequent weighing.

2.7.Determination of egg internal and external quality characteristics

Each treatment group had 75 eggs gathered every 14 days to determine egg weight and shell weight. The egg width and length were used to determine the shape index. The yolk weight was measured by weighing the egg yolk extracted from the egg white by breaking the eggs on a balance (±0.1 g). A Minolta CR-400 colorimeter (Minolta Co. Japan) was used to quantify egg yolk color L* (brightness), a* (redness) and b* (yellowness).

2.8. Statistical Analysis

Statistical analyses were conducted in accordance with randomized plots design with six levels and ten replications. The animals were placed in cages according to the randomization plots experimental design. The analyses were performed with the use of SAS statistical software. Duncan's test was used to assess the differences between the groups. The significance level was set as 5% ($P<0.05$).

CHAPTER 3

RESULTS

3.1. Live Weight And Egg Production Parameters

Live weight (LW) values of the experimental groups are given in Table 3.1. The highest live weight at the end of the experiments (235.96 g) was obtained from 3 mg/kg nano Zn-supplemented group and lowest (234.80 g) from 6 mg/kg inorganic Zn-supplemented group. The differences in final live weights of the treatment groups were not found to be significant ($P>0.0999$). The greatest live weight gain (LWG) (32.36 g) was obtained from 3 mg/kg nano Zn-supplemented group and the lowest (29.20 g) from 6 mg/kg inorganic Zn-supplemented group. The differences in live weight gains of the treatment groups were found to be significant ($P<0.021$). The highest egg yield (24.04 eggs/day/25 quail) was detected in 6 mg/kg nano Zn-supplemented group and the lowest egg yield (21.07 eggs/day/25 quail) was detected in the control group ($P<0.001$). The highest egg yield percentage (96.15%) was detected in 6 mg/kg nano Zn-supplemented group and the lowest (84.29%) was detected in the control group without any Zn supplementations ($P<0.001$).

Table 3.1. Average live weight and egg yield values of the treatment groups

Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
Initial Live Weight, g	204.92	203.76	204.04	205.24	205.32	203,60	206.40	0.659	0.921
Final Live Weight, g	235.40	235.16	234.80	235.28	235.32	235.96	235.60	0.688	>0.999
Live Weight Gain, g	30.48^{abc}	31.40^{ab}	30.76^{abc}	30.04^{bc}	30.00^{bc}	32.36^{a}	29.20^{c}	0.251	0.021
Egg Yield, eggs/day/25 quail	21.07^{b}	23.62^{a}	23.43^{a}	23.71^{a}	23.43^{a}	24.03^{a}	24.04^{a}	0.189	<0.001
Egg Yield, %	84.29^{b}	94.49^{a}	93.72^{a}	94.85^{a}	93.71^{a}	96.13^{a}	96.15^{a}	0.754	<0.001

The means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.2. Feed Consumption Parameters and Feed Conversion Ratios (FCR)

The weekly and overall feed consumptions of the treatment groups are given in Table 3.2. As reported in the table, there were significant differences in weekly feed consumptions of the treatment groups (P>0.05). In the first week of the experiments, the highest feed consumption was determined as 39.48 g in 6 mg/kg nano Zn-supplemented group, while the lowest feed consumption was determined as 38.61 g in 3 mg/kg inorganic Zn-supplemented group (P<0.001). In the 2nd week of the experiments, the highest feed consumption was found to be 39.49 g in 6 mg/kg nano Zn and 6 mg/kg organic Zn-supplemented groups, while the lowest feed consumption was identified as 38.02 g in the control group (P<0.001). In the 3rd week of the experiment, the highest feed consumption was identified as 39.86 g in 6 mg/kg nano Zn-supplemented group, while the lowest feed consumption was determined to be 38.32 g in the control group (P<0.001).

In the 4th week of the experiment, the highest feed consumption was determined as 39.89 g in 6 mg/kg nano Zn-supplemented group, while the lowest feed consumption was determined as 38.34 g in the control group (P<0.001). In the 5th and 6th weeks of the study, the highest feed consumption was determined as 39.93g and 40.03g in 6 mg/kg nano Zn-supplemented groups, while the lowest feed consumption was determined to be 38.36 g and 38.44 g in the control groups (P<0.001).

The average weekly and overall feed conversion ratios (FCR) of the treatment groups are given in Table 3.2. There were significant differences in weekly feed conversion ratios of the treatment groups (P>0.05). In the first week of the study, the highest feed conversion ratio (FCR) was determined to be 3.36 g/g in the control group, while the lowest FCR was identified as 3.03 g/g in 6 mg/kg inorganic Zn-supplemented group (P=0.010).

In the 2nd week of the experiment, the highest FCR value was identified as 3.41 g/g in 6 mg/kg nano Zn-supplemented group and, while the lowest value was determined to be 3.02 g/g in 6 mg/kg inorganic Zn-supplemented group (P<0.001). In the 3rd week of the study, the highest FCR value was identified as 3.40 g/g in 6 mg/kg nano Zn-supplemented group, while the lowest value was determined to be 3.10 g/g in 6 mg/kg inorganic Zn-supplemented group (P=0.016). In the 4th week of the study, the highest FCR value was identified as 3.46 g/g in 6 mg/kg nano Zn-supplemented group, while

the lowest value was determined to be 3.15 g/g in 6 mg/kg inorganic Zn-supplemented group (P=0.012).

In the 5th week of the experiment, the highest FCR value was identified as 3.35 g/g in 6 mg/kg nano Zn-supplemented group, while the lowest value was determined to be 3.05 g/g in 6 mg/kg inorganic Zn-supplemented group (P=0.007). In the 6th week of the experiment, the highest FCR value was identified as 3.46 g/g in 6 mg/kg nano Zn-supplemented group, while the lowest value was determined to be 3.08 g/g in 6 mg/kg inorganic Zn-supplemented group (P<0.001).

Table 3.2. Average weekly and overall feed consumption (FC) and feed conversion ratios (FCR) of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**FC, g**	38.81^{b}	38.61^{b}	38.75^{b}	38.89^{b}	39.33^{a}	38.94^{b}	39.48^{a}	0.056	<0.001
	FCR, g/d	3.36^{a}	3.24^{ab}	3.03^{c}	$3.10b^{c}$	3.33^{a}	3.25^{ab}	3.28^{ab}	0.028	0.010
2nd week	**FC, g**	38.02^{c}	38.72^{b}	39.24^{a}	39.25^{a}	39.49^{a}	39.31^{a}	39.49^{a}	0.076	<0.001
	FCR, g/d	3.11^{b}	3.11^{b}	3.02^{b}	3.03^{b}	3.15^{b}	3.20^{b}	3.41^{a}	0.025	<0.001
3rd week	**FC, g**	38.32^{b}	38.75^{b}	39.85^{a}	39.67^{a}	39.67^{a}	39.74^{a}	39.86^{a}	0.099	<0.001
	FCR, g/d	3.24^{abc}	3.15^{bc}	3.12^{c}	3.14^{bc}	3.10^{c}	3.34^{ab}	3.40^{a}	0.028	0.016
4th week	**FC, g**	38.34^{c}	39.09^{b}	39.80^{a}	39.74^{a}	39.75^{a}	39.83^{a}	39.89^{a}	0.096	<0.001
	FCR, g/d	3.18^{bc}	3.39^{ab}	3.15^{c}	3.17^{bc}	3.18^{bc}	3.39^{ab}	3.46^{a}	0.031	0.012
5th week	**FC, g**	38.36^{c}	39.12^{b}	39.83^{a}	39.77^{a}	39.76^{a}	39.82^{a}	39.93^{a}	0.096	<0.001
	FCR, g/d	3.07^{c}	3.18^{abc}	3.05^{c}	3.13^{bc}	3.12^{bc}	3.29^{ab}	3.35^{a}	0.025	0.007
6th week	**FC, g**	38.44^{c}	39.23^{b}	39.84^{ab}	39.80^{ab}	39.82^{ab}	39.89^{a}	40.03^{a}	0.095	<0.001
	FCR, g/d	$3.15b^{c}$	3.26^{bc}	3.08^{c}	3.13^{bc}	3.09^{c}	3.30^{ab}	3.46^{a}	0.027	<0.001

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.3.Egg Weight (g)

The average egg weights of the treatment groups are given in Table 3.3. In the first week of the study, the lowest egg weight was detected as 11.67 g in the control group, while the highest egg weight was identified as 12.93 g in 6 mg/kg inorganic Zn-supplemented group (P=0.102). In the second week of the study, the lowest egg weight was determined to be 11.62 g in 6 mg/kg nano Zn-supplemented group, while the highest egg weight was detected as 13.10 g in 6 mg/kg inorganic Zn-supplemented group (P<0.001). In the 3rd week of the study, the lowest egg weight was determined to be 11.78 g in 6 mg/kg nano Zn-supplemented group, while the highest egg weight was detected as 12.93 g in 6 mg/kg organic Zn-supplemented group (P=0.010). In the 4th week of the study, the lowest egg weight was determined to be 11.60 g in 6 mg/kg nano Zn-supplemented group, while the highest egg weight was detected as 12.72 g in 6 mg/kg inorganic Zn-supplemented group (P<0.007). In the 5th week of the experiment, the lowest egg weight was determined to be 11.92 g in 6 mg/kg nano Zn-supplemented group, while the highest egg weight was detected as 13.15 g in 6 mg/kg inorganic Zn-supplemented group (P=0.011). In the 6th week of the experiment, the lowest egg weight was determined to be 11.63 g in 6 mg/kg nano Zn-supplemented group, while the highest egg weight was detected as 13.01 g in 6 mg/kg inorganic Zn-supplemented group (P=0.001).

Table 3.3**.** Average egg weights of the treatment groups (g)

Period	**Parameter**	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Egg Weight, g**	11.67^{c}	11.98^{bc}	12.9^{3a}	12.55^{ab}	11.84^{bc}	11.99^{bc}	12.08^{bc}	0.102	0.012
2nd week	**Egg Weight, g**	12.26^{bc}	12.54^{ab}	13.10^{a}	12.97^{ab}	12.58^{ab}	12.30^{bc}	11.62^{c}	0.099	<0.001
3rd week	**Egg Weight, g**	11.89^{bc}	12.35^{abc}	12.84^{a}	12.69^{ab}	12.93^{a}	11.95^{bc}	11.78^{c}	0.111	0.010
4th week	**Egg Weight, g**	12.14^{abc}	11.62^{c}	12.72^{a}	12.56^{ab}	12.58^{ab}	11.82^{bc}	11.60^{c}	0.108	0.007
5th week	**Egg Weight, g**	12.52^{abc}	12.39^{bc}	13.15^{a}	12.75^{ab}	12.77^{ab}	12.17^{bc}	11.92^{c}	0.096	0.011
6th week	**Egg Weight, g**	12.24^{bc}	12.08^{bc}	13.01^{a}	12.72^{ab}	12.95^{a}	12.12^{bc}	11.63^{c}	0.097	<0.001

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.4. Egg Shell weight (g)

The average eggshell weights (g) of the treatment groups are given in Table 3.4. Significant differences were seen in eggshell weights of the experimental groups in the 1st, 2nd, 4th, 5th and 6th weeks (P>0.05). In the first week of the study, the highest shell weight was identified as 1.17 g in 6 mg/kg inorganic Zn and 3 mg/kg organic Zn-supplemented groups, while the lowest eggshell weight was found to be 1.04 g in 6 mg/kg nano Zn-supplemented group (P<0.001). In the 2nd week of the experiment, the highest shell weight was identified as 1.19 g in 3 mg/kg nano Zn-supplemented group, while the lowest eggshell weight was found to be 1.03 g in 6 mg/kg nano Zn-supplemented group (P=0.002).

In the 3rd week of the study, the highest shell weight was identified as 1.18 g in 3 mg/kg organic Zn-supplemented group, while the lowest eggshell weight was found to be 1.09 g in the control, 6 mg/kg inorganic Zn and 6 mg/kg nano Zn-supplemented groups (P=0.147). In the 4th week of the study, the highest shell weight was identified as 1.17 g in 6 mg/kg nano Zn-supplemented group, while the lowest eggshell weight was found to be 1.04 g in 3 mg/kg inorganic Zn-supplemented group (P=0.002). In the 5th week of the experiment, the highest shell weight was determined as 1.18 g in 3 and 6 mg/kg organic Zn-supplemented groups, while the lowest eggshell weight was determined as 1.10 g in the control and 6 mg/kg nano Zn-supplemented groups (P=0.035). In the 6th week of the experiment, the highest eggshell weight was found to be 1.21 g in 3 mg/kg organic Zn-supplemented group, while the lowest eggshell weight was identified as 1.01 g in 6 mg/kg nano Zn-supplemented group (P<0.001).

Table 3.4. Average eggshell weights of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Eggshell Weight, g**	1.05^{b}	1.06^{b}	1.17^{a}	1.17^{a}	1.13^{a}	1.15^{a}	1.04^{b}	0.010	<0.001
2nd week	**Eggshell Weight, g**	1.10^{ab}	1.11^{ab}	1.18^{a}	1.18^{a}	1.12^{a}	1.19^{a}	1.03^{b}	0.012	0.002
3rd week	**Eggshell Weight, g**	1.09^{ab}	1.09^{ab}	1.15^{ab}	1.18^{a}	1.15^{ab}	1.13^{ab}	1.09^{b}	0.011	0.147
4th week	**Eggshell Weight, g**	1.09^{bc}	1.04^{c}	1.13^{ab}	1.15^{ab}	1.17^{a}	1.13^{ab}	1.09^{bc}	0.009	0.002
5th week	**Eggshell Weight, g**	1.10^{ab}	1.12^{ab}	1.16^{ab}	1.18^{ab}	1.18^{a}	1.14^{ab}	1.10^{b}	0.009	0.035
6th week	**Eggshell Weight, g**	1.13^{bc}	1.10^{c}	1.17^{abc}	1.21^{a}	1.17^{ab}	1.16^{abc}	1.01^{d}	0.009	<0.001

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.5. Egg Width (mm)

The average egg widths (mm) of the treatment groups are given in Table 3.5. There were significant differences in weekly widths of the treatment groups (P>0.05). In the first week of the study, the highest egg width was detected as 26.38 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 25.41 mm in 6 mg/kg nano Zn-supplemented group (P=0.015). In the 2nd week of the experiment, the highest egg width was detected as 26.15 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 25.41 mm in 6 mg/kg nano Zn-supplemented group (P=0.018). In the 3rd week of the study, the highest egg width was detected as 26.40 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 25.60 mm in 6 mg/kg nano Zn-supplemented group (P=0.013).

In the 4th week of the study, the highest egg width was detected as 26.48 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 24.97 mm in 6 mg/kg nano Zn-supplemented group (P=0.001). In the 5th week of the study, the highest egg width was detected as 26.50 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 25.44 mm in 6 mg/kg nano Zn-supplemented group (P=0.011). In the 6th week of the study, the highest egg width was detected as 26.70 mm in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg width was detected as 25.14 mm in 6 mg/kg nano Zn-supplemented group (P=0.001).

Table 3.5. Mean egg width (mm) of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Egg Width, mm**	25.66^{bc}	25.85^{abc}	26.38^{a}	26.05^{ab}	25.58^{bc}	25.56^{bc}	25.41^{c}	0.081	0.015
2nd week	**Egg Width, mm**	25.75^{ab}	25.86^{ab}	26.15^{a}	26.03^{a}	25.88^{ab}	25.48^{b}	25.41^{b}	0.066	0.018
3rd week	**Egg Width, mm**	25.65^{bc}	26.16^{ab}	26.40^{a}	26.17^{ab}	$25,99^{abc}$	25.78^{bc}	25.60^{c}	0.071	0.013
4th week	**Egg Width, mm**	25.80^{bc}	25.55^{bc}	26.48^{a}	25.74^{bc}	25.94^{b}	25.32^{cd}	24.97^{d}	0.085	<0.001
5th week	**Egg Width, mm**	25.97^{abc}	25.54^{c}	26.50^{a}	25.94^{abc}	26.23^{ab}	25.77^{bc}	25.44^{c}	0.088	0.011
6th week	**Egg Width, mm**	25.75^{c}	26.07^{bc}	26.70^{a}	26.17^{abc}	26.44^{ab}	25.83^{c}	25.14^{d}	0.089	<0.001

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.6.Egg Length (mm)

The average egg lengths (mm) of the treatment groups are given in Table 3.6. The highest egg length (mm) was observed in 3 mg/kg organic Zn-supplemented group and the lowest egg length (mm) was detected in the control group in the 1st and 3rd weeks of the experiments. In the second week of the experiment, the lowest egg length (mm) was found to be 32.57 g in 6 mg/kg nano Zn-supplemented group ($P<0.001$). In the 4th and 5th weeks of the experiment, the lowest egg length (mm) was detected in 3 mg/kg inorganic Zn-supplemented group. In the 6th week of the study, the lowest egg length (mm) was detected in 6 mg/kg nano Zn-supplemented group ($P=0.002$).

Table 3.6. Mean egg length of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Egg length, mm**	32.14^{c}	32.44^{bc}	33.93^{a}	34.12^{a}	32.89^{bc}	33.24^{ab}	32.84^{bc}	0.138	<0.001
2nd week	**Egg length, mm**	33.26^{bc}	33.87^{ab}	34.21^{ab}	34.60^{a}	33.56^{b}	33.75^{ab}	32.57^{c}	0.133	<0.001
3rd week	**Egg length, mm**	33.10^{b}	33.49^{ab}	33.90^{ab}	34.31^{a}	33.94^{ab}	33.30^{ab}	33.48^{ab}	0.135	0.123
Th week	**Egg length, mm**	33.50^{ab}	32.93^{b}	33.53^{ab}	34.47^{a}	33.88^{ab}	33.25^{ab}	33.39^{ab}	0.138	0.083
5th week	**Egg length, mm**	33.72^{ab}	33.67^{b}	34.42^{ab}	34.94^{a}	34.26^{ab}	33.70^{ab}	34.01^{ab}	0.134	0.094
6th week	**Egg length, mm**	33.24^{b}	33.15^{b}	34.30^{a}	34.66^{a}	34.34^{a}	33.78^{ab}	33.12^{b}	0.135	0.002

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.7.Shape index

The average shape index values the treatment groups are given in Table 3.7. The lowest shape index was detected in 3 mg/kg organic Zn-supplemented group. In the 1st and 5th weeks of the experiment, the highest shape index was detected in the control group with 79.85 and 77.05, respectively, while in the 2nd week of the study, the greatest shape index was determined as 78.08 feeds in 6 mg/kg nano Zn-supplemented group.

In the 3rd and 6th weeks of the experiment, the highest shape index was determined as 78.23 and 78.69, respectively in 3 mg/kg inorganic Zn-supplemented group, while in the 4th week of the study, the greatest shape index was determined to be 79.02 in 6 mg/kg inorganic Zn-supplemented group.

Table 3.7. Average shape index of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Shape Index**	79.85^{a}	79.75^{a}	77.81^{b}	76.36^{b}	77.82^{b}	76.92^{b}	77.38^{b}	0.244	<0.001
2nd week	**Shape Index**	77.46^{ab}	76.45^{abc}	76.51^{abc}	75.26^{c}	77.16^{abc}	75.53^{bc}	78.08^{a}	0.258	0.032
3rd week	**Shape Index**	77.55	78.23	77.88	76.30	76.62	77.47	76.56	0.273	0.405
4th week	**Shape Index**	77.04^{b}	77.72^{ab}	79.02^{a}	74.70^{c}	76.60^{bc}	76.18^{bc}	74.91^{c}	0.283	<0.001
5th week	**Shape Index**	77.05	76.00	77.03	74.29	76.61	76.50	74.94	0.334	0.185
6th week	**Shape Index**	77.50^{ab}	78.69^{a}	77.85^{ab}	75.55^{b}	77.03^{ab}	76.55^{ab}	76.07^{ab}	0.298	0.074

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.8.Yolk Weight (g)

Average egg yolk weights (g) the experimental treatments are provided in Table 3.8. In the first week of the study, the highest egg yolk weight was detected as 3.81 g in 3 mg/kg of inorganic Zn-supplemented group, while the lowest yolk weight was detected as 3.43 g in 3 mg/kg organic Zn-supplemented group (P=0.863). In the 2nd week of the experiment, the highest egg yolk weight was detected as 3.96 g in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg yolk weight was detected as 3.56 g in 3 mg/kg nano Zn-supplemented group (P=0.339). In the 3rd week of the study, the highest egg yolk weight was detected as 4.00 g in 3 mg/kg of inorganic Zn-supplemented group, while the lowest egg yolk weight was detected as 3.56 g in 6 mg/kg inorganic Zn-supplemented group (P=0.794). In the 4th week of the study, the highest egg yolk weight was detected as 3.83 g in 3 mg/kg of organic Zn-supplemented group, while the lowest egg yolk weight was detected as 3.32 in 3 mg/kg nano Zn-supplemented group (P=0.229). In the 5th week of the study, the highest egg yolk weight was detected as 4.06 g in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg yolk weight was detected as 3.88 in 3 mg/kg organic Zn-supplemented group (P=0.777). In the 6th week of the study, the highest egg yolk weight was detected as 3.96 g in 6 mg/kg of inorganic Zn-supplemented group, while the lowest egg yolk weight was detected as 3.55 in 3 mg/kg nano Zn-supplemented group (P=0.200)

Table 3.8. Average egg yolk weights of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	**Yolk Weight, g**	3.67	3.81	3,54	3.43	3.63	3.50	3.80	0.083	0.863
2nd week	**Yolk Weight, g**	3.73	3.98	3.96	3.95	3.85	3,56	3.70	0.056	0.339
3rd week	**Yolk Weight, g**	3.75	4.00	3,56	3.65	3.79	3.86	3.80	0.074	0.794
4th week	**Yolk Weight, g**	3.78^{ab}	3.62^{ab}	3.81^{ab}	3.83^{a}	3.78^{ab}	3.32^{b}	3.62^{ab}	0.058	0.229
5th week	**Yolk Weight, g**	3.97	3.95	4.06	3.88	4.02	3.93	3.85	0.037	0.777
6th week	**Yolk Weight, g**	3,58	3.70	3.96	3.80	3.97	3,55	3.67	0.054	0.200

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

3.9. Egg Yolk Color (L* a* b*)

Egg yolk color (L*, a* and b*) parameters of the treatment groups are given in Table 3.9. It was determined that different zinc sources added to the ration did not affect the egg yolk color L* (brightness) values of layer quail during the experiment. When the mean egg yolk L* value of the groups was evaluated in all periods, except for the 1st week during the experiment, significant differences were not found ($P>0.05$).

Egg yolk color a* (Red-green) of layer quail supplemented with different Zn sources was significantly different at the 2nd and 6th weeks ($P<0.05$). It was not significant in the other weeks of the study ($P>0.05$).

It was determined that the egg yolk color b* (yellow-blue) parameters of the treatment groups were not influenced from different zinc sources added to the rations in all weeks, except for the 1st week ($P>0.05$).

Table 3.9. Average egg yolk color values of the treatment groups

Period	Parameter	Control group	3 mg/kg inorganic Zn supplemented group	6 mg/kg inorganic Zn supplemented group	3 mg/kg organic Zn supplemented group	6 mg/kg organic Zn supplemented group	3 mg/kg nano Zn supplemented group	6 mg/kg nano Zn supplemented group	SEM	P
1st week	L*	69.17^{a}	67.99^{ab}	52.83^{c}	59.76^{abc}	66.48^{abc}	54.09^{bc}	67.24^{abc}	1,847	0.057
	a*	8.46^{a}	8.58^{a}	7.41^{ab}	6.88^{ab}	8.30^{ab}	6.01^{b}	8.87^{a}	0.296	0.088
	b*	63.91^{ab}	64.36^{ab}	51.50^{bc}	57.23^{abc}	64.58^{ab}	47.05^{c}	67.26^{a}	1,930	0.029
2nd week	L*	64.02	57.49	62.29	60.54	59.47	57.82	59.45	1,061	0.662
	a*	7.66^{a}	6.92^{a}	7.98^{a}	7.40^{a}	7.33^{a}	6.99^{a}	5.18^{b}	0.227	0.027
	b*	60.20	53.77	57.48	58.17	56.33	56.16	54.16	1,003	0.648
3rd week	L*	64.31	68.86	60.77	58.93	68.04	67.62	67.76	1,264	0.210
	a*	6.38	6.64	5.72	5.35	6.98	6.05	6.84	0.198	0.239
	b*	62.65^{ab}	63.83^{ab}	58.17^{ab}	56.66^{b}	66.99^{a}	66.02^{ab}	64.75^{ab}	1,193	0.158
4th week	L*	65.93	65.07	66.64	66.45	67.21	61.03	64.91	0.949	0.681
	a*	6.26^{ab}	5.99^{ab}	6.30^{ab}	6.17^{ab}	6.73^{ab}	5.65^{b}	7.15^{a}	0.169	0.305
	b*	65.67	63.62	64.87	65.16	64.06	59.82	61.71	0.915	0.622
5th week	L*	65.37	64.92	66.02	65.01	64.97	64.91	64.06	0.511	0.982
	a*	6.81	6.10	6.58	5.98	6.15	5.91	6.82	0.165	0.583
	b*	61.30^{b}	63.51^{ab}	61.40^{ab}	62.46^{ab}	64.46^{ab}	64.49^{ab}	65.54^{a}	0.471	0.103
6th week	L*	65.35	64.89	64.19	64.26	65.14	58.16	64.57	0.917	0.378
	a*	7.03^{ab}	6.18^{bc}	5.20^{c}	7.61^{ab}	7.59^{ab}	7.44^{ab}	7.94^{a}	0.203	<0.001
	b*	65.16	63.32	63.11	62.35	66.96	62.29	68.27	0.959	0.529

Means indicated with different letters in the same row are significantly different; Zn: Zinc; FCR: Feed conversion ratio; SEM: Standard error of means; P: Significance level.

CHAPTER 4

DISCUSSION, CONCLUSION AND RECOMMENDATIONS

In this thesis, effects of zinc nanoparticles obtained from plant extracts with different zinc sources on live weight, feed consumption, feed conversion ratio, egg weight, eggshell weight, egg width, egg length, shape index, yolk weight and egg yolk color parameters (L*, a* and b*) of laying quail were investigated.

The differences in final live weights of the treatments groups were not found to be significant. Zhao et al., (2014) reported that the addition of nano-zinc to the ration at different rates increased the rate of live weight gain and feed conversion ratio of the broilers. In line with the study, Zhao et al. (2016) reported that there was no difference between the groups in terms of live weight and egg production with the addition of nanoparticle zinc at different rates to the laying hen rations. The greatest egg production was determined in nano Zn-supplemented groups and different nano Zn sources added to the ration increased egg yields.

Tabatabaie et al. (2007) reported that different Zn sources (inorganic and organic) or levels (25 and 50 mg/kg) did not affect egg production, egg weight or feed conversion ratio of laying hens; however, feed consumption was lower in 50 mg/kg organic Zn-supplemented groups than in the other groups. Tsai et al. (2016) reported that feed intake, egg production, egg weight and egg mass of laying hens were not affected by different Zn sources. Mohammadi et al (2015) indicated that the live weight and feed intake of broilers decreased with the addition of nano Zn into the ratios as compared to the addition of Zn-methionine; however, the feed conversion rate was not affected by nano Zn supplementations.

It was determined that the addition of different Zn sources at different rates to quail rations increased the average feed consumption and feed conversion ratio throughout

the experiments. Fathi et al. (2016) reported that nanoparticle ZnO sources increased feed consumption and growth performance of broilers.

The effect of the addition of different Zn sources on the egg weight during the experiment was determined, except for the nano Zn groups, experimental treatments affected egg weights of treatment groups.

Mao and Lien (2017) reported that egg production, egg mass, feed conversion rate and egg weight of laying hens were not affected by different inorganic, organic and nano Zn sources added to the ration.

The addition of different Zn sources to the diets of the experimental groups had the highest effect on eggshell weight during the experiment and it was determined that the groups supplemented with organic Zn had the highest eggshell weights. Nano Zn sources did not have a significant effect on eggshell weights. Cufadar et al. (2020) reported that different levels of nano, organic and inorganic Zn sources added to laying hen rations did not have a significant effect on eggshell weight and eggshell breaking strength. Tsai et al. (2016) reported that eggshell weight and eggshell breaking strength were not affected by different Zn sources; however, the eggshell thickness was improved with the addition of organic Zn and nano Zn. Zhao et al. (2016) indicated that as compared to ZnO_4 (200 mg/kg), nano ZnO (200 mg/kg) reduced eggshell weight. Eggshell breaking strength and eggshell thickness did not change with $ZnSO_4$ or nano ZnO applications. Idowu et al. (2011) also reported that the addition of different Zn sources did not affect eggshell thickness of laying hens. However, Bahakaim et al. (2014) reported that eggshell thickness increased with organic Zn supplementations into laying hen diets.

The effects of addition of different Zn sources to the rations of the experimental groups on egg width were determined and the greatest egg width was observed in 6 mg/kg inorganic Zn-supplemented groups.

The addition of different Zn sources to the rations of the study groups had the highest effect on egg length during the experiment and the highest egg length was observed in 3 mg/kg organic Zn-supplemented group. Except for the 1st and 2nd weeks, effects of different Zn sources on egg lengths were not found to be significant. The addition of different Zn sources to the diets of the study groups did not affect the egg shape index during the experiment. Effects of different Zn sources on egg shape index were found to

be significant in the 3rd and 5th weeks, but insignificant in the 1st, 2nd, 4th and 6th weeks.

The effects of the addition of different Zn sources to quail rations at different rates on weekly and overall egg yolk weights were not found to be significant throughout the experiments.

Effects of different Zn sources on egg yolk color a* (Red-green) values of layer quails were found to be significant only in the 2nd and 6th weeks.

Effects of different Zn sources supplemented into quail diets on egg yolk color b* (yellow-blue) parameter were found to be significant only in the 1st week of the experiments, but insignificant in the other weeks.

Supplementation of different Zn sources into quail diets at different rates (3 and 6 mg/kg) did not significantly influence the live weight change, egg production, % egg production, feed consumption, feed conversion ratio, egg weight, eggshell weight, egg width and egg length, but significantly affected egg length and shape index.

The addition of nano Zn to the diet can reduce Zn excretion and increase intestinal Zn absorption. Therefore, nano Zn addition has positive effects as compared to organic and inorganic Zn. Present findings revealed that nano Zn could be used as effectively as conventional Zn sources (inorganic and organic) to provide optimum performance and egg quality in layer quail. In addition, organic and nano forms of Zn can be preferred in laying quail rations, as it reduces Zn excretion and soil pollution.

It was concluded that nanotechnology could be used in trace mineral supplements and serve as a viable source to replace minerals in laying hen diets. Present findings obtained from quail experiments may also be valid for the other poultry species. Further research is recommended to prove validity of such findings in the other poultry species.

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RESUME

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