The effects of KEPROGAN® as an additive in chicken feed on biochemical tests and the immune systems of the chickens

Abstract

The purpose of this study was to explore the effect that supplementing broiler chickens with Keprogan had on their physiological condition as well as their immunological response. The broiler chickens (Ross) were one day old when they were randomly divided into two food treatments, each of which contained one hundred chicks. Chickens in group 1 were given a food that contained 0.5% keprogan, while chickens in group 2 (the control group) were given a diet that did not contain any supplements. The current study indicated that the supplement Keprogan (T2) provided to broilers resulted in a substantial ($P \leq 0.05$) increase in biochemical tests at day 35 (the conclusion of the experimental period) with (ALP, AST, ALT, T.P, Cholesterol, Triglyceride, VLDL, Bilirubin, albumin, and globulin), as compared to the control. In contrast to the group that served as the control, an increase in ante body titer against ND and IBD virus that was statistically significant ($P \leq 0.05$).

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Introduction

Keprogan is a supplementary premix that can be added to the diets of chickens and calves. It helps to maintain the healthy functioning of the digestive tract as well as the microflora in the digestive tract. Butyric acid, which is present in Keprogan, increases the number of cells that can proliferate and differentiate in the intestinal epithelium, as well as an improved barrier function of the intestines against infections. Keprogan can be found in capsule form. In addition to this, it brings the pH of the intestines down, increases the action of enzymes, and improves the operation of the immune system. According to research conducted by (Ruan et al. 2021), the components carvacrol and thymol found in essential oils encourage feed intake and contribute to the maintenance of a healthy gut flora.

As inexpensive, environmentally beneficial, cost-effective, and non-toxic materials, silicate minerals including attapulgite, zeolite, kaolinite, and sepiolite have demonstrated efficacy on account of their high ion-exchange capabilities and high absorption capacity. (2011; Zhou et al., 2014). This is due to the fact that these minerals are composed of silicate.

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According to Zhou et al. (2014), chemicals of this kind are utilized extensively in a variety of disciplines, including animal nutrition and veterinary care, agriculture, environmental protection and sanitation, and industrial. According to certain reports, silicates have a high concentration of main as well as trace elements, all of which are necessary for the healthy development of aquatic animals, livestock, and poultry. According to Wu et al. (2013), these elements are in an ionic state, which means that they can be released and utilized by the animals.

At this time, herbal products are mostly utilized within the feed industry as sensory enhancers, flavoring components, and attractive components. despite the fact that a comprehension of their mode of operation is necessary for the most efficient application of these methods. (Samant., et al. 2021) One such example is the drug KEPROGAN®.

In order to protect other biological molecules from oxidation, essential oils (EO) in KEPROGAN® are able to do two things: (1) donate a hydrogen or an electron to free radicals, and (2) delocalize the unpaired electron within the aromatic structure. There are natural oils included in KEPROGAN®. It has been proposed that the obtained values can be used to create a ranking order of the aromatic compounds' antioxidant properties (Fernandez-panchon et al., 2008). Several methods have been used to study the antioxidant effects of phenolic compounds in vitro.

Furthermore, Kamel (2001) says that organic hydrocarbons and essential oils can make an animal hungrier and change the kinds of bacteria that live in its surroundings. Different amounts of essential oils may have different antimicrobial effects on germs, such as killing or stopping the growth of bacteria. A number of studies have also shown that essential oils can fight free radicals and have effects on the microbiology of the gut, the physiology of digestion and weaning digestion, and the use of test models in chicken. (Awad et al., 2016).

The aim of this study was to identify KEPROGAN effect on physiological state and immune response at 35 days of broiler chickens.

Materials and methods

Experimental animals designing:

For the purpose of this investigation, two hundred Ross-308 broiler chickens (*Gallus gallus domesticus*) were one day old and randomly assigned to one of two groups, each of which contained four replications (with fifty birds assigned to each replication). Sawdust was used as a litter on the floor of the pens in which all of the replications were kept. The density of the stocking was determined to be ten birds per square meter. During the course of the study, professional methods of management were utilized, and the surrounding temperature was successfully managed. During the course of the experiment, participants received 23 consecutive hours' worth of artificial light. All of the birds were given a vaccination against Gumboro, infectious bronchitis and Newcastle disease in the 2rd week of their lives, after they hatched (Steiner and Syed, (2015)). Ad libitum commercial foods were provided for the birds who comprised the control group (control). The Keprogan was included in the diets of the birds in the 2rd group, who otherwise consumed the same meal as the birds in the 1st group.

two hundred one day old Ross-308 broiler chickens (*Gallus gallus domesticus*) were **:Commented [yr3]**

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Results and Discussions

The results of the experiment are illustrated in Tables 1, 2, 3, and 4, with each table focusing on a different aspect of the immunological response or the physiological condition. There is a correlation between the significant rises in parameter measures and the positive effects on the broiler chickens' states of health.

Table 1. Dietary impact of Keprogan premix according to (Cholesterol, Triglyceride, VLDL, Bilirubin) (means SE)

Parameter Groups	Cholesterol	Triglyceride	VLDL	Bilirubin
Control	131.166± 13.0 A	52.33± 1.50 A	10.66± 0.21 A	0.070± 0.004 B
Treatment	111.833± 5.05 B	34.00± 4.81 B	6.66± 0.98 B	0.090± 0.005 A

Significant differences in treatment at ($p < 0.05$) were denoted by distinct letter in the same raw data.

Table 2. Dietary impact of Keprogan premix according to (ALP, ALT, AST) (means SE)

Parameter Groups	ALP	ALT	AST

Control	2447.166± 307.14 B	24.66± 1.91 A	424.50± 45.22 A
Treatment	3859.666± 516.23 A	17.33± 1.08 B	257.66± 22.93 B

Significant differences in treatment at ($p < 0.05$) were denoted by distinct letter in the same raw data.

Table 3. Dietary impact of Keprogan premix according to (T.P, albumin, globulin) (means SE)

Parameter Groups	T.P	albumin	globulin
Control	1.92± 0.22 B	1.52± 0.12 B	0.52± 0.07 B
Treatment	2.89± 0.18 A	2.10± 0.14 A	1.15± 0.19 A

Significant differences in treatment at ($p < 0.05$) were denoted by distinct letter in the same raw data.

Table 4. Dietary impact of Keprogan premix according to (ND, IBD) (means SE)

Parameter Group	ND	IBD
Control	3325±36.8 B	9593.66±71.63 B
Treatment	5386 ± 77.94 A	12845.14± 84.44 A

Significant differences in treatment at ($p < 0.05$) were denoted by distinct letter in the same raw ~~data.~~data.

Essential oils may affect animal lipid metabolism. For instance, feeding old rats thyme oil or thymol improved superoxide dismutase and glutathione peroxidase levels and tissue polyunsaturated fatty acid composition. In compared to untreated controls, animals given these supplements showed higher enzyme levels and brain phospholipid polyunsaturated fatty acid concentrations. Oregano added to chicken feed at 50–100 mg/kg had antioxidant effects in animal tissues.(Botsoglou, Nicholas A., and others 2002) such as enhancing the flavor of the feed, accelerating the secretion of digestive juices, modifying the structure of the intestines, bringing the intestinal flora back into balance, and decreasing inflammation. According to Ludwiczuk (2017), improving the palatability of animal feed can drive increased feed intake as well as increased appetite. The possibility of bacteria and fungi adhering to the epithelium lining the chicken intestine is eliminated as a result of increased mucus secretion in the gut that is created by chemicals derived from plants (Calo, 2015). Some of the increases in nutrient absorption may be attributable to increased bile and saliva secretions, as well as enhanced specialized biochemical reactions of enzymes; the altered nutrient sucking may permit feed density modifications. Colon epithelial cells' villus healing, adhesion, and intestinal absorption capacity may all

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improve if pathogenic bacteria are reduced (Ludwiczuk, 2017). It has been examined extensively as a feed supplement to improve growth and health in broilers due to its ability to bind toxins and prevent diarrhea(Lambert, 2011). Silica compounds, such as colloidal silica, have been studied for their possible antioxidant effects in broiler chicks (Defoirdt, 2013).

The essential oils and other chemicals in an aromatic compound are what give it its impact (Zhou, 2014). Aromatics used in livestock and meat production have been shown to improve digestion, circulation, and immunity (Wu, 2013). There is evidence that they can fight against bacteria and keep coccidia at bay. Oregano oil, in particular, has been in demand due to the presence of carvacrol and thymol (in Keprogan), as well as y-terpinene, p-cymene, and myrcene, albeit in lower concentrations (Calucci, 2013). Oregano oil has been shown to have beneficial effects in a variety of in vivo and in vitro models. Regardless, feed supplements are used permanently with healthy animals for nutritional and functional objectives, whereas veterinary medications are only used under the supervision of a veterinarian for the treatment of health problems, and even then, only for a limited amount of time.

Considering GIT morphological reactions, Flourou-Paneri (2006) hypothesized that Keprogan's phytogenic impact may include gut tissue irritation, which reduces intestinal surface area. Positive effects on GIT health (such infection reduction). Large-scale chicken production experiments show that essential oil treatment works better (Ebani, 2016). Immunological improvements and intestinal pathogen reduction may explain this behavior.

Essential oil including thymol and carvacrol and boost feed intake and gut bacteria. They also increase intestinal enzyme synthesis and secretion. Due to delayed stomach emptying, intestinal micro biota stabilization, and/or greater enzymatic activity, digestible nutrients may be better absorbed. Keprogan releases targets and is steady during processing.

It has been demonstrated that increased feed digestibility brought about by the addition of EOs leads to enhanced nutrient absorption in pigs and poultry (Ahmed et al. 2013). Botanical have the potential to affect the digestibility of food and the rate at which it moves through the digestive tract. These effects could have an effect on the production of bile, cause an increase in the secretion of saliva, bile, and mucus, and stimulate enzyme activity. According to research done by Hong et al. (2012), an increase in the secretion of mucus in the intestine that is produced by substances that originate from plants eliminates the likelihood of bacterial

and fungal adherence to the epithelium that lines the intestine of chicken. EOs also lessen the impact of disease-causing bacteria thanks to their direct antibacterial activity and their ability to stimulate the establishment of beneficial microflora.

It is possible that the reduced stress on the intestinal immune defense system contributed, at least in part, to the reallocation of nutrients toward development rather than immune defense. Recent experiments conducted in controlled experimental conditions with a better level of sanitation have not been able to replicate the positive effects of EO therapy seen in earlier investigations conducted under the realistic settings of large-scale animal husbandry, as reported by Franz et al. (2012). These experiments were carried out in the laboratory. This could be explained by a decrease in the number of pathogens present in the intestine as well as an improvement in the immunological function.
There is an observed elevation in the rate of lymphocyte proliferation, phagocytosis, and the levels of serum immunoglobulins (IgG, IgA, IgM) as well as complement proteins C3 and C4. have been reported to demonstrate that supplementing EOs improves the immunological state of piglets after weaning (Zeng, et al. 2014). This improvement in immune status was observed in the piglets. According to the findings of Walter et al. (2004), Pigs whose peripheral blood lymphocytes contained 3 g/kg oregano (60 g carvacrol and 55 g thymol per kilogram) displayed greater proportions of CD4:CD8, MHC class II antigens, and non-T/non-B cells compared to pigs fed a control diet.

Most bioactive compounds are metabolized and either removed by the kidneys as glucuronide or exhaled as CO2 after oral, pulmonary, or cutaneous administration (Kohlert et al., 2000). Changes in blood immunological markers may suggest that the absorbed component has stimulated an immune response, while the unabsorbed component may help alleviate the stress on the intestinal immune defense system. However, more study is needed to elucidate the precise mechanisms by which EOs produce their effects.

Blood tests help determine avian health. The current study found that broilers supplemented with carvacrol, EOs, and thymol (in Keprogan) had changes in blood total protein, albumin, globulin, and uric acid. Blood proteins are indicators of an animal's physical health, nutritional status, and homeostasis (Cheng, et al 2022), and no differences showed that EOs did not affect broilers' health and nutrition. IgG participates in humoral immunity and is intimately associated to immunological function (Mohiti and Ghanaatparast. 2017). Our results showed that the serum IgE and IgG concentrations of broilers increased linearly with the addition of

carvacrol and thymol over the course of 21 days. Mohiti-Asli and Ghanaatparast-Rashti (2017) reported that broilers administered 300 mg/kg oregano essential oil had larger IgG and total antibody titers than those fed the control diet (Stamilla, 2020).

We conducted a thorough analysis of KEPROGAN®'s impact on serum biochemical parameters and immunoglobulin content in broiler chickens in the present study. Similar to prior research, our data revealed that supplementing broiler's diets with carvacrol and thymol at varying concentrations significantly improved their health. Adding 150 or 300 mg/kg of oregano essential oil to the diet of yellow-feathered broilers considerably enhanced their average daily gain (ADG) and average daily feed intake (ADFI) (Ruan et al 2021). Broilers supplemented with 25 mg/kg of a mixture of octagon, rosemary, and thyme oregano essential oils gained considerably more weight between 22 and 42 days of age, as reported by Youssef et al. (2021). Supplementing broilers' diets with either natural or synthetic oregano essential oil from 1-21 days of age raised ADG and lowered F:G relative to the control group [Tóthová, et al. 2019]. This was also found by Zhang et al. Although EOs were found to have a beneficial effect in all of the aforementioned research, the ideal supplementation dose varied. This could be because EOs' effective components vary depending on their source or product type.

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