# The Effect of Use of Pistacia Khinjuk on the Amount of Antibody Production in Broilers

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## ABSTRACT

In order to explore the possibility of using Pistacia Khinjukin feeding, 240 pieces of one-day broiler were studied in 4 treatments including diets of different levels of 0, 3, 6, and 9, and the percent of Pistacia Khinjuk with 4 repeats, 15 pieces of broilers in each with a full random plan for 42 days. The analysis of the results has shown that food treatments have significant effect on functional traits (food consumption, increasing of weight and food changes efficiency). Exploring the effect of use of this fruit on antibody titers (the collection titers of IGA, IGG, and IGM) by the use of statistical analysis, a significant difference has been observed in antibody titer and the weight of spleen though there was not any significant difference between the persistence of the treatments. Generally, the findings of this research have indicated that the treatment of 6 percent of Pistacia Khinjuk has the best performance in the production traits and antibody titers.

**Keywords**: Pistacia Khinjuk, Antibody, Immune system, Broiler

## INTRODUCTION

According to food varieties of poultry and the required energy supply, it is essential to substitute the energy supply with new sources of energy supply which are suitable and appropriate in terms of other ingredients. One of the new sources of food which can be used as energy supply in a diet is Pistacia Khinjuk. Pistacia Khinjuk is a kind of turpentine called Pistacia Khinjuk. Different species of Pistacia Khinjuk can be widely found in Iran, Syria, Afghanistan, Pakistan, and India. The wild species can also be found in the areas with the height of 700 to 1900 meters above the sea level [1]. The five species of *Pistacia* called *Pistacia* L., *Pistacia atlantica* Desf., Palestinian Pistacia, mastic tree, and Pistacia Khinjuk are treated naturally around Mediterranean and Middle East areas. Pistacia is originated from middle Asia but it is planted all through Mediterranean region and all over the world. The protein of Pistacia Khinjuk has a large amount of the amino acid threonine, Gluteus, Valine, and Lysine which is very important in poultry feeding. Pistacia Khinjuk contains 40.02% fat with a lot of acid threonine which is a vital fat in poultry feeding. Pistacia Khinjuk contains 2/16% of tannin which its effect on diet is discussable. Paying more attention to this plant – in case of positive effect of its use in diet - it can be used in addition to its nutrition aspect as creating green environment and healthy air which requires a long-term planning. The plants of pistacia species including its medicinal species (*Continue coggyria*) with medicinal qualities have been used for a long time.

The analysis indicates that Pistacia Khinjuk is a rich source of volatile fatty acids (especially Linoleic acid) and partially tannin. The positive effect of volatile fatty acids on the digestion and

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absorption of nutrients and fat-soluble vitamins which have special role in the immunity of body [2]. The analysis of Taran et al. [3] shows the anti-bacterial and anti-fungal effects of Pistacia Khinjuk though it does not show the current research evidences about the species of Pistacia Khinjuk on the immunity system of animals. There has not been any research in this case about poultry but the ant-bacterial traits of tannin and having the least casualties in the poultry utilized Pistacia Khinjuk in various researches [4,5] offered this hypothesis that this fruit can be effective for improving body immune system.

According to the initial information, this research has been conducted for determining the effect of consuming Pistacia Khinjuk on the amount of antibody production and obtaining the most suitable level of using Pistacia Khinjuk for improving body immune system on broilers.

The species of Pistacia Khinjukis observable between cold and subtropical regions. Thus, as we move from the northern latitude of the country toward the southern latitude specifically in higher altitudes, it is added to its plentitude. The slang term for such name is such as Narmavan and it is also Kolkhang. The species of Pistacia Khinjuk has wider and more open crown comparing with the others [6]. It is observable in most parts of the country except the Caspian region and desert areas but its most expansion is alongside Zagros Mountain from northwest toward southeast, northeastern region, east, south and in limited level in central region of the country [6]. The results of some experiments showed that the abnormal consuming food (Atlantica species and Pistacia Khinjuk) which are from different climate have specific attributes while their apparent metabolism value is adaptable to the cereal seeds consumed in diets of poultry [1]. Also, it has been determined in the same diet that atlantica seeds can be used as a replacement for corn [7].

Determining the notorious value of atlantica pistachio and Pistacia Khinjuk, some experiments were conducted from three different climates of southwestern or Zagrous Mountains in an area expanded 500000 hectors by taking samples of these grain foods in Kohkiloye and Boyerahmar province [1]. According to this, the capacity of crude protein of wild pistachio seeds is much like corn and slightly fewer than other creels. The protein of pistachio has high amount of ermine, threonine, valine and leucine which is very important if it is used as food of poultry. The percent of crude fat of Pistacia Khinjuk is 39/1 percent and atlantica pistachio is 26/8 percent along with high level of Linoleic acid - which is very useful in the nutrition of poultry. The percent of crude fiber of atlantica pistachio and Pistacia Khinjuk is high comparing with other cereal seeds. This issue can be a restrictive factor for using of them in diet [1].

In this regard, another experiment was conducted to study the effects of different levels of Pistacia Khinjuk on the performance of laying hens and egg quality characteristics on the 160 white Leghorn hens piece that were 24 weeks. In the first phase and the second laying (weeks 24 to 36) except the weight of egg white and yolk color unit , there was no significant difference [1].

The studies of Zargari et al. [8] showed that there are various plants from pistachio species in Iran which some of them have medical effects and others suitable notorious value. Pistacia Khinjuk is one of these species. The findings of Hosseini [4] research about the use of different level of Pistacia Khinjuk as a substitute of corn showed that treatment 6 has the best percent of performance and the least casualties. Therefore, the purpose of this test is the acceptance or rejection of this hypothesis that the use of Pistacia Khinjuk fruit is effective on the titer of antibody of broilers. The percent of crude fiber of atlantica pistachio and Pistacia Khinjuk is high comparing with other cereals, and this can cause a restrictive factor of using of it in a diet [1]. Of course, crude fiber which includes cellulose and lignin is completely indigestible for poultry. The percent of calcium and potassium of atlantica pistachio and Pistacia Khinjuk seems more than cereal sees while the amount of Sulfur and magnesium is a lot less [1]. The percent of fatty acid of pistachio is several times more than cereal seeds. Due to the high percent of crude fat, the percent of linoleic acid of grain is for instance 83% while for oaks is 1/53 percent and Pistacia Khinjuk is 5/37 percent [1].

## MATERIALS AND METHODS

Exploring the effect of use of Pistacia Khinjuk fruit on the performance, immunity system, and survival power of broilers of four levels of 0, 3, 6, 9 percent of Pistacia Khinjuk fruit with 4 repeats and the number of 15 pieces in each repetition (240 observed) were used for 42 days in the diet of broilers. The amount of wight increase, food consumption, food change efficiency, the weight of splitter, and survival at the end of the period and the titer of antibody in 22 and 35 days old were measured. The required Pistacia Khinjuk fruit of this project were gathered from the region wound Gachsaran city in Kohkiloye and Boyerahmad province in the middle of autumn. After being assured of the dryness of the external skin of fruit (pericarp), the test material (Pistacia Khinjuk) were dumped in bags and transferred to Yasouj. They were preserved in a place away from sun and dry before their consumption for providing the food of broilers (Figure 1).

Figure 1. Pistacia Khinjuk fruit taken for use in diet.

Food diet was set based on the food prospectus requirements booklet of Ross chicken. The Food items were set containing corn, soybean meal, fish meal, di-calcium phosphate, oyster powder, DL-methionine, L-lysine (HCL), salt and vitamin and nutritional requirements based on tables (NRC) and data previous chemical composition includes 91/94% dry matter, protein 11/10, 32/40% fat, ash 38/2, 3/24 percent crude fiber, calcium 26/0, 448/0 percent phosphorus and 05/2 percent of Tannin. The attained metabolism energy for the sample of tested Pistacia Khinjuk was equal to 4240 kcal/kg. Sibbald method was used for determining the metabolism energy of Pistacia Khinjuk [9]. 5000 pieces of broilers were used in this test. The maintained condition was the same from the first day of entrance to the eighth day, and then divided based on the treatments. All the chickens used the dies of initial stage till the end of third week. All the chickens were kept in cages collectively till the seventh day, and they were transferred to the experiment cages on the seventh day. Thus, about 240 pieces of chicken were selected and tested which were equal in size and healthier on the seventh day, and the remained chickens were kept out of the cages in a salon. The selected chicken entered into experimental cages which were selected randomly so that the average weights of the chickens of all cages were 134/5±/5. Water and food were offered to the chickens freely. From the entrance of chickens into the initial diet salon about 2900 kilocalorie of energy metabolism and 20/84 percent of protein were offered to the tested groups so that each group could supply its food.
Studying the effects of Pistacia Khinjuk, this fruit was added to the diet from the 22 day and after initial blood sampling. Exploring the immune system in the interval of 2 days before vaccination with IBD vaccine (22-days) and after 13 days of vaccination (35 days), a blood sample was taken from the chickens from their heart area. The samples were kept stable for some minutes for forming clot, and it was then sent to a Veterinary laboratory for measuring antibody titer (collection of titers IGA, IGG, IGM) by the method of ELISA. The recorded data from the measurement criteria were entered into a computer by statistical program of SPSS through one-way analysis of variance test.

Also, the comparison test of treatments was statistically analyzed by the same program with the method of Duncan's test.

## RESULTS AND DISCUSSION

### *Feed intake, Body weight gain and feed conversion ratio*

The result of the measurement of food intake is shown in table 1. As it is seen, except the treatments of 6 and 9, the percent of all the tested treatments have significant difference. The treatment 6 had the highest and the witness treatment had the lowest food intake in this stage. The findings of variance analysis of body Wight in the 42-days old (Table 1) indicates that there is a significant difference between the witness treatment and 3 percent with others. Since there is anti-nutritional tannin in the percent of 3, 6, and 9 of Pistacia Khinjuk, it is expected that the testing treatments have less weight comparing with the witness treatment but the achieved result is contrary. Treatment 6 had the highest percent and the witness treatment the lowest body weight. The results of measuring FCR final period is shown in table 2. According to the table of variance analysis and comparison of the averages among the treatments, it was observed that there was a significant difference between the FCR control and 3% and 6% treatments in the final stage.

Table 1. The mean and functional traits of different treatments in the test.

| Treatments | Food intake (g) | Weight increase (g) | feed conversion ratio |
|---|---|---|---|
| Witness (0 % PistaciaKhinjuk) | 4053/77±84$^{c}$ | 1983/43±38$^{b}$ | 2/04±0/1$^{bc}$ |
| 3 % PistaciaKhinjuk | 4167/20±68$^{b}$ | 1995/9±89$^{b}$ | 2/08±0/7$^{c}$ |
| 6 % PistaciaKhinjuk | 4278/00±22$^{a}$ | 2183/77±34$^{a}$ | 1/96±0/02$^{a}$ |
| 9 % PistaciaKhinjuk | 4252/22±40$^{a}$ | 2133/5±31$^{a}$ | 1/99±0/05$^{ab}$ |
| Standard deviation | 43/75 | 42/12 | 0/029 |

A, B, C = a significant difference in 1 percent

### *Antibody titer and survival power*

According to table 1, there is an increase in antibody titer in 35-days the witness treatment till 9 percent titer. The analysis of the data shows a significant difference between the witness treatment and treatments of 6 and 9 percent and the treatment of 3 percent with the treatments of 9 percent. Also, there is not considerable difference between the survival of treatments of 0 and 3 but an increase was observed in the treatment of 6 percent and then a quick decrease in the treatments of 9 percent by increasing the level of Pistacia Khinjuk among the chickens. Analyzing the data does not show a significant difference among the treatments.

Table 2. The mean and standard deviation of antibody titer, spleen weight and survival of broiler chickens fed different diets in the experiment.

| Treatments | In 22 days | In 35 days | In the final stage (g) | In the final stage (%) |
|---|---|---|---|---|
| Witness (0 % PistaciaKhinjuk) | 4892/75±292 | 8772±721$^{c}$ | 1/22±0/15$^{c}$ | 87/08±2/08 |
| 3 % PistaciaKhinjuk | 4815±415 | 10337±944$^{bc}$ | 1/26±0/1$^{c}$ | 87/5±2/5 |
| 6 % PistaciaKhinjuk | 5315/75±525 | 11760/75±1055$^{ab}$ | 1/56±0/05$^{a}$ | 90/41±2/92 |
| 9 % PistaciaKhinjuk | 5473/5±492 | 12106±1037$^{a}$ | 1/41±0/05$^{b}$ | 89/58±1/25 |
| Standard deviation | 399/95 | 1017/53 | 0/062 | 1/79 |

A, B, C = significant difference at the level of 1 percent=* in the level of 1 percent of ns =without significance difference

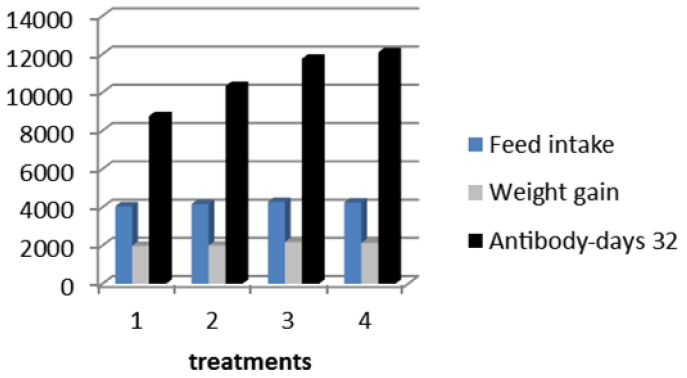


Figure 1. Feed Intake (FI), Body Weight Gain (WG) and Antibody Day32.

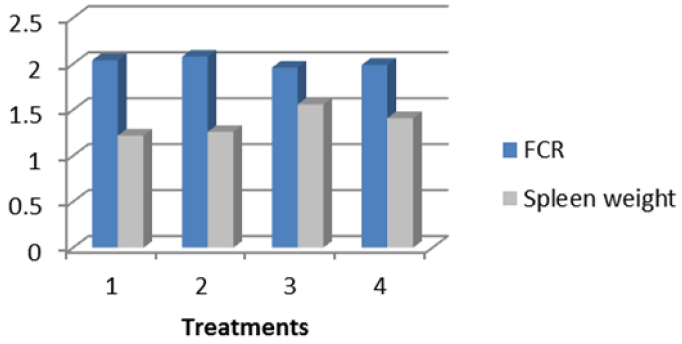


Figure 2. Feed conversion ratio and Spleen weight.

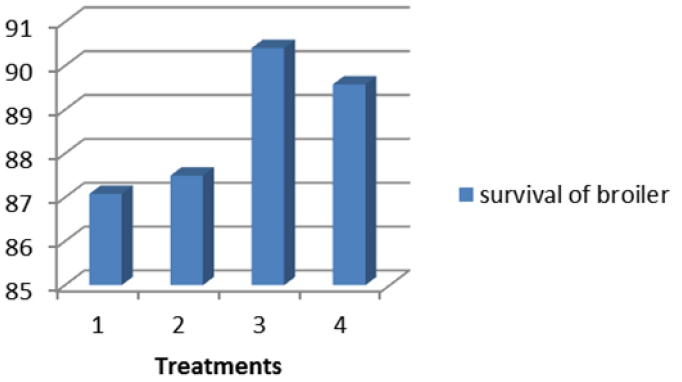


Figure 3. Survival of broiler.

The development and growth of digesting system and the expansion of blind intestines in chickens can be a factor for efficient use of food containing fiber though the restrictive effects of tannin in different levels of Pistacia Khinjuk consumption in a diet can be a factor for distinguishable difference of usage in different treatments. Due to the high level of fiber of Pistacia Khinjuk, the increase use of food of the witnessed treatment to the testing treatment of 6 percent Pistacia Khinjuk can be justified as the attempt of a chicken for getting the required energy by using food with more fiber but the decrease of intake in the treatment of 9 percent can be as the result of eating disorder which is resulted by the increase of tannin of the diet. This subject is in contrast with the findings of Safarzadeh [1] but agrees with the findings of other researchers since tannin of the food increases the metabolism energy of the diet [10], proteins, carbohydrates, and other nutrients due to forming complex with the protein segment of digestive enzymes [11] and deactivating of them in one hand, and forming complex with proteins and other nutrients which results to the indistinctness of them [12]. The decrease of the metabolism energy of the diet should normally increase food intake [13] which the analysis of food intake in this stage and the diagram number 1 is according to this theory. There is evidence that as tannin of the diet of laying chickens is more than 23 percent, the metabolism energy decreases to 40 kilocalories for each 0.1 percent increase [14]. Due to the high growth rate in the first 6 weeks of life and need for nutrients, especially protein, the significant increase in body weight during this period, despite the tannin in Pistacia Khinjuk, can be the development of digestion system in growth period and high fat of Pistacia Khinjuk. On the other hand, since this nutrient is a rich source of unsaturated fatty acids, especially linoleic acid which is the most important fatty acid and the only essential fatty acid, the increase of body weight can be related to this fatty acid. On the other hand, since the diet containing Pistacia Khinjuk is fatter than a diet without Pistacia Khinjuk, and the fat of diet increases by the increasing of replacement percent and as fat is the factor of decreasing the speed of food passing from gastrointestinal tract with more time for food digestion and nutrition intake [13], the effect of creating energy surplus due to the effect of dietary polyunsaturated fatty acids and opposite interaction with saturated fatty acids and help them attract absorption is a justification for the increase in body weight during their rapid growth [15]. The findings of this stage of test are in parallel with Saffarzadeh [1].

Since there is anti-nutrient tannin in the testing diet which effect on the different levels of food digestion and absorption and on the other hand by effecting on the digestive system tissue and damaging Intestinal mucosa [16] decreases nutrition absorption, it is expected that the use of Pistacia Khinjuk due to containing tannin increase FCR. FCR decreased in this test in contract with the initial stage. According to other results of this test indicating that the use of Pistacia Khinjuk did not have negative effect on body weight and it caused the increase of appetite of the chickens of testing treatments comparing with the witnessed group, it can be inferred that the use of Pistacia Khinjuk is permissible to 9 percent level.

However, the tolerance level of broiler chickens is reported 3-4 gram to kilogram against tannin. The sample Pistacia Khinjuk of the test contains 2.16% tannin though the kind of tannin is also effective [17]. According to food intake in the average weight of the chickens in this stage and their significance, and the increase of the weight of chickens which is partially due to saving fat in their body, the significance of difference FCR between treatment 6% with the witness treatment is related to less food intake of treatment 6% comparing with treatment 9% and more weight increase comparing with other testing treatments and the witness treatment itself. However, this can be due to better balance of saturated and unsaturated fatty acids and more linoleic acid comparing with witness treatment and treatment 3% and the amount of tannin and less fiber in treatment 9%. The findings of this test are in parallel with Safari's test results.

### *Antibody titter and survival power*

The significance difference of titters in the 22 days can be as the result of remaining effects of previous vaccine. On the other hand, according to the components of this food, we can understand that this source is rich of polyunsaturated fatty acids specially linoleic acid. According to the improvement of emulsification by bile salts of unsaturated fatty acids and better absorption of saturated fatty acid as the result of synergistic interaction of both types of fatty acids and increase of absorption capacity and the resulted energy of fat and other nutrients, the increase of Pistacia Khinjuk to 6 percent following these effects can help better absorption of nutrients and food balance. However, the more increase of the level of Pistacia Khinjuk can prevent the inhibitory effects of tannin of food absorption and more deaths in higher level of usage of that. Due to some effective factors on the improvement of body immunity system such as vitamin E soluble in fat and synergy of fatty acids mentioned above, the absorption of these factors is better in the presence of sources of unsaturated fatty acid which can be another factor of increasing of antibody titter. This effect is used to its role as a biological anti-oxidant and presence of immune cells from active oxygen mediators produces in inflammatory process [18,19]. The final result of test: the result of this project indicates that in addition to the positive effect of Pistacia Khinjuk fruit on the improvement of performance, this fruit can be effective for production of antibody. This fruit can be used for the diet of broiler chicken to 6 percent level.

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