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Estimate of Coenzyme Q10 on the humeral immune response of infectious bursal disease with different programs recombinant and classical vaccine and liver function in Broilers

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ABSTRACT

The aim of this study was to evaluate the impact of coenzyme Q10 dietary supplements on the humeral immune response (IgY) and certain liver functions including aspartate aminotransferase (AST) and alkaline phosphatase (ALP), in broiler Ross (308). vaccinated with different programs (recombinant HVT-IBD single dose at one day old, classical vaccines D78 at eight days old, and E228 at 16 days old). Two hundred and ten broilers were separated into seven categories, with 30 chicks each group. T1, feed Q10 (20 mg/kg diet) and vaccinated by (D78, E288); T2: feed Q10 (20 mg/kg diet) and vaccinated by (rHVT); T3: feed Q10 (40 mg/kg diet) and vaccinated by (D78, E288); T4; feed Q10 (40 mg/kg) and vaccinated by (rHVT): T5; vaccinated (D78, E288) only: T6; vaccinated (rHVT) only: T7; negative control. Immunoglobulin Y (IgY) titers were measured on day 15, revealing that groups T4 and T2 showed significantly higher titers ($P \leq 0.05$) compared to the other groups. Additionally, data from days 25 and 33 showed a consistent increase in IgY levels across the experimental period. Furthermore, serum antibody titers for viral diseases and the liver enzymes AST and ALP were significantly altered in Q10-treated groups compared to controls ($P \leq 0.05$). In conclusion, dietary supplementation with coenzyme Q10 at levels of 20 mg/kg and 40 mg/kg resulted in a significant enhancement in serum antibody titers against infectious bursal disease (IBD), while liver enzyme activity declined over the course of the experiment.

1. Introduction

Infectious bursal disease (IBD), also known as Gumboro disease, is one of the most widespread immunosuppressive avian diseases, causing high morbidity and mortality in commercial broilers, and in susceptible white leghorn hens, even as high as 100% mortality [1,2, 3]. The IBD virus (IBDV), a single-shelled, non-enveloped virus with a double-stranded RNA genome, is the cause of this disease [4]. The lack of an envelope makes this virus extremely resistant to hostile environments. Young hens with IBD may experience immunosuppression as a result of B lymphocyte reduction [5,6,30]. Moreover, it has been shown that monocytes and macrophages could be vulnerable to IBDV infection [7, 8]. It has been proposed that macrophages transport IBDV from the site of the intestinal infection to the Fabricius bursa and other peripheral organs [9, 10]. Immunosuppression further affects these birds' immune responses to immunizations and lowers their resistance to a number of diseases [11]. Following an IBDV infection, immunosuppression probably results in significant economic loss because of lost productivity, subsequent vulnerability to opportunistic infections, and vaccine failure [12]. To resolve this issue, vaccination is necessary for successfully controlling IBD [13]. Currently, commercially available vaccines against infectious bursal disease included, live attenuated, lethal, vector and immune complex immunizations. Three types of live attenuated vaccines are available, which are the most often utilized IBD vaccines in the field: vaccinations classified as mild, intermediate, intermediate-plus, or hot vaccine [14]. When certain amounts of maternally produced antibodies (MDAs) are present, mild vaccinations are less effective and against extremely virulent IBDV (vvIBDV). [15]. The recombinant turkey herpesvirus (rHVT) vaccine, which has been successfully developed by introducing the IBDV-VP2 gene into the HVT genome (rHVT-IBD), is an alternative to attenuated live IBDV vaccinations [16]. The generation of VP2 ,The VP2 protein is the principal antigen that determines serotypes protein from rHVT triggers an immune response to IBDV known as neutralizing antibodies [17, 18]. Compared to attenuated IBDV live vaccines, rHVT-IBD is thought to be a safer vaccination since it does not cause bursal damage, and the vaccine virus is not expected to be vulnerable to IBDV MDAs. Therefore, rHVT-IBD may be able to allay worries about the effectiveness and safety of IBDV vaccines [19, 20]. Coenzyme Q10 (COQ10) 2, 3-dimethoxy-5-methyl-6-decaprenyl benzoquinone] is a fat-soluble vitamin-like quinone also known as CoQ, ubiquinone, and vitamin Q10 [18]. Coenzyme COQ10 is a component of inner mitochondrial enzyme complexes and plays a role in the synthesis of adenosine 5′-triphosphate (ATP). It is spontaneously produced by mammalian cells. There is evidence that COQ10 possesses strong antioxidant capabilities [21, 22, 23]. By increasing CoQ10, dietary CoQ10 supplementation decreased weight associated with abdominal fat, alanine aminotransferase (ALT), and aspartate aminotransferase (AST) activity. Elevated CoQ10 levels exhibited a quadratic reduction in plasma glucose, cholesterol, and alkaline phosphatase (ALP) activity. [24]. Some studies found that supplementing hatched chickens' meals with coQ10 at different times resulted in significant increase in levels of antibody formation for ND, AI, and IBD [25, 26]. CoQ10 supplementation improves the immunological response to several viruses [27]. Furthermore, CoQ10 supplementation may be able to overcome immunological suppression brought on by age and chronic diseases. The goal of the current study was to investigate how different immunization protocols against infectious Bursal disease were affected by the dietary supplement Coenzyme Q10, which functions as an antioxidant, liver enzyme, immunomodulator, and body performance enhancer [28, 29].

2. MATERIALS AND METHODS

2.1 Experimental design

Two hundred ten (210) one-day old broiler chicks (Ross-308) multi sexes brought under the optimum condition with feed (ad libitum) and lighting program (24hr) period five weeks (35 days old) in the animal house of the University of Baghdad's College of Veterinary Medicine. The chickens are divided into six groups at randomly :

T1: Thirty chicks supplement with (CoQ10) (UK®) 20mg/kg diet during period of study with Classical vaccinal program (D78 at 8th days old & E 228 at 16th days old / drinking water) (MSD, US) with measurement of humoral Ab

(IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 old [31,].

T2: Thirty chicks supplement with (CoQ10) (UK®) 20mg/kg diet during period of study with Recombinant vaccinal program (Poulvac® Procerta® HVT-IBD at 1st days old single dose s/c in.) (MSD –USA), with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 olds.

T3: Thirty chicks supplement with (CoQ10) (UK®) 40mg/kg diet during period of study with Classical vaccinal program (D78 at 8th days old & E 228 at 16th days old / drinking water) (MSD, US), with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 old.

T4: Thirty chicks supplement with (CoQ10) (UK®) 40mg/kg diet during period of study with Recombinant vaccinal program (Poulvac® Procerta® HVT-IBD at 1st days old single dose s/c in.) (MSD-USA) with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 olds.

T5: Thirty chicks without supplements were given a classical vaccination program (D78 at 8th days old & E 228 at 16th days old / drinking water) (MSD, US) with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 olds.

T6: Thirty chicks with no supplement given Recombinant vaccinal program (Poulvac® Procerta® HVT-IBD at 1st days old single dose s/c in.) (MSD-USA), with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 olds.

T7: Thirty chicks with no supplement or vaccination (control negative), with measurement of humoral Ab (IgY) by ELISA Kit (5 serum samples) at (15th, 25th, and 33rd days old) and measurement liver enzyme AST and ALP at 35 old.

2.2 Statistical analysis

The Statistical Analysis System (SAS) (2018) program was used to detect the effect of different factors on study parameters. The least significant difference (LSD) was used to compare the means (ANOVA/Two-way and ANOVA/One-way) significantly in this study [32].

3. Result and discussion

3.1 Detection of ELISA, IgG (humoral immunity)

The results of the IBDV vaccine's serological reaction can be seen in Table (1). The results indicated that after 15 days, the highest proportion of antibody titers had been observed in T4, T6, and T2 (1542 ±76.6, 1119 ±64.9, and 1040 ±67.3), respectively. In contrast to T5, T1, and T3 (517 ±22.9, 556 ±22.8, and 598 ±19.5), which recorded the lowest percentages, correspondingly, exhibiting statistically significant variations (P≤0.05), T7 (1040 ±67.3) had a medium titer.

Results on 25 old days indicated a considerable increase during the following period, with group T4 recording the major value at 9603 ±237.4. T2 (8944 ±187.5) and T6 (7329 ±181.4) came next. In contrast to the non-vaccinated group T7 (132 ±8.5), which had the lowest percentage with a statistically significant difference (P≤0.05) while T3, T1, and T5 (4886 ±108.7, 4471 ±126.2, and 4006 ±175.3)respectively, had recorded medium titers.

On 33 old days, there was a rise in the serum titers of IgG. T4 (15727 ±278.9) had the highest titer, followed by T2 (13867 ±263.8) and T6 (10106 ±182.2). Nonetheless, T3 and T1 did not differ significantly from each other (12178 ±217.5 and 11343 ±167.9). On the other hand, T5 (9788 ±176.4) was higher than T7 (38 ±3.1), with a statistically significant difference (P≤0.05). These findings are consistent with Nemati et al. [2015) and Asadi et al. [2016), who demonstrated that CoQ10 supplementation of hatched chicken diets at various ages resulted in significantly higher levels of antibody production for ND, AI, and IBD compared to the negative and positive control groups [33, 34] and the difference between classical vaccines (D78 E228) compared to Recombinant vaccines (rHVT- IBD) is due to VP2 protein. As the main structural protein, Chickens can be Immunologically protected by VP2 alone by

assembling into virus-like particles (VLPs) or sub-size virus-like particles (s-VLPs) that can produce a high titer of neutralizing antibodies. [35].

Table 1: supplementation of Co-enzyme and different vaccine program on result of ELISA for IBD in chicks

Groups	Mean ±SE of IBD		
	15 days	25 days	33 days
T1	556 ±22.8 D c	4471 ±126.2 E b	11343 ±167.9 D a
T2	1040 ±67.3 BC c	8944 ±187.5 B b	13867 ±263.8 B a
T3	598 ±19.5 D c	4886 ±108.7 D b	12178 ±217.5 B a
T4	1542 ±76.6 A c	9603 ±237.4 A b	15727 ±278.9 A a
T5	517 ±22.9 D c	4006 ±175.3 F b	9788 ±176.4 F a
T6	1119 ±64.9 B c	7329 ±181.4 C b	10106 ±182.2 E a
T7	846 ±35.7 C a	132 ±8.5 F b	38 ±3.1 G b
LSD: 235.84 * Significant differences were found when different big characters were placed in the same column and little letters were placed in the same row. * (P≤0.05).			

3.2 Aspartate Aminotransferase (AST)

The findings showed that, in comparison to the control group, samples from the birds in the Co-enzyme Q10-treated group had significantly lower serum AST activity. We observed a decrease in enzyme activity in the groups T1 and T3 had been recorded the lowest percentage (71 ±3.27 and 81 ±3.93) respectively. Followed by T7, T5, T2 and T4 were recorded the medium percentages (101 ±5.27, 108 ±4.88, 118 ±6.02 and 157 ±8.41). While T1 and T3 had been reached the lowest percentages (71 ±3.27 and 81 ±3.93). There was a significant difference when compared between the groups (P≤0.05) (Table 2). These results were supported by Ashkani-Esfahani et al. [2016] who demonstrated that adding CoQ10 to the diet reduced cholesterol and plasma glucose levels and also reduced the activity of ALT, AST, and ALP. The results indicated that dietary CoQ10 supplementation may have an impact on lipid metabolism and liver function [36]. Furthermore, studies on rats have shown that supplementing the diet with CoQ10 reduced blood AST and ALT activity during metabolic stress [37].

Table 2: supplementation of Co-enzyme and different vaccine program on AST concent. At 35 day old

Groups	Mean ±SE of AST (U/L) 35 days
T1	71 ±3.27 e
T2	118 ±6.02 c
T3	81 ±3.93 de
T4	157 ±8.41 b
T5	108 ±4.88 c
T6	201 ±11.03 a
T7	101 ±5.27 cd
LSD: 24.592 * The means that had distinct letters in the same column varied considerably. * (P≤0.05).	

3.3 Alanine Aminotransferase (ALT)

There was a significant decrease in serum ALT activity of the groups, T1 (1.3 ±0.11) and T2 (1.8 ±0.19) were recorded the lowest value when compared with others. While T3 (2.1 ±0.22) and T4 (2.7 ±0.25) had been recorded the medium percentage. Moreover, T7, T5 and T6 was as the following (7.7 ±0.38, 8.1 ±0.57 and 9.8 ±0.61) respectively. It was no significance when comparing levels of T1, T2, T3 and T4. It was no significance in comparison the results of ALT in T5 and T6, results at T5 and T6 at 35 days old were higher than levels at T1, T2, T3 and T4 days old with statistically significant difference (P≤0.05) in (Table 3). The above results were acknowledged by Raeisi-Zeydabad et al. [2017]. They discovered that dietary CoQ10 reduced serum glucose, cholesterol, ALT, and AST enzyme concentrations, which could be attributed to a CoQ10-induced decrease in oxidative stress. CoQ10 can also decrease serum levels of glucose and lipids in broilers [38, 39]. Moreover, dietary CoQ10 stifles hepatic cholesterogenesis by the hindrance of HMGR action at the posttranscriptional level in chickens, which in turn diminishes plasma VLDL cholesterol concentration [40].

Table 3: supplementation of Co-enzyme and different vaccine program on ALT concent. At 35 day old

Groups	Mean ±SE of ALT (U/L) 35 days
T1	1.3 ±0.11 b
T2	1.8 ±0.19 b
T3	2.1 ±0.22 b
T4	2.7 ±0.25 b
T5	8.1 ±0.57 a
T6	9.8 ±0.61 a
T7	7.7 ±0.38 a
LSD: 3.074 * The means that had distinct letters in the same column varied considerably. * (P≤0.05).	

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تقدير تأثير الإنزيم المساعد Q10 على الاستجابة المناعية الخلطية لمرض الجراب المعدي مع برامج مختلفة للقاح المؤتلف والكلاسيكي ووظائف الكبد في الدجاج اللاحم

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الملخص

تم إجراء هذه الدراسة لتقييم تأثيرات المكملات الغذائية (مساعد الإنزيم Q10) على الاستجابة المناعية الخلطية (IgY) وبعض وظائف الكبد بما في ذلك أسبارتات أمينوترانسفيراز (AST) والفوسفاتاز القلوي (ALT)، في دجاج اللاحم من سلالة روس (308). تم تلقيح الدجاج ببرامج مختلفة (جرعة واحدة من لقاح HVT-IBD المؤتلف في اليوم الأول، واللقاح الكلاسيكي D78 في اليوم الثامن، و E228 في اليوم السادس عشر). تم تقسيم 210 دجاجات إلى سبع مجموعات، تضم كل مجموعة 30 صوصًا. المجموعة T1: تم تغذيتها على Q10 20 ملغ/كغ من العلف) وتم تلقيحها بـ (D78, E288)؛ المجموعة T2: تم تغذيتها على Q10 20 ملغ/كغ من العلف) وتم تلقيحها بـ (rHVT)؛ المجموعة T3: تم تغذيتها على Q10 40 ملغ/كغ من العلف) وتم تلقيحها بـ (D78, E288)؛ المجموعة T4: تم تغذيتها على (Q10) 40 ملغ/كغ من العلف) وتم تلقيحها بـ (rHVT)؛ المجموعة T5: تم تلقيحها فقط بـ (D78, E288)؛ المجموعة T6: تم تلقيحها فقط بـ (rHVT)؛ المجموعة T7: مجموعة السيطرة السلبية. تم قياس عيارات الجلوبيولين المناعي (IgY) في اليوم الخامس عشر، وكشفت النتائج أن المجموعتين T4 و T2 أظهرتا عيارات أعلى بشكل ملحوظ ($P\leq0.05$) مقارنة بالمجموعات الأخرى. بالإضافة إلى ذلك، أظهرت البيانات من اليومين 25 و33 زيادة مستمرة في مستويات (IgY) خلال فترة التجربة. وعلاوة على ذلك، تغيرت عيارات الأجسام المضادة في الدم للأمراض الفيروسية ونشاط إنزيمات الكبد AST و ALP بشكل ملحوظ في المجموعات التي تم علاجها بـ Q10 مقارنةً بالمجموعة السيطرة ($P\leq0.05$). في الختام، أظهرت المكملات الغذائية (مساعد الإنزيم Q10) بمستويات 20 ملغ/كغ و40 ملغ/كغ تحسنًا ملحوظًا في عيارات الأجسام المضادة في الدم ضد مرض التهاب غدة فابريشيا المعدي (IBD)، بينما انخفض نشاط إنزيمات الكبد خلال فترة التجربة.

الكلمات المفتاحية: لقاح الكمبورا ، أسبارتات أمينوترانسفيراز (AST) ، الفوسفاتاز القلوي (ALP).