

## Efficacy Of Some Anticoccidial Agents On Immune Response Of Chickens

Maha E. Awad-Alla<sup>1</sup> and Hanan M. F. Abdien<sup>2</sup>

<sup>1</sup> Diagnostic Lab, Vet. Hospital, Faculty of Vet. , Med., Zagazig Univ., <sup>2</sup>Dept. of Poultry and Rabbit Med., Fac. of Vet. Med., Suez Canal Univ. Egypt

### ABSTRACT

The effect of *Artemisia cina*, Semduramicin and Coccivac-B vaccine as anticoccidial agents on immune response of chickens vaccinated against Newcastle vaccine was studied.

The watery extract of *Artemisia cina* and Semduramicin potentiated immune response resulted in high specific HI antibodies NDV titer at 15<sup>th</sup> and 21<sup>st</sup> day and 28<sup>th</sup> day of age .The immuno-potential effect was clear when HI antibodies titer for Newcastle vaccine in vaccinated treated and vaccinated none treated groups are compared .An increases in the lymphoid organs ratio and index, together with total serum protein was observed either pre or post challenged with sporulated *Eimeria spp* oocysts.

Other extracts of *Artemisia cina*, and Coccivac-B vaccine did not affect significantly on chickens immune response.

This work reflected the role of different Anticoccidial treatments in protection and immuno-potential of these birds and improvement immune response against NDV vaccine.

### INTRODUCTION

Chicken coccidiosis remains one of the economically expensive and most important common diseases in modern poultry production (1, 2). The replication of *Eimeria spp* causes damage to the integrity of the intestine interfering with nutrient absorption. This result in growth depression, mortality, damages the immune system and poultry become more sensitive to different pathogens (3, 4).

Newcastle disease (ND) is the most serious disease attacking poultry populations and causing heavy economical losses in the form of high mortalities reaching 100% at any age (5). Humeral immunity is critical to protect chickens from ND, and an effective vaccine was employed to protect breeder chickens in all countries where ND was endemic (6).

The aim of this work is to investigate the effect of some anticoccidial agents (*Artemisia cina*, Semiduramicin and Coccivac-B) on the immune response of NDV vaccinated chickens.

### MATERIALS AND METHODS

#### Birds

Two hundred and seventy five Hubbard chicks of day- old were obtained from Ommat Company. Egypt. All chicks were fed

commercial ration (El Kahera Company, Zagazig, Egypt) *ad libidum*.

#### Vaccines

- a- Hitchner B1 and IB live bivalent virus vaccine IZO 1000 doses, IZO S.P.A. Brescia, Italy, containing 10<sup>9</sup> EID<sub>50</sub>/ml of live NDV B1 strain and 10<sup>6</sup> EID<sub>50</sub> / ml of live attenuated IBV mass (H120). Birds in all groups were vaccinated via eye drop at 4<sup>th</sup> day of age.
- b -Nobilis Gumboro D78 Intervet 1000 doses Boxmeer Holland , containing 10<sup>6</sup> EID<sub>50</sub> /ml of D 78 strain. All birds were vaccinated at age of 10 and 17 day via drinking water.
- c- La Sota live ND virus vaccine Intervet 1000 doses Boxmeer Holland containing 10<sup>9</sup> EID<sub>50</sub>/ml of live attenuated NDV La Sota strain. Birds were vaccinated at age of 17 days, and followed by a booster dose at age of 27 day via drinking water.
- d- Coccivac-B live oocysts vaccine for broiler vaccination comprised four species of wild type of *Eimeria* (*E.acervulina*, *E.mivata*, *E.maxima*, *E. tenella*), 1000 doses ,Schering Plough Animal Health Intervet Canada Corp .Millsboro, Delaware, USA. It was used as eye drop at 3<sup>rd</sup> day of age

The vaccines were administered according to instruction of the manufacturers.

### Anticoccidial agents

#### *Artemisia cina*

*Artemisia cina* was used as powder, watery and alcoholic extracts prepared as previously described (7).

#### Semduramicin sodium

(Aviastax)® (Pfizer U.S.A) was added in a level of 25 ppm/kg of ration.

#### *Eimeria* species

Sporulated oocysts of mixed field *Eimeria* spp. were obtained from Dept. of Parasit., Fac. of Vet. Med. Zag. Univ. Challenge was carried out at 21 day of age by administration of 7.500 Sporulated oocysts /bird, using crop tube.

### Experimental design:

Two hundred and seventy five chicks were used in this experiment. Five random chicks were slaughtered to detect maternal antibodies in the flock. The remaining chicks (270 chicks) were divided into six equal groups (each group 45 birds). Experimental groups except the 1<sup>st</sup> group (control group) received daily treatment from the 1<sup>st</sup> day up to 42 day of age. The 2<sup>nd</sup> group received powder form of *Artemisia cina* in a dose of 10 gm /kg of commercial ration (1%). The 3<sup>rd</sup> group and the 4<sup>th</sup> group received watery and alcoholic extract of *Artemisia cina* in a dose of 4 ml /kg ration, in a dose equivalent to 1% of *Artemisia cina*. The 5<sup>th</sup> group medicated with Aviastax at a dose of 25 ppm in ration. The 6<sup>th</sup> group was vaccinated with Coccivac-B at the 3<sup>rd</sup> day of age via eye drop.

Five birds from each group were slaughtered weekly for 3 weeks to obtain blood and lymphoid organs. At the 21<sup>st</sup> day of age, each group (contains 30 birds) was subdivided into two sub-groups. One of the subgroup was challenged with sporulated *Eimeria* spp oocysts and the second was left as a negative control.

### Serum samples

Five random samples were taken from sacrificed chicks at 1<sup>st</sup> day of age for the detection of ND maternal antibodies. Five

samples were then collected from the experimental chickens at 7<sup>th</sup>, 15<sup>th</sup> and 21<sup>st</sup> (pre-challenge), 28<sup>th</sup>, 35<sup>th</sup> and 49<sup>th</sup> day (post-challenge) of age. Sera were stored at -20 °C for estimation of HI antibodies against ND.

The total serum protein and albumin was estimated (8, 9). Globulin concentration was calculated from the arithmetic difference between the total protein and albumin concentration.

### Haemagglutination (HA) and Haemagglutination inhibition (HI) tests

HA and HI tests were carried out (10).

### Lymphoid organs Weight

Three birds were randomly slaughtered from each group and lymphoid organs (spleen, thymus and bursa of fabricous) were dissected, weighted and the relative weight of lymphoid organs was calculated according to the following formula:

$$\text{Lymphoid organs weight ratio} = \frac{\text{lymphoid organs weight (gm)}}{\text{Body weight (gm)}} \times 1000 \quad (11)$$

$$\text{Lymphoid organs index} = \frac{\text{lymphoid organs ratio of challenged chickens}}{\text{Lymphoid organs ratio of uninfected control}} \quad (12)$$

### Statistical analysis

The data were analyzed by using one way ANOVA (F-test). Comparison between means by using LSD (least significant difference) (13).

## RESULTS AND DISCUSSION

Anticoccidial drugs are used in chicken industry either for prophylaxis or treatment of chicken coccidiosis. Nowadays, there is no way to control coccidiosis is carried out either through vaccination and medication. Therefore, an insistence question by veterinarians and the owners of poultry farms aroused, which is, what is the effect of administration of anticoccidial agents on the immune response of chickens vaccinated against NDV? This study tried to answer this question.

The immune response to ND vaccination of birds given anticoccidial agents is summarized in Table 1.

Table 1. Mean of GM HI- antibodies titer in N.D vaccinated, treated and non challenged subgroups

Chicken groups	age	Mean of GM HI antibody titer (log2)/ week of age						
		0	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>ed</sup>	4 <sup>th</sup>	5 <sup>th</sup>	7 <sup>th</sup>
G1a	5±0.48*		2.8±0.99	4.8±0.49	6.30±0.91*	6.80±0.99	6.00±0.66	1.67±.33
G2a			3.8±0.99	4±0.58	6.33±0.66*	6.7±0.99	6.33±0.88	2.5±0.91
G3a			4.4±0.85	5.7±0.39*	6.67±0.33*	7±0.96	6.67±0.71	4±0.00*
G4a			4.3±0.66*	4.6±0.25	6.33±0.66*	6.7±0.32	5.33±0.88	3±0.81
G5a			4.5±0.05*	5.5±0.80*	7±0.99**	7.3±0.33	7±0.00	4±0.99*
G6 a			3.8±0.13*	5.33±0.88*	6.30±0.90*	7±0.99 <sup>n,s</sup>	6.30±0.99 <sup>n,s</sup>	3.66±1.45*
			S.D	S.D	S.D	N.S	N.S	S.D

G1a control vaccinated, none treated.

G2a vaccinated, treated with powder of *Artemisia cinia*

G3a vaccinated, treated with watery extract of *Artemisia cinia*

G4a vaccinated, treated with alcoholic extract of *Artemisia cinia*

G5a vaccinated, treated with Aviax

G6a vaccinated, Vaccinated with Coccivac B

All results are mean± S.E., Statistical analysis using F test

S.D: significant difference.

N.S.: non significant difference

\* Value denotes the pre experimental maternal immunity of 5 chicks

Vaccinated chickens showed lower titers one week post 1<sup>st</sup> vaccination compared with HI- titer at day one (maternal antibodies). The decrease in the titer after first vaccination was not only because it neutralize the live virus vaccine (14-16), but also because it exerted a feedback inhibition on the active antibody synthesis (17).

Then an increase in antibodies titer was observed from 2<sup>nd</sup> week up to 4<sup>th</sup> week of age. It was recorded that the antibody production increased after vaccination by 10-15 days (18, 19). The 3<sup>rd</sup> ND vaccinal dose (booster dose of ND vaccine) has led to increase of GMT at 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> week of age in all groups. These results are supported by previous study findings (6) which showed that the fourth ND vaccinal dose increased the GMT of antibodies. Thereafter, the antibodies began to decrease as the birds reached the seven weeks of age and showed titers with variable reading. Titer variations observed were 1.67±0.33, 2.5±0.91, 4±0.00, 3±0.81, 4±0.99 and 3.66±1.45 for control, birds treated with powder of *Artemisia cinia*, watery extract of *Artemisia cinia*, alcoholic extract of *Artemisia cinia*, Aviax and Coccivac-B, respectively.

The maximum GM titer was recorded for groups 3a, 5a and 6a which were treated with watery extract of *Artemisia cinia*, Aviax and

Coccivac B respectively. The increase in titer may be due to the stimulant effect of these treatments which reflected on the immune system. These results were supported by the argument that the different forms of *Artemisia cinia* had a significant protective effects on the integrity of the epithelial lining of the gastrointestinal tract which may lead to increase absorption of amino acids and other nutrients in addition to increase feed intake which improved immune system (20, 21). Similarly the administration of anticoccidial agents before vaccination of chickens against NDV had positive effect on the immune system resulted in potentiation of the immune response leading to increase of HI antibodies titer (22). The GMT HI values of vaccinated birds are considered indicators for immune status (23, 24).

Immunodepression was observed when a dose of 50 ppm of Semduaramicin or 120 mg of Salinomycin was used in ration represented by low HI antibody titers (25, 26). Similar side effect on immune response was obtained when Codrinal (27) and sulphaquinoxalin were used (28).

The effect of coccidial challenge on HI immune response of ND vaccinated chickens previously treated with anticoccidial agents is summarized in Table 2.

**Table 2. Mean of GM HI antibodies titer in N.D vaccinated ,treated and coccidial challenged subgroups**

Challenged sub group	age	Mean of GM HI antibody titer (log2)/ Day of age		
		0 week PC	1 <sup>st</sup> week PC	2 <sup>nd</sup> PC
G 1a		6.3±0.91	6.8±0.99	6±1.63
G 1b		6.3±0.91	3.6±0.88	5±0.00
G 2b		6.33±0.66	4±0.58	5.8±-0.80
G 3b		6.67±0.33	5.10±0.94	5.33±0.88
G 4b		6.33±0.66	4.25±1.03	5.33±0.88
G 5b		7±0.99	5.4±0.33	5±1.63
G 6b		6.3±0.90	4.1±0.88**	5 ±1.53
		N.S	S.D	N.S

G 1a control none treated and non challenged but vaccinated with ND.

G 1b control challenged, none treated, ND vaccinated.

G 2b challenged), treated with powder of *Artemisia cinia*

G 3b challenged treated with watery extract of *Artemisia cinia*.

G 4b challenged, treated with alcoholic extract of *Artemisia cinis*.

G 5b challenged, treated with Aviax

G 6b challenged, vaccinated with Coccivac B

All results are mean± S.E., Statistical analysis using F.test

S.D.: highly significant.

N.S: non significant difference

Pc:post challenge

Challenged subgroups with *Eimeria spp.* at the age of 21 day (G1b-G6b) showed a significant decrease in antibodies titer against ND vaccine at day 28 of age (1<sup>st</sup> week post *Eimeria* challenge) when compared with control non challenged non treated group (G1a). But at two weeks post *Eimeria* challenge increased HI titer was observed in all subgroups. From these results it could be concluded that the anticoccidial agents had immune potentiation effect on the immune system reflected on the immune response of NDV vaccine. The lower GMT values of challenged non treated chickens (G1b) were due to the effect of *Eimeria* on the integrity of the intestinal mucosa and interfering with nutrient absorption leading to impaired health condition. This explanation was supported by the argument that the immunosuppressant effect of coccidia on the immune system is due

to much of the mucosal epithelium is sloughed off and nutrient absorption is compromised which interfering with protein synthesis (1,3).

Their was a significant difference in GMT within treated subgroups at 1<sup>st</sup> week post coccidial challenge in G<sub>3b</sub> and G<sub>5b</sub> which treated with watery extract of *Artemisia cinia* and Semduramicin which showed that these antiocccidial agents had nearly similar immunostimulant effect on birds. These results were supported the finding that chickens treated with watery extract of *Artemisia cina*, had achieved good body performances (5, 21).

The mean weight of lymphoid organs, body weight, lymphoid organs ratio and index in ND vaccinated treated subgroups are shown in Table 3.

**Table 3. Mean Spleen, Bursa, Thymus, body weight and lymphoid organs ratio and lymphoid index in N.D vaccinated, treated non challenged sub-groups**

Group	Lymphoid organs	3 <sup>d</sup> week			4 <sup>th</sup> week			5 <sup>th</sup> week			7 <sup>th</sup> week		
		M.W	ratio	Index	M.W	ratio	Index	M.W	ratio	Index	M.W	ratio	Index
G1a	Bursa	1.13±0.12	1.99	-	1.56±0.09	2.23	-	11.14±0.44*	1.27	-	0.56±0.13	0.50	-
	Thymus	1.90±0.42	3.36	-	2.44±0.22*	3.47	-	2.29±0.42	2.57	-	2.56±0.64	2.28	-
	Spleen	0.63±0.03	1.11	-	0.89±0.11	1.26	-	00.95±0.25	1.06	-	0.83±0.08*	0.74	-
	M.B.W	566±22.60			704±23.13			8892.7±73.49			1123.3±36**		
G2a	Bursa	1.60±0.25	2.91	1.46	1.76±0.46	2.08	0.93	1.55±0.40*	1.66	1.30	0.62±0.15	0.48	0.90
	Thymus	2.23±0.54	4.06	1.21	2.08±0.16*	2.46	0.71	2.70±0.76	2.89	1.12	3.3±0.21	2.53	1.10
	Spleen	0.66±0.03	1.20	1.08	0.75±0.05	0.89	0.71	1.28±0.16	1.39	1.31	1.63±0.23*	1.25	1.60
	M.B.W	549.3±37.33			846±16.69			935±38.11			1300±50**		
G3a	Bursa	1.47±0.35	2.75	1.38	1.57±0.16	2.35	1.05	1.91±0.70*	1.97	1.54	0.58±0.18	0.36	0.70
	Thymus	2.63±0.75	4.92	1.46	2.08±0.120*	3.11	0.96	3.74±0.53	3.86	1.50	3.39±0.56	2.10	0.90
	Spleen	0.75±0.03	1.40	1.26	0.81±0.16	1.21	0.96	1.52±0.18	1.58	1.49	1.48±0.22*	0.92	1.20
	M.B.W	535.3±58.42			668±78.9			970±30.7			1616.7±17**		
G4a2	Bursa	1.47±0.38	2.69	1.35	1.86±0.32	2.77	1.24	1.60±0.44*	1.71	1.34	0.61±0.02	0.48	0.90
	Thymus	2.33±0.33	4.28	1.27	2.47±0.17*	3.68	1.06	3.21±0.04	3.43	1.33	3.68±0.60	2.90	1.30
	Spleen	0.70±0.10	1.29	1.16	0.84±0.08	1.25	0.99	1.28±0.04	1.37	1.21	1.30±0.25*	1.02	1.40
	M.B.W	545±58.43			671±10.53			935±38.11			1271.6±24**		
G5a	Bursa	1.13±0.24	2.11	1.06	1.68±0.14	2.19	0.98	1.48±0.6*	1.73	1.35	0.58±0.04	0.39	0.80
	Thymus	2.07±0.17	3.85	1.15	2.97±0.40*	3.88	1.12	3.17±0.12	3.70	1.44	3.35±0.42	2.23	0.90
	Spleen	0.77±0.03	1.43	1.29	0.75±0.02	0.98	0.78	1.17±0.28	1.37	1.29	1.75±0.19*	1.16	1.60
	M.B.W	537±30.07			765±51			855.7±53			1503±79.6**		
G6a	Bursa	1.13±0.17	2.12	1.07	1.41±0.18	2.14	0.969	0.84±0.16*	0.95	0.74	0.57±0.12	0.46	0.90
	Thymus	1.83±0.08	3.43	1.02	2.38±0.23*	3.63	1.04	3.43±0.29	3.90	1.52	3.2±0.17	2.59	1.10
	Spleen	0.67±0.03	1.25	1.12	0.87±0.06	1.33	1.06	0.97±0.09	1.10	1.04	1±0.01*	0.81	1.10
	M.B.W	534±26.82			655±41.6			879±42.8			1235±52.9**		
		N.S		S			S			S			

G1a control ND vaccinated, non treated.

G2a ND vaccinated, treated with powder of *Artemisia cinia*

G3a ND vaccinated, treated with watery extract of *Artemisia cinia*

G4a ND vaccinated treated with alcoholic extract of *Artemisia cinia*.

G5a ND vaccinated, treated with Aviax

G6a ND vaccinated, Vaccinated with coccivac B

All results are mean± S.E., Statistical analysis using F-test

S.D: significant difference.

N.S: non significant difference

Data obtained showed that ND vaccinated chickens treated with different anticoccidial agents had a significant increase in the weight of bursa at 3<sup>d</sup>, 4<sup>th</sup> 5<sup>th</sup> weeks of age and had a significant decrease at 7<sup>th</sup> week of age. While the weight of thymus and spleen increased gradually from the 3<sup>d</sup> till the 7<sup>th</sup> week of age. The results also showed no significant difference between ND vaccinated subgroups treated with different anticoccidial agents at the 3<sup>d</sup> week of age. While at the 4<sup>th</sup> week of age, there was a significant difference between the different subgroups only between thymus weights when compared with control. At 5<sup>th</sup> week of age, there was a significant difference between the different subgroups only between bursa weights when compared with control. High significant difference were shown between the body weight and the spleen weight in different subgroups

(G1a-G6a) which showed improvement in their values at 7<sup>th</sup> week of age with special references to health condition of birds.

On contrary, other results showed that ND vaccinated chickens treated with semduramicin and salinomycin, evoked significant decrease in the weight of bursa, spleen and thymus at 3<sup>d</sup> week of age (25,26). While at 4<sup>th</sup> week of age there were no significant changes in the weight of lymphoid organs when compared with non treated vaccinated birds

ND vaccinated treated subgroups of chickens were challenged with coccidia the results showed an increased values of lymphoid organs index by values ranged from (0.77-1.63) at 0, 1<sup>st</sup> and 2<sup>nd</sup> week post challenge Table 4. This indicated an initiation of the immune system because if the index is lower than 0.7 it is considered immunosuppression (12,29).

Table 4. Mean lymphoid organs, body weight and lymphoid organs ratio and lymphoid organs index in N.D vaccinated ,treated and challenged subgroups

Groups	Lymphoid organs	0 week P.C			1 <sup>st</sup> week P.C			2 <sup>nd</sup> week P.C		
		Mean weight	Lymphoid organs ratio	index	Mean weight	Lymphoid organs ratio	B.B index	Mean weight	Lymphoid organs ratio	B.B. index
G1a	Bursa	1.13±0.12	1.99	-	1.56±0.09	2.23	-	1.14±0.44	1.27	-
	Thymus	1.90±0.42	3.36	-	2.44±0.22*	3.47	-	2.29±0.42	2.57	-
	Spleen	0.63±0.03	1.11	-	0.89±0.11	1.26	-	0.95±0.25	1.06	-
	M.B.W	566±22.60			704±23.13			892.7±73.49		
G1b	Bursa	1.13±0.12	1.99	1.00	1.63±0.14	2.52	1.13	1.83±0.57	2.06	1.62
	Thymus	1.90±0.42	3.36	1.00	1.73±0.03*	2.68	0.77	3.33±0.68	3.74	1.46
	Spleen	0.63±0.03	1.11	1.00	0.80±0.11	1.23	0.97	1.40±0.40	1.57	1.48
	M.B.W	566±22.60			646±43.85			890±68.73		
G2b	Bursa	1.60±0.25	2.91	1.46	1.57±0.08	2.27	1.02	1.20±0.26	1.08	0.85
	Thymus	2.23±0.54	4.06	1.21	1.92±0.12*	2.74	0.79	3.2±0.45	2.87	1.12
	Spleen	0.66±0.03	1.20	1.08	0.90±0.10	1.09	0.87	0.95±0.24	0.85	0.80
	M.B.W	549.3±37.33			696.7±20.35			1114±15.03		
G3b	Bursa	1.47±0.35	2.75	1.38	1.93±0.39	2.78	1.25	1.17±0.13	1.22	0.96
	Thymus	2.63±0.75	4.92	1.46	2.16±0.37*	3.11	0.90	3.12±0.08	3.25	1.26
	Spleen	0.75±0.03	1.40	1.26	1.43±0.18	2.06	1.63	1.11±0.29	1.15	1.08
	M.B.W	535.3±58.42			694.3±36.13			961.3±56.25		
G4b	Bursa	1.47±0.38	2.69	1.35	1.53±0.08	2.05	0.92	1.82±0.15	1.56	1.23
	Thymus	2.33±0.33	4.28	1.27	2.20±0.26*	2.95	0.85	2.83±0.66	2.43	0.95
	Spleen	0.70±0.10	1.29	1.16	1.07±0.17	1.43	1.13	1.00±0.10	0.86	0.81
	M.B.W	545±58.43			746±0.29			1164.3±55		
G5b	Bursa	1.13±0.24	2.11	1.06	1.60±0.26	2.27	1.02	1.23±0.29	1.47	1.16
	Thymus	2.07±0.17	3.85	1.15	1.93±0.17	2.74	0.79	2.47±0.33	2.95	1.15
	Spleen	0.77±0.03	1.43	1.29	0.77±0.03	1.09	0.87	1.29±0.32	1.54	1.45
	M.B.W	537±30.07			704.3±57.36	2.76		838±42.56		
G6b	Bursa	1.13±0.17	2.12	1.07	1.33±0.14	1.91	0.86	0.83±0.29	1.20	0.94
	Thymus	1.83±0.08	3.43	1.02	2.10±0.35*	3.01	0.87	3.21±0.26	3.85	1.50
	Spleen	0.67±0.03	1.25	1.12	0.77±0.06	1.10	0.61	0.97±0.10	1.16	1.09
	M.B.W	534±26.82			697±34.47		*	834.7±40.25		**
		N.S			S		S			

G 1a control none treated and non challenged but vaccinated with ND.

G 1b control challenged, none treated, ND vaccinated

G 2b challenged), treated with powder of *Artemisia cinia*

G 3b challenged treated with watery extract of *Artemisia cinia*.

G 4b challenged, treated with alcoholic extract of *Artemisia cinia*.

G 5b challenged, treated with Aviax

G 6b challenged, vaccinated with Coccivac B

Means within the same column carrying different letters are significant at ( $p < 0.05$ )

All results are mean± S.E., Statistical analysis using F test.

S.D.: highly significant.

N.S: non significant difference

The B/BW ratio raised until the fourth week and declined from there to the end of the study (30). Plum powder and safflower gave similar results to *Artemisia cinia* extracts when poultry were infected with any parasite by stimulating spleen immune cell production and body weight gain (31).

Data obtained in Table 5 show a significant gradual increase in total serum protein concentration in all subgroups at the 4<sup>th</sup>, 5<sup>th</sup> and 7<sup>th</sup> week of age. This increase was due to the increase in globulin fraction. This improvement of total proteins and globulin values probably reflect the improved digestion

of proteins and absorption of amino acids as well as improved liver function (5, 20, 21).

Similarly the biostimulant effect of Ultra-Natural plus and *Nigella Sativa* and maduramicin induced an increase in gamma globulin and total serum protein were reported (32, 33).

However, the use of Codrinal, Lasalocid, Semduramicin, and Salinomycin as anticoccidial drug post NDV vaccine elicited significant decrease in globulin values but not the serum levels of total proteins, albumin or albumin/ globulin ratio (25- 27,34) .

**Table 5. Serum albumin, globulin and total protein (mg/dl) and albumin/ globulin ratio in ND vaccinated, treated and non challenged subgroups**

Groups	Parameter	3 <sup>d</sup> week	4 <sup>th</sup> week	5 <sup>th</sup> week	7 <sup>th</sup> week
G1a	Albumin	1.42± 0.13	1.46± 0.13	1.54± 0.14	1.63± 0.04
	Globulin	1.66±0.15	1.74± 0.20	1.56± 0.02	2.44± 0.22
	T. protein	3.08± 0.27	3.20± 0.19	3.10±0.19	4.07± 0.37
	A/G ratio	0.86±0.14	0.84± 0.16	0.99±0.08	0.67± 0.05
G2a	Albumin	1.28±0.08	1.40±0.13	1.53± 0.19	1.70± 0.08
	Globulin	1.40± 0.07	1.92±0.05	1.56±0.18	2.27± 0.21
	T. protein	2.68±0.01	3.32±0.25	2.92± 0.24	3.97± 0.01
	A/G ratio	0.91±0.01	0.73±0.08	0.98± 0.11	0.75± 0.09
G3a	Albumin	1.42±0.17	1.70± 0.09	1.81± 0.03	1.92± 0.26
	Globulin	1.52± 0.15	1.72± 0.11	1.92± 0.12	1.98±0.22
	T. protein	2.94± 0.31	3.42±0.12	3.73± 0.24	3.90 ± 0.30
	A/G ratio	0.93± 0.03	0.99± 0.03	0.94± 0.09	0.97± 0.11
G4 a	Albumin	1.28±0.24	1.28± 0.14	1.69± 0.07	1.68 ± 0.10
	Globulin	1.94± 0.08	1.58± 0.09	1.88± 0.15	1.96± 0.16
	T. protein	3.22± 0.25	2.86± 0.28	3.57± 0.35	3.64± 0.01
	A/G ratio	0.66± 0.23	0.81±0.11	0.90±0.07	0.86± 0.12
G5 a	Albumin	1.06± 0.13	1.34± 0.16	1.36± 0.14	1.66± 0.09
	Globulin	1.62± 0.21	1.66± 0.12	1.68± 0.07	1.68± 0.08
	T. protein	2.68± 0.33	3.00± 0.15	3.04± 0.63	3.34± 0.20
	A/G ratio	0.65± 0.08	0.81± 0.12	0.80± 0.14	0.99±0.06
G6a	Albumin	0.96± 0.21	1.10± 0.08	1.58± 0.13	1.52± 0.08
	Globulin	1.38± 0.11	1.30± 0.07	1.58±0.25	1.88± 0.04
	T. protein	2.34± 0.26	2.40± 0.14	3.16± 0.20	3.40± 0.01
	A/G ratio	0.70± 0.17	0.85± 0.04	1.00± 0.0	0.81± 0.05
		N.S	S	S	S

G1a control ND vaccinated non challenged.

G 2a treated with powder of *Artemisia cinia*

G 3a treated with watery extract of *Artemisia cinia*

G 4a treated with alcoholic extract of *Artemisia cinia*.

G 5a treated with Aviax

G 6a Vaccinated with Coccivac B

All results are mean± S.E., Statistical analysis using F-test.

S.D: significant difference.

N.S.: non significant difference

In the current study two weeks post coccidial challenge resulted in a significant increase in the total serum protein concentration and Globulin fraction in all subgroups (G1b-G6b) Table (6).

Similar results were obtained by using narasin-nicarbazin combination in birds

experimentally infected with *E. tenella* (35). The consequently reduction in the protein concentration post *E. tenella* and *E. acervulina* challenge and treated with semduramicin and salinomycin was accused to the decrease in protein retention and changes in the post-absorptive protein metabolism of the host (36).

Table 6. Serum albumin, globulin and total protein (mg/dl) and albumin/ globulin ratio in ND vaccinated, treated and challenged subgroups

Groups	Parameter	0 week PC	1 <sup>st</sup> week PC	2 <sup>nd</sup> week PC
G1a	Albumin	1.42± 0.13	1.46± 0.13	1.54± 0.14
	Globulin	1.66±0.15	1.74± 0.20*	1.56± 0.02*
	T. protein	3.08± 0.27	3.20± 0.19	3.10±0.19
	A/G ratio	0.86±0.14	0.84± 0.16	0.99±0.08
G1b	Albumin	1.42± 0.13	1.24± 0.09	1.38± 0.11
	Globulin	1.66±0.15	1.54± 0.15*	1.60± 0.14*
	T. protein	3.08± 0.27	2.78± 0.16	3.08±0.25
	A/G ratio	0.86±0.14	0.81± 0.11	0.896±0.09
G2b	Albumin	1.42±0.17	1.10±0.13	1.50±0.15
	Globulin	1.52± 0.15	1.18± 0.05*	1.86±0.12*
	T. protein	2.94± 0.31	2.28±0.18	3.36±0.13
	A/G ratio	0.93± 0.03	0.93±0.01	0.81±0.08
G3b	Albumin	1.42±0.17	1.30± 0.07	1.36± 0.17
	Globulin	1.52± 0.15	1.82± 0.15*	2.20± 0.13*
	T. protein	2.94± 0.31	3.12±0.16	3.56± 0.13
	A/G ratio	0.93± 0.03	0.71± 0.11	0.62± 0.09
G4b	Albumin	1.28±0.24	1.28± 0.14	1.68± 0.07
	Globulin	1.94± 0.08	1.58± 0.09*	2.00± 0.15*
	T. protein	3.22± 0.25	2.86± 0.14	3.68± 0.11
	A/G ratio	0.66± 0.23	0.81±0.11	0.84±0.07
G5b	Albumin	1.06± 0.13	1.10± 0.16	1.40± 0.14
	Globulin	1.62± 0.21	1.54± 0.12*	1.60± 0.07*
	T. protein	2.68± 0.33	2.64± 0.17	3.00± 0.24
	A/G ratio	0.65± 0.08	0.71± 0.12	0.88± 0.14
G6b	Albumin	0.96± 0.21	1.00± 0.08	1.40± 0.13
	Globulin	1.38± 0.11	1.10± 0.07*	1.68±0.25*
	T. protein	2.34± 0.26	2.10± 0.16	3.08± 0.21
	A/G ratio	0.70± 0.17	0.91± 0.04	0.83± 0.04
		N.S	S	S

G 1a control none treated and non challenged but vaccinated with ND.

G 1b control challenged, none treated, ND vaccinated.

G 2b challenged), treated with powder of *Artemisia cinia*

G 3b challenged treated with watery extract of *Artemisia cinia*.

G 4b challenged, treated with alcoholic extract of *Artemisia cinia*.

G 5b challenged, treated with Aviax

G 6b challenged, vaccinated with Coccivac B

Means within the same column carrying different litters are significant at ( $p < 0.05$ )

All results are mean± S.E., Statistical analysis using F-test.

S.D.: highly significant.

N.S: non significant difference

From these results we can concluded that the watery extract of *Artemisia cinia* and Semiduramicin have a significant effect on stimulation of the immune response of ND vaccinated chickens. So they could be used as safe anticoccidial agents.

## REFERENCES

1. Conway, D P and Mc Kenzei, M E (1991): Poultry coccidiosis diagnostic

and testing procedures. 2nd Ed., Pfizer Inc. New York.

2. Williams, RB, Carlyle, WW, Bond, W W, Brown, DR (1999): The efficacy and economic benefits of vaccine, in commercial trials with standard broiler chickens in Paracox, a live attenuated anticoccidial the United Kingdom. Internati. Jour. for Parasitol. Oxford, .29: 341-355.

3. **Hyun, S L (2002):** Coccidiosis: This poultry disease's impact is anything but paltry. Agricultural research service. 301:504-508.
4. **Tipu, AM, Pasha, TN, and Ali, Z (2002):** Comparative efficacy of salinomycin sodium and neem fruit (*Azadirachta indica*) as feed additive anticoccidial in broilers. Internati. Jour. Poult. Sci. 1: 91-93.
5. **Alexander, D J (1997):** Newcastle disease and other avian paramyxoviridae infection. In :Diseases of poultry, 10 th ed. B.W.Calnek, ed. Iowa State University Press Ames.
6. **Ali, D S and Abdul-Aziz, T A (1991):** Effect of repeated vaccination on the antibody response of broilers against Newcastle disease. Indian Jour of Animal Sci. 61:357- 359.
7. **Eid, AM, El-Said, AE, El-Dieb, MM and Awad- Alla, ME (2004):** Anticoccidial efficacy of *Artemisia cina* and its extract in broiler chickens. Zag. Vet. J.Vol .32.No2 pp 1-14.
8. **Coles, E (1986):** Veterinary Clinical Pathology, 4<sup>th</sup> ed., W. B. Sanders Com., Philadelphia, London, Toronto, Mexico city, Sydney, Tokyo, Hong Kong
9. **Fukata, T, Komba, Y, Sasai, K, Baba, E and Arakawa, A (1997):** Evaluation of p:asma chemistry and haematolglcal study on chickens infected with *Eimeria tenella* and *Eimeria acervulina*. Veterinary Record .141,44-46.
10. **Villegas, P (1991):** HI test for NDV. In laboratory manual for Avian virus diseases.
11. **Halouzka R J (1991):** Morphological expression of immunosupression in poultry . Acta Vt.Brono. 60:271-27.
12. **Lucio, B, Hütchner, S B (1979):** Infections bursal diseases emulsified vaccine: effect upon neutralizing antibody level in the Dam and subsequent protection of the progeny. Avian Dis.23:466-478.
13. **SPSS (2001):** SPSS computer program (2001).
14. **Solano, W, Giambrone, J J, Williams, J C, Lauerman, L H, Panangala, V S and Garces, C (1986):** Effect of maternal antibody on timing of initial vaccination of young hite leghorn chickens against NDV. Aviaiv Dis. 30:684-652.
15. **Wyeth, P J and Chettle, N J (1990):** Use of NDV vaccines in chicks with maternally derived antibodies. Vet.Rec.126:577-578.
16. **Jong, L, Young, J K, Young, K J and Young, J I (1999):** Studies on the immunization against Newcastle disease virus in ogol chicken. J.Vet.Serv.22:37-42.
17. **Gordan, R F, Jordan, F T W (1982):** Avian immune system. Poult. Dis. 2<sup>nd</sup> edition:328-338.
18. **Bayoumi, HAA (1997):** Immune response in chickens after simultaneous vaccination against Newcastle and Gumboro disease viruses. M V Sc. Tthesis Fac. Of Vet. Med., Zagazig University.
19. **Dashab, G, Sadeghi, H, and Mehri, M (2007):** Performance and humoral immune response to Newcastle disease in broiler chickens. Jour of Animal and Vet Advan.6: 451-453.
20. **Tan, R X; Zheng, W F and Tang, H Q (1998):** Biologically active substances from the genus *Artemisia* ( Review). Planta Med. 64:295-302.
21. **Awad-Alla, M E., El-said, AE., Abdien, H M and El-Deib, MM (2008):** Comparative study on the effect of watery extrct of *Artemisia cina*; Semiduramicin and Coccivac B as anticoccidial agents in broiler chickens. Zag. Vet. Jour .Vol. 36:182-196.
22. **Box, P G and Furamengen, I G S (1975):** Newcastle disease antibody levels in chickens after vaccination with oil emulsion adjuvant killed vaccine. Vet . Rec . 96 : 108-111.

23. **Phillips, J M (1973):** Vaccination against Newcastle disease: An Assessment of haemagglutination inhibition titers obtained from field samples .*Vet. Rec.*93:577-583.
24. **Allan ,W H , Lancaster, J E , and Toth, B (1978):** Newcastle disease vaccines, their production and use. FAO Animal Production and Health series No.10 Rome.
25. **El-Kahky, M A (1991):** Influence of the anticoccidial semduramicin on the immunological response to ND vaccine in chickens .*Athesis . Ph.D (Pharma). Fac. Vet. Med., Zaga.Univ.*
26. **Shalaby, M A , El-Sanousi, A A , Yehia M M and Reda, I M ,(1993):**The effect of salinomycin on the immune response of chicks. *Dtsch. Tieraztl. Wschr.,* 100:182-185
27. **Azeez, M A and Paily, E P (1985):** effect of codrinal on antibody response to Newcastle disease vaccination in chicks. *Kerala Jour. Vt. Sci.* 16:81-88.
28. **Rzedicki, J , Cybulska, R , Trawinska, B and Jaworska, A (1991) :** Effect of some antibacterial agent on the immune response and intestinal mucosa of chickens .*Zeszyty Naukowe Akademii Rolniczej We Wroclawiu, aterynaria.* 49:179-189.
29. **Allam H H, Bayoumie AA Malhat SM and Mohamed IA(2008):** Effect of Toltrazuril and Amprolium plus in broiler chickens infected with coccidiosis . *Assiat Vet.Med.Jour.*54:253-266.
30. **Perozo, F M , Jesús, N , Mavarez, Y, Arenas ,E , Serje, P Y And Briceno , M (2004):** Caracterizaction Morfométrica De Los Órganos Linfoides En Pollos De Engorde De La Línea Ross Criados Bajo.Condiciones De Campo En El Estado Zulla, Venezuela . *Rc,* vol.14: 217-225.
31. **Ann P (2009):** The Poultry Pantry: Plums, Probiotics, Safflower, and Tea. Issue of *Agricultural Research magazine.*
32. **El-Sawah, A A , Tamam, S M ,Mohamed, S H and Kandell, M A (2006):** Immunological and biochemical effects of nigella sativa and ultra natural plus on turkeys vaccinated with Newcastle and turkey rhinotracheitis virus vaccines. *Vet. Med. J. Giza.* 54(2): 461-471.
33. **Abd El-Aziz ,M I and Agag, A E ( 1991):** Effect of lasalocid on immune response to NDV vaccine in chickens. *Vet.Med. Res.,* 1:171-182.
34. **Abd Al-Lateif ,A E (1993):** Side and latent effect of the concurrent use of maduramicin and avotan on broilers. Thesis for Ph. D (pharmacology), *Fac. Vet. Med., Zaga.Univ.*
35. **Mahmoud, M T (2000):** Some pharmacological studies on some recent anticoccidial agents in poultry. A thesis for M.V. Sc .(pharma) *Fac. of Vet.Med.Zag. Univ.*
36. **Conway, D P Johnson J K , Gunonnet V, Long P I and Smothers C D (1993):** Efficacy of semduramicin and salinomycin against different stages of *Eimeria- tenella* and *E. acervulina* in the chickens .*Vet.Parasitol.*45:215-229.

## المخلص العربي

### تأثير بعض مضادات الكوكسيديا على الاستجابة المناعية للطيور

مها عوض الله السيد<sup>١</sup>، حنان محمد فتحي عابدين<sup>٢</sup>  
 (١) مستشفى كلية الطب البيطري- جامعة الزقازيق- دواجن  
 (٢) قسم طب الطيور والأرانب – كلية الطب البيطري- جامعة قناة السويس

نظرا لخطورة مرض النيوكاسل في قطعان الدواجن أجريت هذه الدراسة لمعرفة تأثير الشيخ الخراساني و عقار السيميديوراميسن ولقاح الكوكسيديا (الكوكسيفاك- ب) كمضادات للكوكسيديا على استجابة الجهاز المناعي في بداري التسمين المحصنة ضد هذا المرض.

وقد أدى استخدام المستخلص المائي للشيخ الخراساني و عقار السيميديوراميسن إلى تنشيط الاستجابة المناعية نتج عنه زيادة في معدل الأجسام المناعية في اختبار التلازن ضد مرض النيوكاسل في عمر ١٥ و ٢١ و ٢٨ يوم و قد ظهر هذا التأثير واضحا عند مقارنه معدل هذه الأجسام المناعية بين الطيور المحصنة والمعالجة والطيور المحصنة و الغير معالجه و نتج أيضا زيادة في أوزان الأعضاء الليمفاوية بالإضافة إلى زيادة في نسبة البروتين الكلية خصوصا نسبة الجلوبيولين سواء قبل أو بعد العدوى الأصطناعية بالكوكسيديا. بينما أضافه الشيخ الخراساني و المستخلص الكحولي للشيخ الخراساني ولقاح الكوكسيديا (الكوكسيفاك- ب) لم يسفر عنه تغير معنوي في الاستجابة المناعية أو في أوزان الأعضاء الليمفاوية بالإضافة إلى عدم تأثر نسبة البروتين الكلية تأثيرا معنويا بعد تحصين هذه الطيور ضد مرض النيوكاسل سواء قبل أو بعد العدوى الأصطناعية بالكوكسيديا.

وهذا العمل يعكس دور المعاملات المختلفة لمضادات الكوكسيديا المختلفة المنشأ في حماية و تحفيز الجهاز المناعي وتحسين معدل الأجسام المناعية ضد مرض النيوكاسل لهذه الطيور.