

STUDIES ON SEVERE OUTBREAKS OF INFECTIOUS BURSAL DISEASE

IV-BIOCHEMICAL CHANGES IN THE SERUM OF EXPERIMENTALLY INFECTED CHICKENS

BY

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SUMMARY

Studies on the clinical, pathological and biochemical changes during the course of an experimental infectious bursitis in chickens. Clinical signs included depression, anorexia, and deaths, were first recorded. The bursa of fabricius were slightly enlarged on the 3rd and 4th day post infection. No precipitins against IBDV could be detected by AGPT.

Infected chickens revealed a significant elevation of prothrombin time, alanine aminotransferase, creatinine and uric acid. Other parameters were declined like serum total proteins, alkaline phosphatase, total cholesterol and potassium levels.

INTRODUCTION

Infectious bursal disease (IBD or Gomboro disease), an acute contagious viral disease of young chicks, was first described by Gosgrove, (1962).

Although the bursa of fabricius is acknowledged as the primary target organ for IBDV infection and resulting lesions (Muller et al., 1979 and Kaufner and Weis, 1980), changes do occur in other organs; including spleen, thymus, kidney and liver have been reported by (Cheville, 1967; Helmboldt and Garner, 1964; Henry et al., 1980 and Siller, 1981). Since the emergence of recent outbreaks of infectious bursal disease (IBD) in vaccinated flocks in Egypt (El-Batrawi, 1990), the application of many vaccination programmes, in some instances, failed to protect the vaccinated flocks from field IBDV challenge.

The experienced high mortality, in spite of multiple vaccinations, warranted some attention toward the biochemical changes during the course of the disease that associated with the haemorrhagic and nephropathic lesions of the disease.

The objective of this study was to determine some of the biochemical changes that occur as a result of INCV infection.

MATERIAL AND METHODS

Experimental chickens: Cockerels of an egg-type white layers from parent vaccinated twice with live IBDV vaccines during the period and boosted at 18 weeks of age with an IBD oil adjuvanted vaccine, were used in this study.

IBD field virus (IBD - FV): An IBD - FV, isolated from the bursa collected from naturally infected, 43 - day - old white layers flock (El-Batrawi, 1990) was used for experimental infection of the cockerels. The IBD - FV was detected in the bursa of infected birds using the AGPT as mentioned by El-Batrawi, (1990).

Experimental infection: Two groups of 50 cockerels were kept in separate isolated rooms up till 40 days of age, where birds of the first group were infected intraocularly with 104.6 IBU - FV per

bird (infected group). Birds of the second group were kept to serve as non-infected controls (control group). The birds were kept under observation for 5 days and clinical manifestation, mortalities and gross lesions were recorded (Table 1). When the first signs of the disease appeared, five birds from each group were taken daily at random and individually weighed before being sacrificed for blood collection, post-mortem examination, and bursal weighing.

Blood samples: Individual citrated and uncitrated blood samples were collected daily, from the first signs of the clinical disease. The plasma of the citrated blood were used for determination of prothrombin time using a commercial reagent. "Calcium thromboplastin" according to Quick method (1955), while serum samples were used for detection of precipitins by agar gel precipitation test (AGPT) as reported by Hirai and Shimakura (1972), and also for measuring of other biochemical parameters. Serum total proteins were determined by the method of Weichselbaum (1946), serum alanine aminotransferase and alka-

line phosphatase activities by the methods of Reitman and Frankel (1957) and Kind and Kling (1954) respectively. Creatinine concentration by Jaffe reaction (Seeling and Wust, 1969) and uric acid was determined by the method of Caraway (1955), and total cholesterol according to Zlatkis et al., (1953). Serum sodium and potassium levels were determined by flame photometer 410 complying with a technique described by Varley et al. (1980). Bursa/body weight ratio were calculated as follows:

$$\frac{\text{Bursa weigh} \times 1000}{\text{body weight}}$$

RESULTS AND DISCUSSION

Clinical examinations: Clinical signs and deaths were first recorded 24 and 48 hours respectively after infection. The clinical signs and post mortem lesions were typical to IBD and similar to those described by (Gosgrove, 1962, Faragher, 1972, and Ide and Steveson, 1972, and Becht, 1980). A total of 24 birds (48%) died with the highest

Table (1): Mortality, gross lesions and bursal/body weight ratio in chickens experimentally infected with IBD field virus.

Days post infection	Groups	No of dead birds	Pathological changes in sacrificed birds					Bursal/body wt ratio
			H.M	H.P	K.L	L.L	B.L	
1	Infected	0/50	1/5	0/5	0/5	0/5	2/5	4.5
	Control	0/50	0/5	—	—	—	—	4.6
2	Infected	2/50	3/5	0/5	1/5	2/5	2/5	5.3
	Control	0/50	—	—	—	—	—	4.7
3	Infected	6/50	3/5	2/5	5/5	2/5	3/5	5.9
	Control	0/50	—	—	—	—	—	4.4
4	Infected	9/50	4/5	0/5	4/5	2/5	4/5	5.7
	Control	0/50	—	—	—	—	—	4.7
5	Infected	7/50	2/5	1/5	4/5	1/5	4/5	4.9
	Control	0/50	—	—	—	—	—	4.4

H.M. = haemorrhage on muscles

H.P. = haemorrhage on proventriculus

K.L. = kidney lesion

L.L. = liver lesion

B.L. = bursal lesion

Table (2): Prothrombin time, ALT, ALP, Total proteins and Total cholesterol in serum of infected chickens in comparison with the corresponding control group.

Parameters	Groups	Days				
		day-1	day-2	day-3	day-4	day-5
Prothrombin time(second)	Infected	12.00	12.36	20.20 ^{**}	19.00 ^{**}	17.40 ^{**}
		±0.18	±0.35	±1.26	±0.27	±0.38
	Control	11.80	12.02	12.60	12.70	12.80
		±0.12	±0.36	±0.27	±0.22	±0.23
ALT (U/L)	Infected	12.20	12.60	26.50 ^{**}	21.40 ^{**}	15.80 ^{**}
		±0.20	±0.51	±2.08	±0.80	±1.16
	Control	11.50	11.60	12.40	11.60	11.00
		±0.31	±0.24	±0.24	±0.60	±0.45
ALP (KAU/100ml)	Infected	860.00	848.50	819.80 ^{**}	714.50 ^{**}	639.80 ^{**}
		±28.84	±16.89	± 6.57	±36.38	±17.21
	Control	875.20	878.60	865.00	887.90	870.00
		±17.67	±15.49	±13.70	±22.17	± 5.68
Total proteins(gm%)	Infected	3.44	3.80 [*]	3.30 ^{**}	3.72	3.78
		±0.18	±0.13	±0.13	±0.19	±0.09
	Control	3.60	4.30	3.96	3.94	3.84
		±0.21	±0.16	±0.07	±0.14	±0.15
Total cholesterol (mg%)	Infected	132.60	125.60 ^{**}	90.00 ^{**}	100.00 ^{**}	110.40 ^{**}
		±2.79	±2.06	±3.08	±3.86	±2.98
	Control	140.00	145.00	150.00	136.20	128.80
		± 5.83	± 3.00	± 4.27	± 2.71	± 1.53

* Significant different from control mean (P < 0.05).

** Significant different from control mean (P < 0.01).

Table(3): Creatinine,Uric acid,Sodium and Potassium levels in serum of infected chickens in comparison with the corresponding control group.

Parameters	Groups	Days				
		day-1	day-2	day-3	day-4	day-5
Creatinine (mg%)	Infected	0.078 ±0.006	0.083 ±0.004	0.090 ^{**} ±0.004	0.102 ^{**} ±0.01	0.132 ^{**} ±0.019
	Control	0.054 ±0.012	0.062 ±0.01	0.059 ±0.01	0.062 ±0.009	0.064 ±0.009
Uric acid (mg%)	Infected	5.00 ±0.344	5.70 ±0.261	9.10 ^{**} ±1.07	7.20 [*] ±0.660	5.80 ±0.451
	Control	4.80 ±0.13	5.00 ±0.415	5.00 ±0.540	5.20 ±0.484	4.90 ±0.306
Sodium (mEq/L)	Infected	156.0 ±1.70	155.00 ±1.045	157.00 ±1.303	156.60 ±1.208	159.00 ±0.894
	Control	155.80 ±0.514	154.60 ±1.029	156.20 ±1.655	155.50 ±1.140	157.50 ±1.048
Potassium (mEq/L)	Infected	8.50 ±0.219	8.00 ±0.609	5.20 ^{**} ±0.254	5.60 ^{**} ±0.270	5.70 ^{**} ±0.187
	Control	8.65 ±0.220	8.90 ±0.170	9.00 ±0.281	9.20 ±0.230	8.85 ±0.202

* Significant different from control mean (P < 0.05).

** Significant different from control mean (P < 0.01).

(Table 1). The bursa were slightly enlarged on the 3rd and 4th day post-infection and had the typical haemorrhagic lesion characteristic to IBD. The lesion scores are shown in table 1. No precipitins against IBDV could be detected by AGPT.

Biochemical changes: Tables 2 and 3 show the biochemical changes in sera of sacrificed birds during the five days post infection. The biochemical changes related to liver functions are especially prominent for prothrombin time, alanine amino-transferase, alkaline phosphatase, total proteins and total cholesterol. The changes in the first two parameters, were evident with a highly significant increase ($P < 0.01$) from the 3rd day post infection, while total proteins, ALP and total cholesterol were significantly decreased ($P < 0.01$) from the 2nd day post infection (Tables 2 & 3). Although, Skeeles et al., (1978) recorded an increase in clotting time on the day 3 post infection.

biochemical changes related to kidney functions as indicated by a markedly significant decrease ($P < 0.01$) in serum potassium level from the 3rd day, while uric acid and creatinine were increased markedly in the serum on the day 3 post infection ($P < 0.01$). Although, Ley et al., (1983) reported a increase in serum uric acid on the 3rd day of infection, however according to the available literature, the present study is the first to study the daily biochemical changes in some parameters related to liver and kidney functions during the course of experimental IBD.

The present results indicated that IBD causes severe liver necrosis and kidney damage together with retardation in clotting time and the use of some electrolytes such as potassium and diuretics in the form of bicarbonates or citrates together with some blood coagulants could be beneficial in reducing the losses due to this disease.

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