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Dept. of Poultry Diseases,
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EPIDEMIOLOGICAL STUDIES ON GUMBORO DISEASE IN UPPER EGYPT*

(With 3 Tables and 2 Figures)

By
*M. SAIF-EDIN; M. ALY; AZHAR, M. ABD EL-AZIZ** and FATMA, M. MOHAMED***
** Animal Health Research Institute, Assiut.
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دراسات وبائية على مرض الجمبورو فى صعيد مصر

مصطفى البكري سيف الدين، محمد محمود علي ، أزهار محمد عبدالعزيز ، فاطمة محمد مختار

اجريت هذه الدراسة لاستيضاح وبائية التهاب حويصلة فابريسى المعدى فى كتاكيت السلالات المحلية وكتاكيت انتاج اللحم خلال الفترة من ١٩٩٥-١٩٩٨ فى محافظات المنيا وأسيوط وسوهاج وقد تبين من دراسة ١١٠ وباء أن المرض ظهرفى كتاكيت السلالات المحلية فى عمر مبكر (١٨-٢٨ يوم) عنه فى كتاكيت انتاج اللحم (٢٤-٣٢ يوم) وكانت أعراض المرض أكثر وضوحا ولفترة بقاء فى كتاكيت السلالات المحلية عنها فى كتاكيت انتاج اللحم وكانت نسبة الكتاكيت المريضة تتراوح بين ١٥- ٨٠% وبنسبة نفوق تتراوح بين ١.٥-٣٠% وقد تم تأكيد جميع أوبئة المرض تحت الدراسة باستخدام اختبار الترسيب فى الأجار للكشف عن الفيروس المسبب فى مزيج حويصلة فابريسى. هذا وقد نجحت عملية عزل الفيروس باستخدام الحقن على الغشاء السليى السجقى لأجنة بيض الدجاج مؤدية الى معدلات نفوق منخفضة خاصة فى التمرير الأول وكانت معزولات الفيروس مقاومة للأثير والكلوروفورم والحرارة (٥٦ درجة لمدة ١-٣ ساعات). أما بالنسبة الى قياس مستوى الاجسام المضادة المنحدرة من الامهات التى سبق تحصينها ضد مرض التهاب حويصلة فابريسى المعدى باستخدام اختبار الاليزا فى كتاكيت عمر واحد فكان مستوى هذه الاجسام المضادة عاليا فى كتاكيت انتاج اللحم عنه فى كتاكيت السلالات المحلية وكانت موزعة بنسب متفاوتة فى الافراد المختلفة كما كان معدل انخفاض هذه الاجسام المضادة عاليا فى الاخيرة.

* *Part of thesis, presented to Fac. Vet. Med. Assiut University*

SUMMARY

The present investigation was undertaken to investigate some aspects related to epizootiology of infectious bursal disease (IBD) outbreaks, isolation and identification of IBD virus, waning of maternal antibodies in both broiler and native flocks. The investigation of 110 outbreaks in native breed and broiler chicks during the period of 1995/1998 in El-Menia, Sohag and Assiut Provinces revealed that IBD outbreaks occured at earlier age (18-28 days) in native flocks than in broiler flocks (24-32 days). The clinical symptoms were more pronounced in native flocks than in broilers which demonstrate more prolonged course of the disease. Morbidity and mortality rates were variable ranging from 15-80% and 1.5-30% respectively. All investigated outbreaks were confirmed by IBDV antigen detection in bursal homogenates from acutely affected birds using the AGPT. Chicken embryo inoculation (CAM) resulted in low degree of mortality especially in the initial passage. The IBDV is ether and chloroform resistant and thermostable after1,2 & 3 hours at 56°C. The level of maternal antibodies in one day-old chicks from parent flocks vaccinated with IBD virus vaccines is higher and less uneven in broiler than that in native breed chicks. The titer of maternal antibodies as indicated by ELISA was zero at 5 weeks of age in broiler chicks and at 4 weeks of age in native breed chicks.

Key words: Gumboro Disease Upper Egypt.

INTRODUCTION

Infectious bursal disease (IBD) was first recorded by Cosgrove (1962) who named the disease, avian nephrosis because of the extreme kidney damage of the affected birds. In the same year, Winterfield et al. (1962) isolated and identified the causative virus and justified the name infectious bursal agent (IBA) for it. Hitchner (1970) proposed the term infectious bursal disease as the name of the disease causing specific lesions of the cloacal bursa. Since this time, the disease was reported in many parts of the world.

IBD is caused by a bisegmented RNA virus that is classified as a member of Birnaviridae family (Dobos et al., 1979; Hermann et al., 1979; McDonald, 1979; Muller et al., 1979; Okoye, 1984). There are two serotypes of IBDV, serotype I and II (McFerran et al., 1980). Sertype I is

pathogenic for chickens, whereas serotype II mainly infected turkeys (Ismail et al., 1988). IBDV is very stable. It survived heating up to 56°C for 5 hours, not affected by pH 2, resisted treatment with ether and chloroform (Benton et al., 1967).

Ide and Stevenson, 1973; Okoye, 1984 and Sultan, 1995) reported that IBD usually occurred in chickens between 3-10 weeks of age while Onunkwo, (1995) reported an outbreak in chicks of 9-13 days of age. Moreover, Ley et al. (1979) reported an outbreak of IBD in 14-15 weeks old chickens.

The mortality rates ranging from 4%-21% in commercial broilers, 4%-62% in commercial layer pullets (Sultan, 1995).

In Egypt, IBD was first reported by El-Sergany et al. (1974). Then the disase was studied by many authors, Ayoub and Malek(1976); Bastami (1980); El-Zanaty (1982); Hegazy, (1983); Mousa et al. (1983); Ahlam Farghaly (1989); Khafagy et al. (1990); Mousa and Saif-Edin (1990); Ahmed (1991); El-Sanousi et al. (1992); El-Shorbagy (1992); Madbouly et al. (1992) and Saif Edin et al. (1996).

The aim of this work is to study:

1- The epizootiology of different IBD outbreaks in broiler and native breed chicks.

2- Determination of IBDV maternal antibodies in broiler and native breed chicks.

3- Waning of the IBDV maternal antibody.

MATERIAL and METHODS

Field outbreaks:

During 1995, 1998 IBD outbreaks were investigated in 110 chicken flocks in El-Menia, Assiut and Sohag Governorates (80 broiler flocks and 30 native breed flocks). Data concerninig the history of vaccination, age at onset of the disease, morbidity and mortality rates were collected. Birds were subjeted to clinical and post-mortem examination and samples were collected for virus detection.

Specimens for virus isolation and identification:

Bursae collected from diseased birds were subjected to antigen detection using agar gel precipitation (AGP) test and for virus isolation in chicken embryos. Viral isolates were subjected to physicochemical and serological identification by chloroform and ether sensitivity, heat stability and AGP test using polyclonal and monoclonal antibodies.

IBD reference antisera:

Polyclonal antisera against standard serotype 1 reference vaccinal strain D 78 as well as Winterfield 2512 strain IBDV were prepared in California rabbits by 3 S/C and 2 I/M injections of virus suspensions in incomplete freund's adjuvant at 3 weeks intervals. Antiserum was obtained 3 weeks after the last injection according to the method of Tanimura et al. (1995). Monoclonal antibodies designated 8, 179, 42, 57, R63 and B69 were kindly supplied by Prof. Dr. S.A. Kleven Poultry Disease Research Center, Athens, Georgia, USA.

Preaparation of antiserum against IBDV:

4-5 weeks old, 5 hyline chickens were inoculated 3 times intraocularly, with one week interval, 0.05 ml of a clarified 10% W/V bursal homogenate known to contain 10^3 EID_{50}/dose. 2 chickens were kept uninocualted. 28 days postinoculation, all the inoculated and uninocualted chickens were bled and the sera were inactivated at 56°C for 30 min. and stored at -20°C until used as positive and negative antisera.

Chicken embryos inoculation:

10-days old embryonated chicken eggs, provided by the poultry fram of Fac. of Agriculture, Assiut University were used for virus propagation by inoculation via chorioallantoic membrane (CAM).

Serum samples:

Blood samples were collected from one day old broiler and native chicks from El-Menia, Assiut and Sohag Governorates and serum samples were separated in sterile glass vials and kept in a frozen state -20°C untill subjected to serological examination using ELISA test.

Ether sensitivity test:

Ethyl ether was added to virus suspension to make 20% by volume and incubated at 4°C for 24 hours. The ether was removed by evaporation and the titer of the virus was determined according to the method of Kosters et al. (1972).

Chloroform sensitivity test:

Infectivity titers were determined before and after treatment with chloroform by inoculation of chicken embryo and the drop in infectivity titers were calculated according to the method of Benton et al. (1967).

Thermostability test:

Virus titers were determined after exposure to 56°C for 1,2,3 hours. 2 log drop in titer or more indicated heat sensitivity Kosters et al. (1972).

Serological tests:

Agar-gel precipitation test:

Agar-gel precipitation test procedure according to method of Lucio and Hitchner (1979).

ELISA test:

Serum samples were assayed at a final dilution of 1:500 for antibodies to IBDV using a commercial ELISA system (Flock-chek Agritech system, Porland, Maire). The test procedure followed the directions supplied with the kits and ELISA titers were logarithmically transformed.

Experimental design:

Experiment (1):

(Determination of level and uniformity of IBDV maternal antibody in different chickens types).

Different batches (160 batches) of one-day-old chicks of native breed (45 batchs) and broiler chicks (115 batches) were subjected to determination of the level of IBD maternal antibody - 10 one-day old chicks from each batch were sacrified and serum samples were collected and subjected to ELISA test to determine the maternal antibody level of IBDV.

Experiment (2):

(Waning of IBDV maternal antibody).

The waning of IBD maternal antibody was studied in broiler and native breed chicken flocks. 30 serum samples were collected weekly from either broiler or native breed chicken flocks for 5 weeks. Serum samples were subjected to ELISA test for determination of the IBD antibody.

RESULTS

Data concerning the epizootiology of the IBD outbreaks in 110 chicken flocks (30 native breed and 80 broiler flocks) are presented in table (1). The affeted flocks were previously vaccinated either once or twice with intermediate or less attenuated IBD virus vaccine. The infection usually began at young age (18-28) days in native breed and at

24-32 days in broilers. The clinical signs were non specific and observed as severe depression, ruffling, trembling and reduced feed intake. Post-mortem lesions were more pronounced in native breed than in broilers. The most important lesions were in the bursae of Fabricius which showed a various degree of changes starting from slight sweling up to severe hemorrhagic inflammation and distension of the lumen with large blood clot. In some cases a gelatinous material was seen surrounding the outer surface of the bursa. Haemorrhages on the thigh and breast muscles were also seen on the fourth day after the onset of the disease. Haemorrhages in the proventriculus were observed in 30%of examined cases. Most cases showed severe nephritis with distension of the ureters. Affected broilers showed less severe lesions in the bursa of Fabricius, while in native breeds haemorrhages on the thigh and breast muscles were more prominent.The morbidity rates were high and ranged from 15-80% while the mortality rates ranged from 1.5-30%. The course of disease was usually more prolonged in broiler flocks than in native breed flocks.

Virus assay:

1- Chicken embryos inoculation:

Chicken embryos inoculated via the CAM showed low degree of mortality specially at initial paasage. Dead embryos showed general congestion and haemorrhages on the skin and mottling of liver at third passage.

2- Results of AGP test:

Results of AGP test indicated that affected bursal homogenates as well as reference D78 vaccinal strain of IBDV as standard serotype I refrence virus showed preciptin lines with five representative polycolonal IBD antisera. All isolates and refrence D78 strain showed positive reaction with monoclonal antibodies R 63 and B 69 but not with monoclonal antibody 57. Four isolates did not react with monoclonal antibody 42, two isolates with monoclonal antibody 8, and three isolates with monoclonal antibody 179.

3- Chloroform and ether sensitivity test:

Chloroform and ether sensitivity test revealed that all the tested isolates were ether and chloroform resistant as drop titers before and after treatment did not exceed one log ^{10}

Thremostability:

The effect of heat on the viability of the virus indicated that all isolates were thermostable after 1,2 and 3 hours at 56°C.

Results of Experiment (1):

Determination of level and uniformity of IBDV maternal antibody in different chicken types:-

Tables (2 & 3) summarize the results of ELISA test on 160 batches of one-day-old broiler and native breed chicks. It is clear that broiler chicks possed higher antibody level in comparison with native breeds. The uniformity was uneven in all tested serum samples within the same batch. On the other hand, the uniformity in meat-type chicks was less uneven with coefficient variation (CV) ranged from 31.40%-165.85 % in comparison with native chicks in which CV was (57.45%-178.20).

Results of Experiment 2:

Waning of IBDV maternal antibody:

The results of waning maternal antibodies (Fig. 1 and 2) in broiler and native breed chicks indicated that ELISA test was negative at 5 weeks of age in broiler chicks and at 4 weeks of age in native breed chicks.

DISCUSSION

The epizootiological studies of different IBD outbreaks (Table, 1) in 30 native breed and 80 broiler chicken flocks revealed that outbreaks of IBD were observed in both native and broiler flocks despite their previous vaccination either once or twice with intermediate IBDV vaccine. The disease appeared in broiler flocks at relatively older age (24-32 days of age) than in native flocks in which the disease usually appeared at 18-28 days of age. These results are in agreement with Meulemans et al. (1977) who reported that most of IBD outbreaks are generally occurring in broilers after the fourth week of age. This may be attributed to longer half life of maternal antibodies in broiler flocks (Skeeles et al., 1979).

The clinical signs of investigated IBD outbreaks were nonspecific and observed as severe depression, ruffling, trembling and reduced feed intake. The most important lesions were confined in the bursa of Fabricius in which various degrees of changes were observed starting from slight swelling up to severe haemorrhagic inflammation with or

without filling of the lumen with a large blood clot. In some cases a gelatinous material was seen surrounding the outer surface of the bursa. Haemorrhages on thigh and breast muscles were also seen on fourth day after onset of the disease and the haemorrhages in the proventriculus were observed in about 30% of examined cases. Severe nephritis with distension of the ureters were also seen. On the other hand, post-mortem lesions were more pronounced in native breed chicks than in broilers. These clinical and post-mortem findings are similar to those described by (Box, 1989; El-Batrawy, 1990 and Khafagy et al., 1990). In accordance with the previous studies, the morbidity and mortality rates were greatly variable ranging in the present investigation between 18-80-% and 1.5-30% respectively. The variability in mortality rates in different IBD infected flocks may be requested to variability in the IBDV virulence or the immune status of the birds and the age at which these birds were exposed to infection.

The course of the disease was usually more prolonged in broiler flocks than in native breed flocks. In the same concern, Saif-Edin et al. (1996) mentioned that prolonged course may be attributed to early protection afforded by maternal antibodies together with partial protection by vaccinal immunity.

Concerning the virus assay, IBDV was successfuly propagated in embryonated chicken eggs via CAM inoculation which resulted in low degree of mortality especially at the inital passage. In the 3rd passage, the dead embryos showed general congestion and haemorrhages on the skin and mottling of liver. In the same respect, Mona Ahmed (1998) reported that inoculation of bursal homogenates via CAM route in chicken embryos resulted in variable mortality rates, but trials to detect IBDV antigens in CAM or embryos were unsuccessful.

Results of AGP test revealed that all affected bursal homogenates as well as reference D78 vaccinal strain of IBDV showed preciptin lines with reference polyclonal IBDV antisera. Also isolates and reference strain gave positive reaction with all of the monoclonal antibodies except MA 57 which proved that all tested isolates belonged to the classical serotype 1 of IBVD. On the other hand, four isolates did not react positively with MA 42, two isolates did not react with MA 8 and three isolates did not react with MA 179. These findings support those reported by Snyder et al. (1992); El-Sanousi et al. (1994); Sultan (1995) and Saif-Edin et al. (1996) who concluded that the examined IBDV

isolates were belonged to the classical serotype 1. Additionally, Saif-Edin et al. (1996) suggested a possible partial shift of antigenicity of some field viruses may be existed.

Chloroform and Ether resistance as well as thermostability of IBDV at 56°C for 1-3 hours which proved in the present study were also documented by Benton et al. (1967) and Lukert et al. (1975).

In experiment (1), the determination of IBDV maternal antibody levels in both broiler and native breed chickens indicated that broiler chicks possessed higher antibody level in comparison with that of native breed chicks. The uniformity was uneven in all tested serum samples within the same batch. Moreover, the uniformity in broiler chicks was less uneven with coefficient of variation (CV) ranging from 31.40% to 165.85 in comparison with native breed chicks in which the CV was 57.45%-178.20%. This could be attributed to the uneven distribution of maternal antibodies within the same batch which make the prediction of suitable timing of vaccination is so difficult (Saif-Edin et al., 1996).

The waning of IBDV maternal antibody using ELISA test was studied in experiment (2). Results revealed that ELISA test was negative at five weeks of age in broiler chicks and at four weeks in native breed chicks. These results simulate to those reported by Saif-Edin et al. (1996) who stated that maternal antibodies disappeared at four weeks of age in meat-type chicks and at five weeks of age in egg-type chicks. On the other hand, Tsukamoto et al. (1995) mentioned that waning of maternal antibodies together with their uneven distribution among a flock lead to an immunity gap at the individual and at the flock levels, which represent the period during which chicks could be infected but are still refractory to vaccinal strain. At the flock level, this gap is critical because immune and susceptible birds exist in the same flock.

It could be concluded that in spite of using different IBDV vaccines and vaccination programs severe IBD outbreaks still occurred resulting in severe losses in both broiler and native breed chicken flocks.

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Table 1: Epizootiological data of 110 outbreaks of IBD in broiler and native breed chicks

Flock No.	Type of birds	Age at onset of infection	N. of vaccination	Type of Vaccine	% Morbidity	% Mortality
1-30 B	Broilers	24 days	Twice	IM+ less attenuated	60-80 %	15-30
31-70 B	Broilers	28 days	Twice	IM + IM	30-50%	5-10 %
71-80 B	Broilers	32 days	Twice	IM + IM	20-35%	3-6 %
1-16 N	Native	18 days	Once	IM	40-65%	10-20 %
17-22 N	Native	20 days	Once	IM	25-40%	3-5 %
23-30 N	Native	28 days	Once	IM	15-20%	1.5-3 %

IM = Intermediate **N = Number**

Table 2: Quantification of IBD maternal antibodies in day-old broiler chicks

Batch No.	ELISA titer				
	Minimum	Maximum	Mean (n=20)	GMT*	CV**
1	359	1967	549	359	86.60 %
2	485	1364	569	546	79.60%
3	527	2041	665	596	92.60%
4	1017	2519	1696	1259	43.90%
5	679	1839	889	759	63.40%
6	485	1456	654	589	58.70%
7	355	2009	654	459	84.30%
8	288	1353	544	489	75.40%
9	223	1119	450	356	56.40%
10	491	1256	646	546	71.20%
11	626	1906	758	678	61.50%
12	686	1512	1022	984	56.40%
13	704	2025	1320	1245	39.40%
14	793	2105	1438	1356	27.90%
15	404	1848	750	654	51.30%
16	359	1856	684	489	56.90%
17	223	1954	1088	985	49.80%
18	250	1234	698	514	74.30%
19	324	1256	756	345	68.90%
20	246	1457	696	542	58.60%
21	224	1341	454	354	56.10%
22	292	2071	580	456	68.40%
23	451	1675	952	754	61.50%
24	589	1897	1106	964	38.70%
25	564	1986	1158	973	49.80%
26	50	1267	428	354	146.50%
27	29	1259	566	456	165.85%
28	157	1877	578	454	113.20%
29	104	987	356	321	103.50%
30	225	1327	524	412	98.40%
31	354	1403	651	456	94.10%
32	423	1627	859	754	74.10%
33	455	1553	750	564	71.30%
34	313	1322	640	564	68.40%
35	217	1121	752	591	64.80%
36	181	1256	968	756	61.30%
37	181	1021	746	684	59.60%
38	211	1759	684	598	54.10%
39	153	1654	1360	1243	31.40

Continued Table 2:

Batch No.	ELISA titer				
	Minimum	Maximum	Mean (n=20)	GMT*	CV**
40	185	1803	526	245	84.90%
41	254	1229	464	365	74.30%
42	359	1343	648	468	67.80%
43	408	1623	858	786	64.80%
44	152	1335	434	342	116.50%
45	35	1459	414	336	134.90%
46	59	1568	234	198	129.40%
47	105	952	261	203	84.10%
48	44	858	292	156	96.40%
49	59	985	358	236	78.90%
50	123	955	454	351	65.80%
51	153	1175	556	451	71.50%
52	162	1358	642	458	68.90%
53	234	1409	624	561	75.40%
54	358	1119	422	342	81.55%
55	468	1605	892	756	58.40%
56	383	955	654	451	49.80%
57	401	1609	862	746	59.80%
58	553	1259	764	548	72.60%
59	357	2076	1224	1025	46.80%
60	437	1854	984	789	48.90%
61	552	951	826	694	45.10%
62	619	1155	962	741	34.10%
63	657	1256	944	845	42.10%
64	115	953	542	456	54.10%
65	1128	1855	1628	1591	39.80%
66	253	952	682	584	51.20%
67	352	1257	940	494	56.80%
68	453	1352	786	687	52.80%
69	357	1156	924	875	58.40%
70	313	942	654	546	49.80%
71	423	2028	1542	1123	53.10%
72	554	1858	1324	1236	49.40%
73	135	852	624	546	50.20%
74	158	954	642	564	57.40%
75	253	759	584	489	46.70%
76	356	837	486	401	65.10%
77	659	1255	842	689	56.90%
78	1028	2324	1482	1342	54.50%
79	858	1626	1246	1126	41.30%
80	456	1314	1040	981	41.90%

Continued Table 2:

Batch No.	ELISA titer				
	Minimum	Maximum	Mean (n=20)	GMT*	CV**
81	653	1952	1242	1123	38.70%
82	752	1243	954	781	37.80%
83	852	1355	1124	946	38.80%
84	613	1144	954	894	51.90%
85	554	2045	1642	1236	58.90%
86	634	1855	1352	1154	61.80%
87	657	1649	984	856	58.70%
88	567	1513	876	789	56.10%
89	153	1127	942	856	84.90%
90	257	1387	654	543	96.80%
91	245	1216	754	546	76.40%
92	182	953	558	458	65.20%
93	197	1077	622	459	66.40%
94	356	1815	468	356	67.90%
95	323	1935	682	546	78.90%
96	459	2056	984	745	80.90%
97	523	2145	864	684	86.40%
98	667	2334	1464	1245	87.90%
99	556	1587	988	789	76.80%
100	655	2034	1024	894	64.90%
101	965	2984	1036	839	77.20%
102	1020	1964	985	657	76.40%
103	654	1036	896	812	88.10%
104	588	2036	1089	987	65.80%
105	497	1598	1130	831	69.80%
106	897	1798	1102	789	74.60%
107	566	1698	1230	964	67.90%
108	369	1036	980	893	68.40%
109	597	1895	789	564	86.20%
110	465	1596	1024	634	81.20%
111	589	1793	1103	861	75.10%
112	875	2564	1654	763	71.30%
113	634	2036	1158	751	69.40%
114	599	1893	1105	546	68.40%
115	544	1654	989	468	32.50%

* Geometric mean titer ** Coefficient of variation

Table (3) Quantification of IBD maternal antibodies in day-old native chicks

Batch No.	ELISA titer				
	Minimum	Maximum	Mean (n=20)	GMT*	CV**
1	155	1145	501	389	93.20%
2	65	867	339	278	78.50%
3	45	706	51	49	99.30%
4	175	1236	403	308	116.60%
5	107	954	352	213	143.30%
6	0	659	129	88	63.90%
7	289	1046	541	450	57.90%
8	155	1359	345	258	65.60%
9	181	1265	358	265	60.10%
10	0	960	160	79	160.23%
11	0	1758	416	265	174.30%
12	0	631	295	215	143.20%
13	113	754	154	141	64.20%
14	0	1125	296	156	163.20%
15	0	893	258	196	105.20%
16	112	607	202	156	86.40%
17	101	1011	168	123	74.30%
18	0	850	190	102	64.70%
19	142	1805	548	321	98.40%
20	206	1524	655	432	78.90%
21	0	1198	564	392	178.20%
22	103	3057	265	201	57.45%
23	146	369	236	136	60.30%
24	189	3895	364	296	74.90%
25	146	406	321	264	94.30%
26	183	269	195	156	80.10%
27	165	798	342	236	36.40%
28	0	236	161	164	117.20%
29	0	221	132	98	69.90%
30	185	469	213	130	70.20%
31	164	473	236	139	81.30%
32	179	695	431	179	64.30%
33	0	132	110	46	96.20%
34	102	201	132	102	59.10%
35	89	360	211	165	59.40%
36	1005	116	65	31	58.90%
37	112	4305	264	131	61.30%
38	143	269	169	69	60.10%
39	122	367	133	79	64.50%
40	132	465	302	134	65.50%
41	141	364	210	136	63.30%
42	165	265	169	197	62.50%
43	125	354	165	146	81.20%
44	148	265	134	112	81.30%
45	0	201	112	89	98.90%

*** Geometric mean titer** **** Coefficient of variation**

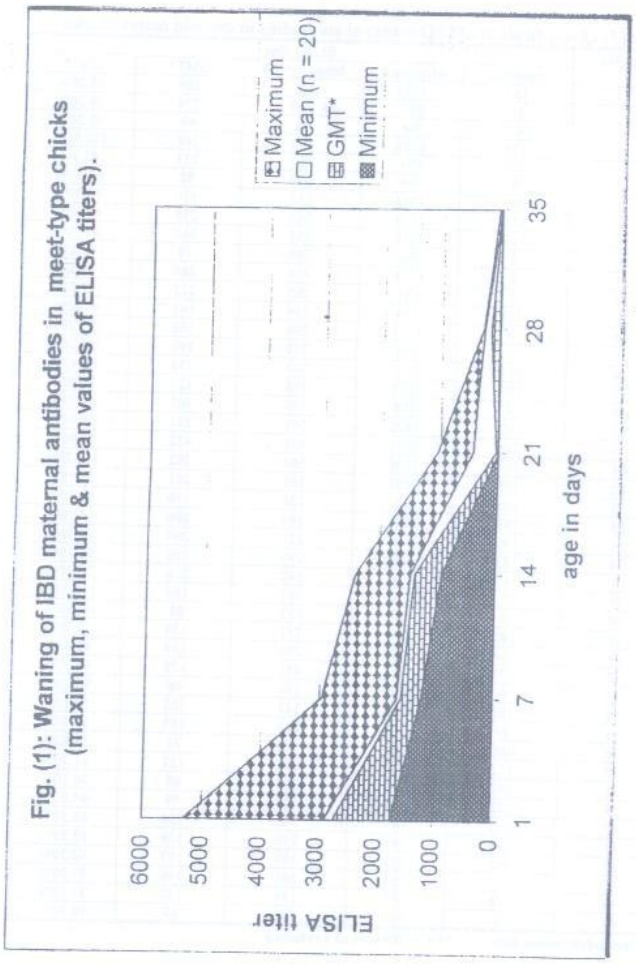

Fig. (1): Waning of IBD maternal antibodies in meet-type chicks (maximum, minimum & mean values of ELISA titers).

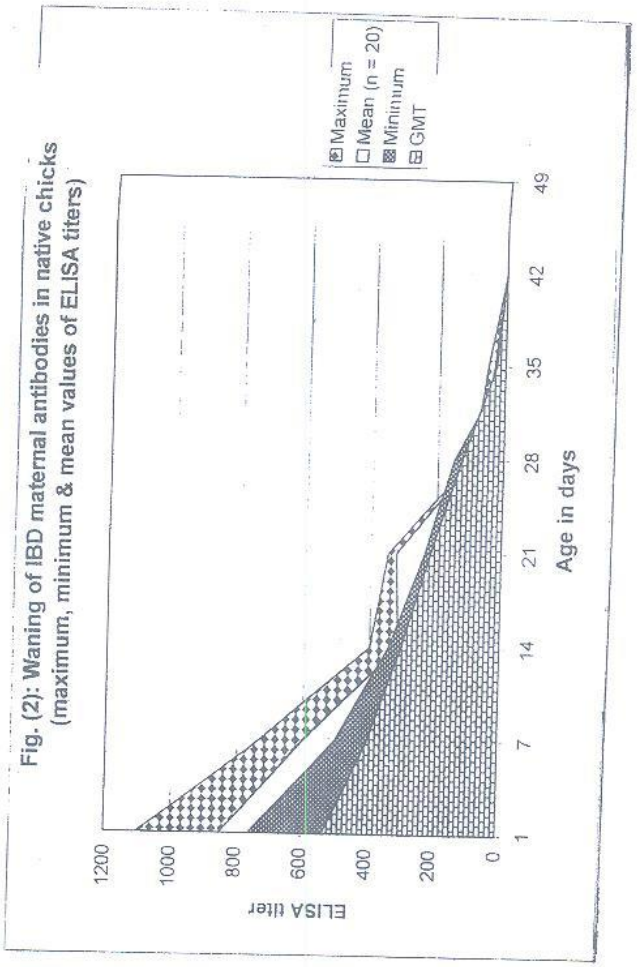

Fig. (2): Waning of IBD maternal antibodies in native chicks (maximum, minimum & mean values of ELISA titers)