ISSN 1682-8356 ansinet.org/ijps



POULTRY SCIENCE

ANSImet

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Serologic Evidence of Chicken Infectious Anemia in Commercial Chicken Flocks in Shahrekord, Iran

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Abstract: CIAV infection in chicken flocks has been described in most countries with a developed chicken industry and can result in economically important clinical or subclinical disease in broiler chickens. In this study sera samples from 46 poultry flocks in Sharekord area, Iran, were tested for the presence of chicken infectious anemia virus (CIAV) antibodies using a commercial enzyme-linked immunosorbent assay kit. All farms were positive for CIAV antibodies and 87.7% of chickens were positive. The prevalence of seropositivity was always over 20% (average 87.7%). Rates of antibody-positive chickens among flocks ranged from 20% to 100%. Seroprevalence was higher within the older flocks than in the younger flock. This is the first report of serologic evidence of CIAV in Shahrekord province, Iran. Since Shahrekord located in the center of Iran and one day chickens imported form other provinces, the infection can probably be found throughout the country and beyond. Further studies are necessary to assess economic losses due to CIAV and the cost benefit of countermeasures.

Key words: Chicken infectious anemia virus, chicken industry, CAV antibody

Introduction

The chicken infectious anemia virus (CIAV) was first described by Yuasa (Yuasa et al., 1979). This is small DNA virus with a circular, covalently closed, single negative-strand genome that has been assigned to the newly created genus; Gyrovirus belong to Circoviridae (Cardona et al., 2000 and Quinn, 2002). The CIAV genome has about 2300 nucleotides with three open reading frames. Until lately, CIAV was known as a very conserved virus of one serotype (McNulty et al., 1990 and Yuasa et al., 1979) with several genetic groups (Islam et al., 2002), but an additional serotype has been reported recently (Spackman et al., 2000a and Spackman et al., 2002b). CIAV infection in chicken flocks has been described in most countries with a developed chicken industry and can result economically important clinical or subclinical disease in broiler chickens (McNulty, 1991; Yuasa et al., 1986). It is difficult to inactivate thermally or with common disinfectants, which limits the utility of normal sanitization practices. The virus is important because of its potential for inducing immunosuppression alone or in combination with other infectious agents like infectious bursal disease or gallid herpesvirus 2. CAV produces a disease following transovarian transmission and causes mortality, severe anaemia, atrophy of the thymus and yellowish bone marrow in young chickens (Goryo et al., 1985; Otaki et al., 1987; Taniguchi et al., 1982; Todd et al., 1995; Yuasa et al., 1979; Yuasa and Imai, 1986). However, chickens become resistant to the disease by 1 month of age (Rosenberger and Cloud, 1998; Yuasa and Imai, 1986). The presence of a maternal antibody is protective in experimental infections with CAV (Yuasa et al., 1980).

Therefore, the disease associated with CAV infection has been prevented by immunization of breeding flocks with live virus vaccines. Serological data has suggested that CIAV appeared to be ubiquitous in all major chicken - producing countries of the world. The virus was isolated from chickens in Japan, China, many European countries, the United States, South Africa, Australia, New Zealand and South Africa (Saif *et al.*, 2003) but there is no published paper about prevalence of CIAV infection in Iran. This paper describes Serologic evidence of chicken infectious anemia in broiler commercial chicken flocks in Shahrekord, Iran

Materials and Methods

The flocks: A prospective study to survey for the presence of Chicken Anaemia Virus (CAV) antibody in meat broiler chickens located on Shahrekord province, centre of Iran, was conducted by collecting blood samples from 46 randomly chosen broiler flocks that ranged in age from 2 day to 9 weeks old at 2003-2004. Flocks were sampled only once at a given age and the number of samples per house was between 5-50 chicks. The flock size ranged from 7000-15000 birds. No clinical signs suggestive of CIA were observed in any of the flocks. The blood was allowed to clot and the serum was poured into small vials. The sera had stored at -20 degree of centigrade until tested.

Enzyme-linked immunosorbent assay (ELISA): A commercial test kit was used to detect specific antibodies against CIAV based on indirect enzyme linked immunosorbent assay (Synbiotics Corporation,

Table 1: Seroprevalence of CAIV infection in broiler chickens in Shahrekord, Iran, 2003

Flock ID	Age (Days)	Size	Samples	No Positive	Positive (%)	Mean Titres	SD
1	20	10000	6	6	100%	8386	2697
2	31	10000	6	3	50%	1482	1699
3	50	10000	11	10	82%	8183	7790
5	22	10000	11	11	100%	7911	5111
6	40	10000	12	9	75%	8909	4792
7	15	10000	10	9	90%	2383	2366
8	22	10000	22	22	100%	7703	4183
9	13	10000	4	2	50%	5096	2345
10	54	10000	20	19	95%	1949	2332
11	2	10000	33	33	100%	4623	2569
12	2	10000	26	26	100%	5749	2026
13	64	10000	10	10	100%	10510	2639
14	47	10000	20	19	95%	4524	2026
15	2	10000	34	32	94%	4670	2686
16	50	15000	20	15	75%	2484	1765
17	35	10000	4	2	50%	2991	3987
18	18	15000	10	7	70%	2450	3042
19	51	15000	50	50	100%	7912	2779
20	2	10000	16	16	100%	4610	3009
21	54	10000	33	26	78%	5223	12545
22	16	15000	37	29	78%	2621	1992
23	37	15000	44	37	84%	3087	2103
Total			450	403	89.6%	5060	4806

ProFlok KPL). A serum dilution of 1:50 was used following the instructions of the manufacturer. Optical density values were read at 450 nm using Anthos 2010[®] automated microplate reader.

Results

Results showed that CAV infection was widespread. All flocks were found to be positive in all ages. The prevalence of seropositivity was always over 20% (average 87.2%%). Rates of antibody-positive chickens among flocks ranged from 20% to 100%. About 87.2 percent of chickens (629/721) had anti-CAV antibody. The means of anti-CIAV antibody titres were 5060 and 83678 in 2003 and 2004, respectively. The minimum and maximum of titers were 0 and 72850, respectively (Table 1). The correlation of age (days) and rates of seropositivity was analyzed by Linear Regression test. The power of the performed test (0.404) is below the desired power of 0.800, so we should interpret the negative findings cautiously. Also, There were no significant relationships between age and mean titres in the correlation table by Pearson correlation test (P > 0.050). The mean titres significantly had increased in 2004 (P < 0.050).

Discussion

High prevalence of Chicken infectious anemia virus infection was found in Shahrekord broiler chickens. This is the first report of a serological survey on the prevalence of CIAV infection in commercial Iranian

chicken populations. The results provide evidence of widespread distribution of the virus and of a considerable high incidence of infection among poultry flocks, as it has similarly been documented to occur worldwide in all major poultry producing countries (Cardona et al., 2000 and Dren et al., 1996-). The presence of high mean titres in two days chickens are due to presence of maternal antibodies and demonstrated that broiler parent flocks were infected with CIAV. Maternal antibodies protect against infection of young chicks when hens are infected well before the onset of lay (Otaki et al., 1987). Infection after the decay of maternal antibody, when the chicks are 2-3 weeks of age, does not result in anemia and hemorrhage (McNulty, 1991). However, infection of chickens older than 2 weeks, although considered subclinical, has immunosuppressive effects (McConnell et al., 1993 and Toro et al., 1997), which are likely resulted in increased susceptibility to diseases caused by other infectious agents. Indeed, a highly significant association between CAV infection and other diseases has been demonstrated in Alabama broiler flocks (Hagood et al., 2000). In addition, clinically normal, CAV-infected broiler flocks in Northern Ireland yielded 13% lower net incomes than uninfected flocks (McNulty et al., 1991). Our results are highly agreed with Owoade et al. (2004) in Nigeria. They found that six out of seven (86%) farms were positive for CIAV antibodies and a positive correlation between age and rate of seropositivity, although we could not found any correlation between these two factors.

Table 2: Seroprevalence of CAIV infection in broiler chickens in Shahrekord, Iran, 2004

Flock ID	Age (days)	Size	Samples	No Positive	Positive (%)	Mean titers	SD
1	36	10000	9	7	78%	2298	1587
2	35	10000	20	18	90%	467940	858964
3	24	7000	15	8	53%	2110	2370
5	25	11500	15	13	86.7%	105243	397616
6	42	10000	16	14	88%	300108	664933
7	21	7000	11	8	73%	499275	720717
8	42	10000	9	8	89%	3665	1924
9	38	10000	5	4	80%	3377	2356
10	19	10000	12	12	100%	5842	2756
11	40	10000	10	10	100%	4250	2269
12	28	10000	12	12	100%	9364	8688
13	37	10000	26	26	100%	6617	3642
14	30	7000	5	4	80%	4353	4257
15	35	11500	17	14	82%	2774	1866
16	30	10000	11	10	91%	3203	2204
17	32	10000	15	12	80%	2681	2145
18	20	10000	5	1	20%	1017	2275
19	15	10000	11	10	91%	5146	3096
20	29	10000	5	4	80%	7055	5796
21	39	10000	6	6	100%	7928	3773
22	42	10000	19	15	79%	4216	3616
23	42	10000	8	5	63%	1935	1800
1	32	10000	9	5	55%	2034	2374
Total			271	226	83.1%	83687	365114

It was concluded that CIAV is widespread in Shahrekord commercial chicken populations and it may be an important cause of immunosuppression in chicken. Thought because of high rate of maternal CIAV antibody among chickens there was a low incidence of clinical disease among them indicating that CIA should be considered endemic to Iran.

Acknowledgements

This research was supported by a grant No P/1042 from the Vice Chancellor for research of Shahrekord University. The authors wish to thank Mr. Safar Pour, the technician of veterinary clinic, for their cooperation during this study

References

Cardona, C., P. B.Lucio, J. O'Conell, Jagne and K.A. Schat, 2000. Humoral immune responses to Chicken infectious anemia virus in three strains of chickens in a closed flock, Avian Dis., 44: 661.

Dren, C.S, T. Farkas and I. Nemet, 1996. Vet. Mic., 50: 7-16

Goryo, M., H. Sugimura, S. Matsumoto, T. Umemura and C. Itakura, 1985. Isolation of an agent inducing chicken anaemia. Avian Pathol.,14: 483-496.

Hagood, L.T., T.F. Kelly, J.C. Wright and F.J. Hoerr, 2000. Evaluation of chicken infectious anaemia virus and associated risk factors with disease and production losses in broilers. Avian Dis., 44: 803-808. Islam, M.R., R. Johne, R. Raue, D. Todd and H. Muller, 2002. Sequence analysis of the full-length cloned DNA of chicken anemia virus (CAV) strain from Bangladesh: evidence for genetic grouping of CAV strains based on the deduced VP1 amino acid sequences. J. Vet. Med., B. 49: 332-337.

McNulty, M.S., T.J. Connor, F. McNeilly, M.F. McLoughlin, and K.S. Kirkpatrick, 1990. Preliminary characterization of isolates of chicken anemia agent from the United Kingdom. Avian Pathol., 19: 67-73.

McNulty, M.S., 1991. Chicken anaemia agent : a review. Avian Pathol., 20: 187-203.

McNulty, M.S., S.G. McIlroy, D.W. Bruce and D. Todd, 1991. Economic effects of subclinical chicken anemia agent infection in broiler chickens. Avian Dis., 35: 263-268.

McConnell, C.D., B.M. Adair and M.S. McNulty, 1993. Effects of chicken anemia virus on cell-mediated immune function in chickens exposed to the virus by a natural route. Avian Dis., 37: 366-74.

Otaki, Y., T. Nunoya, M. Tajima, H. Tamada and Y. Nomura, 1987. Isolation of chicken anaemia agent and Marek's disease virus from chickens vaccinated with turkey herpesvirus and lesions induced in chicks by inoculating both agents. Avian Pathol., 16: 291-306.

- Owoade, A.A., D.O. Oluwayelu, O.A. Fagbohun, W. Ammerlaan, M.N. Mulders and C.P. Muller, 2004. Serologic evidence of chicken infectious anemia in commercial chicken flocks in southwest Nigeria, Avian Dis., 48: 202-5.
- Quinn, P.J., 2002. Veterinary microbiology and Microbial disease. Blackwell Science.
- Rosenberger, J.K. and S.S. Cloud, 1998. Chicken anaemia virus. Poult Sci., 77: 1190-2.
- Saif, Y.M., H.J. Barnes, J.R. Glisson, A.M. Fadley, L.R. McDougald and D.E. Swayne, 2003. Diseases of poultry, 11th ed. Blacwell publishing company, Iowa State Press, Ames, 181-202.
- Spackman, E., S.S. Claud, C.R. Pope and J.K. Rosenberger, 2002a. Comparison of a putative second serotype of chicken infectious anemia virus with a prototypical isolate: I. Pathogenesis. Avian Dis., 46: 945-955.
- Spackman, E., S.S. Claud and J.K. Rosenberger, 2002b. Comparison of a putative second serotype of chicken infectious anemia virus with a prototypical isolate: II. Antigenic and physicochemical characteristics. Avian Dis., 46:956-963.

- Taniguchi, T., N. Yuasa, M. Maeda and T. Horiuchi, 1982. Haemopathological changes in dead and moribund chicks induced by chicken anemia agent. Natl. Inst. Anim. Health, Q., 22: 61-69.
- Toro, H., A.M. Ramirez and J. Larenas, 1997. Pathogenesity of chicken anemia virus (isolate 10343) for young and older chickens. Avian Pathol., 26: 485-499.
- Todd, D., T.J. Connor, V.M. Calvert, J.L. Creelan, B.M. Meehan and M.S. McNulty, 1995. Molecular cloning of an attenuated chicken anaemia virus isolate following repeated cell culture passage. Avian Pathol., 24: 171-187.
- Yuasa, N., T. Taniguchi and I. Yoshida, 1979. Isolation and some characteristics of an inducing anemia in chicks, Avian Dis., 23: 366-385.
- Yuasa, N., T. Noguchi, K. Furuta and I. Yoshida, 1980. Maternal antibody and its effect on the susceptibility of chicks to chicken anemia agent. Avian Dis., 24: 197-202.
- Yuasa, N. and K. Imai, 1986. Pathogenicity and antigenicity of eleven isolates of chicken anemia agent (CAA). Avian Pathol., 15: 639-645.