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Investigation of the Reticuloendotheliosis Virus Infection in Large-Scale Breeder Chicken Farms of China by Serological Survey

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Abstract: To investigate the Reticuloendotheliosis virus infection status in large-scale breeder chicken farms of China, 563 serum samples from 4 great-grandparents breeder farms, 6557 serum samples from 8 grandparents breeder farms, 312 serum samples from parents breeder farms, totally 7432 serum samples were collected and examined using the Reticuloendotheliosis Virus Antibody Test kit. The results showed that REV-antibody positive rates of great-grandparents breeder flocks, grandparent breeder flocks and parents breeder flocks were 0-31, 0-47 and 4.23-42.50%, respectively. REV-antibody positive rate has close relationship with incidence of REV infection history and tumor complaints. This study suggests that REV infection is very common in China large-scale breeder chicken farms and the prevention of REV should be given more attention this is the largest scale serological survey for breeder chickens in China so far.

Key words: Reticuloendotheliosis Virus (REV), serological survey, breeder chickens, prevention, tumor

INTRODUCTION

As a member of the family Retroviridae, Reticuloendotheliosis Virus (REV) has been recognized as one of the most common immunosuppresive viruses which could depress immunity induced by Marek's disease vaccines and Newcastle disease vaccine or humal responses to bacterials (Witter, 2003). REV was often isolated from flocks suffering different syndromes and lessions such as tumors, growth retardness, pericardidis, perihepatitis as co-infection with other viruses such as Marek's Disease Virus (MDV), Avian Leukosis Virus subgroup J (ALV-J) and Chicken Anemia Virus (CAV) (Cui *et al*., 2000; Jin *et al*., 2001; Zhang *et al*., 2003).

More and more attentions were given to contaminations with REV in some live poultry vaccines such as Fowl Pox Virus (FPV) vaccine and MDV vaccine (Yuasa *et al*., 1976; Bagust *et al*., 1979; Fadly and Witter, 1997; Fadly *et al*., 1996). More importantly, more and more studies have proved that REV genome components can be easily integrated into the genome of other virus. As earlier reported *REV* gene fragment can be integrated into the genome of MDV (Isfort *et al*., 1992, 1994; Jones *et al*., 1993; Kost *et al*., 1993) the recombination was also found in the REV with FPV. Researchers have found the co-infection of MDV and REV in Marek's tumor samples and the recombinant field MDV strains GX0101 with partial REV genome were identified (Zhang *et al*., 2004; Zhang and Cui, 2005). The recombinant phenomenon for REV with other viruses make the detection using the molecular biological methods becoming more difficult. As there is no commercial vaccine to control REV in China up to now it is reasonable to reflect the REV infection state with serological survey. Such as, He *et al*. (1998) made a serological survey to REV infection in Beijing area, the results showed that REV-antibody positive rates were 21.4-71.0% among differernt chicken flocks, positive rates to REV in flocks with immunosuppression symptoms were significantly higher than that in flocks without immunosuppression symptoms.

In the last 10 years, it was demonstrated that REV infections is becoming more and more common in chickens in China (Du *et al*., 1999; Cui *et al*., 2000; Jin *et al*., 2001; Jiang *et al*., 2005; Zhang *et al*., 2003). In this study, >7000 serum samples from different breeder chicken farms were examined by the Reticuloendotheliosis Virus Antibody Test kit this is the the largest scale serological survey by far in China.

MATERIALS AND METHODS

Preparation and preservation of serum samples: In this study, 563 serum samples from 4 great-grandparents breeder farms, 6557 serum samples from 8 grandparent breeder farms, 312 serum samples from parents breeder

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Table 1: Detection of REV antibody in serum from great-grandparents breeder chicken farms (random collection)

Farms No.	Generation/Strains	Age (days)	No. of chicks for REV-antibody positive/Total (%)	Tumor lesions and complaint
BJ01	GGP/Local breeder	430	0/98 (0.0)	No typical lesions or tumor complaints
BJ02	GGP/Local breeder	430	0/265 (0.0)	No typical lesions or tumor complaints
HB01	GGP/Hy-line	186	31/100 (31.0)	Tumor complaints severely
SH01	GGP/Local breeder	203	9/100 (9.0)	No typical lesions or tumor complaints
	Total		40/563 (7.1)	

GGP stands for Great-Grandparents

Table 2: Detection of REV antibody in serum from grandparents chicken farms (random collection)

Farms No.	Generation/Strains	Age (days)	No. of chicks for REV-antibody positive/Total (%)	Tumor lesions and complaint
BJ03	Grandparents/Local breeder	175	0/2194 (0.00)	No typical lesions or tumor complaints
BJ04	Grandparents/Local breeder	175	0/772 (0.00)	No typical lesions or tumor complaints
HB02	Grandparents/Hy-line	175	12/100 (12.00)	No typical lesions or tumor complaints
HB03	Grandparents/Hy-line	259	0/228 (0.00)	No typical lesions or tumor complaints
HB04	Grandparents/Local breeder	190	117/840 (13.93)	Tumor complaints severely
HB05	Grandparents/Isa Brown	252	2/144 (1.39)	No typical lesions or tumor complaints
SH02	Grandparents/Local breeder	203	159/1160 (13.71)	No typical lesions or tumor complaints
SD01	Grandparents/Hy-line	280	214/919 (23.29)	Tumor complaints severely
SD02	Grandparents/Hy-line	350	94/200 (47.00)	Tumor complaints severely
	Total		598/6557 (9.12)	

Table 3: Detection of REV antibody in serum from parents breeder chicken farms (random collection)

Farms No.	Generation/Strains	Age (days)	No. of chicks for REV-antibody positive/Total (%)	Tumor lesions and complaint
HN01	Parents/Hy-line	180	11/102 (10.78)	Tumor complaints severely
HN02	Parents/Hy-line	245	3/71 (4.23)	No typical lesions or tumor complaints
HN03	Parents/Hy-line	220	6/59 (10.17)	No typical lesions or tumor complaints
SD03	Parents/Hy-line	195	34/80 (42.50)	Tumor complaints severely
	Total		54/312 (17.32)	

farms, totally 7432 serum samples were collected. The serological survey scale covered more than half breeder support of China. The chicken farms was marked by province and farm numbers according to the requirements of manufacturers as listed in Table 1-3. The sera samples were stored in -40°C.

Detection of REV antibody and judgement standard: The sera were assayed for antibodies to REV by Enzymes-Linked Immunosorbent Assay tests using the Reticuloendotheliosis Virus Antibody Test kit (IDEXX Laboratory) according to the manufacturer's instructions. If the S/p>0.5, the sera was judged as REV-antibody positive. In order to ensure the accuracy of results each sample was testd for twice.

RESULTS

REV-antibody positive rate of great-grandparents breeder chicken farms: Among 563 sera samples 40 were examined positive for REV-antibody, the positive rate was 7.10%, REV-antibody positive rate in 4 great-grandparents chicken flocks is quite different and has close relationship with incidence of REV infection history (Table 1).

REV-antibody positive rate of grandparents breeder chicken farms: Among 6557 sera samples in 9 grandparents breeder chicken farms, 598 were examined positive for REV-antibody, the positive rate was 9.12%. The highest REV-antibody positive rate was up to 47.00% (Table 2).

REV-antibody positive rate of parents breeder chicken farms: All 4 parents breeder chicken farms show positive to REV infection. Among 312 sera samples 54 were examined positive for REV-antibody, the positive rate was 17.32%, the highest REV-antibody positive rate was up to 42.50% (Table 3).

DISCUSSION

There are three tumor viruses in chickens, MDV, ALV and REV. All of them could induce different symptoms from subclinical infection to growth retardation, immunosuppression and tumors and always it is indistigwishable from other infections with similar symptoms and made diagnosis more difficult in the field. More importantly, Australian and American scientists reported that some FPV vaccine and field strains contained the intact REV genome and could produce infectious REV particles (Hertig *et al*., 1997; Fadly and Witter, 1997; Singh *et al*., 2003) indicating that REV has another special transmission way.

The earlier study have proved the co-infection of MDV and REV in Marek's tumor samples and some recombinant field MDV strains with partial REV genome were identified (Zhang *et al*., 2004; Zhang and Cui, 2005). The phenomena of natural genetic recombinations between REV and MDV or FPV warned that the co-infection and recombination of REV with other viruses would speed up evolution of some viruses. So, REV infection not only causes tumors and immunosuppression in chickens but also has other negative potentials by accelerating other viral mutations.

By sero-epidemiological surveys and laboratory studies on field samples in recent years it was recognized that REV infection has become very common in China (Cui *et al*., 1987, 2000; He *et al*., 1998; Du *et al*., 1999; Jin *et al*., 2001; Jiang *et al*., 2005; Zhang *et al*., 2003) but the economic losses caused by natural REV infections were not really recognized. REV caused losses to chicken flocks mainly due to vertical infection and early horizontal infection therefore to understand the REV infection status in breeder chicken flocks is very important and helpful.

In this study, >7000 serum samples from different breeder chicken farms were examined using the Reticuloendotheliosis Virus Antibody Test kit this is the the largest scale serological survey by far in China. The results showed that the REV-antibody positive rates is quite different in different breeder farms, the highest REV-antibody positive rate reached as high as 47.00% while 5 chicken flocks have no REV infection and their REV-antibody positive rate are 0. The survey result has close relationship with incidence of REV infection history, many farms with high REV-antibody positive rate always meet with severe tumor complaints. The study also found many grandparents chickens show negative to REV-antibody but their parents generation chickens has REV infection reports. Most likely is the REV infection to grandparent chickens lead to their immune tolerance they can not produce antibodies to REV. Overall researchers can see the REV infection is very common in China large-scale breeder chicken farms and it is very necessary to take some measures to provent and control REV infection in breeder chickens.

However, it is very difficult to identify where these infections were caused by field virus infection or by the vaccine contamination one by one for the sample quantity is so huge and there is no vaccine preservations to be identified. So, it is necessary for chinese farming enterprises to do well on vaccines preservation and validation.

As reported REV-antibody can be passed to their progenies in prevention early infection among chicks (Sun *et al*., 2006; Wu *et al*., 2009), so it is helpful and useful for many breeder chickens invested in this study to protect their progenies against early REV infection only if the REV is also negative in their body this need more detection. Also, to better understand the pathogenic role of REV infection in these chickens more pathogens detection should be conducted especially for immunosuppressive viruses such as ALV, MDV and so on.

CONCLUSION

This study suggests that REV infection is very common in China large-scale breeder chicken farms and the prevention of REV should be given more attention this is the largest scale serological survey for breeder chickens in China so far.

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REFERENCES

Bagust, T.J., T.M. Grimes and D.P. Dennett, 1979. Infection studies on a reticuloendotheliosis virus contaminant of a commercial Marek's disease vaccine. Aust. Vet. J., 55: 153-157.

Cui, Z., C. Zhu and H. Sun, 1987. Investigation of avian leukosis virus and reticuloendotheliosis viurs infectons. Chin. J. Anim. Poult. Infect. Dis., 1: 37-38.

Cui, Z., Y. Du, W. Zhao, R. Ji and J. Chai, 2000. Reticuloendotheliosis virus infection and immunodepression of chicken flocks. Chin. J. Vet. Drug, 34: 1-3.

Du, Y., Y. Wu, W. Zhu, P. Liu and Y. Wang *et al*., 1999. A reticuloendotheliosis virus isolated from transmssible proventriculitis in Chickens. Chin. J. Vet. Sci., 19: 434-436.

Fadly, A.M. and R.L. Witter, 1997. Comparative evaluation of *in vitro* and *in vivo* assays for the detection of reticuloendotheliosis virus as a contaminant in a live vaccine of poultry. Avian Dis., 41: 695-701.

Fadly, A.M., R.L. Witter, E.J. Smith, R.F. Silva, W.M. Reed, F.J. Hoerr and M.R. Putnam, 1996. An outbreak of lymphomas in commercial broiler breeder chickens vaccinated with a fowlpox vaccine contaminated with reticuloendotheliosis virus. Avian Pathol., 25: 35-47.

He, Y., Z. Zhang and H.C. Yang, 1998. Serological survey of reticuloendotheliosis virus infection in Beijing chicken flocks. Acta Veterina Zootechnica Sinica, 29: 71-78.

Hertig, C., B.E.H. Coupar, A.R. Gould and D.B. Boyle, 1997. Field and vaccine strains of fowlpox virus carry integrated sequences from the avian retrovirus, reticuloendotheliosis virus. Virology, 235: 367-376.

Isfort, R., D. Jones, R. Kost, R. Witter and H.J. Kung, 1992. Retrovirus insertion into herpesvirus *in vitro* and *in vivo*. Proc. Natl. Acad. Sci., 89: 991-995.

Isfort, R.J., Z. Qian, D. Jones, R.F. Silva, R. Witter and H.J. Kung, 1994. Integration of multiple chicken retroviruses into multiple chicken herpesviruses: Herpesviral gD as a common target of integration. Virology, 203: 125-133.

Jiang, S., S. Meng, Z. Cui, F. Tian and Z. Wang, 2005. Epidemic investigation of Co-infection of MDV, CAV and REV in spontaneous diseased chicken flocks in China. Virologica Sinica, 20: 164-167.

Jin, W., Z. Cui, Y. Liu and A. Qin, 2001. Co-infection of MDV, CAV and REV in infectious bursal disease sample. Chinese J. Vet. Sci., 21: 6-9.

Jones, D., R. Isfort, R. Witter, R. Kost and H.J. Kung, 1993. Retroviral insertions into a herpesvirus are clustered at the junctions of the short repeat and short unique sequences. Proc. Natl. Acad. Sci. USA., 90: 3855-3859.

Kost, R., D. Jones, R. Isfort, R. Witter and H.J. Kung, 1993. Retrovirus insertion into herpesvirus: Characterization of a Marek's disease virus harboring a solo LTR. Virology, 192: 161-169.

Singh, P., W.M. Schnitzlein and D.N. Tripathy, 2003. Reticuloendotheliosis virus sequences within the genomes of field strains of fowlpox virus display variability. J. Virol., 77: 5855-5862.

Sun, S.H., Z.Z. Cui and L.X. Qu, 2006. Reactions of maternal antibody to inactivated growth and its immunosuppressive effect by Reticuloendotheliosis virus infection. Scientia Agricultura Sinica, 39: 2335-2340.

Witter, R.L., 2003. Diseases of Poultry. 11th Edn., Iowa Strate University, USA.

Wu, Y.B., M.Z. Zhu and Z.Z. Cui, 2009. Analysis of correlation between maternal antibody titers and resistance to reticuloendotheliosis virus infections in young chickens. Chin. J. Preventive Vet. Med., 9: 671-674.

Yuasa, N., I. Yoshida and T. Taniguchi, 1976. Isolation of a reticuloendotheliosis virus from chickens inoculated with Marek's disease vaccine. Natl. Inst. Anim. Health Q., 16: 141-151.

Zhang, Z. and Z. Cui, 2005. Isolation of recombinant field strains of Marek's disease virus integrated with reticuloendotheliosis virus genome fragments. Sci. China C Life Sci., 48: 81-88.

Zhang, Z., Z. Cui and S. Jiang, 2004. Isolation and identification of reticuloendotheliosis virus from tumors infected with subgroup J Avian Leukosis virus. Chinese J. Vet. Sci., 24: 10-13.

Zhang, Z., Z. Cui, S. Jiang and J. Zhou, 2003. Dual infection of Maker's disease virus and Reticuloendotheliosis virus from tumors in chickens. Chinese J. Preventive Vet. Med., 25: 275-278.