

Investigations on the Presence of Antibodies to Avian Leukosis Virus Subgroup-J (ALV-J) in Broiler Breeders and Broilers

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Abstract: The aim of this study was to investigate the presence of antibodies to ALV-J in broilers and broiler breeders. For this, 5 broiler units and 1 broiler breeding unit in the Marmara region were visited. Seventy 4-6 -week-old chicks from the broiler units, consisting of 14 chicks from each broiler unit, were selected. Seventeen chickens from the broiler breeding unit were also selected. The chicks from the broiler units were necropsied and all internal organs were checked for the presence of tumors. Blood sera collected from all animals were analyzed for the presence of antibodies to avian leukosis virus subgroup-J (ALV-J) by a commercial ELISA (IDEXX). Growth retardation, depression, diarrhea and respiratory disorders were seen in chicks from the broiler units. No tumors were observed in the internal organs. However, pseudomembranes on the liver of 3 chicks and Gumboro-like lesions in the bursa of Fabricius in 5 chicks were seen. No antibodies to ALV-J were detected in any of the broiler chicks' sera. However, antibodies to ALV-J were detected in 13 (76%) of the 17 broiler breeders, which indicates the necessity to apply eradication programs for ALV-J in breeding flocks in Turkey.

Key Words: Avian, leucosis, subgroup-J, ELISA

Damızlık Broiler ve Broilerlerde Avian Leukosis Virus Subgroup-J (ALV-J) Spesifik Antikorların Varlığının Araştırılması

Özet: Bu çalışmanın amacı, damızlık broiler ve broilerlerde Avian Leukosis Virus Subgroup-J (ALV-J) spesifik antikorların varlığının araştırılmasıdır. Bu amaçla, Marmara bölgesinde bulunan 5 broiler ve 1 damızlık broiler ünitesi ziyaret edildi. Her broiler ünitesinden 14 civciv olmak üzere 4-6 haftalık civcivlerden toplam 70 civciv seçildi. Bir broiler damızlık ünitelerden 17 tavuk seçildi. Broiler civcivlere nekropsi uygulandıktan sonra iç organlarda tümör olup olmadığı incelendi. Tüm hayvanlardan kan serumu toplandı ve serumlarda avian leukosis virus subgroup-J (ALV-J) spesifik antikorlarının varlığı ticari ELISA (IDEXX) kiti ile araştırıldı. Broiler civcivlerde büyüme geriliği, depresyon, respiratör bozukluklar ve ishal saptandı. Hiç bir civcivde iç organlarda tümör gözlenmedi. Buna karşın, 3 civcivin karaciğerinde psödomembran ve 5 civcivin bursa Fabriciusunda da Gumboro benzeri lezyonlar görüldü. Broiler civciv serumlarında ALV-J spesifik antikorlar saptanmadı. Buna karşın 17 broiler damızlık tavuktan 13 (% 76) tanesinde ALV-J spesifik antikorlar saptandı ve bu bulgular damızlık ünitelerinde ALV-J ile ilgili eradikasyon çalışmalarının gerekliliğini göstermektedir.

Anahtar Sözcükler: Avian, leucosis, subgroup-J, ELISA

Introduction

Avian leukosis viruses are members of the family Retroviridae and are associated with a variety of neoplasm and production problems in chickens. They are classified into subgroups A, B, C, D, E and J on the basis of envelope glycoproteins and interactions between virus-specific cell receptors (1,3). Lymphoid leukosis is the most common naturally occurring B-cell lymphoma of chickens caused by ALVs. Amongst those subgroups A and B ALVs are the most prevalent and subgroup E is not pathogenic for poultry. However, ALV-J infection was first reported in the United Kingdom in 1991 in meat-

type chickens and is associated with myeloid leukosis (1,2,4). HRPS-103 is the prototype of ALV-J and appears to be a recombinant between ALV and ancient endogenous avian retroviral envelope (E51) sequences (2).

ALVs are spread by vertical and horizontal transmission. Vertical transmission is very important in eradication since virus-tolerant chickens do not develop neutralizing antibodies; transmit the virus to progeny at high rates but are highly prone to develop tumors (2,5). Therefore, an eradication program has been in operation in breeding stocks since 1980. Most studies have focused

on investigating the presence of antibodies to ALV-J in breeders (2) rather than young chickens since sero-conversion does not occur in immuno-tolerant chicks. However, the authors of one study have investigated the presence of antibodies to ALV- in young 4-8 and 14-week old chickens (2). Chickens infected with ALVs shed the virus through their feces as a source of horizontal transmission. However, the virus titer in the feces is low and stability is poor (6). Various methods have been used for the diagnosis of ALV infections to eradicate them from breeding units. These include virus isolation, complement fixation, ELISA and polymerase chain reaction PCR using the blood, organs and feather pulp (2,7-9). ELISA is commonly used in the diagnosis of ALV-J since it is sensitive, convenient and easy to perform. (9-11).

The aim of this study was to investigate the presence of antibodies to ALV-J in broilers and broiler breeders.

Materials and Methods

Collection of samples

Originally it was planned to sample randomly from 5 broiler and 5 breeder units based on the willingness of farmers to cooperate in the study. The farmers were contacted by phone and agreed to cooperate at the beginning of the study. However, later on, farmers at the breeding units did not want to give samples for analysis since the authorities should be notified when the disease is detected. Therefore, 5 broiler units and 1 broiler breeding unit in the Marmara region were visited. In the broiler units, chicks having growth retardation were selected, while random sampling was applied for the breeding unit. Seventy of 4-6- week-old chicks from the broiler units, consisting 14 chicks from each, were selected. Seventeen animals from the broiler breeding unit were also selected. The age and clinical status of the animals were recorded. Blood was collected from all animals and the sera were kept at -20 °C until used. The chicks from the broiler units were necropsied and all the internal organs were checked for the presence of tumors.

ELISA

ELISA was performed to investigate the presence of antibodies to ALV subgroup J as described by the manufacturer (Flock Check IDEXX, 50007.02). Briefly, 100 µl of positive and negative sera was put into antigen-coated wells in duplicate. The same amount of test sera

was used at a dilution of 1:500. The plates were washed after incubation at room temperature for 30 min. After washing, 100 µl of goat anti-chicken HRPO conjugate was added to all wells and the plates were incubated at room temperature for 30 min. After washing, 100 µl of enzyme substrate was added to all wells. The plates were read in 15 min using an ELISA reader (Organon Teknica).

Results

Clinical and Necropsy Findings

Growth retardation, depression, respiratory disorders and diarrhea were seen in chicks from the broiler units. No tumors were seen in the internal organs. Pseudomembranes were observed on the livers of 3 chicks. Gumboro-like lesions were seen in the bursa of Fabricius in 5 chicks.

ELISA

The OD values of the chicks from the broiler units were 0.060-0.123. The mean values of the positive and negative controls were 0.235 and 0.074, respectively. After calculation, antibodies to ALV-J were not detected in any of the broiler chick sera.

The OD values of the chickens from the breeding unit were 0.084-0.831. The mean values of the positive and negative controls were 0.278 and 0.084, respectively. Antibodies to ALV-J were detected in 13 (76%) of the 17 broiler breeders.

Discussion

ALV-J infection has emerged worldwide and is a serious cause of mortality, production problems and economic losses in broilers (12). However, there is no report on ALV-J infection in Turkey at present. Therefore, it was aimed to investigate the presence of antibodies to ALV-J in broilers and broiler breeders. This is necessary for the eradication of ALV-J infections from poultry, particularly from breeders since the virus is vertically transmitted.

Serological studies to investigate the presence of antibodies to ALV-J and antigens of ALV-J have been performed in many countries. However, these studies involved small population groups were experimental studies (7,9). Only a few studies have reported the

frequency of ALV-J infection in some countries (2,11-13). The reason for the limit of comprehensive serological studies on ALV-J could be the reluctance of some breeder units to report the disease. Similar difficulties were encountered in this study when collecting samples, especially from breeding units. Therefore, only one broiler breeding unit was willing to participate in this study. This reflects the difficulty in applying eradication programs. The diagnosis of the disease can also be problematic in eradication programs. ELISA is commonly used but antibodies in 1-day-old chicks may not be developed. In these chicks, antibodies can be detected after 4-5 weeks of age. Therefore, 4-6-week-old chicks were used in this study. In addition, it is important to consider that tests that detect antigens of ALV-J, such as ELISA, may not be specific enough because of the genomic and antigenic differences amongst ALV-Js (2,3,8,10,14). Therefore, as an alternative, PCR can be used to detect viral DNA in blood, organs, and feather pulp, as well as serology (7-9,14).

In Taiwan, ALV-J infection was reported in 1 of 3 breeding units and the mortality rate and decrease in production were 1% and 15%, respectively. The growth of progeny derived from the infected unit was very poor, which indicates the economic losses in broilers (12). In another study performed in Taiwan, ALV-J infection was reported in 5 of 8 broiler units. The genome of strains isolated from these units was analyzed and variations were found (15). In Switzerland, 4 broiler units were investigated for the presence of antibodies to ALV-J by

ELISA. Twenty sera were collected from each unit and the results indicated that there was an increase in antibody titer to ALV-J in 3 of 4 units (11). In the United States, ALV-J suspected broiler units and broiler breeder units were investigated and antibodies to ALV-J were detected in 87% of the units investigated (2). Fadly and Smith (2) have isolated the ALV-J strains, HPRS-103 and ADOL-Hc1. They found that these strains were closely related but not identical. In Korea, 2 ALV-J strains were isolated and found to be antigenically similar to ADOL Hc1 but different from the previous isolates of ADOL Hc1 (13). In this study, none of the broiler sera were positive for ALV-J but antibodies to ALV-J were detected in 13 (76%) of the 17 broiler breeders. Broiler chicks were also investigated for the presence of tumors in the organs but no tumors were observed. However, group-specific antigens of A, B, C, D and E were investigated in Turkey and 15% positivity was recorded (16).

In conclusion, antibodies to ALV-J are present in a broiler breeder located in the Marmara region. Seventy-six percent of the chickens were positive for ALV-J. This is very high although it does not reflect the true prevalence in Turkey. This needs further investigation and precautions to eradicate disease in the Turkish chicken population, particularly in breeding units.

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