To: American Association of Avian Pathologists

From: Toxic and Miscellaneous Infectious Disease Committee -

Aly M. Fadly	R.K. Page
Roger Harvey	Y.M. Saif
Eric Gonder	Sam Schmittle
Chittur V. Radhakrishnan	Mary M. Walser
Phil D. Lukert	David E. Swayne - Chairman

Re: Committee Summary Report on Current Happenings in Toxic and Miscellaneous Infectious Diseases of Avian Species

CHICKEN ANEMIA AGENT:

On Tuesday, July 18 at the AAAP annual meeting the entire morning session will be dedicated to CAA. The beginning presentation will be by invited speaker Dr. Stewart McNulty on current work on CAA, primarily from an international viewpoint. His presentation will be followed by presentations from avian diagnosticians and researchers from Georgia, Delaware and New York concerning isolation of CAA in the USA.

Chicken anemia agent (CAA) and reovirus were isolated from chickens in Sweden affected with blue wing disease (BWD). BWD was experimentally induced only in chickens dually infected with CAA and reovirus. This dual infection of CAA and reovirus resulted in severe disease in the flock with 100% morbidity (anemia) and considerable mortality.

Infection with CAA has interfered with the humoral immune response to killed Newcastle disease vaccine. One report indicated that infection with CAA did not interfere with humoral immune response of infectious bronchitis or infectious bursal disease vaccines. However, some outbreaks of CAA have been associated with concurrent infection with infectious bursal disease virus.

Chemical bursectomy by cyclophosphamide increased pathogenicity of CAA in 2-week-old chickens, suggesting that the pathogenicity of CAA was dependent on production of humoral antibody against CAA in infected chickens.

In a 1988 serological survey in the UK, antibody to CAA was shown to be widespread in broiler breeders and parent and commercial layers. CAA antibody was also demonstrated in 5/11 SPF flocks. Seroconversion in a broiler breeder flock was first noted at 8-9 weeks of age; the majority of birds in the flock developed antibody by 18-24 weeks of age. There was no evidence for CAA antibody in sera tested from ducks or turkeys.

CAA and virulent Marek' disease (MD) virus were isolated from flocks with MD vaccination breaks. The data suggested that CAA may cause early mortality associated with lymphoid depletion and anemia in chickens dually infected with CAA and MD virus.

The physiochemical properties of a typical isolate of CAA (TK-5803) were as follows:

Diameter = 19.1 nm
Density = 1.35-1.36 g/cm^3
50% chloroform for 15 min. at 4°C -resistant
Heating at 60°C for 30 min. -resistant
Heating at 70°C for 30 min. -labile

Most recently (1989), histopathological examination of chickens infected with CAA revealed that lesions appeared first in bone marrow and thymus on day 6 postinoculation (PI), and then in the bursa, spleen and liver. Intranuclear inclusion bodies were seen in hematopoietic precursor cells. Repopulation of hematopoietic and lymphoid cells was noted at day 16-24 PI and survivor chickens had no lesions by 32 day PI.

AVIAN ENCEPHALOMYELITIS:

Avian encephalomyelitis (AE) was diagnosed in 12-to-14-week-old broiler breeder pullets two weeks after vaccination with a commercial AE vaccine. The diagnosis was based on histological examinations and virus isolation.

An antigen capture ELISA was developed for the detection of AE antigen in various tissues. The test could detect 4,000 ng/g tissue. Using his assay, the highest level of AE viral antigen was noted in the brain.

An avian entero-like virus was isolated from feces of 27-day-old broiler chicken. The isolate resembled AE and other enteroviruses in size and morphology, cytoplasmic replication and resistance to pH3 and chloroform treatment. However, using a cross-immunofluorescence test, the new isolate did not react with either AE virus or with other enteroviruses of turkeys, chickens or ducks.

REOVIRUSES

The fact that reoviruses are easy to isolate but difficult to reproduce specific disease syndromes experimentally has been a frustration to avian disease researchers and the recent studies reflect this enigma. Essentially the four areas that have been the subject of recent literature have been: 1) Reoviral arthritis, 2) Pathogenicity studies on isolates of reoviruses, 3) Mixed infections of reoviruses with other agents and 4) Serological studies.

Reoviral Arthritis

Kibenge, et al. (Avian Path., 1987) found that treatment of chickens with cyclophosphamide (Cy) or Cy and thymectomy greatly enhanced the severity of the tenosynovitis. Bursectomy and thymectomy alone had little influence on the outcome of infection, thus it appears that both B and T cell function are necessary for the immune response to the virus.

Pradhan, et al. (Avian Dis., 1987) demonstrated that anti-nuclear antibodies were produced in response to reovirus infections that resulted in arthritis. The antinuclear antibodies were first detected three weeks post infection, reached a peak at 10 weeks and then were gone by 19 weeks. They speculate that the reovirus infection induces an autoimmune disease which is responsible for the arthritis.

Pathogenicity

Several reports addressed the differences in pathogenicity of reovirus strains. One common thread between most of the studies was the fact that the age of the bird, route of inoculation and the dose of the virus has a marked effect on the ability of these viruses to produce clinical disease and mortality. Kibenge and Dhillon (Avian Dis., 1987) compared four strains representing two serotypes. One strain induced 50% mortality in 1-day-old birds, two were moderate (10-20% mortality) and one was apathogenic. The least virulent strain persisted longest in the intestinal tract while the most pathogenic could not be isolated after two weeks. Tang et al. (Avian Dis., 1987) demonstrated the importance of age, route of inoculation and strain difference of reoviruses. They correlated the severity of liver lesions to mortality and demonstrated that the primary site of replication in natural infections is the cloacal bursa. Montgomery and Maslin (Avian Dis., 1988) demonstrated that vaccine strains persist in 1-day-old vaccinates and can be isolated from hock tendon tissue but 1-week-old vaccinates did not exhibit this persistent infection.

Huang et al. (Avian Dis., 1987) found changes in the M and S genes in virus which was attenuated in cell culture as detected by hybridization between wild and attenuated RNAs. They provided evidence that the S_2 and S_4 genes are related to initiation of infection and persistence of the virus while the M_2, S_2 and S_3 genes were related to virulence.

Mixed Infections

Engstrom et al. (Avian Path., 1988) isolated both reoviruses and chick anemia agent (CAA) from clinical cases of bluewing disease in Sweden. Neither the reovirus nor the CAA alone would reproduce the clinical disease. When they coinfected SPF chickens with both cloned reovirus and CAA they produced clinical bluewing with the characteristic thymus, bursa and bone marrow atrophy and petechial hemorrhages of the skin. The CAA alone produce microscopic lesions of the thymus, bursa and bone marrow but not clinical signs were observed.

Guy et al. (Avian Dis., 1988) infected chickens with Cryptosporidium baileyi (Cb) and two strains of reovirus. The Cb alone depressed weight gain for two weeks and Cb and one strain of reovirus depressed weight gain for the three week period of the experiment. Dual infection appeared to promote shedding of both agents for longer periods of time, especially the reovirus.

Serology

Giambrone and Salano (Avian Dis., 1988) compared six strains of reoviruses by virus neutralization (VN) and ELISA. Five of the six were the same serotype and one was a major subtype when compared by cross VN tests. The virus that was different by VN was also different by the ELISA against the S1133 antigen.

Takehara et al. (Avian Dis., 1987) prepared 13 monoclonal antibodies (MAB) against a strain of reovirus. Three of the MAB neutralized virus. Two had broad specificity and one was type specific, neutralizing only homologous virus. The type specific MAB was the only one that reacted in a Western blot and it reacted with a sigma protein.

CHLAMYDIOSIS

C. psittaci is very well adapted to the avian host, and is endemic in all countries where psittaciformes are indigenous. 10% of wild birds are estimated to be carriers. The disease in birds is usually triggered by human interference such as transport, change in feed or environment or other diseases. In latent infections the chlamydia persists in macrophages and epithelioid cells. The organisms are shed through the feces over a period of several months. So the chief mode of transmission is inhalation of dried feces or other materials containing dried feces. The fecal shedding of the organisms is persistent but intermittent. Also birds with no detectable antibodies may shed organisms in feces. The Centers for Disease Control (CDC) has reported an increase in human chlamydiosis cases due to C. psittaci recently due to the increase in pet bird ownership.

Young birds as a general rule are more susceptible than older ones. While there occurs a difference in susceptibility in various species of birds and mammals, the actual condition of an individual and various stress factors are more important than a species specific susceptibility.

A chlamydial glycolysis factor has been demonstrated to be responsible for the destruction of the host cell integrity, providing hydrolytic enzymes and causing cytopathology and/or lysis. A hepatotoxic and nephrotoxic factor does exist but has not been purified. Reports show that C. psittaci of avian origin may colonize the human population and therefore may be transmitted among human population without requiring an avian host. A primary infection in avian species may lead to propagation of the organisms within the host intestinal tract without causing clinical signs of the disease. Production of antibodies is low, and a host surviving a clinical or non-clinical disease is fully susceptible thereafter. Wilson and Grimes (1988) examined 17 isolates of C. psittaci from various avian species and found that the avian isolates can be separated into two groups based on their infectivity and cytopathology for L-929 cells. This separation was confirmed when the major outer membrane proteins (MOMP) of various isolates were examined for molecular weight and antigenicity. Frequent passages through mammalian and avian hosts induces new antigens on the surface of the elementary bodies.

Exotic birds, particularly psittacines (used as pets), enter the market with C. psittaci infections acquired in the wild, during shipment or while in quarantine. Domestically reared birds are also known to harbor Chlamydiae. There are no effective therapeutic control measures or reliable serological diagnostic aids available for the diagnosis of Chlamydiosis. The organism has not been effectively controlled by Chlortetracycline medicated feed during quarantine. The most widely used serological methods are the Direct Complement Fixation test (DFC) and the Latex agglutination test. The Latex agglutination (LA) test is less sensitive than the DCF in detecting antibody activity in birds that are culture positive. However, LA is a useful rapid screening test that can be done easily in any Veterinary Clinic.

DERMATOLOGIC CONDITIONING

A recent review has been published on dermatologic pathology in avian species (Avian Pathology 18:1-72, 1989). This is a good basic review of the topic that has not been addressed in over 15 years. However, it does lack information on rather basic areas: scratches, bruises, and scars encountered at processing.

MYCOTOXINS

Recently, research has focused on the interaction of mycotoxins with other mycotoxins, with nutrient ingredients, and with other feed components. Aflatoxin and ochratoxin, and aflatoxin and T-2 toxin can produce both additive and synergistic toxic responses for certain parameters when fed in combination, whereas aflatoxin and deoxynivalenol in combination cause additive or less than additive toxic effects. The onset and progression of aflatoxicosis and ochratoxicosis in broiler chicks have been characterized, giving new insight into the subtle toxic events that occur, once ingested. Perhaps the most promising new developments in mycotoxicology involve the use of chemically active sorbent compounds to bind aflatoxin in animal feeds, thereby reducing bioavailability, toxicity, and residues. One product, hydrated sodium calcium aluminosilicate, (NovaSil™) is commercially available as an anticaking feed additive, and experimentally it has protected poultry and swine from the adverse effects of aflatoxin and has reduced milk residues in dairy goats and dairy cows.

ASCITES

Ascites or "waterbelly" is a syndrome of increasing concern to the broiler industry. Previously recognized as a condition associated with poultry raised at high altitudes (low atmospheric oxygen), ascites has now begun to appear as a problem at lower altitudes in the U.S., Canada, Europe, and Africa. Factors such as restricted ventilation and lowered temperatures during brooding, improper immunization practices for respiratory diseases, genetic susceptibility of poultry strains and breeds, cardiopulmonary disease, increased dietary intake of sodium, consumption of specific antibiotics and feed additives, and liver disease can contribute to the incidence and severity of ascites in poultry. The sequence of events that lead to the development of ascites are fairly clear, however, a specific etiology has not been determined. In short, chickens develop congestive heart failure due to increased right side rate, volume and pressure. The increased pressure in the lungs and the backflow to the posterior vena cava induce formation of visceral and pulmonary edema.

To date, research has centered on the identification of environmental, management, and genetic factors in geographical areas with a high incidence of ascites in poultry. Recently however, researchers have concentrated on the pathophysiologic differences in normal and ascitic chickens in an attempt to uncover discrete etiologic factors. One such approach has been the development of a high altitude simulation in which ascites is induced in broilers raised in a hypobaric chamber. By use of this model, various environmental and genetic factors can be superimposed to determine their etiologic impact.

At present, the prevention of ascites must rely on improved management practices such as adequate ventilation and heat during brooding, proper vaccination procedures and disease control, continuous monitoring of dietary ingredient concentrations, and elimination of feed-borne toxins. Under conditions of high altitude rearing, an additional management practice may include feed restrictions to reduce oxygen demands by rapidly growing broilers.

SUMMARY

Individual original detailed reports with literature citations can be obtained by contacting: David E. Swayne, Chairman, Toxic and Miscellaneous Infectious Disease Committee, Department of Veterinary Pathobiology, The Ohio State University, 1925 Coffey Road, Columbus, OH 43210.