

Fowl adenovirus infections in Belgian broilers: a ten-year survey

Fowl adenovirus-infecties bij vleeskuikens in België: een overzicht van tien jaar

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ABSTRACT

Between May 2002 and May 2012, fowl adenovirus (FAdV) infections were diagnosed in 38 of 310 diseased Belgian broiler flocks. FAdV isolates were usually derived from pools of multiple organs, predominantly incorporating liver, pancreas and bursa. The FAdV isolates belonged to the serotypes FAdV 1 (five strains), FAdV 2/11 (thirteen strains), FAdV 3 (one strain), FAdV 5 (eight strains) and FAdV 8a (four strains); seven isolates could not be typed with certainty.

The clinical complaints in infected flocks consisted of poor growth, wet litter, respiratory disease and/or lameness in 63%, 37%, 26% and 24% of the cases, respectively. Increased mortality occurred in 39% of the flocks. In 53% of the infected farms, the clinical signs had been showing up in multiple successive production cycles. The most consistent lesions were hepatitis, nephritis, myocarditis, pancreatitis, tracheitis and proventriculitis. Concurrent infections with reovirus, infectious bronchitis virus, avian metapneumovirus, infectious bursal disease virus, chicken anemia virus, *Escherichia coli*, *Enterococcus cecorum* and/or *Eimeria* were observed in 53% of the flocks, predominantly in those that were experiencing increased mortality.

It was concluded that fowl adenoviruses are frequently involved in disease of Belgian broilers, alone or in combination with other infectious agents.

SAMENVATTING

Tussen mei 2002 en mei 2012 werden bij 38 van 310 zieke Belgische vleeskuikentomen fowl adenovirus (FAdV) infecties gediagnosticeerd. De FAdV-isolaten werden meestal bekomen uit mengstalen van meerdere organen waarin vooral lever, pancreas en bursa aanwezig waren. De FAdV-isolaten behoorden tot de serotypes FAdV 1 (vijf stammen), FAdV 2/11 (dertien stammen), FAdV 3 (één stam), FAdV 5 (acht stammen) en FAdV 8a (vier stammen); zeven isolaten konden niet met zekerheid getypeerd worden.

De klinische klachten bij geïnfecteerde groepen bestonden uit groeivertraging, nat strooisel, ademhalings symptomen en/of manken in respectievelijk 63%, 37%, 26% en 24% van de gevallen. Verhoogde sterfte trad op bij 39% van de tomen. Bij 53% van de geïnfecteerde bedrijven waren de symptomen reeds meerdere productiecycli na elkaar opgetreden. De meest consistent waargenomen letsels waren hepatitis, nefritis, myocarditis, pancreatitis, tracheïtis en proventriculitis. Gelijktijdige infecties met reovirus, infectious bronchitis virus, avian metapneumovirus, infectious bursal disease virus, chicken anemia virus, *Escherichia coli*, *Enterococcus cecorum* en/of *Eimeria* werden waargenomen bij 53% van de tomen, vooral bij de groepen die af te rekenen kregen met verhoogde sterfte.

Er werd besloten dat fowl adenovirussen vaak een rol spelen bij ziekte van Belgische vleeskuikens, alleen of in combinatie met andere infectieuze agentia.

INTRODUCTION

Adenoviruses causing infections in birds belong to the family *Adenoviridae*, genus *Aviadenovirus* (Harrach et al., 2012). This genus comprises amongst others the group of fowl adenoviruses (FAdV).

In 1949, the first fowl adenovirus was isolated from non-SPF embryonated chicken eggs that were contaminated with the agent (Van den Ende et al., 1949). One year later, Olson (1950) isolated an adenovirus from diseased quails. This agent was later on called the FAdV 1 serotype. Over the years, eleven additional FAdV serotypes have been discovered (Hess, 2000).

The major structural proteins of fowl adenoviruses are the hexon and two fibres linked to a penton base (Hess, 2000). The hexon protein bears type-, group- and subgroup-specific determinants. The hexon and fibre proteins provoke the formation of antibodies, which can be used for the typing of FAdV into twelve different serotypes using virus neutralization assays. According to the latest classification proposed by the International Committee for the Taxonomy of Viruses (Harrach et al., 2012), the twelve serotypes are named FAdV 1 to FAdV 8a and FAdV 8b to FAdV 11.

Since the end of the nineties of the previous century, efforts have been done to replace the laborious neutralization tests for the typing of fowl adenoviruses by molecular techniques. The hexon gene was chosen as primary target for primer development (Meulemans et al., 2001; Günes et al., 2012). At present, the known FAdV strains are divided into five genotype species, indicated A to E (Benco et al., 2005). There seems to be a rather good correlation between the genotyping- and serotyping results. Molecular type A contains FAdV 1, type B comprises FAdV 5, type C includes FAdV 4 and FAdV 10, type D is composed of FAdV 2, FAdV 3, FAdV 9 and FAdV 11 strains, while type E consists of FAdV 6, FAdV 7, FAdV 8a and FAdV 8b isolates (Adair and Fitzgerald, 2008).

Until recently, fowl adenoviruses in chickens were generally perceived as opportunistic pathogens that only show their pathogenic potential when additional factors, i.e. predominantly concurrent infections, compromise the health of the birds. As early as 1973, it was already known that concurrent infections of FAdV and immunosuppressive agents, such as infectious bursal disease virus (IBDV) and chicken anemia virus (CAV), could lead to a condition called inclusion body hepatitis (IBH) (Winterfield et al., 1973). It is a disease characterized by the sudden onset of high mortality -usually in meat type chickens- resulting in the death of up to 30% of the birds over a period of approximately five days (Adair and Fitzgerald, 2008). Chicks that died from the disease, show pale and swollen livers and the hepatocytes contain viral inclusion bodies in the nuclei. In 2006, Gomis et al. reported cases of IBH in flocks that were not concurrently infected with IBDV or CAV, indicating that at least some strains might act as primary pathogens. In the meantime, IBH has been experimentally reproduced

by inoculation of SPF chicks with strains belonging to different serotypes of the FAdV-E genotype (Adair and Fitzgerald, 2008; Zadavec et al., 2011; Choi et al., 2012; Dar et al., 2012).

In 1987, a new condition was observed in Pakistan, named hepatitis-hydropericardium syndrome (HHS) or Angara disease (Hess, 2011). Later on, the disease was also reported in numerous countries in Asia, South-America and the Middle East (Nakamura et al., 2000; Dahiya et al., 2002). As in IBH, signs of sudden death and liver lesions are common characteristics of HSS but in the latter case, the mortality rate is clearly higher, amounting up to 75% of the flock. Moreover, the filling of the pericardial sac with a clear straw-colored fluid is a typical finding in case of HSS. Especially broiler flocks between three and five weeks of age are hit, although occasionally layers and breeders may also suffer from the disease. From the lesions in affected chickens, FAdV4 isolates have been consistently obtained. The condition has been experimentally reproduced by oral and nasal inoculations with these strains (Hess et al., 1999). Under natural circumstances, both horizontal and vertical spreads have appeared to be important. HSS was the first disease assigned to well-defined strains of fowl adenovirus, acting as a primary pathogen.

Since 2001, FAdV1 strains have increasingly been isolated from lesions of gizzard erosions (GE). This was first observed in Japan, but also seems to be common in Europe nowadays (Okuda et al., 2001; Ono et al., 2003; Marek et al., 2010; Domanska-Blicharz, 2011). Desquamation, degeneration and erosion of the keratinous layer are observed in broilers from one to two weeks of age onwards. This may be accompanied by elevated feed conversions, but it may also lead to gizzard condemnations in the slaughterhouse. The gizzard lesions can be experimentally reproduced in chickens by oral inoculation with FAdV 1 strains (Nakamura et al., 2002). During experimental infection studies, it has been noticed that the virus may also spread to other internal organs (Okuda, et al. 2001).

Besides IBH, HSS and GE, fowl adenovirus infections have been associated with low feed intake, poor growth, increased feed conversion, respiratory disease and tenosynovitis (McFerran et al., 1971; Jones and Georgiou, 1983; Adair and Fitzgerald, 2008). Although these clinical signs were sometimes reproduced through experimental inoculations, they usually remained rather mild. Therefore, it is still unclear whether adenoviruses have a primary role in the origin of these conditions.

Finally, it has been demonstrated that at least some strains of FAdV interact with the humoral and cell-associated functions of the immune system, leading to immunosuppression (Schonewille et al., 2008).

Only little information is available on the epidemiology and significance of FAdV in disease outbreaks of broilers in Belgium. It was therefore the aim of this study to review the data of 310 diseased broiler flocks examined over a period of ten years.

MATERIALS AND METHODS

Flock data and diagnostic procedures

Between May 2002 and May 2012, 310 Belgian broiler farms that experienced increased mortality and/or various signs of clinical disease, were visited to obtain detailed case history data and to collect samples necessary for making a diagnosis. Standard diagnostic procedures included necropsy of four to ten birds per farm and cytological examination of multiple internal organs, most often trachea, lungs, heart, liver, kidneys, pancreas, thymus, bursa, proventriculus and intestinal tract. Impression smears were stained with the Hemacolor (Merck, Darmstadt, Germany) staining reagents, and observed microscopically at a magnification $\times 1000$. When the nature of the clinical problems and/or the gross and microscopic necropsy findings were indicative for an infectious cause of disease, appropriate virological, PCR, bacteriological and/or histological techniques were additionally used. Virological examinations for the isolation and identification of fowl adenovirus were performed at 143 of the 310 broiler farms.

Isolation and identification of fowl adenovirus

Selected organ samples were homogenized either individually or as a pool. The supernatant obtained after centrifugation of the homogenized tissues was filtered (0.8/0.2 μm filters), and inoculated onto freshly prepared primary chicken embryo liver cells. After four to five days of incubation, the monolayers were inspected for the presence of a cytopathic effect (CPE). In case CPE was absent, the cultures were frozen and thawed to make up to three serial passages.

In cultures that showed CPE, FAdV was identified by immunofluorescence using a polyclonal chicken antiserum, and further serotyped through virus neutralization tests. Equal volumes of sera directed to all of the twelve serotypes of FAdV and FAdV positive cultures –both in standardized dilutions– were mixed in 96-well tissue culture plates. Controls consisting of uninoculated wells and wells inoculated with FAdV culture sample only (without serum) were added in every plate. After incubation for 90 minutes at 37 °C, the mixtures of FAdV and serum samples were transferred to chicken embryo liver cells, and were incubated for four days. The FAdV isolates were attributed to the serotype for which the corresponding antiserum was able to prevent CPE development.

RESULTS

Fowl adenovirus isolates

Isolates of FAdV were obtained from samples of broilers in 38 farms (Table 1).

Since culturing had mostly been performed on pools of multiple organs, the prevalence of FAdV

in individual organs could not be determined. Overall, however, the viruses were isolated from –pooled or native– samples containing liver, pancreas, bursa, heart, kidney, lung, trachea, proventriculus, thymus, gastrocnemius tendon, spleen, vertebral column, cecal tonsil, small intestine and gizzard in 25, 17, 13, 9, 8, 6, 5, 3, 2, 2, 1, 1, 1, 1 and 1 of the cases, respectively.

Five isolates belonged to the serotype FAdV 1. Thirteen strains reacted with sera directed to FAdV 2 and FAdV 11, and were therefore indicated as FAdV 2/11. A FAdV 3 strain was obtained once. FAdV 5 strains were isolated from broilers of eight farms, while FAdV 8a isolates were obtained from birds of four farms. In two other cases, the isolates reacted with antisera against both FAdV 8a and FAdV 8b. Five strains remained untypeable, most probably due to the interference resulting from the simultaneous presence of fowl adenovirus and reovirus in the examined organs (Table 1).

Concurrent infections

Concurrent infections were found in 20 of the 38 FAdV infected flocks (Table 1).

Reovirus constituted the most prevalent concurrent infection, found in 11 FAdV positive flocks. Other infections diagnosed at FAdV positive farms were infectious bronchitis (IBV) (four flocks), avian metapneumovirus (aMPV) (one flock), chicken anemia virus (CAV) (one flock) and infectious bursal disease virus (IBDV) (one flock) infections. Bacterial co-infections with *Escherichia coli* (*E. coli*) and *Enterococcus cecorum* (*E. cecorum*) were found in six and one of the flocks, respectively. Two flocks also suffered from coccidiosis.

Clinical signs

The nature and onset of the clinical signs observed in the FAdV infected broiler flocks are summarized in Table 1.

Increased mortality rates were seen in 39% of the flocks. Within these flocks, the mortality increase varied from < 1% up to 5%. Symptoms of poor growth, wet litter, respiratory disease and/or lameness were noticed in 63%, 37%, 26% and 24% of the flocks, respectively. At 20 of the 38 infected farms, the clinical signs had already posed a recurrent problem for multiple production cycles.

For FAdV infected flocks in which no concurrent infections were found, poor growth and wet litter were the most encountered clinical problems, in 72% and 44% of the cases respectively. The signs were recurrent in successive production cycles in more than 70% of these flocks. The most important consequence of additional infections besides FAdV was a sharp rise in mortality. Whereas increased mortality was observed in 22% of the flocks without concurrent infections, it was present in 55% of the flocks with additional infections.

Table 1. Characteristics of fowl adenovirus infections observed in 38 flocks of broiler chicks.

Adenovirus type	Year of isolation	Concurrent infections	Clinical observations					
			First occurrence of signs	Retarded growth	Wet litter	Lameness	Respiratory signs	Increased mortality
FAdV 1	2004	Reovirus	Week 3		x			
	2006	aMPV	Week 6	x*		x	x	
	2011		Week 3	x				
FAdV 2/11	2011	Reovirus	Week 3	x				
	2012	<i>E. coli</i>	Week 2	x				x
	2002		Week 5	x		x		x
	2003		Week 5	x		x		
	2004		Week 1	x				
	2004		Week 4					x
	2004	Reovirus	Week 4		x	x		
	2004	Reovirus	Week 4	x		x		x
	2004	Reovirus, <i>E. coli</i>	Week 5				x	x
	2005		Week 6		x		x	
	2006	IBV, <i>E. coli</i>	Week 4				x	x
	2006	IBV	Week 5	x	x			
	2010		Week 4	x	x			
	2012		Week 5	x	x			
	2012		Week 4	x				
FAdV 3	2004	IBDV, <i>Eimeria</i>	Week 4	x		x		x
FAdV 5	2005		Week 3		x			
	2005		Week 5	x	x			x
	2005	IBV, <i>E. coli</i>	Week 4		x	x	x	x
	2007	<i>E. cecorum</i>	Week 5			x		
	2008		Week 4		x		x	
	2008	Reovirus	Week 5		x			x
	2010		Week 4	x				
	2012		Week 3	x			x	
	2004		Unknown	x				
FAdV 8a	2005		Week 4	x	x			x
	2008		Week 4	x	x			
	2009	CAV	Week 2					x
FAdV 8a/8b	2004	IBV, <i>E. coli</i>	Week 4				x	x
	2007		Week 6				x	
FAdV NT	2005	Reovirus	Week 3	x				x
	2010	Reovirus, <i>Eimeria</i>	Week 4	x				
	2012	Reovirus	Unknown	x				
	2012	Reovirus	Week 3	x	x			
	2012	Reovirus, <i>E. coli</i>	Week 4	x		x	x	x

*x = present

In about 75% of the cases, the first occurrence of clinical signs was between two and five weeks of age. In the remaining flocks, the onset of clinical disease occurred at an earlier or later stage of the growing period.

Gross lesions

When taking into account only the observations in the 18 farms without demonstrated concurrent infections, the following lesions could be related to FAdV infections. In four of the 18 flocks, no or only very

discrete macroscopic lesions were observed. In the other flocks, gross lesions were most frequently found in the livers (twelve farms). Usually, the livers were enlarged, and showed a pale or congested aspect. In three cases, a generalized hepatic necrosis was observed in multiple birds of the flocks (Figure 1). In chicks from eight farms, swollen and pale kidneys were found. In one case, the kidneys even showed extensive bleedings. Reduced sizes of the thymus, the bursa or both were observed in chicks from seven farms. Congestion and/or inflammation of the trachea and/or lungs were seen in six submissions. Femoral



Figure 1. Three-week-old broiler chick infected with fowl adenovirus type 2/11 under field circumstances, showing extensive swelling and necrosis of the liver. Concurrent infections were not demonstrated.

head necrosis was a consistent finding in the birds of six flocks. A common finding in five farms was the presence of watery contents in the intestinal tract. Paleness of the heart muscle and/or hydropericard occurred in broilers from three farms. Congestion of the mucosa of the proventriculus and a cloudy aspect of the pancreas were findings in birds from two farms and one farm, respectively.

In the twenty other FAdV positive flocks, various lesions were found. These findings are not rendered in detail since they most probably resulted not only from FAdV infections but also from various other infectious agents that were concurrently infecting the birds (Table 1). However, it should be mentioned that in one flock infected with a FAdV 1 strain (Table 1, case 21), well-circumscribed areas of necrosis were seen in the gizzard of multiple chicks.

Microscopic lesions

A consistent cytological finding in all FAdV infected flocks was the infiltration of lymphocytes in multiple organs. Considering only the eighteen flocks in which no concurrent infections were found, lymphocytic infiltrates were found especially in the liver (fourteen flocks) but also in the kidneys (eight flocks), the heart (eight flocks), the pancreas (seven flocks), the trachea (five flocks) and the proventriculus (three flocks).

The most prominent lesions were found in the liver, with the presence of viral inclusion bodies in the nucleus of hepatocytes in thirteen flocks (Figure 2). Heterogeneity in the size of the liver cells appeared a characteristic finding in seven flocks. Hepatic necrosis was confirmed microscopically in birds of the three farms in which the necrosis was already obvious from the macroscopic inspections.

Sporadically, nuclear inclusion bodies were also observed in the kidneys (six flocks), bursa (three flocks), pancreas (two flocks), heart (one flock) and proventriculus (one flock).

DISCUSSION

In the present studies, fowl adenoviruses were isolated from 38 of 310 Belgian farms of broiler chicks suffering from various clinical signs and/or experiencing increased mortality. This indicates that FAdV is frequently involved in outbreaks of clinical disease in broilers kept under Belgian field conditions. The isolation of FAdV was only attempted in 143 of the 310 diseased broiler flocks. Since it cannot be excluded that the agent was also present in some of the other flocks, the incidence of FAdV in diseased broilers could be even higher than estimated on the basis of the present isolation rates.

FAdV isolates were serotyped through virus neutralization (VN) assays. VN has been generally accepted for the classification of FAdV isolates (Hess, 2000). However, some conditions might render strains untypable in this test. The concurrent presence of multiple viruses in organs submitted to virological examination might pose problems when these agents are able to grow in the same cell culture systems. Multiple strains of FAdV simultaneously infecting the same animal (Emmy et al., 1995) and/or concurrent infection of

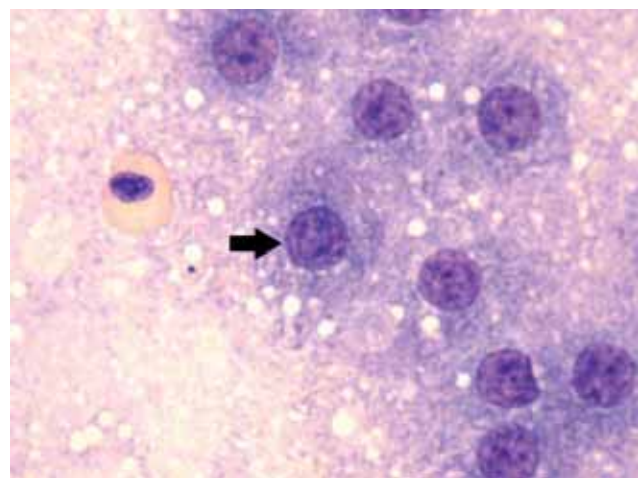


Figure 2. Impression smear of the liver of a five-week-old broiler chick infected with fowl adenovirus type 1 under field circumstances, showing a nuclear inclusion body in a hepatocyte (arrow). Concurrent infections were not demonstrated (Hemacolor stain, magnification 1000x).

birds with FAdV and other agents, such as reovirus (De Herdt et al., 2008b), can be mentioned in this respect. Such interference phenomena form the most likely explanation why seven isolates obtained in the present study could not be classified. Furthermore, FAdV 2 and FAdV 11 strains are very closely related and often difficult to distinguish from each other in VN (Steer et al., 2011). This was also experienced in the present studies. Strains that belonged to either FAdV 2 or FAdV 11 were therefore named FAdV 2/11.

Hydropericardium-hepatitis syndrome constitutes the most devastating disease related to FAdV infections, leading to mortality rates of up to 75% of the broiler flocks (Adair and Fitzgerald, 2008). Up till now, this syndrome has been restricted to South-America, Asia and the Middle East. HHS is typically associated with FAdV type 4 strains. During the present examinations in Belgium, no FAdV 4 strains were isolated, nor were extremely high mortality figures noted. This confirms that HHS does not occur in this region.

Most of the obtained FAdV isolates belonged to various serotypes of the molecular groups D and E. Isolates from these groups are sometimes associated with inclusion body hepatitis (Hess, 2011), a disease characterized by sudden death in up to 30% of the flocks and the presence of viral inclusion bodies in the nuclei of hepatocytes. However, there are differences in virulence between strains isolated from IBH, and concurrent infections are important in the clinical outcome of the infections. Although hepatitis was a prominent finding, and viral inclusion bodies were regularly observed in the present study, the flock mortality rates never exceeded 5%. This may indicate a relatively low virulence of FAdV strains circulating in Belgium and/or a lesser impact of concurrent infections. The latter may underline the importance of taking adequate control measures against infections that occur concurrently and may interact with FAdV. Specific control of FAdV through vaccination is only available for FAdV 4 strains; it is practiced in regions where HHS constitutes a frequent problem.

In recent years, FAdV 1 strains have increasingly been considered important because of their involvement in gizzard erosions. GE were first observed in Japan in 2001, but the syndrome is nowadays also common in Europe (Adair and Fitzgerald, 2008; Marek et al. 2010; Hess, 2011). The Belgian collection of FAdV isolates of the present study included five FAdV 1 strains. Gizzard erosions were observed in one of these FAdV 1 infected broiler flocks. This demonstrates that FAdV 1 related lesions of GE also occur in Belgium.

In the present study, poor growth and wet litter constituted the most prevalent clinical signs in Belgian broilers infected with FAdV, thereby confirming the literature data (Adair and Fitzgerald, 2008). Since FAdV is often recovered from intestinal organs involved in the digestion process, this might not be unexpected. Nevertheless, the exact significance of FAdV in the pathogenesis of these complaints remains

unclear since experimental infections do not necessarily lead to growth retardation and/or wet litter (Adair and Fitzgerald, 2008).

Ten of the 38 FAdV positive broiler flocks of the present study showed respiratory signs. In only four of these ten flocks typical respiratory tract viruses, such as infectious bronchitis virus and avian metapneumovirus, were found. These findings may indicate that FAdV infections may contribute to respiratory disease in chickens. FAdV has frequently been isolated from chickens with respiratory signs (McFerran et al., 1971), but its role in the course of the disease remains a point of discussion (Adair and Fitzgerald, 2008).

Lameness in broilers due to arthritis, osteomyelitis and/or tenosynovitis has been traditionally related to infections with reovirus, *E.coli*, *E. cecorum* and *Ornithobacterium rhinotracheale* (De Herdt et al., 2008a,b; 2012). Lameness appeared a prominent sign in 9 FAdV infected broiler flocks presented in this paper, five of which suffered from concurrent infections with some of the mentioned agents. It thus seems possible that some FAdV strains play a role in the pathogenesis of infectious arthritis and tenosynovitis of broilers. However, experimental infections of SPF layer chicks with FAdV strains isolated from tenosynovitis outbreaks did not result in clinical signs, severe tenosynovitis lesions or prolonged virus persistence in the hock joints (Jones and Georgiou, 1984).

Most of the flocks infected with FAdV under the circumstances of the present study did not present characteristic gross lesions. In most cases, it is not possible to suspect fowl adenovirus on the basis of macroscopic lesions. Microscopic examinations may give additional indications; especially the presence of viral inclusion bodies in the nuclei of hepatocytes and size heterogeneity of these may be suggestive. However, virus isolation remains the conclusive diagnostic test.

In the present study, increased mortality rates were noted in 22% of the broiler flocks in which only a FAdV infection was diagnosed. Flocks that were concurrently infected with other infectious agents experienced increased mortality in 55% of the cases. This may indicate that concurrent infections lead to a mortality increase. As early as 1973, Winterfield et al. described that infection with IBDV and CAV in FAdV infected flocks may lead to IBH related deaths. In the present study, IBDV and CAV each accounted as concurrent infection in one flock only. The most prevalent concurrent infection was reovirus, which was observed in eleven of the 38 FAdV infected farms. Avian reoviruses have been shown to enhance the pathogenicity of at least some of the infectious agents in chickens (Rosenberger et al., 1985; Moradian et al., 1991). The possible ways of interactions between FAdV and simultaneously infecting agents are not all known. The fact that immunosuppression arising from IBDV and CAV multiplication in the bursa and thymus may lead to a more virulent course of FAdV infections (Winterberg et al., 1973) confirms their op-

portunistic nature. However, some strains of FAdV impair the function of the humoral and cellular immune systems themselves, thereby leading to immunosuppression and possibly paving the way for other infections (Schonewille et al., 2008). Moreover, the facilitation of invasion through the primary colonization of the intestinal and/or respiratory tract might also be involved. Further research on this aspect of the pathogenesis is required.

Except for the well-circumscribed clinical syndromes, as HHS, IBH and GE, the pathogenic significance of FAdV infections in chickens remains a point of discussion. FAdV isolates are often perceived as opportunistic pathogens that only show their pathogenic potential when additional factors –predominantly concurrent infections– are involved. The present study associated FAdV and related concurrent infections with clinical disease and mortality in Belgian broilers. Since all examinations were done in broiler farms suffering from severe and/or recurrent disease, it could not be ruled out that FAdV infections also occur in flocks showing milder signs of disease or no signs at all. Taking into account that FAdV most typically infects the intestinal tract and organs contributing to the digestion process, such as liver and pancreas, it seems obvious that FAdV infections may interfere with digestion. Suboptimal digestion leads to retarded growth and/or increased feed conversion. Hence, FAdV infections might also have economic consequences in the absence of prominent clinical signs.

REFERENCES

- Adair B.McC., Fitzgerald S.D. (2008). Group I Adenovirus infections. In: Saif Y.M., Fadly A.M., Glisson J.R., McDougald L.R., Nolan L.K., Swayne D.E. (editors). *Diseases of Poultry*. 12th Ed., Blackwell Publishing Professional, Ames, IA., 252-266.
- Benko M., Harrach B., Both G.W., Russell W.C., Adair B.M., Adam É., de Jong J.C., Hess M., Johnson M., Kajon A., Kidd A.H., Lehmkuhl H.D., Li Q.G., Mautner V., Pring-Akerblom P., Wadell G. (2005). Family Adenoviridae. In: Fauquet C.M., Mayo M.A., Maniloff J. (editors). *Virus Taxonomy: VIIIth Report of the International Committee on Taxonomy of Viruses*. Elsevier Academic Press, New York, NY, 213–228.
- Choi K.S., Kye S.J., Kim J.Y., Jeon W.J., Lee E.K., Park K.Y., Sung H.W. (2012). Epidemiological investigation of outbreaks of fowl adenovirus infection in commercial chickens in Korea. *Poultry Science* 91, 2502-2506.
- Dahiya S., Srivastava R.N., Hess M., Gulati B.R. (2002). Fowl adenovirus serotype 4 associated with outbreaks of infectious hydropericardium in Haryana, India. *Avian Diseases* 46, 230-233.
- Dar A., Gomis S., Shirley I., Mutwiri G., Brownlie R., Potter A., Gerds V., Tikoo S.K. (2012). Pathotypic and molecular characterization of a fowl adenovirus associated with inclusion body hepatitis in Saskatchewan chickens. *Avian Diseases* 56, 73-81.
- De Herdt P., Defoort P., Van Steelant J., Swam H., Vanrobbaeys M. (2008a). *Enterococcus cecorum* osteomyelitis and arthritis in broiler chickens. *Vlaams Diergeneeskundig Tijdschrift* 78, 44-48.
- De Herdt P., Paul G., Koopman R., Van De Zande S. (2008b). Prevalence and significance of ERS type reovirus infections in diseased broilers reared under Western European field circumstances. *Vlaams Diergeneeskundig Tijdschrift* 78, 168-172.
- De Herdt P., Broeckx M., Vankeirsbilck W., Van Den Abeele G., Van Gorp S. (2012). Improved broiler performance associated with *Ornithobacterium rhinotracheale* vaccination in breeders. *Avian Diseases* 56, 365-368.
- Domanska-Blicharz K., Tomczyk G., Smietanka K., Kozaczynski W., Minta Z. (2011). Molecular characterization of fowl adenoviruses isolated from chickens with gizzard erosions. *Poultry Science* 90, 983-989.
- Emmy K., Pallister J., Sheppard M. (1995). Immunological and molecular comparison of fowl adenovirus serotypes 4 and 10. *Archives of Virology* 140, 491-501.
- Gomis S., Goodhope A.R., Ojick A.D., Willson P. (2006). Inclusion body hepatitis as a primary disease in broilers in Saskatchewan, Canada. *Avian Diseases* 50, 550-555.
- Günes A., Marek A., Grafl B., Berger E., Hess M. (2012). Real-time PCR assay for universal detection and quantitation of all five species of fowl adenoviruses (FAdV-A to FAdV-E). *Journal of Virological Methods* 183, 147-153.
- Harrach B., Benko M., Both G.W., Brown M., Davison A.J., Echavarría M., Hess M., Jones M.S., Kajon A., Lehmkuhl H.D., Mautner V., Mittal S.K., Wadell G. (2012). Family Adenoviridae. In: King A.M.Q., Adams M.J., Carstens E.B., Lefkowitz E.J. (editors). *Virus Taxonomy: IXth Report of the International Committee on Taxonomy of Viruses*. Elsevier Academic Press, San Diego, CA, 125-141.
- Hess M., Raue R., Prusas C. (1999). Epidemiological studies on fowl adenoviruses isolated from cases of infectious hydropericardium. *Avian Pathology* 28, 433-439.
- Hess M. (2000). Detection and differentiation of avian adenoviruses: a review. *Avian Pathology* 29, 195-206.
- Hess M. (2011). Fowl adenovirus infections in chickens: current status and control approaches. In: *Proceedings of the XVII World Veterinary Poultry Congress, Cancun, Mexico*, 41-48.
- Jones R.C., Georgiou K. (1984). Experimental infection of chickens with adenoviruses isolated from tenosynovitis. *Avian Pathology* 13, 13-23.
- Marek A., Schulz E., Hess C., Hess M. (2010). Comparison of the fibers of fowl adenovirus A serotype 1 isolates from chickens with gizzard erosions in Europe and apathogenic reference strains. *Journal of Veterinary Diagnostic Investigation* 22, 937-941.
- McFerran J.B., Gordon W.A.M., Taylor S.M., McParland P.J. (1971). Isolation of viruses from 94 flocks of fowl with respiratory disease. *Research in Veterinary Science* 12, 565-569.
- Moradian A., Thorsen J., Julian R.J. (1991). Single and combined infection of specific-pathogen-free chickens with infectious bursal disease virus and an intestinal isolate of reovirus. *Avian Diseases* 34, 63-72.
- Meulemans G., Boschmans M., Van den Berg T.P., Decaesstecker M. (2001). Polymerase chain reaction combined with restriction enzyme analysis for detection and differentiation of fowl adenoviruses. *Avian Pathology* 30, 655-660.
- Nakamura K., Mase M., Yamaguchi S., Yuasa N. (2000). Induction of hydropericardium in one-day-old specific-pathogen-free chicks by adenoviruses from inclusion body hepatitis. *Avian Diseases* 44, 192-196.
- Nakamura K., Ohyama T., Yamada M., Abe T., Tanaka H., Mase M. (2002). Experimental gizzard erosions in

- specific-pathogen-free chicks by serotype 1 group I avian adenoviruses in broilers. *Avian Diseases* 46, 893-900.
- Okuda Y., Ono M., Yazawa S., Imai Y., Shibata I., Sato S. (2001). Pathogenicity of serotype 1 fowl adenovirus in commercial broiler chickens. *Avian Diseases* 45, 819-827.
- Olson N.O. (1950). A respiratory disease (bronchitis) of quail caused by a virus. In: *Proceedings of the 54th Annual Meeting of the US Livestock Sanitary Association*, 171-174.
- Ono M., Okuda Y., Yazawa S., Imai Y., Shibata I., Sato S., Okada K. (2003). Adenoviral gizzard erosions in commercial broiler chickens. *Veterinary Pathology* 40, 294-303.
- Rosenberger J.K., Fries P.A., Cloud S.S., Wilson R.A. (1985). In vitro and in vivo characterisation of avian *Escherichia coli*. II. Factors associated with pathogenicity. *Avian Diseases* 29, 1094-1107.
- Schonewille E., Singh A., Göbel T.W., Gerner W., Saalmüller A., Hess M. (2008). Fowl adenovirus (FAdV) serotype 4 causes depletion of B and T cells in lymphoid organs in specific pathogen-free chickens following experimental infection. *Veterinary Immunology and Immunopathology* 121, 130-139.
- Steer P.A., O'Rourke D., Ghorashi S.A., Noormohammadi A.H. (2011). Application of high-resolution melting curve analysis for typing of fowl adenoviruses in field cases of inclusion body hepatitis. *Australian Veterinary Journal* 89, 184-192.
- Van den Ende M.P., Don P.A., Kipps A. (1949). The isolation in eggs of a new filterable agent which may be the cause of bovine lumpy skin disease. *Journal of General Microbiology* 3, 174-182.
- Winterfield R.W., Fadly A.M., Gallina A.M. (1973). Adenovirus infection and disease. I. Some characteristics of an isolate from chickens in Indiana. *Avian Diseases* 17, 334-342.
- Zadravec M., Slavec B., Krapež U., Kaján G.L., Racnik J., Juntos P., Cizerl L.J., Benko M., Zorman Rojs O. (2011). Inclusion body hepatitis associated with fowl adenovirus type 8b in broiler flock in Slovenia – a case report. *Slovenian Veterinary Research* 48, 107-113.

Uit het verleden

DR. WILLEMS

Jeneverstoker en pionier van de vaccinatie tegen besmettelijke runderpleuropneumonie (zie katern in het VDT 2000, nr. 3).

Bronzen beeld van Luc Steegen in de Hasseltse Witte Nonnenstraat naast het Jenevermuseum. Bemerkt de 'koetjes' op het voetstuk.

