

## Summary and conclusion

### A) Field cases:

- ❖ A total of 100 broiler chickens suspected for avian infectious bronchitis were collected from 33 farm flocks (from Fayoum and Beni-Suef) during the period between February 2012 to May 2014.
- ❖ A thick fibrinonecrotic or caseous plug was found at tracheal bifurcation and may extend to involve the primary bronchi that appeared microscopically fibrinous exudate mixed with desquamated necrotic epithelium, in addition to inflammatory cells mainly of mononuclear type
- ❖ Degenerative changes, hyperplastic proliferation of tracheal epithelium and necrosis with desquamation of necrotic epithelium were seen.
- ❖ The lung was enlarged and congested. Concerning the conducting system showed degeneration and necrosis of lining epithelium. The wall of both primary and secondary bronchi may be thickened due to edema and leukocytes infiltration of their lamina propria.
- ❖ Microscopically, lung tissue showed interstitial pneumonia in the form of diffuse mononuclear cell infiltrations.
- ❖ Swollen pale kidneys with focal hemorrhagic spots were observed macroscopically. Hypercellularity together, tubular degenerative changes, mononuclear cell infiltration (interstitial nephritis) were seen.
- ❖ Bursa of Fabricius showed depletion and atrophy of lymphoid follicles. Interstitial edema, congestion, infiltration and fibroplasia were remarked.

- ❖ Proventriculitis was evidenced by degeneration, necrosis and some time complete loss of the covering mucosal epithelium. Proventricular glandular epithelium showed necrobiotic changes from vacuolar degeneration till necrosis and desquamation which most common with degree from mild to moderate. Mononuclear cell infiltration with few heterophils and congestion of blood vessels were seen inter and intra glandular.
- ❖ Remarkable histopathological alterations in pectoral muscle were characterized by necrosis and hyalinosi with muscle fibers fragmentation and mild mononuclear cell infiltration
- ❖ Positive immunohistochemical labeling of IBV antigen was detected in tissues of trachea, kidney, lung, bursa of Fabricius and proventriculus.

#### **B) Experimental study:**

- ❖ One hundred and five -1- day old SPF chickens were used in this study divided into 3 groups of 35 chicks. First and second infected groups were inoculated intranasally and intraperitoneally with 0.2 ml of  $10^5$  EID<sub>50</sub> of classical and variant virus isolates respectively, while third control group was inoculated with 0.2 ml of 0.01 M PBS. Clinical signs were recorded daily for 15 DPI. 5 chicks from each group are killed at 2, 4, 6, 8, 10 and 12 DPI with all surviving chicks were euthanized at 15 DPI. Tissue specimens were collected from, trachea, kidneys, lung, proventriculus, bursa of fabricius pectoral muscle, esophagus and small intestine for histopathological and immunohistochemical studies and RT-PCR.
- ❖ No mortalities, in addition to the presence of respiratory signs in the form of mild sneezing developed at 3 DPI and lasted till 8 DPI in both inoculated groups.

- ❖ The common histopathological lesions detected on examined tracheal sections from all infected groups at the different sacrifices included tracheitis, deciliation and necrosis of the lining epithelium, hyperplasia in epithelial lining, and finally evidences of epithelial regeneration. IBV antigen detected as fine brown granules in the lining epithelium of trachea, mucous glands and sometime attached to cilia that confirmed by RT-PCR detection.
- ❖ Most commonly detected lesions in lungs included degeneration, desquamation and hyperplasia of epithelium lining conducting system with congestion, edema and infiltration of thier lamina propria of conducting system and lung tissue
- ❖ The most common lesions in kidneys were paleness and enlargement in both infected groups. Microscopically, vacuolar degeneration in renal tubular cells and podocytes, nuclear pyknosis with epithelium sloughing were seen. Interstitial tissue infiltrated with mononuclear inflammatory cells. The viral antigen was detected in the cytoplasm of proximal, distal convoluted tubules and glomerular epithelium; in addition in case of classical IBV-inoculated chicks the antigen was detected in the embryonic rests and Henle's lope, these results were confirmed by RT-PCR.
- ❖ The most common histopathological lesions in proventriculus of both inoculated group were hyperplasia and hypersecretion of mucosal epithelium, focal necrosis and desquamation in mucosal epithelium of some proventricular glands. Mild pyknotic changes of epithelium lining adenomeres and moderate vacuolar degeneration with hyalinosis in the musculosa. Focal intraglandular infiltration was also observed. IBV was detected by RT-PCR and appeared as fine brown granules in adenomere lining epithelium, proventricular

musculosa and in the cytoplasm of lamina propria infiltrated macrophages.

- ❖ The main histopathological changes of pectoral muscle of both classical IBV-inoculated and variant IBV-inoculated chicks were necrosis of myofibers and degenerative changes in blood vessels. Edema and solitary inflammatory cells between myofibers with detection of IBV antigen inside myofibers of both classical and variant IBV-inoculated chick that confirmed by RT-PCR.
- ❖ The main histopathological changes of esophagus of both classical IBV and variant IBV-inoculated chicks were vacuolar degeneration and nuclear pyknosis of mucosal epithelium with detection of IBV antigen as fine brown granules in the mucosal epithelium, lamina propria and musculosa of both groups
- ❖ Most commonly detected lesions in the small intestine were necrosis and desquamation from tips of villi with focal mononuclear cells infiltration with IBV detection in the epithelial cells of intestinal villi and inflammatory cells.

### ***Conclusions***

- ❖ The main cause of death in IBV infection is suggested to be a combination between respiratory failure, asphyxia (due to suffocation) and renal failure.
- ❖ The most affected organs were respiratory and urinary organs in both field cases and experimental study with detection of viral antigens replicating in these organs causing histopathological alterations
- ❖ The results of the experimental study indicated that the the classical and variant IBV isolates did not cause either mortalities or severe clinical signs and gross lesions in experimentally infected SPF chickens.

- ❖ Using IHC, antigens of IBV were detected in trachea, lung, kidney, bursa and proventriculus. Most importantly, we found infection of duodenum, jejunum, esophagus and pectoral muscle, these results were consistent with RT-PCR results and denote the wide tissue tropism of Egyptian IBV field isolates in SPF inoculated chicks.
- ❖ Further studies should be carried out to study the role of these strains in co-infection with other bacterial and fungal infections.