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SOME INVESTIGATION ON INFECTIOUS BOVINE RHINOTRACHEITIS (IBR) INFECTION IN FRIESIAN CALVES

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SUMMARY

Respiratory distress was shown to be problem in 1200 friesian calves belonging to military farm, El-Tall El-Kabeer. Ismailia Governorate, Egypt. The problem began two weeks after importation and transportation of calves from western Nobaria to El-Tall El-Kabeer farm. The clinical signs observed were fever, dyspnea, nasal and ocular discharge, conjunctivitis, corneal opacity and occasionally coughing. Some complicated cases were emergency slaughtered or died. The signs were more serious in young calves (6-10 months) than in older calves (11-15 months). The morbidity rate was 77.16% while, the mortality rate was 6.59%.

Nasal and ocular swabs were taken for virological and bacteriological examinations. Also paired serum samples were collected from diseased calves for serum neutralization test (SNT). Bovine herpesvirus type 1 (BHV-1) was isolated on MDBK cell line and confirmed by virus neutralization test (VNT). SNT revealed high titer which reached up to 1/320 . In complicated cases *Pasteurella haemolytica* was isolated Trasportation, overcrowding, and bad weather were considered to be predisponing factors for the disease. Injection of Enrofloxcin 10% (cidotryl-cid, Egypt, 1ml/40Kg. B. W.) and tonics were given to mininize the course of the disease and secondary bacterial complication.

INTRODUCTION

IBR plays a prominent role among causes of undifferentiated respiratory diseases of feedlot cattle especially in intensive farm production. IBR virus can produce a mild inapparent infection or a wide variety of clinical infection, some of these have profound effects on feed efficiency, milk production and reporduction. (Anonymous 1979).

IBR was first described as a new respiratory tract disease of feedlot cattle in western United States (Miller, 1955). Isolation of the virus was accomplished soon thereafter (Madine et al., 1956). It is called bovine herpesvirus 1 (Gibbs and Rweyemamu 1977). The outcome of natural infection is probably determined multifactorially by the strain of virus, dose and route of exposure or inoculation, immunologic status of the exposed animal, and enviroumental influences. (Schultz et al., 1976). The respiratory form of IBR is manifested by fever, reduced appetite, rapid respiration, dyspnea, nasal discharge and dilated nostrils (Kahrs and Smith, 1965). Some cattle

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with respiratory form of IBR have conjunctivitis (Ferris et al., 1964). In the respiratory tract, the lesions are most common in the trachea and nasal passages, the characteristic lesion on mucosal surfaces is adherent whitish necrotic material raised above the mucosal surface (Studdert et al., 1964). Laboratory confirmation in suspected cases of IBR can be obtained by serologic tests or virus isolation (Carbrey et al., 1972). IBR virus was isoalted from an outbreak in herd of fattening calves. The virus was identified by clinical symptoms, postmortem lesions, electron microscope and serolgoically.

Bovine herpesvirus 1 was isolated in bovine fetal kidney cell monolayers from lungs of 2 lambs showing hepatization of the lungs (Cavalli et al., 1994). Winkler et al. (1995) studied the neutralizing monoclonal antibodies to bovine herpesvirus type 1 by immunoperoxidase and immunoelectron microscopy. Serum neutrization test was used for the presence of antibodies to bovine herpeswirus 1 (BHV 1) on serum samples collected from dairy herds in Brazil (Lovato et al., (1995) . Lu and Kwang (1989) propagated IBR virus in MDBK cells. Lu et al. (1989) isolated IBR virus during outbreak from nasal discharges.

Pneumonic pasteurellosis is a primarily disease of recently weaned newly housed calves (Dalgleish 1990). There is often a history of movement, mixing and confinement of cattle.

The present study was planned to investigate the causative agents of respiratory distress to control the problem.

MATERIAL AND METHODS

Animals:-1200 friesian calves imported fc production to military farm No. 9 E El-Kabeer, Ismailia Governorate, Egyp calves aged from 6-15 months. The farn subjected to many outbreaks of respi distress and eye affections. Samples (nas ocular swabs as well as serum samples) taken from diseased as well as emer slaughtered or dead animals.

Table (1). Age distribution of calves

Age of calves	No.
6 -10 months	740
11 -15 months	460
Total	1200

Serum samples

A total of 942 paired blood samples collected 2 weeks apart. The sera were sepa and kept at -20 °C until examined.

Nasal and ocular Swabs:- 320 nasal swab 320 ocular swabs were collected at an early of the disease on maintenance media and sto -20°C until used for virological isolation identification.

Samples from emergency slaughtered or dead animals:-

Nasal and tracheal swabs, lung exudate, citrated blood, lymph nodes and liver samples were taken for virological and bacteriological examination under aseptic conditions and examined at laboratory as soon as possible.

Cells:- MDBK cell line was obtained from Veterinary Serum and Vaccine Production and Research Institute, Abbassia, Cairo.

Media:- Maintenance and growth media were obtained from Animal Health Research Institute Dokki, Giza.

Virus strain:- IBR virus Abou-Hammed strain which was isolated by Hafez (1973), kindly supplied by Animal Health Research Institute, Dokki.

Positive-Immune sera:- IBR hyperimmune serum was kindly supplied by Animal Health Research Institute, Dokki.

Serology:- All serum samples were tested for presence of IBR antibodies by serum neutralization test (SNT).

Serum - Neutralization test (SNT):- was carried out according to Cabrey and Lee (1966).

- Serum samples were inactivated at 56°C for 30 minutes and serially diluted two fold in microtitration plates.

Virus in the amount of 100 TCID50 was added to each serum specimen dilution. After incubation for 1 hour at 37°C MDBK cell were added in an amount sufficient to form a monolayer . The plates were incubated for 3 days and then examined miroscopically. The reciprocal of the highest serum dilution that completely inhibited the appearance of the viral cytopathic effect (CPE) was taken as the SN titer.

Virus isolation:

All nasal and ocular swabs firstly centrifuged at 3500 r. p. m. for 15 minutes then filtered before using for viral isolation on cell cultures.

Attempts of viral isolation from nasal and ocular swabs:-

Monolayer cultures (MDBK cells) were prepared in microtiter paltes and incubated at 37°C in humidified 5% CO_2 atmosphere until confluent monolayer was formed, then growth medium was removed and a volume of 0.025 ml. from samples were inoculated into the monolayer using 3 wells per each sample.

The inoculum was evenly distributed over the cell sheet. Cultures were incubated in CO_2 incubator at 37°C for 1 hour to permit viral adsorption. Then maintenance medium was put on each well of microtiter plate, incubated at 37°C in humidified 5% CO_2 incubator. Each well was checked under inverted microscope daily to show cytopathic effect (CPE) if positive.

Islation and identification of secondary bacterial invasion:

61 samples taken from emergency slaughtered or dead animals were examined bacteriologically.

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These samples were streaked over Petri plates containing blood agar base (Difco laboratories, Detroit, Michigan) with 5% bovine blood, incubated overnight at 37°C. Plates were then examined for pasteurella-like colonies. These were streaked for reisolation on blood agar incubated at 37°C ovenight and examined for pasteurella-like colonies by morphology and haemolysis. Growht on MacConkey was tested by Gram-staining and biochemical reactions. The isolation, purification and identification of bacterial isolates was carried out according to Cruickshank et al. (1978).

RESULTS

A problem of respiratory distress and eye affection occurred in military farm No. 9 El-Tall El-Kabeer. Ismailia, Egypt, where the calves were kept for intensive beef production. The clinical signs including fever, nasal and ocular discharge, anorexia, depression, rapid respiration, cough, salivation, dyspnea, conjunctivitis and occasionally corneal opacity. Hyperemia, oedema and hemorrhagic focal necrosis of the na[sal] mucosa and diarrhea. Some animals show[ed] severe clinical signs with serious complicatio[ns] and were emergency slaughtered or died. Th[e] morbidity rate reached 77.16% (926 casses) whi[le] the mortality rate was 6.59% (61 cases).

Virus isolation:

IBR virus has been isolated from nasal and ocula[r] swabs. Cytopathic effect (CEP) was observed i[n] MDBK cells, there was the characteristi[c] herpesviral CPE in cell line. The isoalte was als[o] neutralized by bovine hyperimmune serum o[f] IBRvirus by VNT.

Serological test (SNT):

SNT revealed IBR antibody, the titer reached 1/320.

Table (2). Clinical cases from which virus was isolated .

Swabs from diseased animals	Total	No. of positive
Nasal swabs	320	204
Ocular swabs	320	178

Complicated cases which showed secondary bacterial invasion:

Pasteurella haemolytica was isolated from 24 samples taken from camplicated cases. Table (4).

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Table (3). Morbidity and mortality rate .

Animals	Total No.	No. of +ve	Morbidity%	No.of E.S.or dead A.	Mortality %
Calves (6 - 10 months)	740	594	80.27%	45	7.58%
Calves (11-15months)	460	332	72.17%	16	4.82%
Total	1200	926	77.16%	61	6.59%

E.S. = emergency slaughtered A= Animals

Table. (4). Results of isolation from complicated cases .

No. of Animal exam.	Total +ve cases to viral isolation	+ve cases to viral isolation only	+ve cases to pasteurella & virus	- ve cases
61	51	27	24	10

DISCUSSION

An outbreak of severe upper respiratory tract infection occurred in 6-15 months friesian calves intended for intensive beef production at military farm, El-Tall El-Kabeer, Ismailia. The calves showed fever, respiratory distress, nasal and ocular discharge, conjunctivitis and corneal opacity. Some complicated cases were emergency slaughtered or died. The signs were serious in young calves (6-10 m), than older calves (11-15 m.). The morbidity rate was 77.16%, while the mortality rate was 6.57%, similar results were recorded by Kahrs and Smith (1965).

The IBR virus was isolated from nasal and ocular discharge, tracheal and lung exudate from diseased, emergency slaughtered and dead animals Bovine herpes virus (BHV-1) was isolated on MDBK cell line and this isolate was confirmed by VNT. These results coincide with those of Lu et al. (1989) and Cavalli et al. (1994). Pasteurella haemolytica was the secondary bacterial invasion that caused serious disease. So, some calves were emergency slaughtered or died. Similar results were obtained by many authors (Ferris et al., 1964., Rosner 1968, Dalgleish 1990).

Transportation, overcrowding, mixing and bad weather were considered the predisposing factors for the disease.

The examination of paired serum samples revealed 1/320 titer of antibodies against IBR virus, similar results were obtained by Lovato et

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al. (1995).

The majority of calves subjected to the disease had received Enrofloxcin 10% (cidotrylcid, Egypt 1 ml/40 Kg. B. W.), tonics and electrolyte to minimize the course of the disease and prevent the secondary bacterial complications.

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