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Preliminary Investigation of IBR in Buffaloe (Bubalusbubalis) and Cattle (Cross Bred) in Baghdad/ Iraq.

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Abstract: *Infectious bovine rhinotracheitis (IBR) is an economically important disease of cattle and buffaloes, and causes abortions, infertility, meningeal encephalitis, and keratoconjunctivitis. The aim of the study was to determine seroprevelence of the IBRantibodiesin two herds, 26buffaloes and 21 cow related agriculture research office and veterinarymedicine college respectively in Baghdad, these animals did not take any vaccination program against the disease.For the serum samples tested with enzyme-linked immunosoarbentassay(ELISA) used specifically for detection IBR antibodiessurvelence, which was apartof survey for detection of the disease in cattle &buffaloe in Baghdad. The results showed the highest seropositivity percentage in buffaloes 65.36%(17:26), while in cattle showed the lowest seropositivity 4.7%(1:21). These results showed that buffaloes are more susceptable than cattle to the infection in this study.*

I. Introduction:

Infectious bovine Rhinotraciatis1s(IBR) is caused by bovine herpes virus-1(BHV-1) of the family Herpes viridae and the subfamily Alphaherpesviridae, known to cause several diseases worldwide incattle, including Rhinotraciatis, vaginitis, balanoposthitis, abortion, conjunctivitis, and enteritis. BHV-1is also a contributed factor in shipping fever, also known as Bovine Respiratory disease (BRD,)respiratory and genital tract infections. It is spread horizontally through sexual contact, artificial insemination, and aerosol transmission and it may also be transmitted vertically across the placenta.infectious Bovine Rhinotracheitis is a highly contagious disease caused by the bovine herpes virus-1 (BoHV-1), resulting in significant losses to livestock around the world (Nardelli et al 2008). The disease is noticeable in many, but not all countries. Bovine herpesvirus 1 (BoHV-1), classified as an alphaherpesvirus, is a major infection of a seronegative cow (Ackermann & Engels 2006). Naturally occurring BoHV-1 abortions are usually observed at 4 to 8 months of gestation, although experimental virus inoculation of heifers prior to 3 months induces embryonic death (Muylkens et al 2007). Like other alphaherpesviruses, an essential intrinsic characteristic of BoHV-1 is that the virus remains in a latent state in ganglionic sensory neurons following infection and can be reactivated by different stress stimuli, It is one of the most widespread respiratory/reproductive viral diseases of bovines in India (Kiran et al,2005). There are many serological tests for detection BoHV-1 antibodies, the virus neutralization test (VNT), gB-ELISAs and indirect enzyme-linked immunosorbent assays (ELISAs), cannot differentiate between infected and vaccinated animals (Kramps et al 2004). Serum neutralization tests and various ELISA are routinely used for BoHV-1 antibody detection (Van Oirschot, 2000.)As ELISA is more sensitive (Payment et al, 1976).

IBR has been known to exist in India since 1976 (Mehrotra et al. 1976). The reported seroprevalence in buffaloes varied from 2.75 to 81.0 percent while in cattle ranges from 50.9 to 60.46% in India (Renukaradhya et al ., 1996; Renukaradhya et al ., 2002, Malmurugan et al ., 2004, Trangadia et al ., 2009). & (Sinha et al. 2003, Malmurugan et al. 2004).In Iraq a study was perfumed by Al-Bena et al, 1985) : who reported isolation of the virus for the first time and diagnosed the disease clinically in cattle at a dairy farm station, Baghdad. The affected cattle were showing rise of body temperature, severely congested nasal mucous membrane and conjunctivae(Al-Bena et al,1985).The aim of this study is to investigate antibodies against Bovine herpes virus(BOHV-1) seroprevalence of infectious bovine rhinotracheitis based on avid in-biotin ELISA in buffaloes& cattle herds.

II. Materials and methods:

Samples Collection:(21) cattle &(26) buffaloes blood samples were collected from the cows and buffaloes herds of Veterinary Medicine College/ Baghdad University and Agriculture Research Office respectively. This study was conducted at the veterinary directorate/ Department of Central Veterinary Laboratories and Researches CVL(serology unit).The blood samples were centrifuged at 2000 × g at room temperature for five minutes to separate sera. Serum samples were stored at (-20c) until used inIndirect ELISA assay. Serum samples were tested for the presence of anti-gB antibodies using Antibody Test kit manufactured by IDEXX (HerdChek, IDEXX Laboratories, Westbrook, ME, USA), in a 96-well micro titration plates. Tests were carried out in duplicate, according to the manual, serum samples were diluted (1:1) by wash solution,and 100µl of diluted sera was loaded into wells and incubated for 2 hours at 37 ºC. Positive and negative control sera were used as indicated in the kit. Thewells were washed five times with 300µl of wash solution. Following the final washing, the plates slapped vigorously, well down on a bench top which covered with paper towels. Then, 100 µl of anti IBR-gB Horseradish Peroxidase (HRP) conjugated was loaded into all the wells and incubated for one hour at room temperature. The plates were washed as described above to remove the excess conjugate. For color development, 100 µl of 3.3', 5.5'-Tetramethyl benzidine (TMB) substrate solution (TMB/H2O2 solution) was added to each well and incubated for 10 minutes at room temperature at darkness. The reaction was terminated by the addition of 100 µl of stop solution to each well. The absorbance at 450 nm was monitored in ELISA reader Calculation. Calculations for test samples were analyzed as follow for BoHV1 antibody: The presence or absence of antibody to IBR-gB in the sample is determined by the blocking percentage for each sample. NCx^{-} A450- (OD Sample) A450 x100(%)/NCx^{-} A450).NCx^{-} represent negative control mean. OD represents absorbance of each sample in 450 um. According to manual, samples with blocking less than 55% were classified as negative, samples with blocking equal or greater than 50% but less than 55% were classified as suspected and must be retested, and samples with blocking equal or greater than 55% were considered as positive for IBR antibody.

III. Results:

Table (1) showed that the positivity percentage of IBR in buffaloes was 17 out of 23; (73.9%), while the positivity percentage of IBR in cattle was 1 out of 13 ;(7.6%)

***Table (1): the detection of IBR by ELISA in Buffaloes**

Final Results	OD	Animal number
+ve6.067747	1.735	4
+ve 6.951399	1.753	32
+ve 6.480118	1.753	40
+ve 7.481591	1.753	28
+ve 5.832106	1.753	8
+ve5.655376	1.753	32
+ve5.007364	1.753	50
-ve 96.02356	1.753	53
-ve 94.23626	1.753	76
+ve 11.07511	1.753	22
-ve 90.83947	1.753	6
+ve 5.366082	1.753	54
-ve 65.86156	1.753	1
+ve 6.597938	1.753	14
-ve 70.7511	1.753	36
+ve 6.539028	1.753	68
+ve 4.418262	1.753	46
+ve 7.18704	1.753	65
+ve7.481591	1.753	66
+ve 6.421208	1.753	62
-ve 85.71429	1.753	6
+ve 3.949681	1.75	68
-ve94.25626	1.753	67

***Table (2): detection of IBR by Elisa in Cattle**

Final Results	OD	Animal Number
-ve 108.1591	1.753	497
-ve 62.21077	1.753	2711
+ve 27.86451	1.753	2620
-ve 93.72607	1.753	2707
-ve 94.02062	1.753	81
-ve 88.7187	1.753	1716
-ve 73.28424	1.753	2720
-ve 93.49043	1.753	2705
-ve 93.25479	1.753	1419
-ve 92.37113	1.753	2716
- ve 95.49337	1.753	4015
-ve 80.23564	1.753	3981
-ve 105.1546	1.753	4046

***The two tables analyzed by student t- test**

IV. Discussion:

In this study, the presence of antibodies against BoHV-1 in serum samples of cattle's&buffaloes was tested by Indirect ELISA and the seroprevalence of IBR was 17 out of 23;(73.9%) in buffaloes which is more than cattle1 out of 21;(4.76%) and this was agree with other researchersperformed in Andhra & Gujarat.(Renukaradhyaetal., 1996; Sinhaet al, 2002, Malmuruganet al.,2004).

The Variation in the prevalence of the disease between buffaloes and cattle's may be due to several factors as methods of sampling, source of samples, intensity of dairy farming, inter-mixing of animals under same husbandry practices, unrestricted movement of infected animals and the extend of control measures adopted(Sinhaet al, 2002). Regarding the seroprevelense of IBR in buffalo was higher(73.9%) and these results agree with (Renukaradhyaet al. 1996,Malmuruganet al. 2004). The present study confirmsthat IBR has established in buffalopopulation warrants efforts to control this enzootic disease asthe infection has a direct impactionproduction bycausing infertility and abortion leading to huge economic loss to farmers and visa versa ourstudy disagree with other research reported by (Sinnha et a l(2003) who found low prevalence of IBR in buffalo,the low prevalence is due to thedifferent geographical location of the animalsscreened (Ardhanaet al. 2004) and less movementof the animals in and out of the unorganized farmsor small farmers from whom most of the animalsusually came for slaughter. The serological presence of IBR with low antibodies titers and no clinical history is available could be interpreted as evidenceof past infection with IBR virus or as slow antibodyresponse following primary infection. The absence of antibodies in the remaining cases also not be freedom from infection as poor antibody response was noticed incows with infectious pustularvulvovaginitis from which the IBR- pustularvulvovaginitis virus was isolated (Snowdon 1964).Regarding the seroprevelencepf IBR in cattle the present study disagree with other studies which were done by (Wang et al (2005)& (Xiao et al 2004)who found the seroprevelence of IBR in cattle was high and these results was implicated to sampling, limited sampling area and size of the herd(Yan et al 2008).

References:

[1]. Al-Bena AS; Shony MO; Mohymen NA., (1985). Association of infectious bovine rhinotracheitis virus in an outbreak of respiratory tract infection in cattle in Iraq. Indian Journal of Comparative Microbiology, Immunology and Infectious Diseases..; 6(1): 5-10.

[2]. Ackermann, M., and Engels, M. (2006). Pro and contra IBR-eradication, Veterinary Microbiology 113**:**293–302.

[3]. Ardhana, Sharma D R, Dhand N K, Singh J and Gumber S. (2004). Status of infectious bovine rhinotracheitis (IBR) in Punjab state. IndianJournal of Animal Sciences 74(3): 264-66.

[4]. Sinha, B.K., Mishra, K.K., Singh, A.P., Kumar, R., (2002),.Seroprevalence of infectious bovine rhinotracheitisinBihar. In Proc. 4th Asian Buffalo Congress on Buffalofor Food Security and Rural Employment, 22-25 February2002, CIRB, Hisar, India, pp 17.

[5]. Trangadia, B.J., Rana, S.K., Mukherjee, F., Srinivasan, V.A.,2009. Prevalence of brucellosis and infectious bovinerhinotracheitis in organized dairy farms in India. TropicalAnimal Health and Production 42 (2), 203-207

[6]. Snowdon W A. (1964). Infectious bovinerhinotracheitis and infectious pustularvulvovaginitis in Australian cattle.Australian Veterinary Journal 40: 277-288.

[7]. Malmurugan S, Raja A, Saravanabava K andDorairajan N. (2004). Seroprevalenceofinfectious bovine rhinotracheitis in cattle andbuffaloes using Avidin- Biotin ELISA.Cheiron33(5&6): 146-49.

[8]. Sinha B K, Mishra K K, Singh A P and Kumar R.(2003). Seroprevalence of infections bovinerhinotracheitis in Bihar. Proceedings of the4th Asian Buffalo Congress on Buffalo forFood Security and Rural Employment. 2: 17.

[9]. Kramps J.A., Banks M., Beer M., Kerkhofs P., Perrin M., Wellenberg G.J., and van Oirschot J.T., (2004). Evaluation of tests for antibodies against bovine herpesvirus 1 performed in national reference laboratories in Europe. Veterinary Microbiology 102: 169-181.

[10]. Mehrotra M L, Rajya B S and Kumar S. (1976).Infectious bovine rhinotracheitis (IBR)kerato-conjuctivitis in calves. IndianJournal of Veterinary Pathology 1:70-73.

[11]. Muylkens, B., Thiry, J., Kirten, P., Schynts, F., and Thiry, E. (2007). Bovine herpesvirus 1 infection and infectious bovine rhinotracheitis. Veterinary Research 38: 181–209.

[12]. Nardelli, S., Farina. G., Lucchini. R., Valorz, C., Moresco, A., Dal Zotto R., and Costanzi, C. (2008). Dynamics of infection and immunity in a dairy cattle population undergoing eradication programme for Infectious Bovine Rhinotracheitis (IBR). Preventive Veterinary Medicine 85: 68-80

[13]. Renukaradhya, G.J., Isloor, S., Rajasekhar, M.,(2002).Epidemiology,zoonotic aspects, vaccination andcontrol/eradication of brucellosis in India. VeterinaryMicrobiology 90, 183-195.

[14]. Renukaradhya, G.J., Rajasekhar, M., Raghavan, R., (1996) .Prevalence of infectious bovine rhinotracheitisinsouthern India. Review Scientific Techniques 15 (3),1021-1028**.**

[15]. Kiran K K, Ravi P and Prabhudas K. (2005). Infectiousbovinerhnotracheitis- National survey ofIBR antibodies by AB-ELISA kit. AnnualReport of Project Directorate on AnimalDisease Monitoring and Surveillance, ICAR,Bangalore 7-10.

[16]. Van Oirscho, J.T. (2000). Infectious bovine rhinotracheitis/infectious pustularvulvovaginitis. In: Manual of Standards for Diagnostic Tests and Vaccines. 4th ed. Paris: Office International des Epizooties 381–391.

[17]. Wang, H.Y., Zhu, Y.M., Xue, F., Tong, G.Z., Zhao, L.P., Xiang, W.H. and Han, W.Y. (2005). Indirect Enzyme-linked Immunosorbent Assay with Recombinant Glycoprotein E of Infectious Bovine Rhinotracheitis Virus. Biotechnology Progress 25**:** 29–33.

[18]. Xiao, D.H., Li, L.H., Jiang, H.X., and Wang, L.Y. (2004). Research in Prevention and Cure of Infectious Bovine Rhinotracdeitis, China Dairy Cattle 4**:** 43–45.

[19]. Yan, B.F., Chao, Y.J., Chen, Z., Tian, K.G., Wang, C.B., Lin, X.M., Chenand, H.C. and Guo, A.Z. (2008). Serological survey of bovine herpes virus type 1 infection in China. Veterinary Microbiology 127: 136-141.