PREVALENCE OF ANTIBODY AGAINST INFECTIOUS BOVINE RHINOTRACHEITIS VIRUS IN SENTINEL CATTLE IN WEST AND EAST NUSA TENGGARA

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ABSTRAK

WIYONO, A., A. SAROSA, M. GEONG, dan S. UTAMI. 1995. Prevalensi antibodi virus *infectious bovine rhinotracheitis* pada sapi sentinel di Nusa Tenggara Barat dan Timur. *Jurnal Ilmu Ternak dan Veteriner* 1 (2): 99-104.

Sejumlah 4.977 serum yang berasal dari sapi sentinel di Nusa Tenggara Barat dan Timur diperiksa kandungan antibodi virus *infectious bovine rhinotracheitis* (IBR). Serum ini dikumpulkan dari bulan Juni 1990 hingga Juni 1993. Uji yang dipergunakan adalah uji serum netralisasi (SN). Dari seluruh serum yang ada, sebanyak 3.713 serum bisa dipergunakan untuk uji SN IBR, dan sebanyak 349 serum (10%) merupakan serum reaktor. Reaktor IBR lebih prevalen di Nusa Tenggara Timur dibandingkan dengan di Nusa Tenggara Barat. Berdasarkan hasil pemeriksaan disimpulkan bahwa antibodi virus IBR sudah terdapat di Propinsi Nusa Tenggara Barat dan Timur.

Kata kunci: Uji SN, BHV-1, IBR, sapi sentinel

ABSTRACT

WIYONO, A., A. SAROSA, M. GEONG, and S. UTAMI. 1995. Prevalence of antibody against infectious bovine rhinotracheitis virus in sentinel cattle in West and East Nusa Tenggara. *Jurnal Ilmu Ternak dan Veteriner* 1 (2): 99-104.

A total of 4,977 sera from a sentinel cattle in West and East Nusa Tenggara were tested for antibody against BHV-1, the causal agent of infectious bovine rhinotracheitis (IBR). These sera were collected between June 1990 and June 1993, and were tested by using serum neutralization test (SNT). Out of these sera, 3,713 were suitable for IBR SNT. A total of 349 sera (10.4%) reacted. IBR reactors were more prevalent in East Nusa Tenggara (NTT) than in West Nusa Tenggara (NTB). Based on this survey, it is concluded that antibodies against IBR virus are present among cattle in East and West Nusa Tenggara.

Keywords: SNT, BHV-1, IBR, sentinel cattle

INTRODUCTION

Infectious bovine rhinotracheitis-infectious pustular vulvovaginitis (IBR-IPV) is an acute and contagious viral disease affecting cattle and buffaloes. The causal agent is Bovine Herpesvirus-1 (BHV-1) (GIBBS and RWEYEMAMU, 1977). IPV is a genital form of BHV-1 infection which was recognized in Europe long before IBR was known as a respiratory form of BHV-1 infection. GIELESPIE *et al.* (1959) observed a close antigenic relationship between IBR and IPV viruses. On the basis of serological results, an antigenic cross-reactivity between two viruses was identified, and these viruses were named IBR-IPV virus and classified as BHV-1 (LUDWIG, 1984).

A wide range of clinical signs of IBR-IPV have been reported. It has been associated with respiratory, reproductive tract, central nervous system, intestinal tract and skin disease (GIBBS and RWEYEMAMU, 1977). KAHRS (1977) suggested that IBR-IPV infection plays a role in unknown respiratory disorder and abortion.

Infection of IBR-IPV is transmitted by direct contact especially under crowded conditions. Large quantities of virus are shed in respiratory, ocular and reproductive secretions of infected cattle (KAHRS, 1977). The disease is worldwide distributed. However, the introduction of artificial insemination decreases the occurrence of clinical IPV (LUDWIG, 1984).

Clinical manifestation of the respiratory form of IBR infection varies from animals to animals including fever, increased respiratory rate, anorexia, depression, drop in milk production for milking cows, serous to mucopurulent nasal and ocular discharge and hyperemic of nasal mucosa. In mild cases, clinical signs may only include serous nasal and ocular discharge (GIBBS and RWEYEMAMU, 1977).

The diagnosis of IBR infection is based on viral isolation, history of the disease, serological tests where there is a seroconversion from negative to positive or a fourfold increase in titre, and histopathological findings (KAHRS, 1977).

With regard to serological study of IBR, antibody against IBR virus is prevalent among Indonesian buffaloes and cattle. In North Sumatra, NOOR *et al.* (1984) found that among buffaloes in this province, the prevalence rate was between 15 and 20%. In Lampung Province, antibody against IBR virus was also prevalent in Bali cattle (MARFIATININGSIH, 1982; ARJONO *et al.*,1984; WIYONO, 1993). A broader survey conducted by SAROSA (1985) indicated that the prevalence rate of antibody against IBR virus among cattle and buffalo from the islands of Sumatra, Java and Bali was 20%.

A survey was conducted as part of a project on animal health and production (Cattle Health and Productivity Survey, CHAPS) in sentinel cattle in the Provinces of West and East Nusa Tenggara from June 1990 to June 1993. This paper describes the prevalence rate of antibody against IBR virus in cattle during the survey.

MATERIALS AND METHODS

Sera

A total of 4,977 sera were received from the CHAPS project in the period from June 1990 to June 1993. They were received in 13 different consignments which were called CHAPS 1 to CHAPS 13. All sera were heat inactivated at 56°C for 30 minutes prior to testing.

Virus stocks

An IBR virus (Colorado strain) was used. The virus was grown in African green monkey kidney (VERO) cell lines in a minimal essential medium (MEM) supplemented with 10% foetal bovine serum (FBS). It was titrated in microtitre plates using 10 fold dilutions to determine the titre of the viral stock. The viral concentration which was used for the serum neutralization test (SNT) was 100 tissue culture infective dose ($TCID_{50}$) per 0.025 ml.

Screening test using a serum neutralization (SN) test for IBR

The serum neutralization test was used as recommended by DENNETT (1976) with some modifications. The test was conducted in 96 well microtitre plates (Nunc), where 0.025 ml of tested sera were mixed with 0.025 ml of MEM containing 100 $TCID_{50}$ of virus. The microtitre plates were sealed with plastic and incubated at 37°C for one hour. Each serum was tested in duplicate. After incubation, 0.100 ml of a suspension of VERO cells with a concentration of $3x10^5$ cells per ml were added to each well. Each test was consisted of tested sera, cell controls, reactor and non-reactor serum controls, and back titration of the virus. The plates were re-sealed with plastic and incubated at 37°C, and observed daily for cytopathic effect (CPE) for five days. A tested sera which showed a titre of equal to 1:2 or greater than 1:2 was considered to be a reactor sera.

Data recording and analysis

Sentinel data was taken from a PANACEA data base (PAN LIVESTOCK) which consisted of serum bank data at Balitvet, Bogor. However, there were difficulties in matching some samples due to coding problems. Regarding this, reactive sera in each CHAPS collection were screened and identified manually. The percentage of reactor animals in East and West Nusa Tenggara were compared and statistically analyzed using Stat-Sak program (The Statistician's Swiss Army Knife-Version 2.40).

RESULTS

A total of 4,977 sera were received and tested for IBR SNT in the study. These were consisted of 3,713 sera (74.6%) which were suitable for IBR SNT, and 1,264 sera (25.4%) which were unsuitable for IBR SNT due to bacterial contamination and toxication to the cell culture system of IBR SNT (Table 1c).

The sera which were not suitable for IBR SNT came mainly from the first four CHAPS collection (CHAPS 1 to CHAPS 4). The unsuitable sera occurred in all collection periods except for sera collected at CHAPS 6, CHAPS 7, CHAPS 9, and CHAPS 13 in NTT (Table 1a) and at CHAPS 6, CHAPS 7, CHAPS 8, and CHAPS 11 in NTB (Table 1b).

Out of 3,713 sera which were suitable for IBR SNT, 349 sera (10.4%) contained antibody against IBR virus (Table 1c). However, the number of sera which had antibody against IBR virus in NTT was higher than in NTB. Table 1a shows the prevalence of sera from NTT which had antibody against IBR virus was 12.6% (287 of 1,988 sera) with range from 4.8% (CHAPS 4) to 25% (CHAPS 3). In comparison, in NTB the prevalence was 4.3% (62 of 1376 sera) ranging from 0% (CHAPS 1, CHAPS 2 and CHAPS 12) to 17.8% (CHAPS 4) (Table 1b). The comparison between the prevalence of IBR reactor in NTT and NTB was

conducted using a Mantel-Haenszel chi-square statistic ana- ysis (Stat-Sak program), it was concluded that there was a highly significant different (χ^2 =70.351 and P<0.01).

Table 1. Sera originated from the sentinel cattle in West and East Nusa Tenggara during observation period

a. East Nusa Tenggara (NTT)

CHAPS	Suitable sera for IBR SNT			Unsuitable sera for IBR SNT	Total
	Reactor	Nonreactor	Total		
1	1 (6.3%)	16	17	234	251
2	4 (4.8%)	79	83	97	180
3	25 (25.0%)	75	100	224	324
4	NA				
5	21 (8.8%)	218	239	63	302
6	35 (12.4%)	248	283		283
7	42 (13.7%)	264	306		306
8	24 (11.2%)	179	203	100	303
9	44 (15.5%)	239	283		283
10	39 (12.0%)	285	324	8	332
11	12 (8.7%)	125	137	134	271
12	36 (13.7%)	227	263	14	277
13	4 (10.8%)	33	37		37
Sub-total	287 (12.6%)	1,988	2,275	874	3,149

b. West Nusa Tenggara (NTB)

CHAPS	Suitable sera for IBR SNT			Unsuitable sera for IBR SNT	Total
	Reactor	Nonreactor	Total		
1	0	18	18	156	174
2	0	86	86	84	170
3	3 (3.8%)	76	79	61	140
4	13(17.8%)	60	73	35	108
5	5 (2.7%)	181	186	2	188
6	11 (7.2%)	142	153		153
7	3 (2.0%)	150	153		153
8	7 (4.4%)	151	158		158
9	4 (3.5%)	110	114	25	139
10	5 (3.5%)	139	144	4	148
11	11 (7.1%)	145	156		156
12	0	118	118	23	141
13	NA				
Sub-total	62 (4.3%)	1,376	1,438	390	1,828

c. East and West Nusa Tenggara

Province	Suitable sera for IBR SNT			Unsuitable sera for IBR SNT	Total
	Reactor	Non-reactor	Total		
NTT	287	1,988	2,275	847	3,149
NTB	62	1,376	1,438	390	1,828
Total	349	3,364	3,713	1,264	4,977
	(10.4%)		(74.6%)	(25.4%)	100%

In terms of the distribution of IBR reactor sera, they were found in thirteen locations, five in NTB and eight in NTT (Table 2). The prevalence was high in six locations i.e. Lewa, Waihabur and Raknamo of NTT, and Taliwang, Kananga and Ndano of NTB. Each reactor animal was identified and arranged individually to examine consistency of reactions during collection periods. This was done manually. On the basis of this, 349 sera which were reacted (Table 2) were derived from 171 animals (data not shown). The animals appeared to be reacted between once and six times over thirteen collections period (data not shown). In order to simplify, each individual animal was grouped into one to six reactions during the observation period (Table 3). Table 3 shows a total of 84 (49.1%) animals reacted once, 41 (24.0%) twice, 31 (18.1%) three times, 11 (6.4%) four times, 3 (1.8%) five times, and 1 (0.6%) six times.

DISCUSSION

The results of the survey indicated that antibody against IBR virus occurred among the sentinel cattle with an average prevalence of 10.4%. This findings is similar to the results previously reported from western of Indonesia (NOOR *et al.*, 1983, ARJONO *et al.*, 1984; SAROSA, 1985; and WIYONO, 1993). Results from NTT and NTB indicate that the sentinel cattle were infected previously with IBR virus (LUDWIG and GREGERSEN, 1986). The prevalence of reactors in NTT and NTB were 12.6% and 4.3% respectively. KAHRS (1977) stated that cattle with antibody prevalence between 10% to 96% are potential sources of IBR infection. Sentinel cattle in NTT could therefore be potential reservoir of IBR virus. Cattle are believed to be the principle reservoir of IBR virus, although buffalo, goats and swine can be infected but their role is unknown (KAHRS, 1977). The principle problem of IBR infection is viral latency and reactivation (LUDWIG and GREGERSEN, 1986). The virus may persist in the trigeminal ganglion in the form of DNA (ACKERMANN *et al.*, 1982). At this stage, antibody against IBR virus is believed to control the amount of activated virus but it does not prevent IBR virus latency (LUDWIG and GREGERSEN, 1986).

Although antibody against IBR virus occurs among large ruminants in Indonesia, there are few reports on outbreak of disease due to IBR virus infection.

Table 2. Number of IBR reactor sera in each area during observation period

Village	CHAPS													Total
	1	2	3	4	5	6	7	8	9	10	11	12	13	
Pengandangan						1								1
B2														1.4%
Semayam			1											1
B3														1.4%
Taliwang			2	8	6	3	1	3	1					24
B4														32.4%
Kananga			2	3	7	5	1	2	3	5	8			36
B5														48.6%
Ndano		2			1	2	2	1			4			12
B6														16.2%
Sub-total		2	5	11	14	1	4	6	4	5	12			74
														100%
Naukae			8											8
T2														2.9%
Kringa					1	1								2
T3														0.7%
Lewa			1			10	15	13	16	14		11		80
T4														29.1%
Waihabur		1		2	1	12	11		13	12	3	10	4	69
T5														25.1%
Raknamo		1	9		9	11	14	11	13	10	8	11		97
T6														35.3%
Talibura												1		1
T7														0.4%
Benlutu	1	1				1	1	1	2	3		3		13
T8														4.7%
Lili			5											5
T1														1.8%
Sub-total	1	3	23	2	11	35	41	25	44	39	11	36	4	275
														100%
Total	1	5	28	13	25	46	45	31	48	44	23	36	4	349

MARFIATININGSIH (1982) in Lampung Province reported an IBR-like disease outbreak. The outbreak was diagnosed clinically and serologically. Another IBR infection occurrence was reported by WIYONO *et al.* (1989; 1990) in a diarrhoeal disease outbreak in Bali cattle in West Kalimantan. The outbreak was diagnosed on the basis of historical, clinical, serological and histopathological findings, but not viral isolation.

During the observation period, only a few animals consistently had antibody against IBR virus. This may have been due to three different reasons. Firstly, it may have been due to an error of classification of individual animals as this was done manually. Secondly, the animals had no antibodies against IBR virus at those collection period (GIBBS and RWEYEMAMU, 1977). However, BITSCH (1984) reported that bulls with a naturally acquired genital infection will be latently infected for life. Lastly, the antibody could not be detected by the test i.e. false negative results, since YORK (1968) reported that antibody titre to IBR virus are generally low. Moreover, DEAN and BURGESS (1976) found that SNT is relatively insensitive. In this case, DURHAM and SILLARS (1986) and WIYONO (1994) suggested to use an enzyme-linked immunosorbent assay (ELISA) for detecting antibody against IBR virus.

In conclusion, antibody against IBR virus was detected in cattle in East and West Nusa Tengggara but no clinical disease was found in the sentinel cattle.

Table 3. Frequency of sentinel cattle which had antibody against IBR virus during observation period

Village	Area Code	Frequency of sentinel cattle as IBR reactor						No. of animal
		1 x	2 x	3 x	4 x	5 x	6 x	
Pengandangan	B2	1						1
Semayam	B3	1						1
Taliwang	B4	10	2			1		13
Kananga	B5	7	4	2	2		1	16
Ndano	B6	4	1	2				7
Naukae	T2	8						8
Kringa	T3	1	1					2
Lewa	T4	12	11	12	3			37
Waihabur	T5	19	8	7	1			35
Raknamo	T6	11	13	7	5	2		38
Talibura	T7	1						1
Benlutu	T8	4	1	1				6
Lili	T1	5						5
Total		84	41	31	11	3	1	171

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