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Risk Analysis of Concurrent Occurrence of Infectious Bovine Rhinotracheitis and Brucellosis in Organized Dairy Farms

Keywords: Seroprevalence; Epidemiology; IBR; Brucellosis; Organized dairy farms

Abstract

In the present study, epidemiology and risk analysis of Infectious Bovine Rhinotracheitis (IBR) and brucellosis in organized dairy farms was done. Sera samples (cattle (421) and buffalo (160)) were collected randomly from dairy farms viz., Bijapur (Farm A), Dharwad (Farm B) in Karnataka, Chennai (Farm C), Cuddalore (Farm D) in Tamilnadu state, India. Apparent and true prevalence were 63.2% and 67.9%, 28.2% and 30.3%, 18.8% and 20.2% for IBR, Brucella and both antibodies, respectively. Higher seroprevalence of IBR and brucellosis were in Farm B (81.3%) and Farm D (60.6%), respectively. Higher age groups and crossbred bovines showed increased IBR and brucellosis seroprevalence. Seropositivity for brucellosis was high in Females (29.0%) whereas, IBR was high in males (82.6%). Relative risk in cattle were 1.30 (brucellosis) and 1.15 (IBR) times more than buffaloes. Attributable risk of IBR and brucellosis were 23.34% and 13.25% in cattle than buffaloes. Risk difference of IBR and brucellosis were 9/100 and 7/100 cattle, than buffaloes. There was association for occurrence of IBR and brucellosis based on age (brucellosis only), sex, breed, animal health status, indicating influence of host factors in concurrent occurrence of these diseases. There is need for IBR vaccination, zoosanitary measures and strengthening of brucellosis control in organized dairy farms for better productivity and profitable dairying in India.

Introduction

The reproductive problems and diseases are the main impediments for the profitable dairying in India (abortions, repeat breeding, retained placenta, anoestrus, metritis, etc.). There are several bacterial and viral infectious agents like Brucella abortus, Bovine herpes virus-1 (BoHV-1), Leptospira spp. etc. may be responsible for causing abortions in dairy animals [1,2]. Infectious bovine rhinotracheitis (IBR), a viral disease caused by BoHV-1, which causes respiratory signs and abortions in dairy animals. It causes latent infection and transient immunosuppression which may lead to infectious rhinotracheitis and infectious pustular vulvovaginitis in bovines. It also causes huge economic losses due to abortions, repeat breeding and low milk production in affected animals. A high prevalence of antibodies against BoHV-1 in cattle (50.9%) and buffaloes (52.5%) has been recorded in India [3] and BoHV-1 was also isolated from bovine semen samples [4]. Presently, there is no IBR vaccination for cattle and buffaloes being used in India as reported earlier [5].

Brucellosis is a zoonotic bacterial disease of livestock predominantly caused by *Brucella abortus*, having high zoonotic impact. Its transmission to humans occurs mainly by the consumption

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of raw dairy products and by direct contact with the skin or mucosa during parturition and abortion. Cattle (*Bos taurus*) are natural hosts for *Brucella abortus*, while sheep (*Ovis aries*) and goats (*Capra hircus*) for *B. melitensis* and *B. ovis*. In particular, the disease is characterized by abortions, infertility and reduced milk yield in cattle and buffaloes. A high seroprevalence of bovine brucellosis (17%) has been reported in Indian dairy herds with the history of abortions [1] and the organism could also be isolated from aborted material in cattle [6]. Brucellosis is a self limiting disease but causes abortions in cattle and buffaloes to the dairy farmers in India [5].

In a recent study, the seroprevalence of IBR, brucella and both antibodies were 61.54%, 11.63% and 10.58%, respectively in organized dairy farms in southern India [5]. The IBR seroprevalence in organized dairy farms in India was reported to be 61.6% [7]. Many studies are available on the seroprevalence of IBR and brucellosis in organized dairy farms separately [7] and in a recent study on concurrent prevalence of IBR and brucellosis based on age, sex, species, breed and animal health status has been reported [5]. There are limited studies on the epidemiological and risk analysis of IBR and brucellosis in bovines of organized dairy farms. These studies are important for better understanding of these disease and their relationships. Hence, the present study was undertaken to know the epidemiology and risk factors for IBR and brucellosis seroprevalence in organized dairy farms in Karnataka and Tamilnadu states in India.

Materials and Methods

Selection of farms

A cross-sectional study was conducted during 2014-15 in four organized dairy farms, comprising two farms each in Karnataka (Farm

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S No.	Details	No. of animals	IBR Positive (AB ELISA)	Brucella Positive (iELISA)	Both IBR and Brucella positive	
Farm wise						
1	Farm A-Bijapur					
	Cattle	59 (28.6)	47 (79.7)	5 (8.5)	5 (8.5)	
	Buffalo	147 (71.4)	78 (53.1)	37 (25.2)	31 (21.1)	
	Subtotal	206 (100.0)	125 (60.7)	42 (20.4)	36 (17.5)	
2	Farm B-Dharwad					
	Cattle	115 (89.8)	91 (79.1)	32 (27.8)	28 (24.3)	
	Buffalo	13 (10.2)	13 (100.0)	0 (0.0)	0 (0.0)	
	Subtotal	128 (100.0)	104 (81.3)	32 (25.0)	28 (21.9)	
3	Farm C-Chennai					
	Cattle	138 (100.0)	88 (63.8)	24 (17.4)	17 (12.3)	
	Buffalo	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Subtotal	138 (100.0)	88 (63.8)	24 (17.4)	17 (12.3)	
4	Farm D-Cuddalore					
	Cattle	109 (100.0)	50 (45.9)	66 (60.6)	28 (25.7)	
	Buffalo	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
	Subtotal	109 (100.0)	50 (45.9)	66 (60.6)	28 (25.7)	
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)	
	Confidence Interval at 95%		62.0 - 64.4	26.6 - 29.8	18.3 - 19.2	
Species wise						
1	Cattle	421 (72.5)	276 (65.6)	127 (30.2)	78 (18.5)	
2	Buffalo	160 (27.5)	91 (56.9)	37 (23.1)	31 (19.4)	
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)	
	Confidence Interval at 95%		62.7 - 63.7	27.8 - 28.6	18.6 - 19.0	

Table 1: Seroprevalence of IBR and Brucella antibodies based on farms and species.

Note: Values in parenthesis are expressed in percentage

A-Bijapur; Farm B-Dharwad) and in Tamilnadu (Farm C-Chennai; Farm D-Cuddalore) in southern India, which were selected randomly for this study. Organized dairy farms means the cattle or buffaloes were reared under intensive system of management with systematic production management systems and maintained mainly for milk production. The exact geographical locations of four dairy farms was collected and mapped by using Quantum GIS software version 2.10. The history of the animals like species, age, sex, breed, animal health status and managemental practices, were collected from these organized dairy farms. The history included occurrence of abortions, post abortion complication, infertility within the past three years were also collected.

Sample collection

For serological investigations, a total of 581 serum samples, from cattle (421) and buffaloes (160), were collected from animals *via* jugular venipuncture for screening against IBR and brucella antibodies. The blood samples were collected by using sterile serum vacutainer tubes coated with silicone and micronized silica particles to accelerate clotting, and multi-sample vacutainer needles (Becton and Dickenson Company, USA). The serum was separated by centrifugation at 560 g for 20 minutes. All serum samples were

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stored at -20 °C until used for testing. Based on sex, 23 male and 558 female samples were collected. The animals were classified based on age in years for bovines having date of birth and bovines not having date of birth, the number of lactations was considered to implicitly derive the age. Based on breeds in cattle, Jersey cross (158), Holstein Friesian cross (138), Deoni (36), Sahiwal (29), Gir (28), Kangayam (16), Rathi (8), Tharparkar (8) and among buffalo breeds, Murrah (157), Jaffrabadi (3) were tested. Based on animal health status and reproductive history, the animals were classified as apparently healthy (421), pregnant (48), repeat breeding (78), mastitis (16), abortions (13) and metritis (5).

Infectious bovine rhinotracheitis screening

The seroprevalence of IBR was carried out by detection of antibodies against BHV-1 virus from serum using Avidin Biotin Enzyme Linked Immunosorbent Assay (AB-ELISA) kit developed by ICAR - NIVEDI, Bengaluru, Karnataka. The sensitivity and specificity of the AB ELISA were 98 and 95 per cent respectively. This is the only indigenously developed kit as per World Organisation of Animal Health standards [8] and available for screening IBR in India.



Figure 1: Geographical location of organized dairy farms in Karnataka and Tamil Nadu states in India.

Brucellosis screening

Diagnosis of brucellosis from serum on the basis of detection

of antibodies against Brucella abortus was carried out by using an indirect ELISA (iELISA) kit developed by ICAR-NIVEDI, Bengaluru, Karnataka. The test has been standardized using smooth lipopolysaccharide antigen from a standard strain of Brucella abortus S99 and recombinant protein G conjugated with Horse radish peroxidase as per World Organisation for Animal Health standards [8]. The sensitivity and specificity of the test were 98 and 95% in cattle and 92 and 98% in buffaloes, respectively. The use of smooth lipopolysaccharide antigen from Brucella abortus S99 strain in iELISA instead of crude antigen helps to detect antibodies against other smooth species such as B. melitensis and B. suis. The use of protein G conjugate, a common conjugate was advantageous over using different conjugates, protocols for different livestock species and can be used for screening brucellosis in cattle, buffalo, sheep, goat, pigs and humans. The details of the iELISA kit and test protocol are described earlier [9].

Statistical analysis

The apparent prevalence and true prevalence was estimated as per the following formula [10]. (i) Apparent prevalence = number reds in organized dairy farms

S No.	Details	No. of animals	IBR Positive (AB ELISA)	Brucella Positive (iELISA)	Both IBR and Brucella positive	
Age wise (in Lactation numbers)						
1	1 Calving	100 (21.8)	58 (58.0)	32 (32.0)	27 (27.0)	
2	2 Calvings	151 (32.9)	86 (57)	51 (33.8)	33 (21.8)	
3	3 Calvings	125 (27.2)	83 (66.4)	41 (32.8)	26 (20.8)	
4	4 Calvings	49 (10.7)	32 (65.3)	22 (44.9)	15 (30.6)	
5	5 Calvings	22 (4.8)	16 (72.7)	8 (36.4)	4 (18.2)	
6	6 Calvings	6 (1.3)	3 (50.0)	2 (33.3)	0 (0.0)	
7	7 Calvings	4 (0.9)	4 (100.0)	0 (0.0)	0 (0.0)	
8	8 Calvings	2 (0.4)	2 (100.0)	0 (0.0)	0 (0.0)	
	Sub total	459 (100.0)	284 (61.9)	156 (34.0)	105 (22.9)	
	Confidence Interval at 95%		60.2 - 63.6	32.5 - 35.5	21.7 - 24.1	
	Chi Square	test value	8.67 ^{ns}	6.00 ^{ns}		
			Age wise (in Years)			
9	1 year	26 (21.3)	17 (65.4)	1 (3.85)	0 (0.0)	
10	2 years	35 (28.7)	24 (68.6)	0 (0.0)	0 (0.0)	
11	3 years	22 (18.0)	16 (72.7)	0 (0.0)	1 (4.5)	
12	4 years	10 (8.2)	8 (80.0)	1 (10.0)	0 (0.0)	
13	5 years	8 (6.6)	5 (62.5)	2 (25.0)	0 (0.0)	
14	6 years	9 (7.4)	6 (66.7)	0 (0.0)	0 (0.0)	
15	7 years	8 (6.6)	4 (50.0)	2 (25.0)	1 (12.5)	
16	8 years	3 (2.4)	2 (66.7)	1 (33.3)	1 (33.3)	
17	9 years	1 (0.8)	1 (100.0)	1 (100.0)	1 (100.00)	
	Sub total	122 (100.0)	83 (68.0)	8 (6.56)	4 (3.3)	
	Confidence Interval at 95%		65.6 - 70.4	0.9 - 12.3	1.4 - 5.2	
	Chi Square	test value	2.75 ^{ns}	36.53**		
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)	

Sex wise							
1	Female	558 (96.0)	348 (62.4)	162 (29.0)	107 (19.2)		
2	Male	23 (4.0)	19 (82.6)	2 (8.7)	2 (8.7)		
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)		
	Confidence Interval at 95%		58.1 - 68.3	25.9 - 30.4	17.3 - 20.3		
	Chi Square test value		3.89*	7.47**			
Breed wise (Cattle)							
1	Jersey cross	158 (37.5)	89 (56.3)	69 (43.7)	30 (19.0)		
2	Holstein Friesian cross	138 (32.8)	97 (70.3)	36 (26.1)	30 (21.7)		
3	Deoni	36 (8.6)	20 (55.6)	1 (2.78)	3 (8.3)		
4	Sahiwal	29 (6.9)	20 (69.0)	4 (13.8)	3 (10.3)		
5	Tharpakar	8 (1.9)	7 (87.5)	6 (75.0)	6 (75.0)		
6	Gir	28 (6.7)	26 (92.9)	6 (21.4)	6 (21.4)		
7	Rathi	8 (1.9)	6 (75)	3 (37.5)	0 (0.0)		
8	Kangayam	16 (3.8)	11 (68.8)	2 (12.5)	0 (0.0)		
	Sub total	421 (100.0)	276 (65.6)	127 (30.2)	78 (18.5)		
	Confidence Interval at 95%		64.3 - 66.9	28.0 - 32.5	16.2 - 20.8		
	Chi Square	test value	20.40*	42.50**			
Breed wise (Buffalo)							
9	Murrah	157 (98.1)	88 (56.1)	37 (23.6)	31 (19.7)		
10	Jaffrabadi	3 (1.9)	3 (100.0)	0 (0.0)	0 (0.0)		
	Sub total	160 (100.0)	91 (56.9)	37 (23.1)	31 (19.4)		
	Confidence Interval at 95%		52.1 - 61.7	20.5 - 25.7	17.2 - 21.6		
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)		

Note: Values in parenthesis are expressed in percentage; ns-non significant; *-Significant at 5% level; **-Significant at 1% level.

of positive animals/number of tested animals. (ii) True prevalence = [apparent prevalence + (specificity -1)] / [(sensitivity + specificity) - 1]. The data analysis was carried by calculating percentage positivity for IBR, Brucellosis and both these diseases. The one way analysis of variance was used for analysis between the farm wise seroprevalence for IBR, brucellosis and both positivity. The Chi square test of independence was calculated to measure the association between the age, breed, sex, animal health status with IBR and brucellosis occurrence as per earlier method described [11] and by using Statistical Analysis System (SAS) Software version 9.3 (SAS India Ltd, Mumbai, India). The risk analysis between species and, IBR, brucellosis positive number of animals, like relative risk, risk difference, attributable risk and Odds ratio was calculated as described method earlier [10].

Results

The location of the four organized dairy farms in Karnataka and Tamil Nadu state in India is depicted in Figure 1. The seroprevalence of IBR and brucellosis based on farm and species wise in the four organized dairy farms is given in Table 1. The overall apparent prevalence of IBR, brucellosis and both the diseases in organized dairy farms were 63.2%, 28.2% and 18.8%, respectively, where as true prevalence were 67.9%, 30.3% and 20.2 %, respectively. No significant difference was observed between the farms for seroprevalence of IBR, brucellosis and both positivity. The farm wise analysis showed Farm B had high percentage positivity for IBR (81.3 %) and Farm D for brucellosis (60.6 %) and both positivity (25.7 %). Based on species, cattle and buffalo showed 65.6% and 56.9% positivity for IBR antibodies, and 30.2% and 23.1% positivity for brucella antibodies, respectively. There was no significant difference between the species for seroprevalence of IBR and brucellosis.

The seroprevalence of IBR and brucella antibodies based on age, sex and breed wise in the organized dairy farms is given in Table 2. On the basis of age, the bovines with higher lactation numbers, above 4 calvings (73.53%) and above 4 years of age (69.89%) showed increased percentage positivity for IBR. There was no significant difference between different age groups indicating association of age of animals with occurrence of IBR and brucellosis in dairy animals. Based on sex, the males revealed higher positivity (82.6%) for IBR whereas, females were positive for brucellosis (29.0%) and positive for both IBR and brucellosis (19.2%). Breed wise analysis revealed high percentage positivity for IBR and brucellosis were observed in Gir and Tharparkar breeds in indigenous breeds of cattle, respectively. In crossbred cattle, more percentage positivity for IBR and both positivity were observed in Holstein Friesian cross and for brucellosis in Jersey cross than the indigenous cattle breeds. In buffalo breeds, Murrah showed more percentage positivity for brucellosis and both positivity. There was significant difference between sex, breed with

S No.	Details	No. of animals	IBR Positive (AB ELISA)	Brucella Positive (iELISA)	Both IBR and Brucella positive
1	Apparently Healthy	421 (72.5)	260 (61.8)	130 (30.9)	83 (19.7)
2	Pregnant	48 (8.3)	42 (87.5)	12 (25.0)	16 (33.3)
3	Abortion	13 (2.2)	9 (69.2)	2 (15.4)	1 (7.7)
4	Repeat Breeding	78 (13.4)	47 (60.3)	8 (10.3)	3 (4.2)
5	Metritis	5 (0.9)	2 (40.0)	1 (20.0)	0 (0.0)
6	Mastitis	16 (2.7)	7 (43.8)	11 (68.8)	6 (37.5)
	Total	581 (100.0)	367 (63.2)	164 (28.2)	109 (18.8)
	Confidence Interval at 95%		58.1 - 68.3	25.9 - 30.5	17.3 - 20.3
	Chi Square test value		16.89**	27.50**	

Table 3: Seroprevalence of IBR and Brucella antibodies based on animal health status in organized dairy farms.

Note: Values in parenthesis are expressed in percentage; ns-non significant; *-Significant at 5% level; **-Significant at 1% level.

Table 4: Risk analysis of IBR and Brucella positivity in bovines of organized dairy farms in India.

A. Species	IBR positive (Numbers)	IBR negative (Numbers)	Total	Parameters	Values
Cattle	276	145	421	Relative risk	1.15
Buffalo	91	69	160	Risk diff	0.09
Total	367	214	581	Attributable risk (%)	13.25
	Chi S	quare test value=3.8 ^{ns}	Odds ratio (OR)	1.44	
			CI at 95% of OR	0.94-2.19	
			P-value of OR	0.092 ^{ns}	
B. Species	Brucella positive (Numbers)	Brucella negative (Numbers)	Total	Parameters	Values
Cattle	127	294	421	Relative risk	1.30
Buffalo	37	123	160	Risk difference	0.07
Total	164	417	581	Attributable risk (%)	23.34
	Chi S	quare test value=2.9 ^{ns}	Odds ratio (OR)	1.44	
			CI at 95% of OR	0.99-2.09	
			P-value of OR	0.053 ^{ns}	
C. Category	Brucella positive (Numbers)	Brucella negative (Numbers)	Total	Parameters	Values
IBR positive (Numbers)	109	258	367	Relative risk	1.16
IBR negative (Numbers)	55	159	214	Risk difference	0.04
Total	164	417	581	Attributable risk (%)	13.47
	Chi S	quare test value=1.1 ^{ns}	Odds ratio	1.22	
			Kappa value	0.033	
				P-value	0.302

Note: ns-non significant

IBR, brucellosis seroprevalence by Chi square test, which indicated that there was association between these factors and occurrence of IBR and brucellosis in bovines.

The seroprevalence of IBR and brucellosis based on animal health status in the organized dairy farms is given in Table 3. The animals with history of repeat breeding were high in number in these dairy farms. IBR seroprevalence was high in aborted animals when compared with animals of other reproductive disorders. There was significant difference between animal health status and IBR, brucellosis seropositivity, indicating association between them. The measures of association between species and IBR, brucellosis seropositivity is given in Table 4. The relative risk for occurrence of IBR and brucellosis were 1.15 and 1.30 times in cattle more than the buffalo. Risk difference was 9 per 100 animals for IBR and 7 per 100 animals for brucellosis in cattle than the buffaloes. The attributable risk was 13.25% and 23.34% more for IBR and brucellosis in cattle when compared to buffaloes. The Odds ratio for IBR and brucellosis seropositivity was 1.44 times more in cattle than buffaloes. However, the Odds ratio of IBR seropositivity occurrence was 1.22 times more than brucellosis in the dairy animals. The Chi square test results revealed no significance difference between species and occurrence

of IBR and Brucellosis, which indicated that the cattle and buffaloes having no dependency for acquiring the IBR and brucellosis infections in organized dairy farms. The Kappa value was 0.033 for IBR seropositive to become brucellosis seropositive, which indicated that there was slight agreement between these two diseases in cattle and buffaloes. The risk analysis indicated that there was variation between the host factors and occurrence of these two diseases in dairy animals and needs to take up the preventive and control measures for these diseases.

Discussion

Earlier literature indicated that the seroprevalence of IBR in organized dairy farms in India ranged from 51 to 65% [3,5,12] and concurred with the present study for IBR (63.2%). In the present study, the seroprevalence of IBR is maintained within the range, however the seroprevalence showed slight increase from 51.4% during 1996 [3] to 63.2% in 2015 which might be due to non-availability of IBR vaccine in India. The high prevalence indicates the importance of the disease in bovines in India. Across farms, Farm B showed higher seroprevalence of IBR (81.3%) indicating variation in prevalence levels. Further, in the all the farms screened in this study practiced artificial insemination, procured from government agencies whereas in Farm B, regularly practiced natural breeding which might be the reason for spread of IBR in this farm. The higher seroprevalence of IBR observed in buffaloes across the farms studied and might due to the purchasing of Murrah buffaloes from northern states of India viz., Haryana and Punjab by these farms, which have higher seroprevalence of IBR. The seroprevalence of brucellosis was higher in Farm D which might be due to inflow of animals from farms with unknown history on disease status, the poor management practices and zoo sanitary measures adopted in these farms. The seroprevalence of brucellosis observed in Farm C-Chennai was 17.4% and it was higher than the previous report which indicated seroprevalence of 6.7% by iELISA in an organized dairy farm with cattle in Chennai [13]. This might be due to the spread of the brucella infection from animals to animals over the period of time, since the culling of infected animals is not practiced in the farm. This is also due to the social customs and beliefs prevailing in India, in which the people worship cow as god and slaughter of cow are banned in these two states. Perusal of the literature indicated that the prevalence of brucellosis in India varied across the states. In Gujarat, it was ranged from 8.98 to 44.00% [14,15], whereas 17% in southern India [1], 6.3% in Madhya Pradesh and 12.9-57.3% in Punjab [16-18]. The seroprevalence of brucellosis in dairy animals varied across the geographical regions in India. In other countries, seroprevalence of brucellosis was reported to be 3.1% in Ethiopia, 6.5% in Jordan, 8.4% in Cameroon, 32.92 to 39.45% in Turkey, 8% in Pakistan, 35.8% in China, 42.5% in England and 62.5 to 85.7% in Egypt [19-26].

The highest number of seropositivity was observed in cattle for both IBR and brucellosis compared to buffaloes. Seroprevalence of IBR in cattle and buffaloes were 50.9% and 52.5%; 60.46% and 62.39%; 59.30% and 67.08%, respectively as per earlier reports and corroborated with the present study [3,5,12]. In India, seroprevalence of antibodies against BoHV-1 was reported to be 50.9% in cattle and 2.75 to 81.0% in buffaloes [3,27,28] and concurred with the present study. The age wise analysis showed increased seropositivity in higher age group for IBR and brucellosis which might be due to reduced immune response in aged animals. There was association between the age of the bovines with occurrence of brucellosis in dairy farms indicating the susceptibility for the diseases across the age groups. The seroprevalence was higher in males for IBR and in females for brucellosis. The male and female animals showed significant (P<0.05) difference in test for independency for IBR and highly significant (P<0.01) difference for brucella antibodies and indicating association in susceptibility to these diseases among males and females. The crossbred cattle showed increased seroprevalence for IBR, brucella and both antibodies when compared to indigenous breeds of cattle. This might be due to increased susceptibility to infections in crossbred cattle and also chance of spread of diseases by infected bull semen as majority of the farms are artificially inseminated [5]. Further, the genetic nature of indigenous breeds which may be resistant to the diseases than the crossbred animals. Based on Animal health status, the bovines with history of abortion (69.2%) and mastitis (68.8%) showed increased seroprevalence for IBR and brucellosis, respectively. There was highly significant (P<0.01) difference for animal health status and IBR, Brucellosis seroprevalence indicating the association with occurrence of this diseases with reproductive problems. Previous report indicated that there was association of IBR with bovine abortion and it was recorded as 55.4% from aborted crossbred cows [3] and corroborated with the present study.

The relative risk of occurrence of IBR and brucellosis was higher in cattle than buffaloes which might be due to the resistance nature of indigenous breeds of buffalo than the cattle (Jersey and Holstein Friesian cross) reared by organized dairy farms. The risk difference showed chance of occurrence of IBR (9 in 100) was more than brucellosis (7 in 100) in cattle compared to buffaloes. The attributable risk also revealed the more chance of occurrence of these diseases in cattle than buffaloes. The Odds ratio was 1.44 times high in cattle implying the more chance of occurrence of IBR and brucellosis in this species. The Odds ratio between the occurrence of IBR and brucellosis showed the chance of IBR is 1.22 times more than brucellosis in animals and indicated there was no association for occurrence of these two diseases in dairy animals. However, the Kappa statistics showed that there was a slight agreement between the IBR and Brucellosis diseases occurrence in dairy animals. This indicated that these two disease occurrence might be slightly related. The measures of association between age and brucellosis; sex, breed and animal health status with IBR, brucellosis occurrence showed dependency which might be the reasons for occurrence these diseases in particular age, sex, breed and animal health status. There are no previous studies available on risk analysis for IBR and brucellosis seroprevalence in organized dairy farms to compare the results obtained in the present study.

There is a limitation in this study that the AB-ELISA kit used for detection of IBR is having antigen for the detection of BoHV-1 but used for detection of bubaline Herpes virus-1 in water buffaloes. The IBR antibodies was high compared to Brucella antibodies and it might be due to awareness on management practices followed by dairy farm owners and also by implementation of brucellosis control programmes by central and state Governments. After

BoHV-1 infection the virus becomes latent in ganglions and the animals remain seropositive for rest of the life, the virus can be reactivated upon stress and such animals are likely to shed the virus intermittently into the environment [12]. There is no vaccination for IBR in India and the seropositivity indicates the present or latent infection in animals. The higher percentage of seropositivity of IBR in all age groups and breeds indicated the natural circulation of the BoHV-1 virus in the animal population and as reported earlier [7]. Further, the bovines in these farms are reared by intensive system and head to head system of housing which may also favours the spread of these disease in the bovines. The review of literature on the status of brucellosis in organized dairy farms with a history of abortions using RBPT and ELISA revealed seropositive animals were 13.78 and 22.18%, respectively [12], however, slightly higher prevalence (28.2%) was observed in this study. There are several factors playing a possible role for the spread of disease viz unrestricted movement of animals, procurement of animals without proper screening, absence of quarantine before entry to the main herd, lack of prophylactic measures like vaccination, etc. Eradication of brucellosis in the organized dairy farms may be possible by application of test-andremove principle, but non-acceptance of this strategy will hinder its control in India. However, to control brucellosis, a nucleus of brucellosis free bovines may be formed with less contact of reservoirs and continuous monitoring of the health status of all bovines reared for breeding purposes. Many measures to control brucellosis and IBR in organized dairy farms has been described earlier [5] but scrupulous following of the same is important. Further, brucellosis is self-limiting infection, its prevalence in the organized dairy farms mainly depends upon the protocol adopted for procuring animals by the farms as well as zoo-sanitary measures followed. Variation in the incidence of brucellosis in different farms might be due to various levels of biosecurity measures adopted by dairy farm owners.

The high prevalence of IBR than brucellosis in organized dairy farms warrants immediate attention to control the disease by adoption of IBR vaccination strategies. The complementary measures like regular screening of animals against these diseases, culling of positive reactors, strict vaccination, quarantine of animals at the time of procurement, use of semen free from infectious agents and strict implementation of zoo sanitary and biosecurity measures should be adopted in the organized dairy farms to reduce the prevalence level of these diseases. For successful control of brucellosis, implementation of various control regimens including test and removal of affected animals, calfhood vaccination, use of semen obtained from brucella free bull and general hygienic measures need to be adopted in the organized dairy farms. Thus, IBR and brucellosis are important diseases that limits the productivity and hence they have to be effectively controlled for more profitable dairying by organized dairy farms in India.

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