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Prevalence of Infections Bronchities Virus at Four Selected Districts (Damot Gale, Sodozuri, Humbo and Kindokoy) In Southern Nations, Nationalities and Peoples

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ABSTRACT

Infectious bronchitis is an important virial disease affecting poultry production which causes significant economic losses. A cross-sectional study was carried out from November 2019 to January 2020. To estimating the prevalence of infectious bronchitis and determines the risk factors. A total 420 poultry were by a simple random sampling procedure. Data was analyzed using STATA version 11. Chi square test and logistic regression analysis were used to determine the association between prevalence of infectious bronchitis and potential risk factors. From the total 420 poultry examined for infectious bronchitis were found positive. There was significance difference between the risk factors and bronchitis infections. More infectious bronchitis infected poultry were found in kindokoy 90.4%, odd ratio: 0.3 Confidence Interval :0.1-0.7, p = 0.00 than humbo, sodozuri and damotgale. There was high prevalence of infectious bronchitis in gerater than 6 months than less than 3 months and between 3 and 6-month age of poultry. The presence of infectious bronchitis in poultry production might entail morbidity and mortality. Therefore, further detailed molecular epidemiological studies are warranted. Good hygienic and husbandry practices are essential to limit the spread of infection.

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Introduction

Poultry production is an immediate growing sector in world. Previously market share poultry meat is about 2 to 2.5 in 1971 that has been increase to 25% in 2010 [1]. Poultry production economical to stockholders due to small farming size, quick output, genetic improvement and improvement in feed stuff [2,3].

Know a day despite world the common viral infections such as Newcastle disease, avian influenza and infectious bronchitis influenced by various pathogens [4]. From those common viral pathogens Infectious bronchitis virus (IBV) is an acute and highly contagious virus causing respiratory disease in poultry [5,6]. Outbreaks of IBV have been increased in last few decades in countries such as Tunisia, Egypt and Asia that caused heavy economic losses [7]. It is associated with sneezing, gasping, tracheal rales, coughing, puffy swollen eyes and inflamed sinus with poor weight gain [8].

The infection of IBV is responsible for causing morphological and histopathological changes in oviduct which influence lying of eggs [9]. Additionally, the important clinical case of the disease was; Drop in egg production, poor quality eggs, misshapen, broken and weak shelled eggs followed by reduction in egg production up to 50% and loss shell color [8]. The Prevalence of infectious bronchitis virus has been increased time to time because it leads heavy morbidity and mortality in poultry industry. In the world the prevalence of infectious virus in commercial poultry is-*+ about 82.43% from the others [10].

The most controls of IBV is essentially based on the use of live attenuated and killed vaccines. However, the low level of cross protections between vaccines of different serotypes and coexistence of several genotypes in the same regions are the major obstacles to controls of the disease. Therefore, study is designed to investigate the prevalence and associated risk factors of infectious bronchitis (IBV)

Materials and Methods

Study population

All chickens young and adults of age found in the study areas constitute the study populations.

Study animals

The sampled animals were exotic and indigenous poultry aged between 3 and 6, less than 3and greater than 6 months.

Study Design

A cross-sectional study was conducted in different poultry farms from November 2019 to January 2020 to study the prevalence of IBV.

Sample size determination and sampling technique

As there was no previous report on the prevalence of IBV infection in the study area, the sample size was calculated by considering the

expected prevalence of 50% according to Thrusfield and Christley (2018). n= 1.962*Pexp (1-Pexp) d2
Where: n = required sample size; Pexp = expected prevalence; d =desired absolute precision. With 5% desired precision, at 95% confidence level is considered. Accordingly, the total calculated sample size was 384 however to increase the precision the sample size was increased to 420. Using a simple random sampling technique all samples were collected from all study areas.

Sample collection and transportation
A total of 420 blood sample from poultry were collected from flocks from four selected districts (demote gale, sodozuri, humbo and kindokoy) from November 2019 to January 2021. In order to investigate prevalence of IBV samples were taken from poultry farms (broiler and layers) Samples were transferred in test tubes containing sterilized phosphate-buffered saline (PBS) and shifted to research and development laboratory and preserved at -80°C.

Sampling Procedure and Sample Analysis
Blood samples were collected from the brachial vein in 3-mL disposable syringes and left horizontally for 3 hr, and then vertically for the serum to ooze out. Serum was decanted into 1.8-mL volume cryovial tubes and kept at −20°C until testing. NCD virus antibody works on the principle of indirect nucleoprotein ELISA and is developed to detect specific antibodies against PMV-1 in serum. Serum samples were analyzed using commercial ELISA kits for the presence of antibodies to NCDV (IDVet NCDV-Ab ELISA, Veterinary Innovative diagnostic, France), according to the manufacturers' instructions. Briefly, allow all reagents to come into room temperature (21oC + 5°C) before use. Homogenize all reagents by inversion or vortex.

The negative and positive controls are supplied ready-to-use and no need of adding dilution buffer to the control wells (A1, B1, C1 and D1). Samples however, are tested at a final dilution of 1:100 in dilution buffer 14 (1:50 pre-dilution, followed by 1:2 dilutions in the microplate). In a pre-dilution plate, set aside wells A1, B1, C1 and D1 for the controls, and add: 5µl of each sample to be tested, 245 µl of dilution buffer 14 to all wells except control wells. Then, in the ELISA microplate, add: 100µl of the negative control to wells A1 and B1. 100µL of the positive control to wells C1 and D1. 50µl of dilution buffer 14 to each wells except control wells, 50µl of the pre-diluted samples as prepared above.

Cover the plate and incubate 30min + 3min at 21oC + 5oC. Prepare the conjugate 1x by diluting the concentrated conjugate 10x to 1:10 in dilution buffer3. Empty the wells. Wash each well 3 times with at least 300 µl of the wash solution 1x. Avoid drying of the wells between washings. Add 100 µl of the substrate solution to each well and incubate at 21oC + 5oC in the dark for 15min+ 2min. Add 100 µl of the stop solution to each well to halt the reaction. The sample and control optical density (OD) values were read using an ELISA reader (ELX800 ELISA Plate reader, BioTek instrument, USA) at 450 nm wave length. From OD values, the sample/positive values (S/P) were calculated using the following formula: S/P = [(OD sample- OD negative control)/ (OD positive control- OD negative control) × 100]. So, that S/P values <0.3 were considered negative and S/P values >0.3 were positive. Similarly, the antibody titer was calculated using the formula; log10 (titer) =1.00x log10 (S/P) +3.520. The antibody titer result can be interpreted as titer < 993 were considered as negative and titer >993 were positive.

Data management and analysis
The data generated were stored in Microsoft excel spreadsheet and analyzed using STATA version 11.0. Percentages, Chi-square test, univariable logistic regression were performed, to summarize and quantify the association between risk factors. Only the independent variables showing colinearity<50% and P<0.25 were included to final multivariable model analysis. These variables were categorized during data analysis. The categories of the variables were as follows: species of animals, age, sex, breed, body condition and origin of animals. An odds ratio was used to quantify the degree of association between risk factors and the disease and confidence level was held at 95%and significance was at $P < 0.05$.

Results
Prevalence infection bronchitis virus(IBV)
In the current study, 420 chicken were examined and the overall prevalence was 82.62%.

Association of risk factors with infectious bronchitis bursa infections
The results of the association of risk factors with the prevalence of infectious bronchitis bursa chi square(X2) testis is shown in Table 1. Prevalence of infectious bronchitis was significantly associated with sex, Keble's, district, and number of flocks.

Tables 1: The association between prevalence of infectious bronchitis and the risk factors

Risk factors	No of examined	No of positive	Prevalence (%)	Chi square	p-value
Sex					
females	279	223	79.9	4.1	0.04
male	141	124	87.9		
Age					
between 3 and 6	161	130	80.7	1.05	0.59
less than 3	180	149	82.7		
greater than 6	79	68	86		
Breed					
Exotic	170	133	78.2	3.82	0.051
Indigenous	250	214	85.6		
Kebles					
ade Aro	8	4	50	52.4	0.00
wandarboloso	16	9	56.2		
kokote	28	16	57.1		
Ade Sibaye	10	6	60		
Ampo Koysha	17	11	64.7		
Zalashala	16	6	72.7		
hanaza	27	21	77.7		
dembakoysha	31	26	83.8		
ambe shoya	28	24	85.7		
shochoraogodoma	24	21	87.5		
waja shoya	25	22	88.0		
chocha	25	22	88.0		
gulgula	35	31	88.5		
Harto kontola	20	18	90.0		
koysha Ogodoma	13	12	92.3		
dalbo	20	19	93.0		
borkoshe	22	21	95.4		
Sere finchewa	49	48	97.6		
Districts					
damot gale	78	59	75.6	8.02	0.04
sodozuri	130	104	80.0		
humbo	113	94	83.1		
kindokoy	99	90	90.4		
Previous outbreak					
yes	130	103	79.2	1.79	0.4
no	290	244	84.1		
Production					
intensive	59	46	77.9	1.03	0.3
extensive	361	303	83.3		
No of flocks					
less than 5	107	85	79.4	11.0	0.01
greater than 5	262	313	87.3		

Univariable logistic regression for IBV risk factors

During the Univariable logistic regression analysis, for all the risk factors, the first level of each independent variable (the category of a risk factor with lowest prevalence) was used as a reference category for measuring the degree of association between the disease and risk factors. The prevalence of IBV was significant different ($p < 0.05$) between the various risk factors investigated. Accordingly, the odd of infection in male was 1.4 times higher than female. The odd of IBV infection in less than 6 was 1.6. times higher than

between 3 and 6 and greater than 6. The odd of IBV infection in indigenous of chicken was 2.6 times than exotic. The odd of IBV infection of chicken based on keble's shows Sere finchewa was 9.8 times higher others. Univariable logistic regression analysis also showed significant association ($p<0.05$) between IBV positivity and sex,

Table 2: Unvariable logistic regression analysis of potential risk factors of IBV

Risk factors	No of examined	No of positive	Prevalence (%)	OD ratio (95 CI)	p- value
Sex					
females	279	223	79.9	1	
male	141	124	87.9	1.4(0.3-0.9)	0.04
Age					
between 3 and 6	161	130	80.7	1	
greater than 3	180	149	82.7	1.2(0.5-15)	0.6
less than 6	79	68	86	1.6(0.3-1.4)	0.3
Breed					
Exotic	170	133	78.2	1	
Indigenous	250	214	85.6	2.6(0.3-1)	0.05
Kebles					
ade Aro	8	4	50		
wandarboloso	16	9	56.2	1.2(0.03-1.8)	0.17
kokote	28	16	57.1	1.4(0.01-1.6)	0.12
Ade Sibaye	10	6	60	1.4(0.1-1.7)	0.9
Ampo Koysha	17	11	64.7	1.4(0.02-1.7)	0.15
Zalashala	16	6	72.7		
hanaza	27	21	77.7	1.5(0.5-3)	0.5
dembakoysha	31	26	83.8	1.6(0.04-1.4)	0.15
ambe shoya	28	24	85.7	1.6(0.06-2)	0.24
shochoraogodoma	24	21	87.5	1.6(0.05-1.9)	0.22
waja shoya	25	22	88.0	1.7(0.01-1.1)	0.06
chocha	25	22	88.0	1.7(0.1-4.8)	0.9
gulgula	35	31	88.5	1.7(0.03-1.25)	0.08
Harto kontola	20	18	90.0	1.7(0.04-1.9)	0.2
koysha Ogodoma	13	12	92.3	1.7(0.2-7.3)	0.7
dalbo	20	19	93.0	1.8(0.06-0.9)	0.04
borkoshe	22	21	95.4	18(0.03-1.2)	0.09
Sere finchewa	49	48	97.6	1.9(0.08-2)	0.2
Districts					
damot gale	78	59	75.6	1	
sodozuri	130	104	80.0	0.7(0.3-1.5)	0.4
humbo	113	94	83.1	0.6(0.3-1.2)	0.2
kindokoy	99	90	90.4	0.3(0.1-0.7)	0.00
Previous outbreak					
yes	130	103	79.2	1	
no	290	244	84.1	0.7(0.4-1.2)	0.2
Production					
intensive	59	46	77.9	1	
extensive	361	303	83.3	0.7(0.4-1.2)	0.3
No of flocks					
less than 5	107	85	79.4	1	
greater than 5	262	313	87.3	0.7(0.4-1.3)	0.3

Multivariable logistic regression analysis of risk factors

For multivariate analysis all variable was entered into the multivariate model because the independent variables were showing collinearity<50% and P<0.25. Multivariate logistic analysis showing all independent variables were statistically not significant (Table 3).

Table 3: Multivariable logistic regression analysis of risk factors in chicken IBV

Risk factors	No of examined	No of positive	Prevalence (%)	OD ratio (95 CI)	p-value
Sex					
females	279	223	79.9	1	
male	141	124	87.9	1.4(0.3.1.05)	0.07
Age					
between 3 and 6	161	130	80.7	1	
greater than 3	180	149	82.7	1.2(0.4-1.5)	0.6
less than 6	79	68	86	1.6(0.3-1.6)	0.3
Breed					
Exotic	170	133	78.2	1	
Indigenous	250	214	85.6	1.7(0.4-1.3)	0.3
Kebles					
ade Aro	8	4	50	1	
wandarboloso	16	9	56.2	1.2(0.02-1.8)	0.1
kokote	28	16	57.1	1.3(0.01-1.6)	0.1
Ade Sibaye	10	6	60	1(0.1-1.7)	0.9
Ampo Koysha	17	11	64.7	1.3(1-1.2)	0.1
Zalashala	16	6	72.7	1.3(0.1-1.2)	0.1
hanaza	27	21	77.7	1.6(1.3.6)	0.5
dembakoysha	31	26	83.8	1.7(1.01-3.1)	0.1
ambe shoya	28	24	85.7	1.72(1.4-4)	0.1
shochoraogodoma	24	21	87.5	1.8(1.6-9)	0.2
waja shoya	25	22	88.0	1.81(0.6-1)	0.06
chocha	25	22	88.0	1.9(0.1-4.8)	0.9
gulgula	35	31	88.5	1.92(4.1-9)	0.2
Harto kontola	20	18	90.0	1.92(4.0-9.1)	0.2
koysha Ogodoma	13	12	92.3	10(02-7.2)	0.7
dalbo	20	19	93.0	10.7(0.2-11)	0.04
borkoshe	22	21	95.4	18(0.03-1.2)	0.08
Sere finchewa	49	48	97.6	19(1.6-9.5)	0.2
Districts					
damot gale	78	59	75.6	1	
sodozuri	130	104	80.0	1.6(0.2-1.6)	0.3
humbo	113	94	83.1	1.7(0.2-1.8)	0.4
kindokoy	99	90	90.4	1.8(0.1-9)	0.01
Previous outbreak					
yes	130	103	79.2	1	
no	290	244	84.1	0.7(1.3-4.2)	0.4
Production					
intensive	59	46	77.9	1	
extensive	361	303	83.3	1.7(0.2-1.2)	0.6
No of flocks					
less than 5	107	85	79.4	1	
greater than 5	262	313	87.3	1.7(0.4-1.3)	0.3

Discussion

The overall prevalence in the current study was found to be 82.6% in poultry production at study area. The difference in prevalence of rotavirus infection between studies could be attributed to the number of samples investigated, livestock management style, geographical variations, and differences in the test method applied Despite expeditious growth of poultry in Ethiopia, this is the first study on overall prevalence of IBV in study area. The current study had determined that IBV is a major respiratory pathogen that causes catastrophic morbidity; mortality leads to heavy economic loss.

The study investigated that IBV infected birds characterized by signs, that is, severe conjunctivitis, lacrimation, gasping, sneezing, watery eyes, severe tracheal rales and cough. Correspondingly, severe conjunctivitis, lacrimation, sneezing, mild tracheal rales and cough have been reported in 2013 in Egypt [11].

Additionally, IBV infected poultry were depressed, lethargic, reluctant to move and take feed these finding was in agreement with [12]. Main lesions in respiratory tract were reddish streaks ranging from mild to severe, increased concentration of mucin in trachea with accumulation sero-mucus exudates in trachea and bronchi. These findings are correlated with [8,12]. However, lungs were congested, discolored, infiltrated with mucus leading to pneumonia that were similar with previous reports [8].

It has been found that sero-mucus exudates in trachea are due to degeneration of cilia by viropexin enzyme produced by IBV (Ashraf et al., 2010). Similarly, infiltration of inflammatory cells in the lamina propria and submucosa, activation of goblet cells, oedema in the submucosa, epithelial lymphoid infiltration, epithelial hyperplasia in trachea have been reported [13,12,8].

Consultation Recommendation

Infectious bronchitis virus is an acute and highly contagious disease of poultry industry. It causes respiratory distress, heavy morbidity and mortality. Prevalence of IBV is slightly higher in layers (61.2%) than broilers (52 %). Therefore, studies must be conducted on the prevalence, identify associated risk factors of infection. Hence based on the current findings, the following points were recommended.

- ✓ Public education and awareness creation among the communities of poultry farms about the method of transmission, risk factors, prevention, and control methods and the effect of the disease are essential.
- ✓ Proper hygienic practices and good husbandry or management practices should be exercised.
- ✓ Further large scale epidemiological and molecular studies are warranted to determine economic importance of infectious bronchitis in Ethiopia

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