Research article

Seroprevalence of infectious bursal disease and its potential risk factors in backyard chicken production of Waliso district, South Western Shoa Zone, Ethiopia

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Abstract

A cross sectional study on infectious bursal disease was conducted in apparently healthy backyard chicken at Waliso district of Southwestern Shoa, central oromia, Ethiopia from from November, 2018 to October, 2019. A total of 282 chickens were randomly selected to estimate seroprevalence of IBD infection and to identify the likely potential risk factors for the disease. Serum samples collected and serological test conducted in laboratory at National Animal Health Diagnosis and Investigation Center Sebeta, Ethiopia. Out of 282 serum samples tested 224 were positive for indirect ELISA technique and the overall seroprevalence of IBDV in the study area was found to be 79.43% at individual level. Educational level of owners, kebeles and flock size significantly affect seroprevalence of IBD in the study area. The effect of difference in managements like source of replacement, frequency of house cleaning, use of disinfectant and isolation practice has a significant effect on IBDV sero-prevalence. A lower seroprevalence of IBDV was reported in good hygienic level of house (26.7%) than poor level of chicken house hygiene (96.4%) with statistically significant difference (P < 0.05). The seroprevalence of IBDV in the present study associated with chicken management, flock size, owner education level and other animal related risk factors for occurrence of the disease. Therefore, awareness on chicken health management, and importance of immunization would help to minimize the prevalence of the disease and play crucial role in the control of the disease. Furthermore, characterizing virus strains circulating in the area in future study is recommended.

Keywords: Backyard chickens, Infectious bursal disease, Risk factors, Seroprevalence

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INTRODUCTION

Chickens are the most important species, adapted globally to different ecological conditions where human beings live and are important to subsistence, economic and social livelihoods of a large human population (Bettridge, 2014). Ethiopia has 50.38 million chickens population from which 96.9% of the chicken populations are indigenous chickens, while the remaining 2.56% and 0.54% consists of exotic and hybrid breeds (CSA, 2015).

Despite, Ethiopia owned huge chicken flock; there are different constraints like poor nutrition, poor management and prevalent diseases like Infectious bursal disease that hinder the productivity of the chicken in most area of the country. Infectious bursal disease (IBD) has been a great challenge to the poultry industry world-wide for a long time, particularly for the past two decades following emergence of new pathotypes; variant and very virulent strains (Mazengia, 2008). The greatest economic losses are due to sub clinical disease in chickens from one to twenty-one days of age. At this stage the virus impairs the immune response and renders the chick susceptible to various infections. The disease is spread through orally via contaminated feed and water (Sharma et al., 2000; Sun and Gao, 2001).

In Ethiopia the occurrence of IBD was first reported in 2002 at privately owned commercial poultry farm in which from 45-50% mortality rate was documented (Zeleke et al., 2003). In addition to this report Mariam and Abebe (2007) reported seropositivity of 98.90% by Agar Gel Immuno diffusion test in Amhara region (Andasa farm). Other published reports of Zeleke et al. (2005) and Hailu et al. (2010b) also documented incidence rates of 17.40% in Dabre zeit and 38.4% in Bahirdar areas. The seroprevalence of IBD in chickens were also documented as 72.70% in Gondor (Kassa and Molla, 2012), 38.39% in Bahirdar (Sinidu et al., 2015), and 38.30% in Sebeta district (Asamenew et al., 2016).

Several factors like vaccination status, biosecurity measure and management practice may play an important role in the seroprevalence of IBD. Knowledge on infectious bursal disease epidemiology is necessary for successful prevention-control program at backyard chicken. Information on status of the disease and awareness of chicken owner on IBD in and around Waliso district, south west shoa zone, Ethiopia is limited and need to determine the status of IBD in the area. Therefore, the objectives of this study was to estimate seroprevalence of IBDV infection and its potential risk factors in backyard chickens production systems in Waliso district of south western Shoa of Oromia, Ethiopia.

MATERIALS AND METHODS

Study Area

This study was conducted in Waliso district of South Western Shoa Zone of Oromia Regional state, Ethiopia. The district is located at 116 km South West of Addis Ababa on the highway leading to Jimma at an altitude of 1500 to 2900 m above sea level. The area is located at longitude of 37°58’16.3”E and latitude of 8°32’23.0”N. It was characterized by mild sub-tropical weather,
with average minimum and maximum temperatures of 5.5°C and 23°C respectively. This area experiences a binomial rainfall pattern with along rain season from June-September and short rain season from March-April (CSA, 2015). The area has 35 rural and 2 urban administrative Kebeles (small administrative unit of government in a given area). The total human population of the district was 165,391, of which 50% was reported to be Female. The livestock resource of the study district comprises of 224,334 cattle, 39,543 sheep, 51,042 goats, 7,625 horses, 2101 mules, 16,320 donkeys and 147,679 chickens (115,814 local and 31,865 hybrids) (Waliso Woreda Finance and Economic Development Office, 2018).

Study Population and Management

The study was conducted in chickens raised under backyard production system. Feeding systems in the backyard poultry production system was not purposeful and scavenging was almost the only source of diet.

Housing systems in the backyard poultry production system is rudimentary and mostly built with locally available materials. The bio-security of the backyard poultry production system was very poor, as scavenging birds live together with people and other species of livestock. Cleaning of chicken houses usually included with manual removal of manure and bedding, which was subsequently used as fertilizer. There was practice of isolating sick birds from the household flocks with most of dead birds were left for either domestic or wild predators.

Study Design

A cross sectional study was implemented from November, 2018 to October, 2019 to assess seroprevalence of IBDV with its associated risk factors in backyard chickens production of selected kebeles. Semi structured questionnaire interview of selected household owner was used to assess management practice and other factors associated to the occurrence of IBDV.

Sample size and sampling technique

From ca total of 37 kebeles in the study district, 6 kebeles which include Dire dulet, Bedese koricha, Fodu gora, Obi koji, Gurura Beka and Tombe Anchabi were selected purposively based on easy of accessibility and chickens population per household.

The sample was collected from non-vaccinated and apparently healthy chickens from 3 weeks up to 6 months. The selection of household and samples was proportionally allocated between selected kebeles of study area. Flock size in the study animals were range from 9-15 which includes only local breeds of chickens. About 10-13 household per kebeles and 3-5 chickens per households were selected proportionally for sampling. Therefore, a total of 282 samples were collected to determine seroprevalence of IBDV in the study area.
Data collection

Data on potential risk factors for the occurrence of IBDV was collected during sample collection from chicken on format prepared for this purpose. Housing system, kebeles, flock size, sex, age in weeks and hygienic level of house were emphasized as risk factors of the disease.

Semi-structured questionnaire was conducted to interview 70 selected owners of households to assess the impact of owners educational level, experience of keeping chicken (year), isolation practice of sick chickens, disposal of dead carcass and source of replacement practices for the occurrence of IBDV infection in the study area.

About 1.5-3ml of blood samples were collected from brachial veins using 21G inch needle and 3ml syringes. The blood was allowed to clot overnight (24 hrs) in the syringe and then separating the serum to sterilized cryovials and transported by icebox to National Animal Health Diagnosis and Investigation Center (NAHDIC) Sebeta.

For purpose of ethical issues the study was reviewed and approved by Jimma University College of Agriculture and Veterinary Medicine, Ethiopia on February 07, 2019.

Serological test

Samples were tested using a commercial ELISA kit (ProFLOK® PLUS, IBD Coated ELISA, Symbiotic Corporation, San Diego, USA) at NAHDIC, Sebeta Ethiopia. This commercial ELISA kit detects IBD antibody and demonstrates excellent correlation with the virus neutralization test. All conditions were standardized according to the kit manufacturer and conditions described for poultry disease monitoring using ELISA. Samples were tested for IBDV specific antibodies using a commercial IBDV ELISA kit following manufacture’s direction. Serum was prediluted to1:500 in dilution buffer, added to an antigen coated plate. Specific IBD antibodies in the serum form antigen -antibody complex with antigen bounded to the plate. After washing the plate, anti- chicken horse radish peroxidase conjugate was added to each well and the formed antigen- antibody bind to the conjugate. After incubation period unbounded conjugate was removed by washing and substrate which contains chromogenis added which form a clear to green blue color in the presence of enzyme, after incubation for 15 minute stop solution is added to terminate reaction and plate was read using ELISA reader at 450nm. Row absorbance data was transferred to a personal computer for further calculation and analysis.

After reading of the ELISA results, the test validity was checked for each plate based on two criteria set by the kit manufacturer; the mean optical density (OD) of the positive controls and normal controls on each plate. The test was considered valid of when the mean OD405 of the positive control value range between 0.250 and 0.900 and when the mean OD405 of the normal (negative) control serum is less than 0.250. The sample to positive (SP) ratio of each test serum was calculated as:

$$ SP = \frac{\text{Sample absorbance} - \text{Average normal control}}{\text{Corrected positive control absorbance}} $$

Hence, SP value ≤ 0.25 was Negative while SP value > 0.25 was considered Positive
Data management and Analysis

Data obtained from questionnaire and laboratory test (Indirect ELISA) were inserted into Microsoft Excel for Windows 2010. Analyzes were performed using STATA software version 12. Descriptive statistical methods were used to summarize seroprevalence of IBDV and characteristics of the study animals. Odd ratio (OR) was used to examine the strength of association between risk factors and outcome. Chi-square ($\chi^2$), Uni-variable and Multivariable logistic regression was conducted to examine the association of the risk factors with occurrence of IBDV. A 95 % confidence intervals were calculated and P-value <0.05 was used for significance.

RESULTS

From a total of 282 serum samples examined 224 were positive to IBDV using indirect ELISA test with an overall seroprevalence of 79.43% in the study area. The present study showed that sex of study animals have an effect on seroprevalence of IBDV. The seroprevalence of IBD virus slightly decreases with increasing age of chicken.

Table 1  Seroprevalence of IBDV in association with animal related risk factors in the study area.

| Factors       | Number of Sample | Number of Positive (%) | Uni-variable analysis |
|---------------|------------------|------------------------|-----------------------|
|               |                  |                        |                      |
| Sex           |                  |                        |                      |
| Female        | 156              | 128(82.05)             | 0.70                  |
| Male          | 126              | 96(76.19)              | 0.22                  |
| Age/week      |                  |                        |                      |
| <4            | 207              | 173(83.57)             | 0.95                  |
| >4            | 75               | 51(68.00)              | 0.89                  |
| Total         | 282              | 224 (79.43)            |                      |

The higher seroprevalence was recorded in female chickens (82.05%) than males (76.19%). Female chickens became more seropositive to IBDV with odds of 0.70 as compared to Male with statistically non-significant difference (P > 0.05). Chickens of less than 4 weeks age had chance to become seropositive with odds of 0.95 as compared to more than 4 weeks age. The statistical test indicated non-significance difference with P > 0.05 (Table 1).
Table 2 Seroprevalence of IBDV across the study Kebeles

| Kebele          | Number (N) | Positive (%) | Odd  | SE  | P-Value | 95% CI    |
|-----------------|------------|--------------|------|-----|---------|-----------|
| Fodu Gora       | 51         | 46 (90.20)   | 0.21 | 0.10| 0.002   | 0.08-0.56 |
| Gurura Beka     | 52         | 45 (86.54)   | 1.89 | 1.15| 0.29    | 0.57-6.24 |
| Direduleti      | 47         | 39 (82.98)   | 1.32 | 0.74| 0.62    | 0.44-3.97 |
| Tombe Anchabi   | 44         | 36 (81.82)   | 0.89 | 0.49| 0.84    | 0.30-2.64 |
| Obi Koji        | 43         | 35 (81.40)   | 0.92 | 0.50| 0.88    | 0.31-2.71 |
| Bedesa Qoricha  | 45         | 23 (51.11)   |      |     |         |           |
| **Total**       | **282**    | **224 (79.43)**|    |     |         |           |

In study kebeles the highest seroprevalence of IBD was found at Fodu gora (90.2%) and the lowest was recorded at Bedese Qoricha (51.11%). Fodu gora kebele had more chance to be come seropositive by IBDV with odds of 0.21 with reference to Bedese Qoricha kebele and showed statistically significant difference (P < 0.05) as shown in Table 2.

The factors that affect chicken rearing like owner age, Educational level and Experience of keeping chicken (year) has been seen for its association. Difference in age group of farm owner and their educational level showed no significance difference (P > 0.05) on seroprevalence of IBDV. The owner of age >30 years was indicated for higher seroprevalence of IBDV (85%) than those <25 years old age (74.19%). Slightly higher seroprevalence was indicated in illiterate group than educated persons rearing backyard chickens. Concerning experience of keeping chicken >4 years was significantly lower seroprevalence of IBDV (63.64%) than in those of < 1 year experience (93.75%) with P=0.01 (Table 3).

Table 3 Relationships of age, education level and experience of farm owner with seroprevalence of IBDV in the flock

| Factors                        | Number of respondents | Number of Positive flock (%) | \( \chi^2 \) | P-value |
|--------------------------------|-----------------------|-----------------------------|-------------|---------|
| **Owner age**                  |                       |                             |             |         |
| <25                            | 31                    | 23 (74.19)                  | 0.85        | 0.06    |
| 25-30                          | 19                    | 15 (78.95)                  |             |         |
| >30                            | 20                    | 17 (85.00)                  |             |         |
| **Education level**            |                       |                             |             |         |
| Illiterate                     | 17                    | 15 (88.24)                  | 1.82        | 0.40    |
| Grade 1-4                      | 33                    | 26 (78.79)                  |             |         |
| Grade 5-9                      | 20                    | 14 (70.00)                  |             |         |
| **Experience of keeping chicken (year)** |               |                             |             |         |
| < 1                            | 16                    | 15 (93.75)                  | 7.69        | 0.01    |
| 2                              | 15                    | 14 (93.33)                  |             |         |
| 3                              | 17                    | 12 (70.59)                  |             |         |
| > 4                            | 22                    | 14 (63.64)                  |             |         |
Experiences of rearing chickens have inverse relationships with seroprevalence of IBDV as prevalence decreases with increasing experiences of owners (Figure 1).

In questionnaire survey 70 respondents were interviewed on management practice and owner characteristic for rearing the chickens in the study area. The effect of source of replacement, isolation practices and disposal of dead carcass on seroprevalence of IBD indicated that chickens replacement from market had higher seroprevalence of IBDV (80%) than those from near neighbors (76.6%). The isolation practice indicated lower seroprevalence of IBDV (72.92%) than those not practiced sick chicken isolation from health one (90.91%). Properly disposal of dead carcass (Buried) indicated lower seroprevalence of IBDV (65.71%) than that of group practicing to thrown dead carcass on the field (91.43%) (Table 4).
Table 4 Seroprevalence of IBDV in association with management factors using Multivariable logistic regression.

| Factors                        | Number (N) | Number of Positive (%) | OR   | SE  | P-Value | 95% CI       |
|--------------------------------|------------|------------------------|------|-----|---------|-------------|
| Source of replacement          |            |                        |      |     |         |             |
| Market                         | 40         | 32(80)                 | 0.82 | 0.48| 0.74    | 0.26-2.59   |
| Neighbors                      | 30         | 23(76.67)              |      |     |         |             |
| Isolation practice             |            |                        |      |     |         |             |
| Practiced                      | 48         | 35(72.92)              |      |     |         |             |
| Not practiced                  | 22         | 20(90.91)              | 3.71 | 3.00| 0.11    | 0.76-18.16  |
| Disposal of dead carcass       |            |                        |      |     |         |             |
| Buried                         | 35         | 23(65.71)              |      |     |         |             |
| Thrown on the field            | 35         | 32(91.43)              | 5.56 | 3.90| 0.01    | 0.41-0.99   |
| Hygienic level of house        |            |                        |      |     |         |             |
| Good                           | 15         | 4(26.7)                |      |     |         |             |
| Fair                           | 27         | 24(88.9)               | 29.31|145.82|0.09    |41.11-59.79 |
| Poor                           | 28         | 27(96.4)               | 88.00|12.91|0.00    |0.07-0.89   |
| Housing system                 |            |                        |      |     |         |             |
| Cage                           | 19         | 16(84.9)               | 0.80 | 0.37|0.03    |0.33-0.96   |
| With family                    | 32         | 28(87.5)               | 0.16 | 0.07|0.04    |0.07-0.37   |
| Separate                       | 19         | 11(57.9)               |      |     |         |             |
| Flock size                     |            |                        |      |     |         |             |
| <10                            | 19         | 13(68.4)               |      |     |         |             |
| 10-12                          | 30         | 25(83.3)               | 2.68 | 0.92|0.01    |0.36-0.25   |
| >12                            | 21         | 16(76.2)               | 3.08 | 1.21|0.01    |0.42-0.67   |

Hygienic condition was significantly associated with the occurrence of IBD when Multivariable logistic regression analysis was carried out (Table 4). When hygienic condition of house was observed; the highest seroprevalence of flocks maintained in poor hygienic conditions (96.4%) than those maintained under good hygienic condition (26.7%) with significant difference (P < 0.05).

Housing system was significantly associated with the occurrence of IBD when Multivariable logistic regression analysis was carried out. High seroprevalence was reported in chickens kept in house with family with odds of 0.16 as compared to those kept in separate house with significant difference (P < 0.05). Household flock size had significant effect on the seroprevalence of IBD in the study area. Flocks of chicken with size of >12 animals per flock had an odd 3.08 of having IBD higher seropositivity than flocks with size less than or equal to 10 animals per flock.
DISCUSSION

An overall seroprevalence of 79.43% of IBDV in chickens kept under backyard poultry production system was indicated in the present study area. This report is comparable with findings of Hailu et al. (2010a) who reported 76.64% seroprevalence from three districts of West and South West Shoa, 72.7% by Kassa and Molla (2012) in Gonder, Tesfaheywet et al. (2012) who reported 82.2% from Central Ethiopia. Serological studies conducted in different parts of the country, 100% in DebreZeit (Woldemariam and Wossene, 2007) and 90.3% in Mekele (Shiferaw et al., 2012) reported higher prevalence than current study. However, lower seroprevalence of 38.39% was reported by Sinidu et al. (2015) in Bahirdar and 38.3% by Asamenew et al. (2016) in Sebeta hawas Ethiopia. The variation in seroprevalence of IBDV in these studies attributed to the difference in poultry management systems in backyard poultry production such as poor vaccination practice, poor sanitary condition, nutritional deficiencies or frequent contact of wild birds.

In this study relatively higher seroprevalence of IBDV was recorded in female chickens (82.05%) than male (76.19%) with non-significant difference. This finding was agreed with the report of Sinidu et al. (2015) in Bahirdar. The lower seroprevalence of IBDV was also recorded in male chickens by Shiferaw et al. (2012) and Tadesse and Jenbere (2014) in different parts of Ethiopia.

The seroprevalence of IBDV was found high (80.46%) in age group ≤4 weeks, than higher age groups. A comparable result of seroprevalence 86.6% and 87.26% of IBDV in young chicken was reported by Shiferaw et al. (2012) and Hailu et al. (2010a) respectively. This is due to the fact that, at early age the virus impairs the immune response and renders the chick susceptible to various infections and different genetic backgrounds of chicken breeds also may have different impacts on the early immune responses to IBDV infection (Aricibasi et al., 2010; Tippenhauer et al., 2013).

In the present study, study sites indicated a statistical significant difference in seroprevalence of IBDV. Fodu gora kebele had more chance to become seropositive by IBDV with odds of 0.21 with reference to Bedese Qoricha kebele. Similar findings were reported by Hailu et al. (2010a) in backyard local chicken of three district of Oromia region regional state of Ethiopia. These differences in seroprevalence of IBDV in the study area are generally attributed to difference in management and environmental settings.

Experience of farm owner has importance in chicken health management and easy of understanding problems occurring in the flocks of chicken. In this study the level of IBDV infection was decreasing with increasing experiences of owners. This implies that owner might be able for early identification of the problems, take measures for its control and provide good management to prevent the disease as become more experienced. Significant variations in practicing disposal of dead carcass were indicated in the present study. A 91.43% seroprevalence of IBDV occurred in those groups thrown dead carcass on the field and 65.71% in flocks where owners dispose dead carcass and waste products by burning or buried it. This can associated with frequent movement of backyard chicken and constant contact with infected environments.
Hygienic condition of the environment has a significant effect ($P < 0.05$) on seroprevalence of IBDV. Higher seroprevalence was observed in flocks maintained under poor hygienic conditions (96.4%) than good hygienic condition (26.7%). The lower prevalence in good hygienic conditions could probably be due to the favorable and healthy environmental condition. This might be due to variation in predisposing factors such as improper cleaning, keeping used litter in house, poor ventilation and crowding of wastes as these factors influence spread of the infection from house to house and from flock to flock.

A statistical significant difference in seroprevalence of IBDV was observed in different flock size of present study. The highest seroprevalence of IBDV (83.3%) were found in largest flock size ($\geq 10$) than the lower flock size. This difference might be due to the fact that increased chicken population number is a factor for stress, transmission and widely occurring of the diseases (Farooq et al., 2003).

**CONCLUSIONS**

This study determined high seroprevalence of IBDV and slight susceptibility difference with respect to sex and age of studied chickens. Experiences of rearing chickens improve ability of owners to provide proper management that lowers the seroprevalence of IBDV. A lower seroprevalence of IBDV was obtained in good hygienic level of houses than fair and poor level of chicken house hygiene. Risk factors such as flock size and study sites have significant effect on seroprevalence of the disease. Therefore, attention should be given in improvement of village chicken management practices with special emphasize on predisposing factors of IBDV through community training and further studies to characterize virus strains circulating in the study area.

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**AUTHOR CONTRIBUTIONS**

Chala B.: Design the research work, sample collection and processing, manuscript writing.
Ararsa D.: Design, data analysis, manuscript editing and review.
Asamenew T.: Provided materials and reagents, sample processing and manuscript review.
Tadele T.: Concept, design, manuscript editing and review.
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