

Project ID: 448

Competitive Research Grant (CRG)

Sub-Project Completion Report

on

Monitoring and Surveillance of Duck Diseases in Hakaluki and Tanguar Haor and Development of Suitable Vaccination Models and other Preventive Strategies

Project Duration

October 2017 to March 2019

Department of Pathology and Parasitology
Chittagong Veterinary and Animal Sciences University
Khulshi, Chittagong-4225



Submitted to
Project Implementation Unit-BARC, NATP 2
Bangladesh Agricultural Research Council
Farmgate, Dhaka-1215



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Project Implementation Unit
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Bangladesh Agricultural Research Council (BARC)
New Airport Road, Farmgate, Dhaka – 1215
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Edited and Published by:

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Acknowledgement

The execution of CRG sub-project has successfully been completed by Department of Pathology and Parasitology, Chittagong Veterinary and Animal Sciences University using the research grant of USAID Trust Fund and GoB through Ministry of Agriculture. We would like to thank the World Bank for arranging the grant fund and supervising the CRGs by BARC. It is worthwhile to mention the cooperation and quick responses of PIU-BARC, NATP 2, in respect of field implementation of the sub-project in multiple sites. Preparing the project completion report required to contact a number of persons for collection of information and processing of research data. Without the help of those persons, the preparation of this document could not be made possible. All of them, who made it possible, deserve thanks. Our thanks are due to the Director PIU-BARC, NATP 2 and his team who have given their wholehearted support to prepare this document. We hope this publication would be helpful to the agricultural scientists of the country for designing their future research projects in order to technology generation as well as increasing production and productivity for sustainable food and nutrition security in Bangladesh. It would also assist the policy makers of the agricultural sub-sectors for setting their future research directions.

Published in: September 2018

Printed by:

Acronyms

Abbreviations and symbols	Elaboration
AI	Avian influenza
AIV	Avian influenza virus
BARC	Bangladesh Agricultural Research Council
BDT	Bangladesh taka
bp	Base pair
CRG	Competitive Research Grant
CVASU	Chittagong Veterinary and Animal Sciences University
DLS	Department of livestock services
DNA	Deoxyribonucleic acid
DPP	Department of pathology and parasitology
DVE	Duck viral enteritis
DVH	Duck viral hepatitis
e.g.	For example
ELISA	Enzyme-linked immunosorbent assay
F	Frequency
GI	Gastrointestinal
ml	Milliliter
Min	Minute
NATP	National Agricultural Technology Programme
NDD	New duck disease
OR	Odds ratio
PCR	Polymerase chain reaction
PIU	Project Implementation Unit
RNA	Ribonucleic acid
RT-PCR	Reverse transcription polymerase chain reaction
Tk	Taka
VTM	Viral transport medium
RPM	Rotation per minute
%	Percentage

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Executive Summary

Bangladesh has the third largest duck population (38.1 million) in the world. A significant number of ducks are reared in the swampy land areas (e.g. haor) that play a vital role in the livelihood of poor people in the areas. Infectious and non-infectious diseases in haor areas are considered one of the major constraints in duck rearing. Current study was designed to determine the prevalence of different infectious and non-infectious diseases in the domestic ducks of Hakaluki and Tanguar haor as well as to recommend a compatible vaccination strategy/schedule for disease control and prevention. Various demographical data were collected from the farmers during sample collection using a pre-structured questionnaire. Then, a total of 200 fecal, blood, tracheal and cloacal swabs were collected in each season (winter, summer and monsoon) from household and free-ranged ducks of Hakaluki and Tanguar haor. Different parasitological, microbiological and molecular techniques were used to identify gastrointestinal (GI) parasitic, bacterial and viral diseases in ducks. Results demonstrated that ducks in the haor areas were suffered from six genera of GI parasitic infections. The rates of occurrence of such infections were 3.33, 4.67, 17.83, 9.33, 11.33, 5.83 and 3.33% for *Ascaridia* spp, *Capillaria* spp, *Prosthogonimus* spp, *Amidostomum* spp, *Tetrameres* spp and *Hymenolepis* spp respectively. The prevalence of blood protozoan such as *Haemoproteus*, *Leucocytozoon* and *Plasmodium* spp was 13.17, 8.33 and 7.67% respectively. Among the viral diseases identified, the highest prevalence was recorded for avian influenza (AI; 90.5%) compared to duck viral hepatitis (4%) and duck viral enteritis (3.33%). Among the bacterial diseases, infections caused by *Escherichia coli*, *Salmonella typhimurium*, *Staphylococcus aureus* were found to be 55.33, 20.5 and 44.1%, respectively in the ducks of the haor areas. Furthermore, using PCR assay this study documented first time in Bangladesh the presence of Duck septicemia also known as New duck disease (NDD) with a prevalence rate of 10%. The occurrence of *Mycoplasma gallisepticum* infection was found in 5.3% duck samples. Analysis of various demographic data identified statistically significant risk factors and protective factors that are associated with the duck rearing in the haor areas. Ducks those scavenges in 'pond' and 'wetlands' areas were found more protective ($P=0.007$ and $P=0.01$ respectively) for *Haemoproteus* spp infection compared to household premise, rice paddy field and river. 'Flock size between 520 to 4000' ($P=0.02$) and 'muddy type housing' ($P=0.001$) were found more protective ($P=0.02$ and $P=0.001$ respectively) for *Capillaria* spp infection in ducks compared to other factors (e.g. bamboo, wooden type housing). Through considering the disease prevalence data as well as risk factors identified in this study, a suitable vaccination schedule has been recommended. The vaccination schedule developed on the basis of the outcome of this study is needed to be applied in future to justify its efficacy through a proper and active monitoring system. This study also recommends taking special attention to prevent and control AI in duck population in the haor areas, as significantly higher prevalence of the disease in the areas may contribute to the endemicity of such disease in the poultry population of Bangladesh. The first time documented NDD in haor areas also needs special attention to introduce vaccination program in the breeder flocks to prevent such disease in ducks.

CRG Sub-Project Completion Report (PCR)

A. Sub-project Description

1. Title of the CRG sub-project:

Monitoring and surveillance of duck diseases in Hakaluki and Tanguarhaor and development of suitable vaccination models and other preventive strategies

2. Implementing organization:

Department of Pathology and Parasitology, Chittagong Veterinary and Animal Sciences University

3. Name and full address with phone, cell and E-mail of PI/Co-PI (s):

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4. Sub-project budget (Tk):

Total: Taka 22,00,000.00

5. Duration of the sub-project:

- 1.1 Start date (based on LoA signed) : 9 October, 2017
1.2 End date : 30 September 2018

6. Justification of undertaking the sub-project:

Ducks contribute about 22-25% of the total annual egg production and their contribution remains constant throughout the year. Despite tremendous opportunities of duck farming in the haor areas (Hakaluki and Tanguarhaor), the wet land ecosystem in Bangladesh, the challenges and opportunities of duck farming in the haor areas have remained unexplored. The main obstacles have so far been identified include extreme remoteness, lack of basic knowledge of duck rearing and the poverty. The other factors that are strongly associated in developing commercial duck farming are high mortality due to various diseases and shortage of adequate supply of vaccines. Farmers those are lacking sufficient knowledge of disease management, prevention and control are always under threat of disease outbreaks in their duck farms. In the haor region there are also inadequate healthcare services from certified, skilled and experienced veterinarians.

Duck farmers in the haor region do not have sufficient knowledge of disease management. They do not know the symptoms of duck diseases and are unable to decide what to do. Therefore, careful

planning and identification of existing diseases and their epidemiology is crucial for their effective control and prevention. To our knowledge, no such empirical research was conducted in the haors for duck diseases.

In such a situation, monitoring of the diseases will identify the challenges of duck farming in haor areas and the development of strategic plans relating to prevention and control of duck disease will ultimately result in a revolutionary change in poultry production to meet the protein demand, generate employment opportunities for the local communities and as a whole help to achieve economic development in the country.

7. Sub-project goal:

To identify the challenges of duck farming in the Haor areas and to develop strategic plans for the prevention and control of duck diseases in the areas.

8. Sub-project objectives:

- i) To develop a database of existing infectious diseases of ducks and their risk factors in the selected areas of Hakaluki and Tanguar haor.
- ii) To monitor the duck diseases through active surveillance and GIS mapping in the selected haor areas.
- iii) To identify the challenges of duck farming and development of strategic plans to prevent duck diseases in the haor areas.

9. Implementing Location(s):

Hakaluki haor of Moulovibazar district and Tanguar haor of Sunamganj district in Sylhet division were selected for the study (Figure 1 and 2).

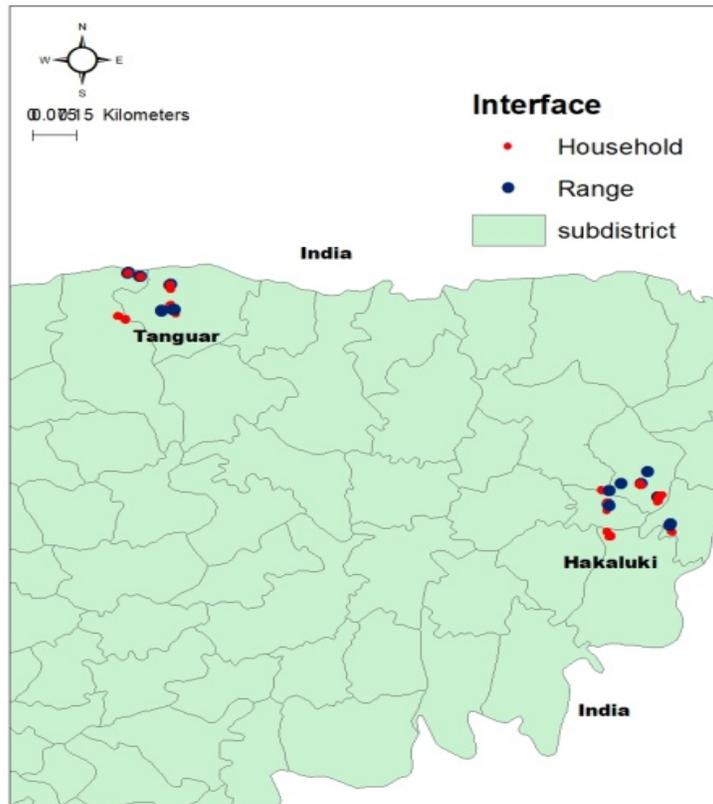


Figure 1: Geographical location of Hakaluki and Tanguar haor

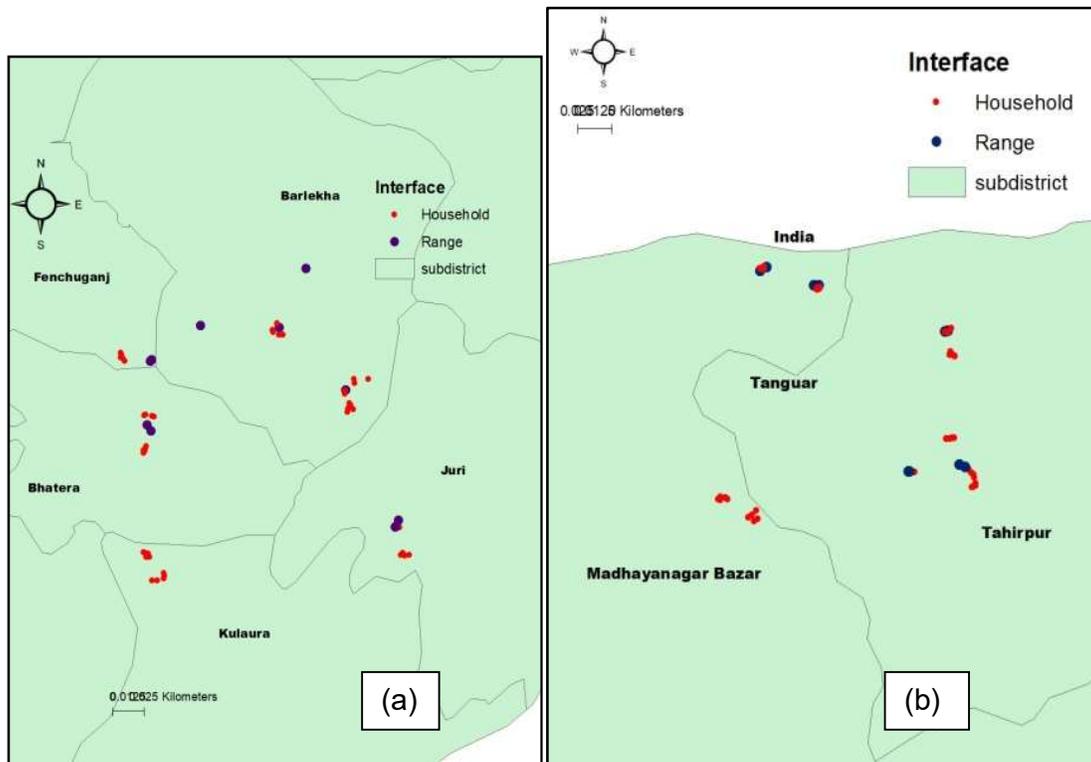


Figure 2: Geographical distribution of household and range farms
 (a) Hakaluki haor of Baralekha upazilla, (b) Tanguar haor of Tahirpur upazilla.

10. Methodology:

10.1 Development of a database of existing infectious diseases of ducks and their risk factors in the selected areas of Hakaluki and Tanguar haor

Sampling strategy

Toward meeting the surveillance requirements in Hakaluki and Tanguar haors any scavenging or captive ducks within the geographic location irrespective of age were included in this study.

Study design

Three surveillance visits for monitoring of duck diseases in and around the villages of Hakaluki and Tanguar haor (Figure 2). In this cross-sectional study, five villages in and around each haor areas were randomly selected for household and free range duck sample collection.

Collection of data for risk factor analysis of duck farming in haor areas

An active face to face survey was performed during all the three seasons. A questionnaire was developed with a view to identifying the challenges of duck farming in haor areas based on the available information from previous studies (Rahman *et al.*, 2009) and suspected challenges of duck rearing in haor areas. Several factors (e.g. Social status, status of the duck farms, managerial data like cleaning of duck houses, biosecurity condition, vaccination & deworming status, etc. were included in the questionnaire (Appendix figure 1). A total of 100 farmers who rear household ducks (Figure 3a and b) and 100 farmers who rear free-range ducks (Figure 3c and d) were selected for interview in time point of sampling. Altogether, 600 farmers were interviewed in three seasons.

Study population

Both household and free-range ducks were considered for sample collection in this study.

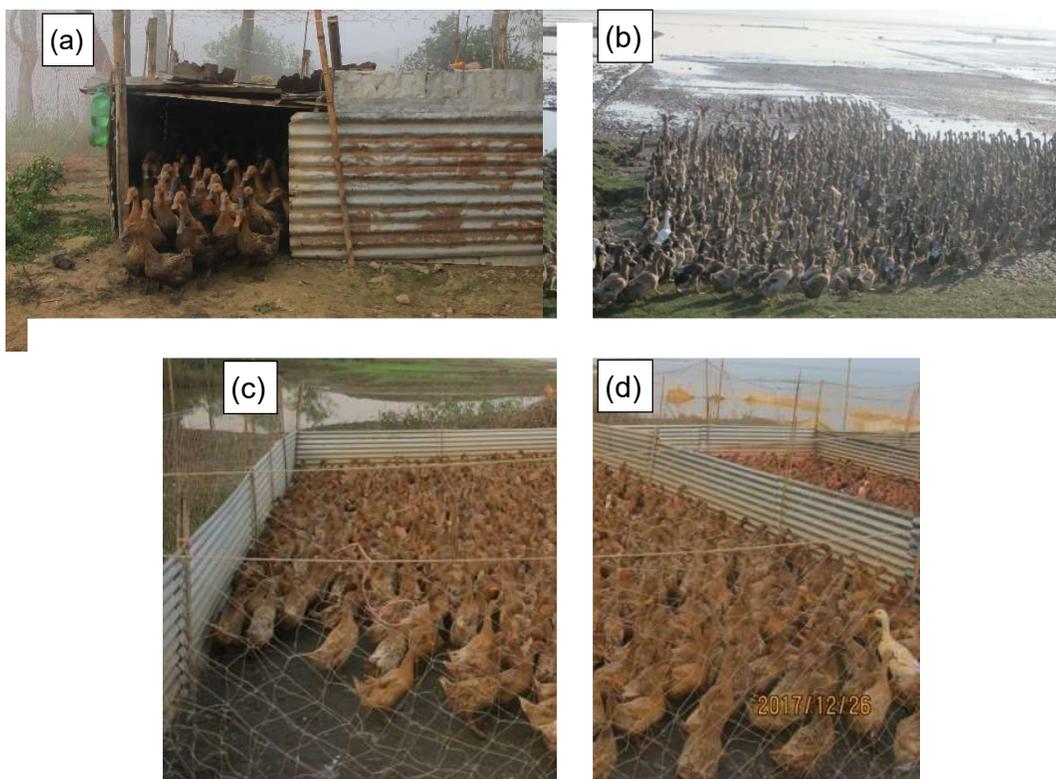


Figure 3: Household and free-range duck rearing at haor areas

Figures show household ducks rearing at Hakaluki haor (a) and Tanguar haor (b) and Free-range ducks at Hakaluki haor (c) and Tanguar haor (d).

Calculation of sample size

For sampling of household ducks, 10 ducks (one duck from each household) were selected from each village (from randomly selected 5 villages) targeting the sample size of 50 from each haor area. A minimum of 5 ducks in each household were considered as the inclusion criteria.

For sampling scavenging ducks, a total of 10 free-range duck flocks were randomly selected from each haor while the minimum of ≥ 500 ducks per flock was considered as inclusion criteria. From each selected flock 5 ducks were randomly selected and therefore a total of 50 duck samples were collected from each haor areas. Thus, the total sample size was 100 for household ducks and 100 for range ducks in winter visit. The same sampling size was repeated in the summer and monsoon seasons and thus altogether there were 600 samples.

Sample collection and shipment

In every season (winter, summer and monsoon), a total of 200 fecal samples, 200 tracheal swabs, 200 cloacal swabs as well as 200 blood samples were collected from both Tanguar and Hakaluki haor (Figure 4). Freshly voided pooled droppings were considered from each of the households and free range shelter areas. All the fecal samples were collected in sterile specimen container where 10% formalin was added for preservation. Cloacal and tracheal swabs were collected from individual duck using sterile swab sticks and immediately kept separately in transport media. For viral isolation, collected swabs were kept in VTM followed by shifting them to liquid nitrogen (-196°C) until preserved in -80°C freezer. For bacterial isolation, all the collected swabs were kept in nutrient broth (Oxoid™) and carried them with a cool box and preserved in refrigerator (4°C). Blood samples from individual birds were taken directly from wing vein using sterile needles and syringes. At least

two thin blood smears were prepared immediately after collection and fixed with 100% methanol. Specialized FTA cards (Whatman® FTA® card, Sigma) were used to collect blood samples for DNA extraction (FavorPrep Blood Genomic DNA Extraction Mini Kit™. Cat.No.: FABGK 001-2) followed by subsequent molecular studies. Furthermore, blood samples (2 ml/duck) were also collected in vacutainer tube (without anticoagulant) for the collection of serum. All samples carried to Clinical and Molecular Pathology laboratory under the Department of Pathology and Parasitology, CVASU and preserved appropriately until examined.



Figure 4: Sample collection from household and free-range ducks of Hakaluki and Tanguar haor. Figures show (a) Face to face interview while collection of demographic and farm related data, (b) Collection of samples in summer (from free range duck), (c) in monsoon (from household ducks) and (d) in winter season (from household duck), (e) Collection of blood directly from wing vein and (f) preparation and fixation of thin blood smear, (g) Collection of swab sample (e.g. tracheal swab) and (h) transferring of collected tracheal and cloacal swab into liquid nitrogen container.

Laboratory diagnosis of diseases

Different laboratory tests (e.g. parasitological, microbiological and molecular techniques) were used for the confirmation of the parasitic, bacterial, mycoplasmal and viral diseases. All the tests were performed at parasitology, clinical and molecular laboratories under the Department of Pathology and Parasitology, CVASU.

Diagnosis of parasitic diseases

For the identification of eggs of gastro-intestinal parasitic infections, three different types of tests, namely direct smear, flotation and sedimentation were performed according to previously described procedure (Hendrix and Robinson, 2006). At least, two smears were prepared from each sample for each technique to identify the morphological characteristics of helminths' eggs (Hendrix and Robertson 2006; Urquhart *et al.*, 1996 and Soulsby, 1982).

Haemoprotozoan diseases (e.g. *Haemoproteus* spp, *Plasmodium* spp and *Leucocytozoon* spp) were identified by examining Giemsa stained thin blood smear according to published methods (Afifi *et al.*, 2014). Further confirmation of these blood protozoans were done using PCR assay according to previously described methods (Benschet *et al.*, 2000; Hallergen *et al.*, 2004).

Diagnosis of bacterial diseases

For the diagnosis of different bacterial diseases, cloacal and tracheal swabs were used. Standard microbiological procedures were followed for the isolation using culturing (Figure 5a) and identification of bacteria (e.g. *E. coli*, *Salmonella*, *Staphylococcus*) (Wirth *et al.*, 2006; Rahn *et al.*, 1992; Brakstad *et al.*, 1992). MacConkey, EMB agar for the culturing of *E. coli*, XLD for *Salmonella* and Manintol salt agar for *Staphylococcus* were used as selective media.



Figure 5: Laboratory activities during identification of different bacterial diseases of ducks
Figure shows culturing of *Salmonella* spp on XLD agar (a) and detection of *Mycoplasma gallisepticum* in ELISA (b)

Polymerase chain reaction (PCR) was conducted for further confirmation of *E. coli*, *Salmonella typhimurium*, and *Staphylococcus aureus* according to published procedures (Wirth *et al.*, 2006; Rahn *et al.*, 1992; Brakstad *et al.*, 1992). Briefly, DNA was extracted from the pure cultures by boiling methods. Detail information of the primers for particular gene of each mentioned bacteria used in this study are shown in Table 1. PCR conditions followed were according to the previously published works (Wirth *et al.*, 2006; Rahn *et al.*, 1992; Brakstad *et al.*, 1992). For the confirmation of *Riemerella anatipestifer*, *Pasturella multocida* and *Borrelia anserine*, only PCR assay was performed using the primers shown in Table 1 (Tsai *et al.*, 2005; Miflin *et al.*, 2001; Barbour *et al.*, 1996), after DNA extraction from swab sample using commercial kit.

Diagnosis of mycoplasmal diseases

Blood serum samples were collected by centrifuging the blood samples at 8000 rpm for 20 min. Enzyme-linked immunosorbent assay (ELISA) technique was used to identify *Mycoplasma gallisepticum* (Figure 5b). Furthermore, PCR assay was used for all the ELISA positive samples (Garcia *et al.*, 2005). The specific primer set used for identification of mycoplasmal disease are presented in Table 1.

Diagnosis of viral diseases

Viral diseases were diagnosed directly from the DNA extracted from all the cloacal swab samples those preserved in VTM at -80°C. PCR was carried out using specific set of primers designed from GenBank database (Table 1). The PCR conditions (Doley *et al.*, 2013; El-Samadony *et al.*, 2016; Munch *et al.*, 2001) and specific primers for detection of duck viral enteritis, duck viral hepatitis and avian influenza virus are described in Table 1.

Table 1. Oligonucleotide primers used for identification of different duck diseases using PCR

Gene (Organism)	Primer name	Primer sequence (5' - 3')	Amplicon size (bp)	Reference
Primer for Haemoprotozoan diseases				
<i>haem2</i> (<i>Haemoproteus</i> spp)	HaemFL	ATGGTGTTTTAGATACTTACATT	358	Bensch <i>et al.</i> , 2000
	HaemR2L	CATTATCTGGATG AGATAATGGIGC		
<i>haem1</i> (<i>Leucocytozoon</i> spp, <i>Plasmodium</i> spp)	HaemNFI	CATATATTAAGAGAA ITATGGAG	587	Hallergen <i>et al.</i> , 2004
	HaemNR3	ATAGAA AGATAAGAATACCATTC		
Primers for bacterial disease				
<i>invA</i> (<i>S. typhimurium</i>)	ST11	GTGAAATTATCGCCACGTTTCGGGCAA	301	Rahn <i>et al.</i> , 1992
	ST15	TCATCGCACCGTCAAAGGAACC		
<i>Adk</i> (<i>E. coli</i>)	Adk-F	ATTCTGCTTGGCGCTCCGGG	583	Wirth <i>et al.</i> , 2006
	Adk-R	CCGTCAACTTTCGCGTATTT		
<i>nuc-A</i> (<i>S. aureus</i>)	nucA-F	GCGATTGATGGTGATACGGTT	447	Brakstad <i>et al.</i> , 1992
	nucA-R	GCCAAGCCTTGACGAACTAAAGC		
<i>flab</i> (<i>B. anserine</i>)	FP	ACATATTCAGATGCAGACAGAGGT	372	Barbour <i>et al.</i> , 1996
	RP	GCAATCATAGCCATTGCAGATTGT		
NDD (<i>R. anatipestifer</i>)	RA20F2	CAGCTTAACTGTAGAACTGC	662	Tsai <i>et al.</i> , 2005
	RA20R4	TCGAGATTTGCATCACTTCG		
PM 23-2 (<i>P. multocida</i>)	PM23F1	GGCTGGGAAGCC AAATCAAAG	1432	Miflin <i>et al.</i> , 2001
	PM23R2	CGAGGGACTACAATTAAGTGA A		
Primers for viral diseases				
DVE (Duck enteritis virus)	DVE-F	GAAGGCGGGTATGTAATGTA	450	Doley <i>et al.</i> , 2013
	DVE-R	CAAGGCTCTATTCGGTAATG		
DVH (Duck hepatitis virus)	DVH-F	AAGAAGGAGAAAATYAAGGAAGG	280	El-Samadony <i>et al.</i> , 2016
	DVH-R	TTGATGTCATAGCCCAASACAGC		
AIV (Avian influenza virus)	MMU19	AGAGCTCTTCTGATAGGTG	218	Munch <i>et al.</i> , 2001
	MMU39	CATCCCAGTGCTGGGAARGAYCCTAAGAA		
Primer for mycoplasmal disease				
<i>mgc2</i> (<i>M. gallisepticum</i>)	MG-14F	GAGCTAATCTGTAAAGTTGGTC	182	Garcia <i>et al.</i> , 2005
	MG-13R	GCTTCCTTGCGGTTAGCAAC		
DVE= duck viral enteritis, DVH= duck viral hepatitis, FP= forward primer, RP= reverse primer, NDD= new duck disease, AIV=avian influenza virus				

Data Analysis

After completion of the laboratory tests data was entered and stored electronically in Microsoft Office Access 2007. Choropleth maps were produced using ArcGIS version 10.1 (ESRI, Redlands, California) to show the distributions of specific diseases. Obtained data were analyzed by using the software STATA/IC-13. Positive and negative results of each disease were compared based on the categories of data. Prominent statistical analysis was done by descriptive analysis, univariate and multivariate logistic regression, χ^2 -test, t-test, etc. Significance of variance between and within the variables was measured when the P value was <0.05.

10.2. GIS mapping:

The location of each collected sample was recorded. Latitude and longitude of each sample area was represented through ArcGIS mapping (ArcGIS version 9.1).

10.3. Identification of the challenges of duck farming and development of strategic plans to prevent duck diseases in the haor areas:

A workshop entitled “Essential Vaccination and Biosecurity Measures for Preventing Duck Diseases in Haor Areas” was held in the Conference Room of CVASU, Chittagong on 18th September 2018 (Figure6). A total of 25 participants including practitioners, researchers and faculty members were present in the workshop and gave their suggestions in relation to developing a suitable vaccination schedule, deworming schedule, biosecurity measures for the prevention and control of duck diseases in the haor areas.



Figure 6: Participants in the workshop on “Essential Vaccination and Biosecurity Measures for Preventing Duck Diseases in Haor Areas”.

11. Results and discussion:

11.1. Database of infectious diseases of ducks and their risk factors in the selected areas of Hakaluki and Tanguar haor

Flock Comparison

Figure 7 shows the average flock sizes for the household and free-ranged ducks at Tanguar haor and Hakaluki haor. Larger flocks were located at Hakaluki haor in free-ranged interface. But, in case of household ducks average flock size was higher in Tanguar haor. The largest free-ranged flock of duck was found to be as big as 2500 in Tanguar haor

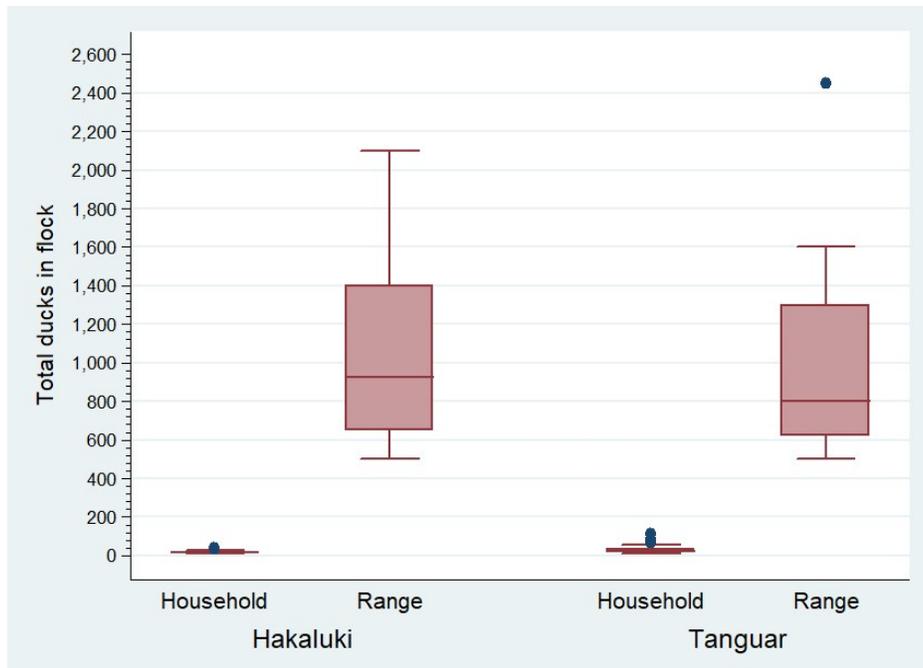


Figure7: Flock size comparison in Hakaluki and Tanguar haor areas

Social status of farm owner

The social status of the duck farmers is shown in Table 2. Out of the 600 duck farmers studied 55.5% were male and 49.5% of the farmers were found to have primary education followed by secondary education (27%) and illiterate (21%). Most of the farmers were found to rare indigenous ducks (57.33%) followed by Khaki Campbell (41.5%).

Table 2: Demography of duck farm owner (n=600) at Hakaluki and Tanguar haor areas

Variable	Category	Frequency	Percentage (%)
Gender	Female	267	44.5
	Male	333	55.5
Educational status	Illiterate	126	21
	Primary	297	49.50
	Secondary	162	27
	Others	15	2.5
Primary source of income	Crop production	80	13.33
	Livestock rearing	100	16.67
	Poultry rearing	420	70
Duck breed reared	Indigenous (Pati Duck)	344	57.33
	Khaki Campbell	249	41.5
	Muscovy	7	1.17

Health management, housing and feeding status of ducks in the duck farms:

The status of duck farms in relation to vaccination, deworming, scavenging places, housing materials, ventilation, etc. are mentioned in Table 3. Out of the 600 farmers interviewed, 59.5% were found not to vaccinate their ducks and 70.5% farmers did not use any anthelmintic for deworming their ducks. Around 37.83% farmers informed that their ducks scavenge in the ponds followed by paddy field (34.17%) and wet lands (24%).

Table 3: Status of duck farms in relation to disease control, housing and feeding of ducks in the haor areas

Variable	Category	Frequency	Percentage (%)
Sex of duck	Female	489	81.5
	Male	111	18.5
Vaccination status	Yes	243	40.5
	No	357	59.5
Deworming status	Yes	177	29.5
	No	423	70.5
Presence of sick duck	Yes	51	8.5
	No	549	91.5
Places of scavenging	Household premises	18	3
	Pond	227	37.83
	Rice paddy field	205	34.17
	Rivers	6	1
	Wetlands	144	24
Mixing with neighboring bird	Yes	152	25.33
	No	367	61.17
	Don't know	81	13.5
Duck housing type	Bamboo	228	38
	Metallic	18	3
	Muddy	294	49
	Wooden	60	10
Location of duck houses	Wetlands	210	35
	Within house	154	25.67
	Yard	236	39.33
Ventilation of house	No ventilation	168	28
	Open air	402	67
	Wall opening	30	5

Status of farm hygiene:

The status of farm hygiene practiced by the farmers in the haor areas are shown in Table 4. Out of the 600 farmers interviewed 92% (555) were found to clean their farms regularly and among them 95% (528) responded to clean their farms daily. Around 17.33% were found to practice hand and foot washing before handling ducks however, 44.5% of the farmers did not practice the said hand and foot washing measures.

Table 4. Status of duck farm hygiene practices at Hakaluki and Tanguar haor areas

Variables	Categories	Frequency	Percentage (%)
Cleaning of duck house	Regularly	555	92.5
	Uncleaning	45	7.5
Frequency of cleaning	Daily	528	95
	Once a week	24	4
	Once a month	3	1
Cleaning agent used	Ash	326	54.33
	Paper	1	1.67
	Sand	256	42.67
	Shipment of houses	17	28.33
Hand and foot washing before duck handling	Always	104	17.33
	Never	267	44.5
	Often	18	3
	Sometimes	211	35.17

Prevalence of different duck diseases at haor areas and their causal agents

The prevalence of infectious agents in the ducks during winter, summer and monsoon seasons in the haor areas are depicted in Table 5. Ducks from the study areas were found to harbor different species of parasites that were identified by direct microscopic examination of the parasitic eggs. The rates of occurrence of the parasites were 3.33%, 4.67%, 17.83%, 9.33%, 11.33%, 5.83% and 3.33% for *Ascaridia*, *Capillaria*, *Prosthogonimus*, *Amidostomum*, *Tetrameres* and *Hymenolepis* respectively. Microscopic examination of blood smears revealed the presence of three blood parasites named *Haemoproteus* spp, *Leucocytozoon* spp and *Plasmodium* spp (Figure 8) occurring all over the year at the rate of 13.17%, 8.33% and 7.67% respectively. However, the occurrence of the blood parasites could not be confirmed in PCR assay.

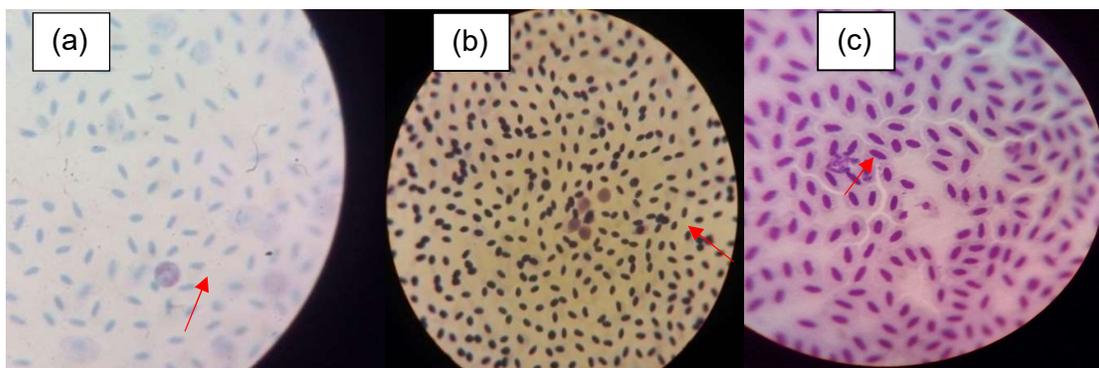


Figure 8: Identification of hemoprotozoan parasites under microscope (100X magnification). Red arrows on the figures show diagnostic stage (e.g. gametocyte) of *Haemoproteus*spp (a), *Leucocytozoon*spp (b) and *Plasmodium*spp (c)

Analyzing the culture properties (Figure 9) and PCR assay (Figure 10 -13) bacterial infectious agents were found to occur in the samples collected covering the said three seasons (total 600 samples), at an average rate of 55.33% for *Escherichia coli* (Figure 9b, 9c and 11), 20.50% for *Salmonella typhimurium* (Figure 9a and 10), 44.17% for *Staphylococcus aureus* (Figure 9d and 12) and 10% for *Riemerella anatipestifer* (Figure 13).

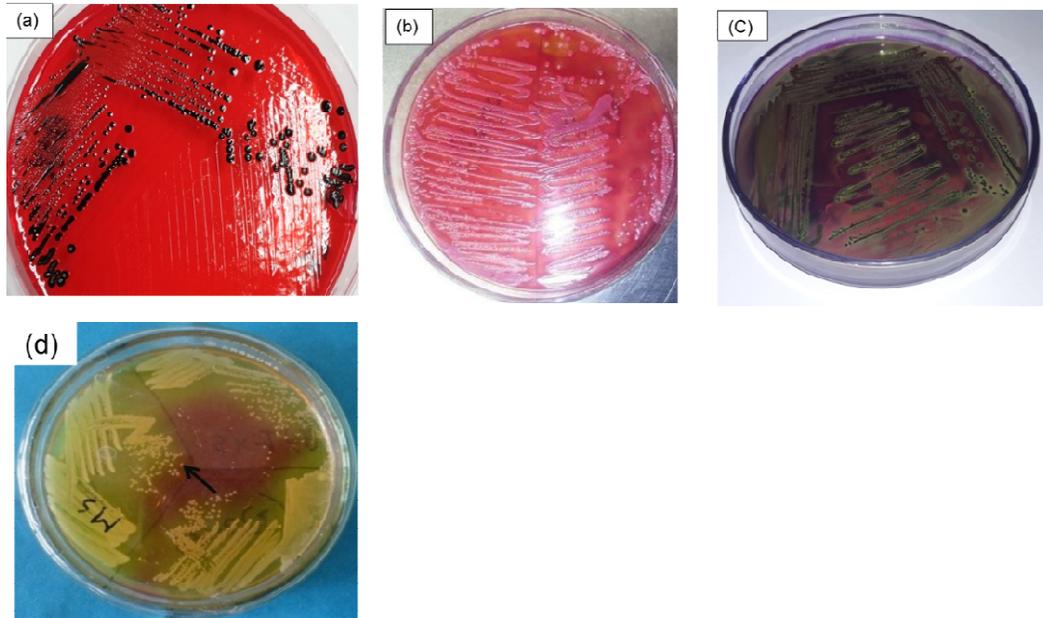


Figure 9: Identification of bacteria using different selective media

Figures show that typical colonies of *Salmonella spp* onto XLD agar medium (a), *E. coli* on MacConkey agar (Large pink colonies) and onto EMB agar (Greenish metallic sheen) and *Staphylococcus aureus* onto Mannitol Salt Agar (d).

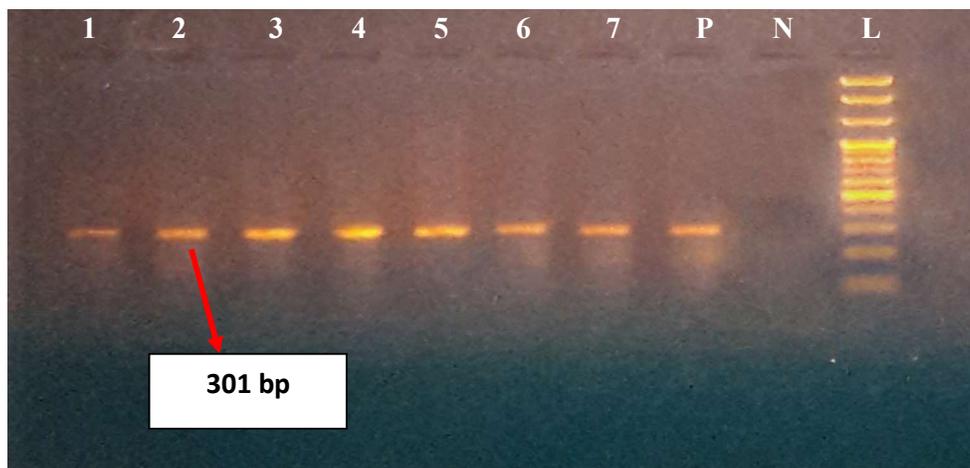


Figure 10: Electrophoresis of PCR products containing *Salmonella typhimurium* specific *invA* gene Lane L = 1 kb plus DNA ladder marker. Lane N = negative control, Lane P = positive control Lanes 1–7 = positive flocks (positive band at 301 bp).

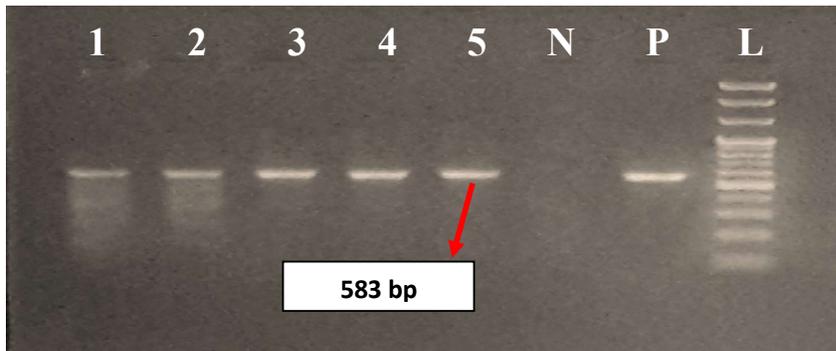


Figure 11: Electrophoresis of PCR products containing *Escherichia coli* specific *adk* gene
Lane L = 1 kb plus DNA ladder marker. Lane N = negative control, Lane P = positive control. Lanes 1–5 = positive flocks (positive band at 583 bp).

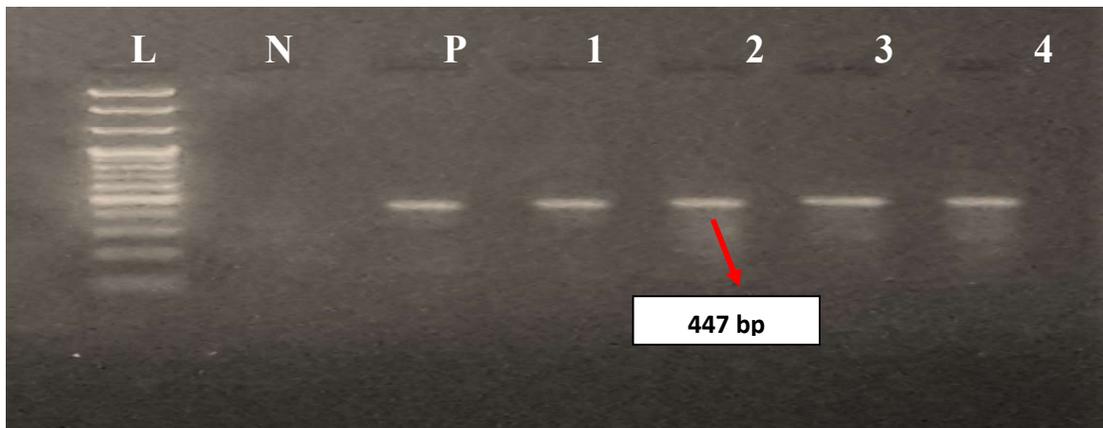


Figure 12: Electrophoresis of PCR products containing *nuc-A* gene for *Staphylococcus aureus*
Lane L = 1 kb plus DNA ladder marker. Lane N = negative control, Lane P = positive control. Lanes 1–4 = positive flocks (positive band at 447 bp).

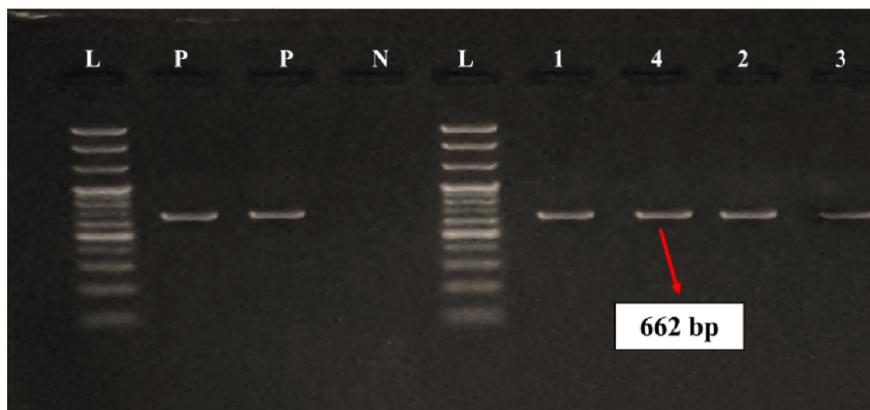


Figure 13: Electrophoresis of PCR products for *Riemerella anatipestifer*
Lane L = 1 kb plus DNA ladder marker. Lanes 1–4 = positive flocks (positive band at 662 bp). Lane P = positive control, Lane N = negative control.

Table 5: Prevalence of different duck diseases at Hakaluki and Tanguar haor areas

Sl.	Infectious agents	Season			Total N=600
		Winter (n=200)	Summer (n=200)	Monsoon (n=200)	
		% (positive)	% (positive)	% (positive)	% (positive)
Helminths infections					
1	<i>Ascaridiaspp</i>	4 (8)	7.5 (15)	2.5 (5)	4.72 (28)
2	<i>Capillariaspp</i>	19 (38)	15 (30)	19.5 (39)	17.83 (107)
3	<i>Prosthogonimus</i> spp	5.5 (11)	15 (30)	7.5 (15)	9.33 (56)
4	<i>Amidostomum</i> spp	8.5 (17)	10 (20)	15.5 (31)	11.33 (68)
5	<i>Tetrameresspp</i>	-	7.5 (15)	10 (20)	5.83 (35)
6	<i>Hymenolepisspp</i>	-	5 (10)	5 (10)	3.33 (20)
Haemoprotozoandiseases					
7	<i>Haemoproteusspp</i>	17 (34)	14 (28)	8.5 (17)	13.17 (79)
8	<i>leucocytozoon</i> spp	10.5 (21)	11.5 (23)	3 (6)	8.33 (50)
9	<i>Plasmodium</i> spp	5.5 (11)	6.5 (13)	11 (22)	7.67 (46)
Bacterial diseases					
10	<i>Escherichia coli</i>	59 (118)	53 (106)	54 (108)	55.33 (332)
11	<i>Salmonella typhimurium</i>	26 (52)	11.5 (23)	24 (48)	20.50 (123)
12	<i>Staphylococcus aureus</i>	12 (24)	66 (132)	54.5 (109)	44.17 (265)
13	<i>Riemerella anatipestifer</i>	16 (32)	0.5 (1)	13.5 (27)	10 (60)
14	<i>Pasturellamultocida</i>	-	-	-	-
15	<i>Borrelia anserine</i>	-	-	-	-
16	<i>Mycoplasma gallisepticum</i>	-	4.5 (27)	11.5 (69)	16 (96)
Viral diseases					
17	Duck enteritis virus	3 (6)	3 (6)	4 (8)	3.33 (20)
18	Duck hepatitis virus	12 (24)	-	-	4 (24)
19	Avian influenza virus	91 (178)	92.3 (184)	90.5 (181)	90.5 (543)

F= frequency, %= percentage, n=sample size, N= total population/total samples

Using PCR assay, this study confirmed for the first time in the haor areas of Bangladesh, the presence of *Riemerella anatipestifer*, the causal agent of the disease duck septicemia also known as goose 'flu, riemerellosis, new duck disease and polyserositis (Figure 13). The disease was found to occur throughout the year at a rate of 16% during winter, 0.5% during summer and 13.5% during monsoon (Table 5). It is assumed that due to being misdiagnosed with duck cholera that has the same clinical signs and pathological lesions the presence of the disease had ignored by the scientists and veterinarians in the country. *Pasturella multocida* and *Borrelia anserine* were not present in any of the samples collected during the said three seasons. *Mycoplasma gallisepticum* was found positive using PCR assay (Figure 14) in the summer and monsoon samples at the rate of 4.5% and 11.5% respectively.

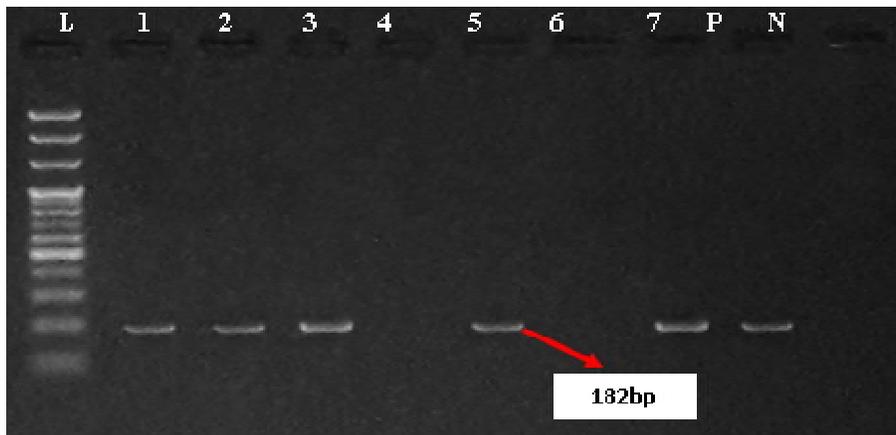


Figure 14: Electrophoresis of PCR products for *Mycoplasma gallisepticum*
 Lane L = 1 kb plus DNA ladder marker. Lanes 1–7 = *Mycoplasma gallisepticum* positive flocks (positive band at 182 bp). Lane P = positive control, Lane N = negative control.

In case of viral diseases, the presence of duck viral enteritis (DVE), duck viral hepatitis (DVH) and avian influenza (AI) were confirmed by polymerase chain reaction (PCR). Causal agents for DVE (Figure 15) and AI (Figure 17) were detected in all the samples (at the rate of 3.33% and 90.5% respectively) collected throughout the year covering winter, summer and monsoon seasons but, the causal agents for DVH were found only in the samples collected during winter season (Figure 16) at the rate of 12% (out of 200 winter samples 24 were positive). The reason behind the high rate of occurrence of AIV throughout the year (winter 91%, summer 92.3% and monsoon 90.5%) in the ducks might be due to that ducks act as natural reservoir for avian influenza virus. This news is considered to be very important for the policy makers and veterinarians, those who are working hard to control avian influenza in Bangladesh, for taking necessary actions immediately.

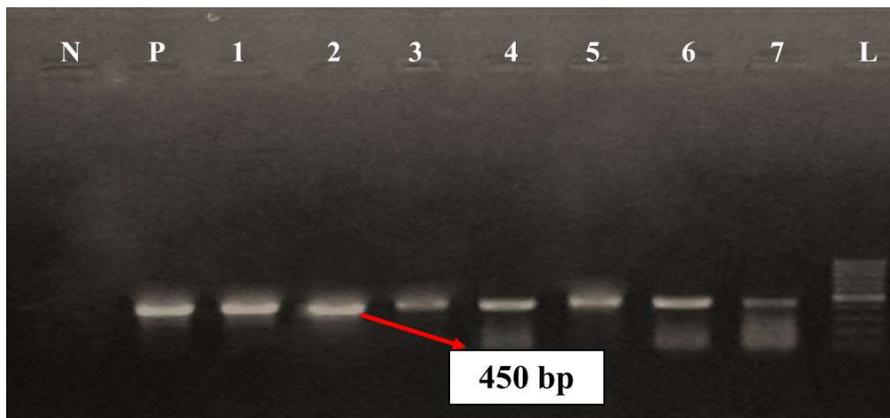


Figure 15: Electrophoresis of PCR products for duck viral enteritis virus
 Lane L = 100 bp to 1 kbp DNA ladder marker. Lanes 1–6 = duck viral enteritis virus positive flocks (positive; band at 450 bp). Lane P = positive control, Lane N = negative control.

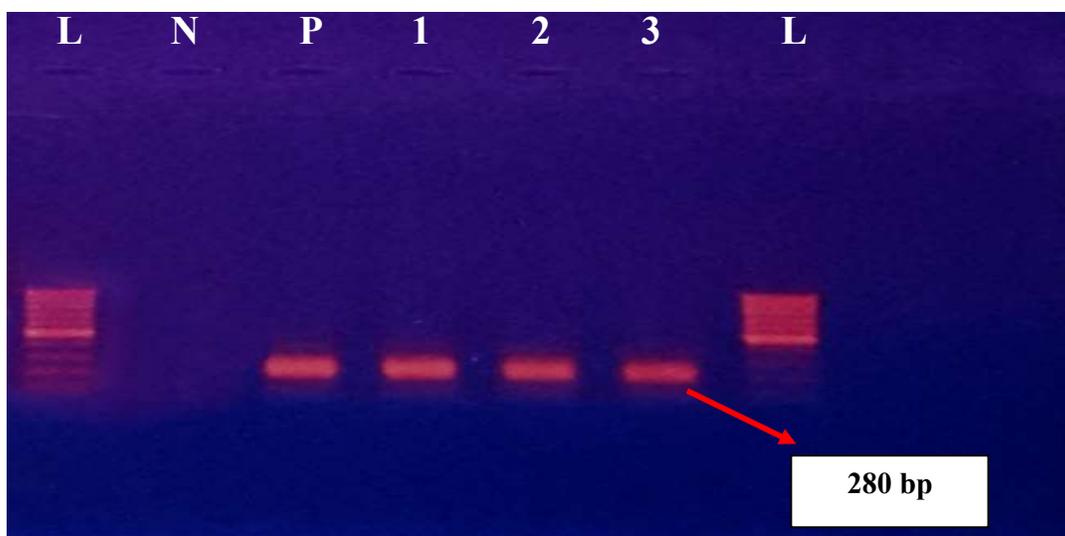


Figure 16: Electrophoresis of PCR products for duck viral hepatitis virus
 Lane L = 100 bp to 1 kbp DNA ladder marker. Lane P = positive control. Lane N = negative control. Lanes 1–3 = duck viral hepatitis virus positive flocks (positive band at 280 bp).

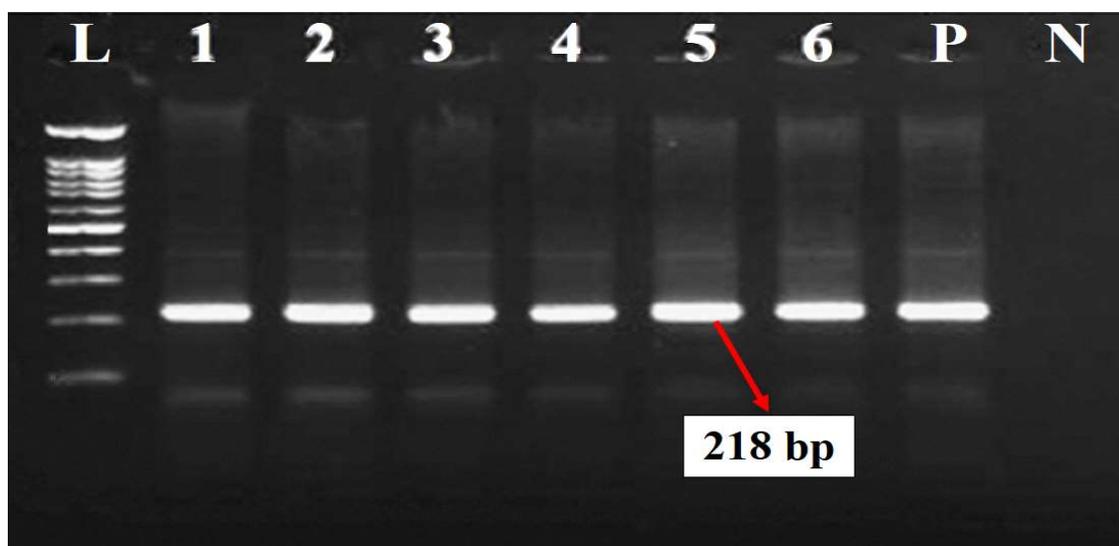


Figure 17: Electrophoresis of PCR products for avian influenza virus
 Lane L = 100 bp to 1.5 kbp DNA ladder marker. Lanes 1–6 = avian influenza A virus positive flocks (positive band at 218 bp). Lane P = avian influenza A virus positive control, Lane N = negative control.

Risk assessment by univariate logistic regression analysis

Univariate logistic regression analysis was done to evaluate the associations between several explanatory variables with some infectious agents. These results revealed that there are many risk factors as well as protective factors at 95% confidence interval. The results of univariate logistic regression analysis are given below (Table 6-8). Univariate logistic regression analysis revealed that 'pond' and 'wetlands' are statistically significant ($P=0.007$ and $P=0.01$ respectively) protective factors for *Haemoproteus* infection.

Table 6: Univariate logistic regression analysis to determine the protective factors for *Hemoproteus* spp

Explanatory variable	Category	N	Total positive (%)	OR	P-value
Places of scavenging	Household premise	18	7 (33)	1	
	Pond	227	19 (7)	0.15	0.007
	Rice paddy field	205	35 (15)	0.34	0.10
	River	6	6 (75)	5.99	0.17
	Wetlands	144	13 (7)	0.15	0.01
N= total population, OR= odds ratio, %= percentage					

In case of *Capillaria* spp, univariate logistic regression analysis revealed that 'flock size in between 520 to 4000' ($P=0.02$) and 'muddy type house' are the protective factors with statistical significance ($P= 0.001$).

Table 7: Univariate logistic regression analysis to determine the protective factors for *Capillaria* spp

Explanatory variable	Category	N	Total positive (%)	OR	P-value
Flock size	10-29	300	67 (22)	1	
	520-4000	300	40 (13)	0.54	0.02
Duck housing type	Bamboo	228	60 (26)	1	
	Metallic	18	0	0	0
	Muddy	294	33 (11)	0.34	0.001
	Wooden	60	14 (23)	0.84	0.68
Location of house	Wet land	210	11 (5)	1	
	Within house	155	26 (22)	5.4	0.001
	Yard	235	60 (25)	6.27	0.001
N= total population, OR= odds ratio, %= percentage					

In case of *Riemerella anatipestifer* infection, 'flock size in between 520 to 4000' was found as protective factors in univariate logistic regression analysis which was statistically significant ($P=0.05$). At the same time, farmers who washed hand and feet sometimes was found as risk factor ($P=0.009$) for *Riemerella anatipestifer* infection in ducks of haor areas.

Table 8: Univariate logistic regression analysis to determine the protective factors for *Riemerella anatipestifer*

Explanatory variable	Category	N	Total positive (%)	OR	P-value
Flock size	10-29	300	41 (10)	1	
	520-4000	300	19 (5)	0.44	0.05
Washing hands and feet	Never	267	15 (4)	1	
	Always	104	6 (4)	1.11	0.88
	Often	18	2 (8)	2.22	0.47
	Sometimes	211	37 (12)	3.34	0.009
N= total population, OR= odds ratio, %= percentage					

11.2. GIS mapping of bacterial and viral diseases

Distribution bacterial and viral diseases of ducks at Hakaluki and Tanguar haor are shown in the figures 18-21 by using GIS map.

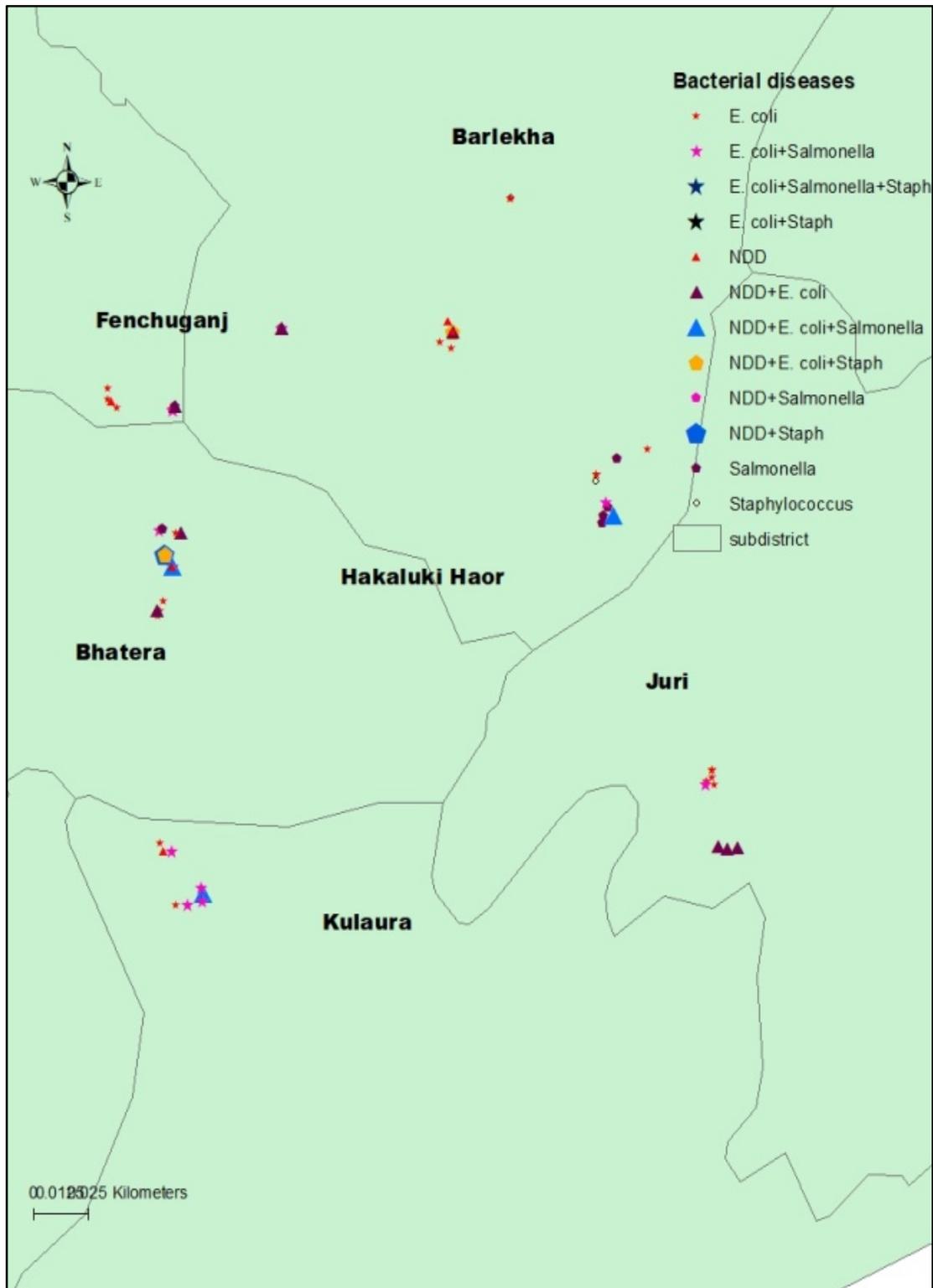


Figure 18: Distribution of bacterial diseases of ducks at Hakaluki haor

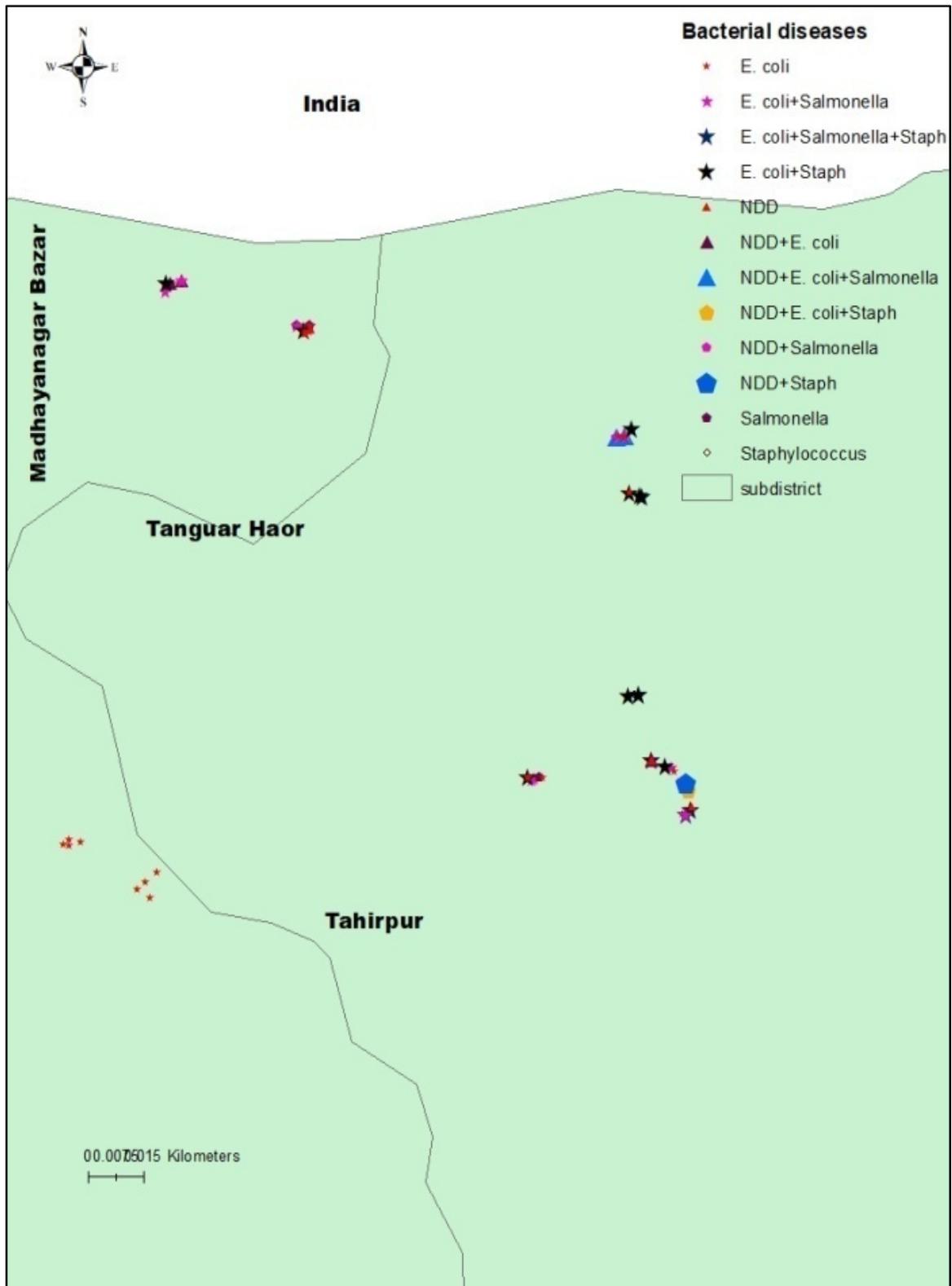


Figure 19: Distribution of bacterial diseases of ducks at Tanguar haor

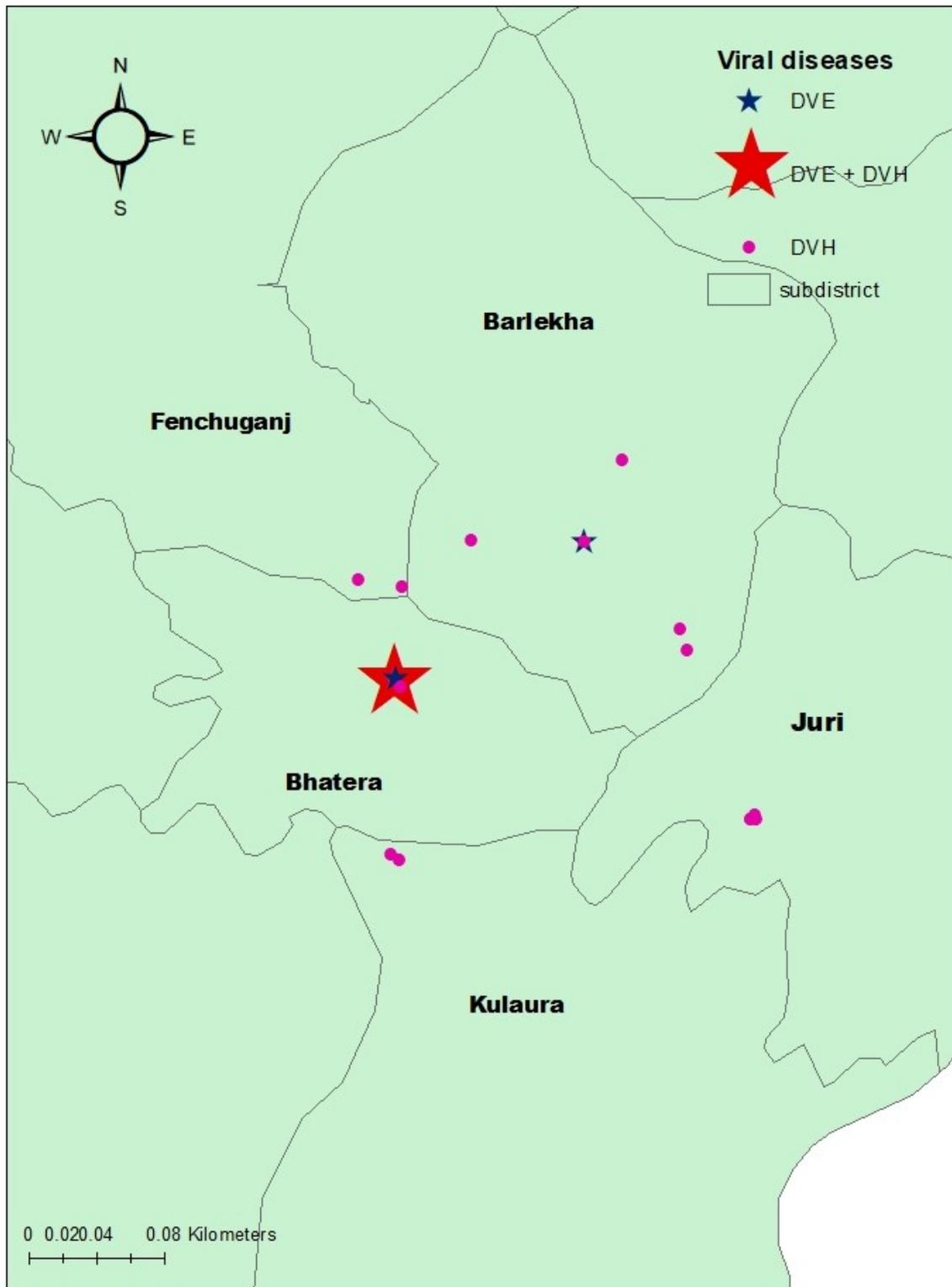


Figure 20: Distribution of viral diseases of ducks at Hakaluki haor

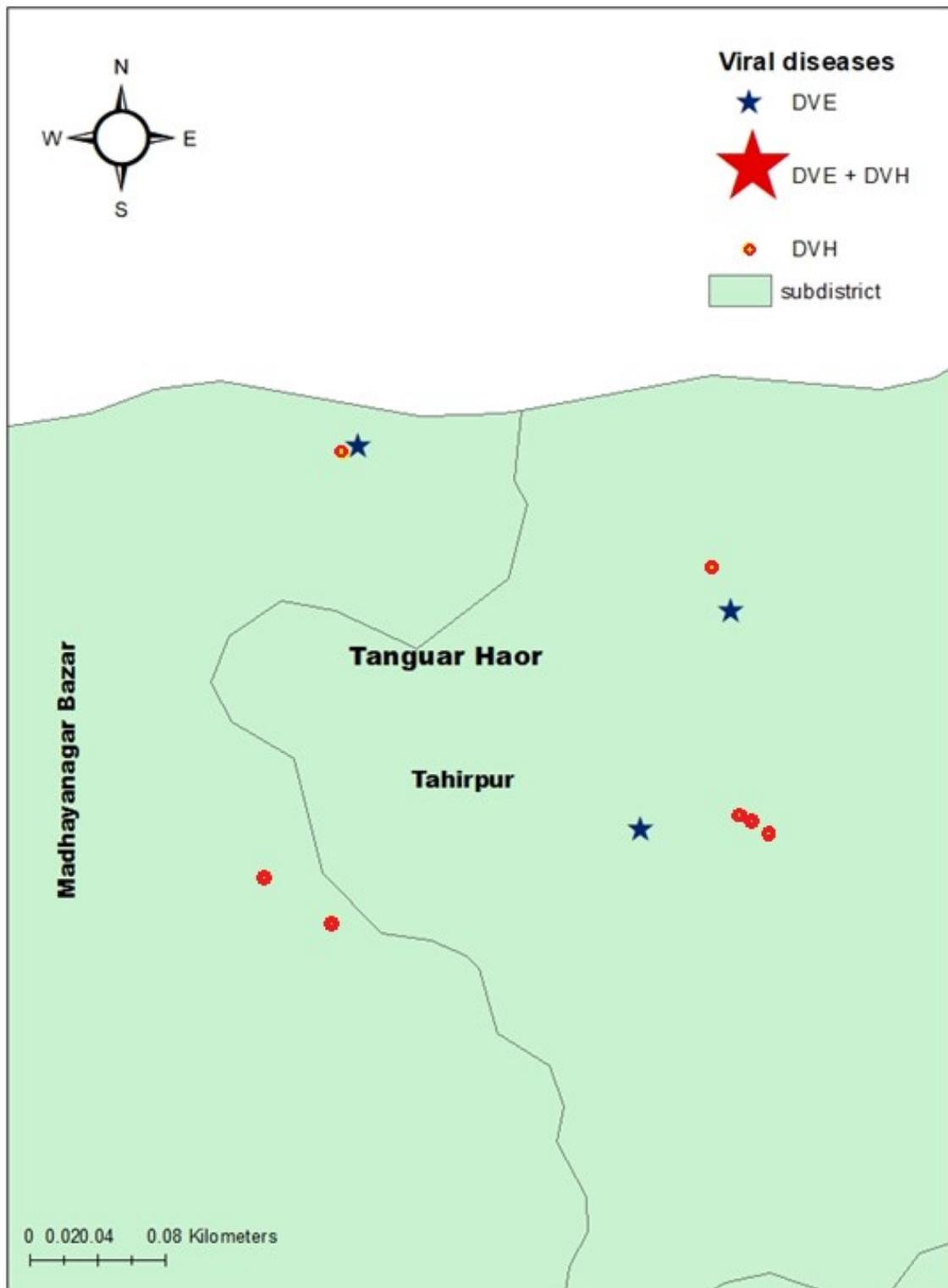


Figure 21: Distribution viral diseases of ducks at Tanguar haor

11.3. Challenges of duck farming and development of strategic plans for preventing duck diseases in the haor areas

Challenges of duck farming

Figure 22 shows the farmers' perception about the challenges of duck farming in the haor areas. Out of the 600 farmers interviewed most of the farmers (93.5%) identified the disease and mortality of ducks to be the number one problem of duck farming in haor areas followed by predators (34%), scarcity of feed and water (32.8%), flood (25%) and poisons in the paddy field (23.8%).

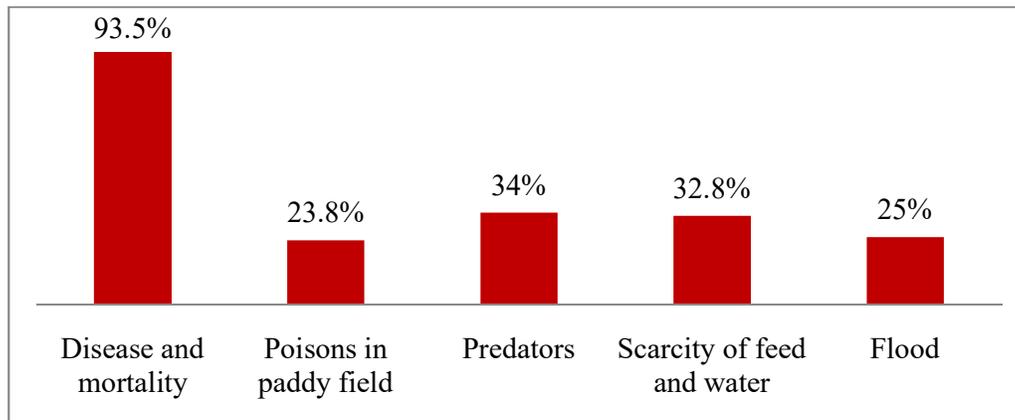


Figure 22: Challenges of duck farming (farmers' perception) in the haor areas

Strategic plans for prevention and control of duck diseases in the haor areas

On the basis of availability of the infectious agents in the haor areas some specific decisions were made from the workshop "Essential Vaccination and Biosecurity Measures for Preventing Duck Diseases in Haor Areas". The proposed suitable vaccination schedule for ducks in the haor areas is given in Table 8. Adoption of biosecurity measures, farmers' awareness building, regular deworming of ducks, ensuring veterinary medical services was recommended in the workshop for prevention and control of duck diseases in the haor areas.

Table 9. Suggested vaccination schedule for ducks at Hakaluki and Tanguar haor areas

Name of vaccines	Comments
Duck plague	Compulsory vaccination at the interval of 6 months.
Duck cholera	Circulating serotype should be identified first, then vaccine should be used accordingly
Duck viral hepatitis	Vaccination was suggested only for breeder ducks (intensively reared for hatching eggs)
Avian influenza	Investigation should be conducted towards clinical form of AI in ducks. Feasibilities of vaccination against AI should be piloted in commercial ducks
Salmonella infection	Vaccination was recommended
<i>Riemerella anatipestifer</i>	Vaccine should be developed

Recommendations of the workshop:

Biosecurity measures:

- a. Cost effective and practical use of disinfectants should be incorporated in duck housing. The participants emphasized regular use of desiccated Calcium Oxide (Choon) and bleaching powder.
- b. If possible, the ducks should be housed in elevated bamboo stages at night.

Farmers' awareness about the duck diseases:

- a. The participants advocated for different extension activities towards awareness building among farmers for important biosecurity measures in duck farming. Among different means distribution of poster and leaflets, application of mobile SMS gateway, farmers training and community engagement meetings were highlighted.
- b. Community engagement should be initiated through duck hatcheries and duckling and pullet distributors.

Deworming:

Deworming of the ducks was recommended to be performed at 4 months' interval.

Veterinary medical services:

- a. All duck farms should be registered in the government database.
- b. Department of livestock services should ensure regular vaccination of all flocks through a vaccination card.

12. Research highlight/findings:

- Among the different species of parasites *Ascaridia*, *Capillaria*, *Prosthogonimus*, *Amidostomum*, *Tetrameres* and *Hymenolepis* were found to occur at a rate of 3.33%, 4.67%, 17.83%, 9.33%, 11.33%, 5.83% and 3.33%, respectively.
- Among the blood parasites *Haemoproteus* spp, *Leucocytozoon* spp and *Plasmodium* spp were found to occur at the rate of 13.17%, 8.33% and 7.67%, respectively all over the year.
- Among the bacterial diseases prevalence of *Escherichia coli*, *Salmonella typhimurium* and *Staphylococcus aureus* infection was found to be 55.33%, 20.50% and 44.17%, respectively.
- The causal agent of duck septicemia, *Riemerella anatipestifer* was detected for the first time in the haor areas of Bangladesh. The disease was found to occur at an average rate of 10% all through the year.
- *Mycoplasma gallisepticum* was found positive in the summer and monsoon samples at the rate of 4.5% and 11.5%, respectively.
- Among the virus diseases the presence of duck viral enteritis (DVE), duck viral hepatitis (DVH) and avian influenza (AI) were confirmed by PCR. The ducks in the haor was found to harbor the causal agent of AI at a very high rate of 90.5% throughout the year.
- For the prevention and control of duck diseases in the haor areas regular vaccination against the prevailing viral and bacterial diseases along with regular deworming at the interval of 4 months, was suggested.
- Farmers' awareness development through training and community engagement meetings in relation to biosecurity measures and health management in duck farming was suggested for controlling duck diseases in the haor areas.

B. Implementation Position

1. Procurement:

Description of equipment and capital items	PP Target		Achievement		Remarks
	Phy (#)	Fin (Tk)	Phy (#)	Fin (Tk)	
(a) Office equipment	9	165,000	9	164,760	
(b) Lab & field equipment					
(c) Other capital items					

2. Establishment/renovation facilities:

Description of facilities	Newly established		Upgraded/refurbished		Remarks
	PP Target	Achievement	PP Target	Achievement	
N/A					

3. Training/study tour/ seminar/workshop/conference organized:r

Description	Number of participant			Duration	Remarks
	Male	Female	Total		
(a) Workshop	21	4	25	1 day	Workshop on Essential Vaccination and Biosecurity Measures for Preventing Duck Diseases in Haor Areas
(b) Seminar	82	18	100	1 day	Project completion seminar

C. Financial and physical progress

Items of expenditure/ activities	Total approved budget	Fund received	Actual expenditure	Balance/ unspent	Physical progress (%)	Reasons for deviation
A. Contractual staff salary	365220	287357	294142	71078	13.37	
B. Field research/lab expenses and supplies	990005	971887	1057780	-67775	48.08	
C. Operating expenses	220000	202529	203595	16405	9.25	
D. Vehicle hire and fuel, oil & maintenance	195000	183944	195000	0	8.86	
E. Training/ workshop/ seminar etc.	100000	93155	100000	0	4.55	
F. Publications and printing	95000	34965	89900	5100	4.08	
G. Miscellaneous	69775	25000	63600	6175	2.89	
H. Capital expenses	165000	164760	164760	240	7.49	
Total	2200000	1963597	2168777	31223	98.57	

D. Achievement of Sub-project by objectives:

Specific objectives of the sub-project	Major technical activities performed in respect of the set objectives	Output (i.e. product obtained, visible, measurable)	Outcome (short term effect of the research)
<p>i) Development of a database of the infectious diseases of ducks in the selected areas of Hakaluki and Tanguar haor and their risk factors</p>	<p>- Sample collection -Detection bacterial infection (Culture and isolation) -DNA/RNA extraction for molecular tests -ELISA test -PCR and RT-PCR -Identification of parasitic eggs by direct microscopic examination -Detection of blood protozoan disease by direct microscopic examination followed by PCR -Interview of farmers for data collection using a structured questionnaire and risk factor analysis</p>	<p>-600 samples (swab, blood, feces and blood smear slides) -Parasitic infestation with <i>Ascaridia</i>, <i>Capillaria</i>, <i>Prosthogonimus</i>, <i>Amidostomum</i>, <i>Tetrameres</i> and <i>Hymenolepis</i> were found to occur at a rate of 3.33%, 4.67%, 17.83%, 9.33%, 11.33%, 5.83% and 3.33%, respectively. -Blood parasites <i>Haemoproteus</i> spp, <i>Leucocytozoon</i> spp and <i>Plasmodium</i> spp were found to occur at the rate of 13.17%, 8.33% and 7.67%, respectively. -Bacterial disease prevalence of <i>Escherichia coli</i>, <i>Salmonella typhimurium</i> and <i>Staphylococcus aureus</i> infection was found to be 55.33%, 20.50% and 44.17%, respectively. -The causal agent of duck septicemia, <i>Riemerella anatipestifer</i> was detected for the first time in the haor areas of Bangladesh with the prevalence rate of 10%. -<i>Mycoplasma gallisepticum</i> was found positive in the summer and monsoon samples at the rate of 4.5% and 11.5%, respectively. -The presence of duck viral enteritis (DVE), duck viral hepatitis (DVH) and avian influenza (AI) were confirmed by PCR. The ducks in the haor was found to harbor the causal agent of AI at a very high rate of 90.5% throughout the year. -Farmers who washed hand and feet irregularly have been found as risk factor. Flock size in between 520 to 4000, muddy type house, pond and wetlands for scavenging have been found as protective factors.</p>	<p>PCR, RT-PCR protocols developed could be used for routine diagnosis of duck diseases. Information generated on the prevalence of duck diseases along with the challenges and risk factors identified will help policy makers to Develop suitable disease prevention and control plan for the duck farms in the haor areas. This in turn will help reducing the occurrence of diseases in the ducks resulting in decreasing mortality and increasing profitability of duck farms in the haor areas.</p>

Specific objectives of the sub-project	Major technical activities performed in respect of the set objectives	Output (i.e. product obtained, visible, measurable)	Outcome (short term effect of the research)
ii) Monitoring of duck diseases through active surveillance and GIS mapping	3 consecutive monitoring and GIS mapping	GIS maps of the bacterial and viral diseases of ducks in the Tanguar haor and Hakaluki haor have been generated	
iii) Identification of the challenges of duck farming and development of strategic plans for the prevention and control of duck diseases in haor areas.	Data analysis for identification of challenges of duck farming in the haor areas	Challenges of duck farming identified were disease and mortality (93.5%), attack of predators (34%), scarcity of feed and water (33%), etc.	
	Workshop with the experts to develop strategic plans for prevention and control of duck diseases	<ul style="list-style-type: none"> - A suitable vaccination schedule with the suggestion for vaccine development was proposed. - A suitable deworming schedule was advised. - Adoption of proper biosecurity measures, farmers' training, intensification of veterinary medical services, etc. was recommended. 	

E. Materials Development/Publication made under the Sub-project:

Publication	Number of publication		Remarks (e.g. paper title, name of journal, conference name, etc.)
	Under preparation	Completed and published	
Leaflet	1		Duck Rearing and Management (Appendix figure 2)
Thesis publication		2	<ol style="list-style-type: none"> 1. Occurrence of enteric parasites in household and free range ducks at Hakaluki and Tanguar haor of Sylhet division, Bangladesh 2. Prevalence of blood protozoa in duck at Hakaluki and Tanguar haor of Sylhet division in Bangladesh
Newspaper publication		1	Project outcomes and suggestion to the farmers in The Daily Samakal (Appendix figure 3)

F. Technology/Knowledge generation/Policy Support:

i. Generation of technology (Commodity & Non-commodity)

Molecular techniques of different diseases of ducks were established in Clinical Pathology Laboratory of CVASU. Primers and thermal profiles were adjusted and configured for future detection of such diseases named duck viral enteritis, duck viral hepatitis, avian influenza,

Escherichia coli, *Salmonella typhimurium*, *Staphylococcus aureus*, new duck disease, *Mycoplasma gallisepticum*, *Haemoproteus* spp, *Leucocytozoon* spp and *Plasmodium* spp.

ii. Generation of new knowledge that help in developing more technology in future

- a) The causal agent of duck septicemia, *Riemerella anatipestifer* was detected for the first time in the haor areas of Bangladesh. The organism could be used in the production of vaccine in future.
- b) The ducks in the haor was found to harbor the causal agent of AI at a very high rate of 90.5% throughout the year. This information is very important in planning control and eradication strategy for avian influenza in the poultry in Bangladesh.

iii. Technology transferred that help increased agricultural productivity and farmers' income

Prevention of diseases will promote duck production and duck egg production in haor areas. This project was designed to find out circulating fatal diseases in ducks and their distribution and temporal pattern. Vaccination model can be developed by piloting suggested vaccination schedule in haor areas of Bangladesh.

iv. Policy Support

Through current project, circulating infectious diseases were identified in ducks of Hakaluki and Tangar haor areas. A logical vaccination schedule was designed through a fruitful workshop with the participation of different stakeholders, practitioners and researchers. However, piloting of the suggested vaccination schedule along with the adoption of proper biosecurity measures and suggested farmers' training is needed to be conducted for suggesting any policy support.

G. Information regarding Desk and Field Monitoring

i) Seminar

Project completion seminar was held on 6th December 2018 in the conference room of CVASU. Around one hundred participants including veterinary practitioners and researchers attended the seminar.



Figure 23: Project completion seminar

i) Field Monitoring (time& No. of visit, Team visit and output):

Single time visit was performed by the technical team of BARC at 31st March 2018. The results of the laboratory test till the visit were presented. After the field monitoring the team suggested confirming the bacterial species by PCR.

H. Lesson Learned

- a. Investigation on clinical outbreaks and mortality by avian influenza in haor ducks should be carried out, not just virus identification as reservoir host.
- b. A pilot study should be carried out for up gradation of the local breeds by systematic genetic approach.
- c. Active surveillance should be carried out to monitor seasonal disease prevalence and risk factors.

I. Challenges

- 1. The farmers were reluctant to provide information and cooperation for sampling unless they were paid or subsidized by medicine/gifts.
- 2. In practical, the sampling period (1week) was insufficient because of the remoteness of the marsh land and unavailability of the ducks from 8 AM to 5 PM.

Signature of the Principal Investigator
Date

Seal

Counter signature of the Head of the
organization/authorized representative
Date

Seal

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Appendix

Questionnaire of Monitoring and surveillance of duck diseases in Hakaluki and Tanguar haor and development of suitable vaccination models and other preventive strategies

Part 1: Interviewee Details and Farm Locations

1. Farm identity number: H-255
2. Name: Fulea Begum
3. Age: 40
4. Gender: 1=Male 2=Female
5. Educational Status: 1=~~Illiterate~~ 2=Primary 3=Secondary
4=Higher education 5=Tertiary 6=Other (explain)
6. Length of time in duck farming: 9 years
7. Main source of income (rank sources from 1-5 with 1 being the primary source and 5 being the smallest source):
Poultry rearing:
Livestock rearing: (3).....
Crop production:
Daily labor:
GO/NGO job:
Other (explain):
8. Farm location:
House/Bari/Para: Bhambanpara
Village: Bhatna.....
Union: Bhatna.....
Upazilla (Sub-district): Banalkha.....
District: Moulvibazar.....
Latitude (N): 24.624562
Longitude (E): 91.973950
9. Farm Size: 11.....

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Part 2: Duck Flock

10. What breed, ages, sex, number, and vaccination and dewormed status of duck do you have today?

Breed	Age	Sex: M/F/DN	Number	Vaccination: Y/N If "Y" include approximate date	Dewormed: Y/N If "Y" include approximate date
<u>Manday</u>	<u>28</u>	<u>F</u>		<u>N</u>	<u>N</u>

11. Do you currently have any sick Ducks or other birds? If Yes, explain. Yes, Disease

Part #: Farm Management Practices

12. Where did your Ducks scavenge in the last 12 months?
1= Household premises 2= Rice Paddies
3=Rivers/Ponds/Wetlands 4= No scavenging 5= Other (explain)
13. Where are they currently scavenging? Household premises
14. Are the Ducks mixed with neighboring poultry or other poultry such as ducks or pigeons?
1=Yes 2=No 3= don't know
15. Duck housing type: 1= wooden 2= Bamboo
3= Muddy 4= concrete
5= Metallic 6= Other (explain)
16. Location of Duck Housing : 1= Yard 2= Within house
3= Other (explain)

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17. How is the Duck house ventilated? A

- 1= Wall openings 2=Open air 3=No ventilation
4= Other

18. Duck feed: 1= Rice bran 2= Rice polish 3=Paddy

- 4=Whole rice 5=Cooked rice 6=Food scraps
7=Commercial feed 8=Grain 9=Slaughter remnants
10= Only what they find

20. Cleaning Particles

	Poultry House	Feeder	Waterer	Nest Box
20.1 Do you clean and disinfect?	1=Clean only 2=Disinfect 3=Both 4=Neither 5=Other(specify)	1=Clean only 2=Disinfect 3=Both 4=Neither 5=Other(specify)	1=Clean only 2=Disinfect 3=Both 4=Neither 5=Other(specify)	1=Clean only 2=Disinfect 3=Both 4=Neither 5=Other(specify)
20.2 How frequently do you clean?	1=Daily 2=Once a Week 3=Twice a week 4=Once a month 5=Other(specify)			
20.3 How frequently do you disinfect?	1=Daily 2=Once a Week 3=Twice a week 4=Once a month 5=Other(specify)			
20.4 How and what do you use to clean ?	Band	H ₂ O	H ₂ O	H ₂ O
20.5 How and what do you use to disinfect?				

21. Do you or your family wash hands and feet before handling Ducks?

- 1=always 2= Often 3= Sometimes 4= Never

22. Do you use soap?

- 1=Yes 2=No

23. How often are litter / droppings cleaned/ removed?

- 4= Monthly 5= Never 6= Other (explain)

24. How is litter disposed?

- 1= Spread on Fields 2= Compost 3= Bury
4= Throw in Bushes 5= Throw in pond 6=Left in yard
7=Throw in bushes 8= Roadside 9= Other

25. How are dead birds disposed of?

1. Bury 2. Feed to other animals 3. Throw in pond
4. Throw in canal 5. Throw in bushes 6. Throw on roadside 7. Burn
8. Other

26. What is the biggest challenges to Duck farming?

- Predators
- Disease and mortality
- Food/Water availability
- Flood/Tide water/Heavy rainfall
- Toxins/ Poisoning
- Poultry house
- Other

Appendix figure 1: Questionnaire used to collect the data from farmers during sampling

ডিম সংরক্ষণ

ডিম সংরক্ষণের কয়েকটি পদ্ধতি আছে যেমন :

১. ঠাণ্ডা জায়গায় বাতের কাঁচা মেখেতে পাঠ করে মাটির হাঁড়ি বসাতে হবে। হাঁড়ির চারপাশে দাঁঠি কলসী ভিড়িয়ে দিলে হাঁড়ির মধ্যে বেশ ঠাণ্ডা হবে। হাঁড়ির মধ্যে ডিম বেখে মাটির সন্নিবিষ্ট করে রাখতে হবে। এ পদ্ধতিতে বেশ কিছুদিন ডিম ভালো থাকবে।
২. ১৪০° ফা. তাপমাত্রার গরম পানিতে ১৫ মিনিট ডিম পিঙ্ক করে বেশ কিছুদিন ডিম সংরক্ষণ করা যাবে।
৩. ডিমের উপরিভাগের অসংখ্য ছোট ছোট হিংস্র কদার দানা এক মিনিট খাঁটি সর্ষিয়ার তেলের মধ্যে ডুবিয়ে রাখতে হবে। এ পদ্ধতিতে ডিম বেশ কিছুদিন ভালো থাকবেও এ ডিমের সর্ষিয়ার তেলের ঝাঁক পাওয়া যাবে। তুলের পানিতে ডিম ডুবিয়ে বেখে ডিমের হিং বন্ধ করা যাবে।

একদিকে নিচের ধাপগুলো অনুসরণ করতে হবে :

- * একটি পাতে ১ লিটার পানি নিয়ে তার মধ্যে ১০০ গ্রাম লবণ গুলে গরম করতে হবে।
- * লবণ পানি ঠাণ্ডা হলে তার মধ্যে ২৫০ গ্রাম চুন ভালোভাবে ওলাতে হবে।
- * পাতটি একদিন বেখে দিলে তখন ডুবানি জমবে।
- * নিচের তরানি না নেড়ে উপরের পরিষ্কার গার্মি আলোয় পাতের তেলের দিতে হবে।
- * এই পদ্ধতির পানিতে ২০ মিনিট কাচের বাঁচার করে ডিম ডুবিয়ে রাখতে হবে।
- * বাঁচাসহ ডিম ছায়ায় শুকিয়ে নিতে হবে।

রোগবাহী ও চিকিৎসা

১. গোপের হাত থেকে বাঁচাতে হাঁসকে নিয়মিত গুণ্ড ও টিকা দিতে হবে।

টিকার নাম	১ম ডোজ	২য় ডোজ	সুটোর ডোজ
ডাক টিকা	২-৩০ দিন	৩০ দিন	প্রতি ৩ মাস অন্তর
ড্রাক কলেরা	৩০ দিন	৬৫ দিন	প্রতি ৬ মাস অন্তর

২. হাঁসকে সবসময় পরিষ্কার-পরিচ্ছন্ন আলো ও টাটকা খাবার দিতে হবে।
৩. নতুন হাঁস কিনলে কয়েকদিন আলস্য রাখতে হবে।
৪. অমুহু হাঁসকে চিকিৎসা করার জন্য ভালো/দুহু হাঁস বেছে আলাদা রাখতে হবে।
৫. খারেসাল, ফিনাইল, নাইকল ইত্যাদি দিয়ে ঘর ও জিনিসপত্র ধুতে হবে এবং মাঝে মাঝে গোপের গর্তিয়ে গোপনুক করতে হবে।

প্রশিক্ষণ

হাঁস পালন করতে হলে এ বিষয় অধিক কারো কাছ থেকে হাঁস পালনের বিস্তারিত জেনে নিতে হবে। হাঁস পালন সংক্রান্ত কোন তথ্য জানতে হলে স্থানীয় প্রসিদ্ধ অধিদপ্তরের ইউনিটের পর্যবেক্ষণ কর্মকর্তা অথবা উপজেলা প্রশাসন/অফিসে যোগাযোগ করা যেতে পারে। এছাড়া বাংলাদেশের প্রতিটি জেলা ও উপজেলা পর্যায়ে যুব উন্নয়ন অধিদপ্তরের প্রশিক্ষণ কেন্দ্র আছে। এদের প্রশিক্ষণ কেন্দ্র নির্ধারিত ফি এর বিনিময়ে পড়াগোপন বিষয়ক প্রশিক্ষণ দিয়ে থাকে। হাঁসের হাঁস এবং ডিম জনপ্রিয়। পরিবারিক বামাের অল্প মূলধন নিয়ে যেটি পরিপূর্ণ হাঁসের খামার স্থাপন ও পরিচালনার মাধ্যমে হাঁস পালন করলে পরিবারের খানা চাহিদা পূর্ণ করার পাশাপাশি বাজার আয়ের সুযোগ সৃষ্টি করা সম্ভব।

আরো বিস্তারিত জানতে যোগাযোগ করুন

১. প্যাথলজি ও প্যারাসাইটোলজি বিভাগ, চট্টগ্রাম ভেটেরিনারি ও এনিম্যাল সাইন্সেস বিশ্ববিদ্যালয়, খুলশী, চট্টগ্রাম-৪২২৫।
২. উপজেলা প্রশাসন/স্বাস্থ্য ও ভেটেরিনারি হাসপাতাল, তাহিরপুর, সুনামগঞ্জ, সিংগাই।
৩. উপজেলা প্রশাসন/স্বাস্থ্য ও ভেটেরিনারি হাসপাতাল, বকুলগঞ্জ, মৌলভীবাজার, সিংগাই।

Acknowledgement

“Monitoring and Surveillance of Duck Diseases in Hukaluki and Tangaur Haor and Development of Suitable Vaccination Models and other Preventive Strategies” শীর্ষক Competitive Research Grant (CRG) উপ-প্রকল্পের গবেষণাকর্ম ফলাফলের ভিত্তিতে প্রস্তুতকৃত লিফলেট

অর্থায়নে : প্রকল্প বাস্তবায়ন ইউনিট (পিআইউ-বিএআরসি), ন্যাশনাল এগ্রিকালচারাল টেকনোলজি প্রোগ্রাম-ফেজ II, গবেষণা (এনএটিপি-২) বাংলাদেশ কৃষি গবেষণা কাউন্সিল, ফার্মগেট, ঢাকা।

“হুকালুকি ও টাঙ্গুর হাওড়ের হাঁসের রোগ-বাহী পর্যবেক্ষণ ও নজরদারি এবং উপযুক্ত টিকাদানের মডেল ও অন্যান্য প্রতিরোধক কৌশলের উন্নয়ন”



প্যাথলজি ও প্যারাসাইটোলজি বিভাগ
চট্টগ্রাম ভেটেরিনারি ও এনিম্যাল সাইন্সেস বিশ্ববিদ্যালয়
খুলশী, চট্টগ্রাম-৪২২৫

হাঁস পালন

আমাদের দেশে গ্রামীণ এলাকায় অনেক বাড়িতেই হাঁস পালন করতে দেখা যায়। হাঁসের লালন-পালন খরচ কম এবং খামেনো বিহীন। হাঁসের মাংস সুস্বাদু এবং ডিম আকারে বড়। হাঁসের মাংস ও ডিমের বাজারের চাহিদা রয়েছে। হাঁসপালন করলে পরিবারের পুষ্টি চাহিদা পূরণ করার পাশাপাশি বাড়তি আয়ের সুযোগ সৃষ্টি হয়। বর্তমানে শহর, উপশহর এবং গ্রামেও বাণিজ্যিক ভিত্তিতে হাঁসের খামার গড়ে উঠেছে। বসত বাড়িতে হাঁস চাষ একটি সহজ ও লাভজনক কাজ। বাড়ির গৃহিণী এবং ছেলে-মেয়েরা অল্প মূলধন নিয়ে ছোট পরিসরে হাঁসের খামার স্থাপন ও পরিচালনা করতে পারে। পরিবারিক এই খামারে দেশি জাত ছাড়াও উন্নত জাতের হাঁসপালন করা যায়। এ অভিজ্ঞতা পরবর্তীতে বাণিজ্যিক খামার প্রতিষ্ঠার কাজে লাগানো যায়।

হাঁস পালনের উপকারিতা

১. হাঁসের ডিম ও মাংস গ্রামীণ আয়ের অন্যতম উৎস।
২. হাঁসের ডিম ও মাংস বিক্রি করে পরিবারে বাড়তি আয়ের সুযোগ সৃষ্টি করা সম্ভব।
৩. হাঁসের রোগ প্রতিরোধ ক্ষমতা বেশি এবং প্রতিকূল পরিবেশে মানিয়ে চলতে পারে।
৪. হাঁস ও মাছের চাষ একসাথে করা যায়।
৫. হাঁস হাওর-বিল, ডোবা-নালা ও ধানের জমিতে নিজেরা চরিয়ে খেতে পারে। এর ফলে হাঁসের প্রাকৃতিক খাবারের অভাব হয় না।
৬. হাঁসের বিষ্ঠা ভালোমানের জৈব সার।

বাজার সন্ধান

স্থানীয় বাজার ছাড়াও বড় বড় হাট-বাজারে হাঁস বিক্রি করা যায়। হাঁস জবাই করে পালক খসিয়ে বাজারে বিক্রি করা যায়। সম্পূর্ণ হাঁস বিক্রি করা তুলনামূলক সহজ। ডিম সরাসরি বাজারে বিক্রি করা যায়।

হাঁসের জাত নির্বাচন

দেশি হাঁস আকারে ছোট এবং ডিমও কম দেয়। এদের মধ্যে রয়েছে নাগেশ্বরী, মাটি হাঁস, সাদা হাঁস ও গাজ হাঁস। খালি ক্যাম্পবেল, ওরোডো, রোজিৎ হাঁস ও ইন্ডিয়ান রানার ডিমের জন্য ভালো। খালি ক্যাম্পবেল ও ওরোডো বহুরে ভালো পরিবেশে ২৫০-৩৫০টি পর্যন্ত ডিম দেয়। মাংসের জন্য পেকিন, মেসোডি ও সাদা পিঙ্ক ভালো। এরা বছরে ৮০-১২০টি ডিম দেয়। এদের মাংস বেশ সুস্বাদু। হাঁস পালনের ক্ষেত্রে যে কোন একটি জাত বাছাই করতে হবে। একই খামারে নানা জাতের হাঁস রাখা ঠিক হবে না।

ডিম হেঁটানো

হাঁসীর বয়স ৬ মাস হবার আগেই ডিম দিতে পারে। একটি সাধারণ আকারের হাঁসী ১০-১৫টি ডিম নিয়ে ৩০-৩৫ দিন তাপে বসতে পারে। হেঁটানোর জন্য ডিম উর্ধ্ব দিক সোটা বাতি দিয়ে দেখতে হবে। ডিমে ১৫ দিন তাপ হলে পরীক্ষা করতে হবে। অন্ধকারে আলো জ্বালিয়ে অথবা চর্কলাইটের মাধ্যমে ডিম পরীক্ষা করতে হবে। ডিম উর্ধ্ব দিক সোটা মত পোচনা ভাল দেখা যাবে। ডিম অন্দুর হলে তার কুসুম পরিষ্কার দেখা যাবে এবং কোন ধরনের জাল বা চিহ্ন দেখা যাবে না।

হাঁসের বাজা পালন

১. হাঁসের বাজাকে সবসময় উষ্ণ ও শুষ্ক স্থানে রাখতে হবে।
২. হাঁসের বাজাকে তুড়ি বা মাটির উপর রাখলে ১-২ সপ্তাহ খানের তুষ বা কাটা খড় দিতে হবে। কিছুটা বড় হলে মেঝের উপর বালি এবং ছাই দিতে হবে।
৩. নতুন বাজাকে প্রথমে মিহি খুঁদি, ভাত দিতে হবে। কয়েকদিন গুলে অন্যান্য খাদ্য মেঘন ছোট কেঁচো, শামুকের মাংস কাটা এবং সবজির পাতা ইত্যাদি দিতে হবে।
৪. বিড়াল, কুকুর, চিঁচা, বেড়ী ও কানের আক্রমণ থেকে রক্ষা করার জন্য বেড়া বা কাপি, পলো দিয়ে আটকিয়ে রাখতে হবে। ৬-৮ সপ্তাহ বয়স হলে আর আটকাতে হবে না।

আধুনিক পদ্ধতিতে হাঁসের বাজা পালন ও ব্যবস্থাপনা

আধুনিক পদ্ধতিতে হাঁসের বাজা পালনের ক্ষেত্রে উন্নতমানের বাজার জাত বাছাই করতে হবে।

ক্রমচার প্যান

১. হাঁসের ঘরকে কয়েক প্যানে ভাগ করে প্রতি প্যানে ১/২টি ক্রমচার স্থাপন করতে হবে।
২. ক্রমচারটির পরিধি প্রয়োজনে ছোট/বড় এবং উপরে/নিচে নামানো/উঠানো যায় সেই ব্যবস্থা করতে হবে।

তাপ ব্যবস্থা

সাধারণত বাতের মাধ্যমে অথবা গ্যাস দিয়ে ক্রমচার তাপ দিতে হবে। ক্রমচারের ভিতরের তাপ ব্যবস্থা

সপ্তাহ	তাপমাত্রা
১ম	৩৫° - ২৪° সে.
২য়	২৪° - ১৮° সে.
৩য়	১৮° - ১৭° সে.

বাতির চরার ব্যবস্থা থাকলে ২য় সপ্তাহ পরে শাভাবিক প্রাকৃতিক তাপমাত্রা বজায় রাখলেই হবে।

মেঝের ধরণ ও পরিমাপ

১. মেঝে অবশ্যই পাকা হতে হবে। তাছাড়া হাঁচ বা পাখর দিয়ে শক্ত করে সমতল রাখতে হবে।
২. বাজার দৌড়ানো ও চরার জন্য বয়সামুখী যে পরিমাপ জায়গা লাগবে তা নিচে উল্লেখ করা হলো :

বয়স (সপ্তাহ হিসেবে)	বর্গমিটার (প্রতিটির জন্য)
১-২	০.০৯
২-৩	০.১৩
৩-৪	০.১৯
৪-৫	০.২৩
৬-৮	০.৩৭

খাবার ও পানির ব্যবস্থা

১. খাবার ও পানির পাত্র প্রাচীর ও আলুমিনিয়াম দিয়ে তৈরি করতে হবে।
২. সুস্থ ও মানসম্মত খাবার যেমন- পিলেট অথবা গুড়া খাবার দিতে হবে। পীচা, বসি ও ছাত্বাখা খাবার দেয়া যাবে না।
৩. খাবার পাতের মধ্যে অথবা মেঝেতে ছিটপেস দিতে হবে।
৪. হাঁসের বাজাকে পানির পাত্রে পরিষ্কার পানি দিতে হবে।

হাঁসের ঘর

১. হাঁসের ঘর হবে সাধারণ এবং একদিক খোলা থাকতে হবে।
২. ১/২ মি. x ৫ মি. x ১ মি. মাপের একটি ঘরে ৪০-৫০ টি বড় হাঁস রাখা যাবে।
৩. হাঁসের ঘরটি শুষ্ক থাকবে এবং ঘরে তুষ, খড় অথবা বালি দিতে হবে।
৪. হাঁসের ঘর হেঁচির করার জন্য বাঁশ, বেতন, শন নল-বাগড়া, ইকড়া বা কালামাটি ইত্যাদি উপকরণ ব্যবহার করা যাবে।
৫. ঘরের উপরে অংশ সবসময় জালি দিয়ে রাখতে হবে।
৬. শাক-সবজির পাত, শসা, পুতুরের পানি ইত্যাদি যেন হাঁস নষ্ট করতে না পারে সেজন্য হাঁসকে বেড়া দিয়ে আটকে রাখতে হবে।
৭. হাঁসের ঘর ও বেড়া শক্ত হতে হবে।

হাঁসের খাবার

১. হাঁস সবকিছু খেতে পছন্দ করে। তবে হাঁসের বাজা ভালো খাবার জন্য চালের খুঁদি, গম ভাঙ্গা, ভুট্টা, ভট্টা, মাছের ওঁড়া, ঝেল, ইপিল-ইপিলের শুকনো পাতা এবং শামুক, বিম্বক, গুপলী, কেঁচো, কলারমোটা, সবজির ছাল, সবজির পাতা ইত্যাদি দেয়া যায়।
২. হাঁসকে সবসময় পরিষ্কার পানি দিতে হবে।
৩. পুরাতন টায়ার, বাঁশ এবং বাসনপত্রের মধ্যে গ্রামীণ পরিবেশে হাঁসকে খাবার দেয়া যাবে।

Appendix figure 2: Leaflet on duck rearing and management

সমকাল



হাওরাঞ্চলে হাঁস মারা যাওয়ার কারণ অনুসন্ধান করছেন চট্টগ্রাম ভেটেরিনারি বিশ্ববিদ্যালয়ের গবেষকরা

■ সমকাল

আট রোগে মরছে হাওরের হাঁস

■ আবু সাঈদ, চট্টগ্রাম

হাওর-বাওড়ে হাঁস পালন খুবই জনপ্রিয়। দেশের প্রায় সোয়া কোটি হাঁস পালন করা হয় এ অঞ্চলে। প্রকৃতি থেকে ৭০-৮০ শতাংশ খাবার সংগ্রহ করে বলে হাঁস লালন-পালনে খরচ হয় না খুব বেশি। ফলে হাওর অঞ্চলের অনেকেই হাঁস পালনের মাধ্যমে জীবিকা নির্বাহ করে থাকেন। অনেকে স্বাবলম্বীও হয়েছেন এর মাধ্যমে। কিন্তু হাঁস পালনে সবচেয়ে বড় ঝুঁকি হচ্ছে রোগব্যাধি। সাম্প্রতিক সময়ে রোগাক্রান্ত হয়ে মহামারী আকারে মরছে হাঁস। এর সঙ্গে মিহিয়ে যাচ্ছে খামারিদের স্বপ্ন। ২০১৮ সালে হাওরে অকাল বন্যায় ক্ষতিগ্রস্ত হয়েছেন তারা। কী কারণে এসব হাঁস মারা যায়, সে বিষয়ে জানা নেই তাদের। সম্প্রতি হাঁস মারা যাওয়ার কারণ নিয়ে গবেষণা করেছেন চট্টগ্রাম ভেটেরিনারি অ্যান্ড অ্যানিম্যাল সায়েন্স বিশ্ববিদ্যালয়ের (সিভাস) একদল গবেষক। গবেষণায় আটটি রোগের কথা জানা গেছে। সমন্বিত টিকা প্রদান ও সচেতনতার মাধ্যমে মারা যাওয়া রোধ করা যাবে বলে মনে করছেন গবেষকরা।

প্রান্তিক খামারিদের হাঁস পালনে উৎসাহিত করতে সিভাসুর অধ্যাপক ড. মো.

মাসুদজ্জামানের তত্ত্বাবধানে বাংলাদেশ কৃষি গবেষণা কাউন্সিলের অর্থায়নে 'হাকালুকি ও টাস্ফায়ার হাওর অঞ্চলে হাঁসের রোগ-ব্যাদি

গবেষণায় মিলেছে তথ্য

পর্যবেক্ষণ এবং সমন্বয়পযোগী রোগ প্রতিরক্ষা ও টিকাদান সময়সূচির উন্নয়ন' শীর্ষক এ গবেষণা করা হয়।

এ প্রসঙ্গে অধ্যাপক ড. মো. মাসুদজ্জামান বলেন, হাঁস মারা যাওয়ার প্রধান কারণ হচ্ছে 'ডাক প্লেগ'। এ ছাড়া কলেরার কারণেও অনেক হাঁস মারা যায়। সাধারণত হাঁস পচা ও নষ্ট খাবার খাওয়ার কারণে বিভিন্ন ভাইরাসজনিত রোগে আক্রান্ত হয়। নিয়মিত টিকা ও সচেতনতার মাধ্যমে হাঁসের মারা যাওয়া রোধ করা সম্ভব হবে। অনেকেই জানেন না হাঁসের টিকা আছে। তাই এ বিষয়ে মাঠ পর্যায়েও বিভিন্ন ধরনের সচেতনতা প্রয়োজন।

২০১৭ সালের নভেম্বর থেকে ২০১৮ সালের অক্টোবর পর্যন্ত এ গবেষণা পরিচালনা করা হয়।

এক বছরে তিনটি ধাপে হাকালুকি ও টাস্ফায়ার হাওর ও হাওর সংলগ্ন এলাকা থেকে ৬০০ হাঁসের নমুনা সংগ্রহ করে বিভিন্ন রোগের উপস্থিতি ও বিতৃতি শনাক্ত করা হয় সিভাসুর ক্লিনিক্যাল প্যাথলজি ল্যাবরেটরিতে। গবেষণায় হাওরের হাঁসের বিভিন্ন সংক্রামক প্রাণঘাতী রোগ-ব্যাদির চিত্র শনাক্ত করা হয়। যার মধ্যে উল্লেখযোগ্য হলো- ডাক প্লেগ, ডাক হেপাটাইটিস, কলিব্যাসিলোসিস, সালমোনেলোসিস, নিউ ডাক ডিজিসহ অনেক ভাইরাল, ব্যাক্টেরিয়া ও পরজীবীঘটিত রোগ। খামারিদের সঙ্গে সরাসরি সাক্ষাৎ ও তথ্য সংগ্রহের ভিত্তিতে হাঁস পালনের ক্ষয়ক্ষতির প্রধান কারণ হিসেবে চিহ্নিত করা হয়- সময় মতো টিকা না দেওয়া, হাঁস পরিচর্যা অজ্ঞতা ও অসচেতনতা।

জানা যায়, সারাদেশে রয়েছে প্রায় চার শতাধিক হাওর। দেশের উত্তর পূর্বাঞ্চলের সাতটি জেলার প্রায় দুই কোটি মানুষ প্রত্যক্ষ ও পরোক্ষভাবে হাওরের ওপর নির্ভরশীল। বোরো মৌসুমের ধান আবাদ শুরু হওয়ার পর থেকে হাওরাঞ্চলের সম্পদের তালিকায় ধান আর মাছ চাষের পর আসে হাঁস

■ পৃষ্ঠা ১৫ : কলাম ৩

Appendix figure 3: News of research results in one of the famous daily news papers