ANNUAL TECHNICAL REPORT
FISCAL YEAR 2079/080 (2022/23)

Government of Nepal
Ministry of Agriculture and Livestock Development
Department of Livestock Services
Central Veterinary Laboratory
Tripureshwor, Kathmandu

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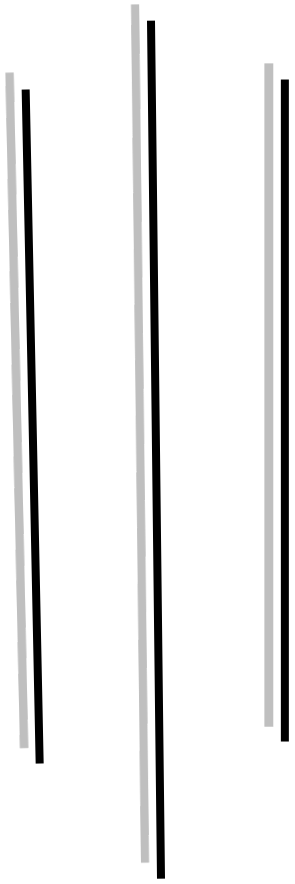

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FOREWORD

It is with great pleasure and enthusiasm that I present to you the Annual Bulletin of the Central Veterinary Laboratory for the fiscal year 2079/80 (2022/23). This bulletin highlights our activities, accomplishments, and overall progress in improving animal health.

As the referral veterinary diagnostic center in the country, CVL plays a crucial role in diagnosing animal diseases, conducting disease investigation, and ensuring the safety of animal and its products. we have established advanced molecular diagnostic techniques that enable routine diagnosis of diseases such as AI, ND, IBD, ASF, LSD, PPR. We also utilize multiplex PCR technology, alongside other methods like ELISA and Fluorescent Antibody Tests (FAT), to ensure accurate and efficient diagnostics for both livestock and poultry. In addition to routine testing in bacteriology, CVL actively participates in an Antimicrobial Resistance (AMR) surveillance program in collaboration with other veterinary laboratories and the National Avian investigation Disease Investigation Laboratory (NADIL) in Chitwan. The public health section of CVL has also initiated the surveillance of antibiotic residues in meat and milk employing ELISA methods to ensure food safety. Our dedicated team works tirelessly to provide accurate and reliable diagnostic services, conduct disease surveillance to improve animal health.

Nepal, as a member of the World Trade Organization (WTO), implements Sanitary and Phytosanitary (SPS) measures for which our laboratory actively works on conducting regular diagnostics and surveillance to uphold food safety and animal health standards in compliance with international guidelines.

We are focused on upgrading the Central Veterinary Laboratory (CVL) and the veterinary laboratories under its umbrella, while collaborating with the National Avian Disease Investigation Laboratory (NADIL) to provide reliable and prompt diagnostic services nationwide. We maintain strong coordination among veterinary laboratories, provincial veterinary offices, and local units to ensure quality sample flow for advanced diagnosis, reinforcing CVL's role as the National Reference Laboratory for animal health.

I sincerely thank the Food and Agriculture Organization (FAO), the Australian Center for Disease Preparedness (ACDP), and the International Atomic Energy Agency (IAEA) for collaboration and continuous support to upgrade and strengthen the quality of diagnostic services of the CVL. I am also grateful to FHI 360 (Fleming Fund) and FIND for their assistance in antimicrobial resistance and laboratory data management, as well as to Nepal Livestock Sector Innovation Project (NLSIP) for their technical support. Finally, I appreciate the hard work of all CVL staff in preparing this annual technical report.

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Dr. Barun Kumar Sharma
Chief Veterinary Officer

ABBREVIATIONS

AGID	Agar Gel Immuno diffusion
AI	Avian Influenza
ALC	Avian Leucosis Complex
AMR	Antimicrobial Resistance
ASF	African Swine Fever
AST	Antibiotic Sensitivity Test
BVD	Bovine Viral Diarrhoea
CCHF	Crimean Congo Haemorrhagic Fever
CCPP	Chronic Caprine Pleuoropneumonia
CCRD	Complicated Chronic Respiratory Disease
CFT	Complement Fixation Test
CMT	California Mastitis Test
CRD	Chronic Respiratory Disease
CSF	Classical Swine Fever
CVL	Central Veterinary Laboratory
CVRH	Central Veterinary Referrral Hospital
DLC	Differential Leucocyte Count
DLS	Department of Livestock Services
ELISA	Enzyme Linked Immunosorbent Assay
EPG	Egg per Gram
ESBL	Extended Spectrum Beta Lactamase
ESR	Erythrocyte Sedimentation Rate
FFCGN	Flehming Fund Country Grant Nepal
FMD	Foot and Mouth Disease
FY	Fiscal Year
HA	Haemagglutination
HI	Haemagglutination Inhibition
HPAI	Highly Pathogenic Avian Influenza
IB	Infectious Bronchitis
IBD	Infectious Bursal Disease
IBH	Inclusion Body Hepatitis
ILT	Infectious Laryngotrachitis
LIMS	Laboratory Information Management System
LSD	Lumpy Skin Disease
MD	Marek's Disease

NADIL	National Avian Disease Investigation Laboratory
NE	Necrotic Enteritis
NPHL	National Public Health Laboratory
PAT	Plate Agglutination Test
PCR	Polymerase Chain Reaction
PCV	Packed Cell Volume
PLT	Platelets
PPR	Peste des petits Ruminants
PRRS	Porcine Respiratory and Reproductive Syndrome
PT	Proficiency Testing
RBPT	Rose Bengal Plate Agglutination Test
RDT	Rapid Diagnostic Test
SOP	Standard Operating Procedure
SPS	sanitary and phyto sanitary
TB	Tuberculosis
TLC	Total Leucocyte Count
VHLSEC	Veterinary Hospital and Livestock Service Expert Center
WOAH	World Organization for Animal Health
WTO	World Trade Organization

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CHAPTER I: INTRODUCTION

1. Background

Central Veterinary Laboratory (CVL), referral laboratory in animal health sector in Nepal, focuses program with the objective of securing healthy national herd of livestock throughout the country by mitigating the occurrence of diseases of livestock and poultry. CVL and the laboratory functioning under it works on epidemic investigation as well as surveillance and investigation on various diseases. The direct benefit of the performance of various laboratories has been experienced in the field of veterinary medical care based on valid laboratory test results. To achieve these multidimensional activities, CVL works with a series of laboratory test procedures through its various laboratory sections; Pathology, Parasitology, Microbiology, Serology, Laboratory management and teaching laboratory and Molecular Biology with a considerable progress in the later.

CVL has Standard Operating Procedures (SOPs), test protocols and quality guideline manual. CVL is gradually practicing the biosafety/biosecurity measures so that Good Laboratory Practice is followed in all diagnostic laboratories. CVL is working to build knowledge and skills on biosafety and biosecurity to lab staff of all laboratories in animal health sector. CVL has already been participating in proficiency testing (PT) program with World Animal Health Organization (WOAH)–reference laboratories and has been adopting test verification system through international reference laboratories which will support to achieve different test for accreditation for international certification in future.

To provide diagnostic facilities throughout the country, CVL works through its five Veterinary Laboratories (VLs) located in different provinces of the nation; Koshi Province Veterinary Laboratory (Biratnagar), Madhesh Province Veterinary Laboratory (Janakpur), Gandaki Province Veterinary Laboratory (Pokhara), Karnali Province Veterinary Laboratory (Surkhet) and Sudurpashim Province Veterinary Laboratory (Dhangadhi). Diagnostic services are provided throughout the country in collaboration with provincial veterinary hospital, local veterinary unit. Specimens that cannot be processed in the aforementioned laboratories or needed to be further tested for confirmation are referred to the CVL. Provincial, veterinary hospital and veterinary units of local level also send samples to the CVL in coordination with Veterinary Laboratories for confirmatory diagnosis.

CVL has been actively involved in developing laboratory capacity of provincial and local level veterinary units. In this CVL is supporting with laboratory placement of newly recruited staff and also bridging through on site mentoring to build local capacity. This has also contributed to build functional network which has supported in sample collection and dispatch.

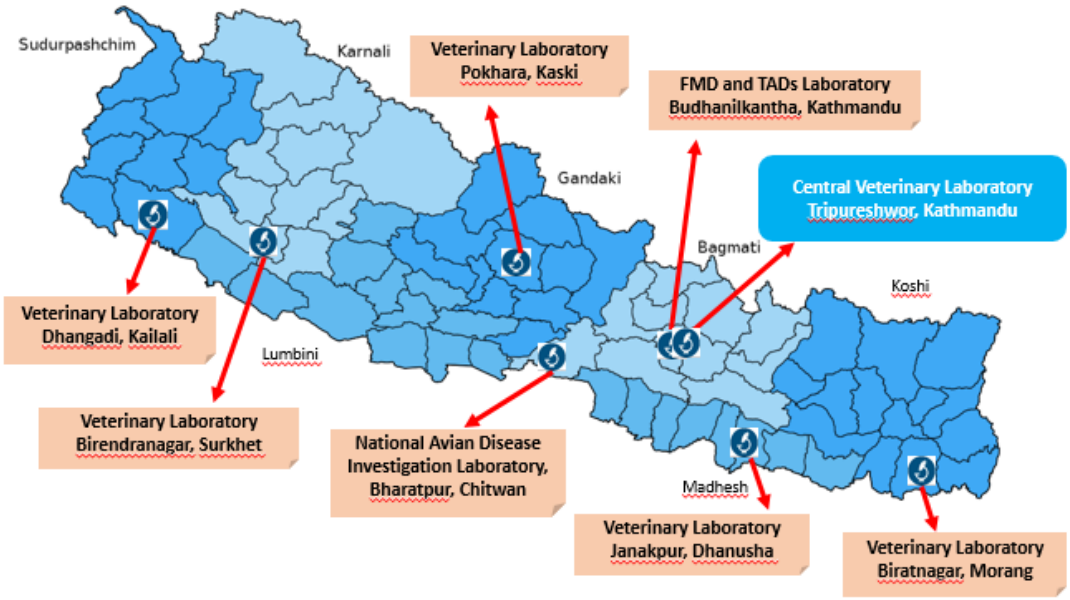


Figure 1: Map showing veterinary laboratories in Nepal.

2. Objectives

The role of veterinary laboratory system has become dynamic with advent of emerging diseases, public health, food safety issues, economic liberalization and trade globalization. Nepal joined as a World Trade Organization (WTO) member in 2004. Therefore, Nepal follows the guidelines provided by WOAH for the provision of Sanitary and Phytosanitary (SPS) agreement under WTO that seeks scientific procedures and evidence in the course of disease diagnosis as well as production chain. The roles of veterinary diagnostic laboratories are now therefore expanded and challenging in the new context. Moreover, CVL works with the following objectives in the country.

- Provide laboratory diagnostic services for animal health and veterinary public health.
- Conduct disease surveillance and monitoring programs.
- Ensure the quality and reliability of diagnostic tests and laboratory procedures by adhering to national and international standards and guidelines.
- Conduct epidemiological investigations to understand disease patterns, causes, and effects, and to control their spread and impact.
- Collaborate with national and international reference laboratories and institutions on laboratory diagnosis.
- Acquire, adopt, upgrade, and share diagnostic methods for livestock and poultry diseases to the laboratories working under CVL.
- Recommend on animal health policies and disease control programs to Department of Livestock Services (DLS) for effective disease control.
- Provide training in laboratory diagnostic for veterinarians and veterinary paraprofessionals.
- Provide technical support to strengthen veterinary laboratories at three tiers government.

To achieve the aforementioned objectives, there are a series of approved annual activities

carried out by different laboratory sections of the CVL and five VLs.

3. Organization structure:

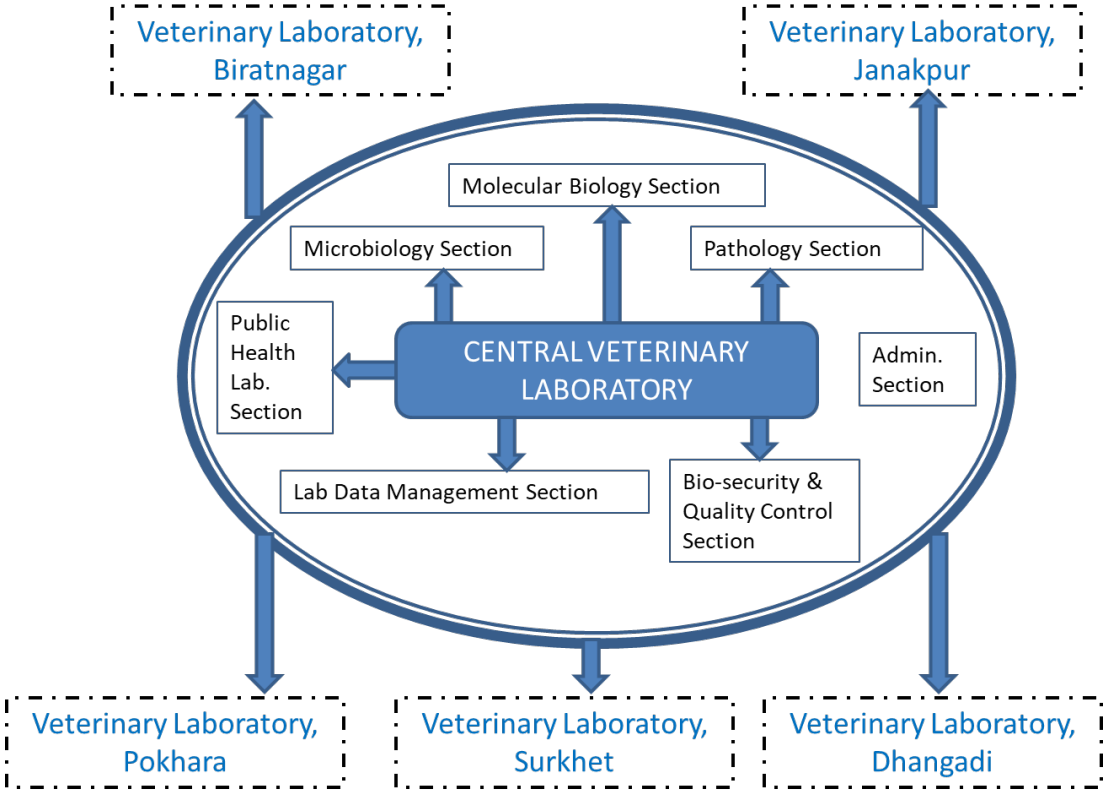


Figure 2: Organizational structure of CVL.

4. Human resources

There are total 39 permanent position in CVL, which is led by Chief Veterinary Officer. Under Chief Veterinary Officer, there are four Senior Veterinary Officer, tweleve Veterinary Officer, eight Animal Health Technicians and four Junior Animal Health Technicians. In the administration part, there are one accountant, one non-gazetted first class, two driver and six office assistants. The details of staffs at CVL is given in Table 1.

Table 1: List of Staffs working at CVL (F/Y 2079/80)

S.N	Name	Position	Total Number	Fulfilled
1	Dr Sharmila Chapagain	Chief Veterinary Officer	1	1
2	Dr. Pragya Koirala	Senior Veterinary Officer	4	4
3	Dr. Sita Rijal			
4	Dr. Ram Chandra Sapkota			
5	Dr. Tulsi Ram Gompo			
6	Mr. Purna Bahadur Budha	Veterinary Officer	12	12
7	Mr. Prakash Devkota			
8	Mr. Bal Bahadur Kunwar			
9	Mr. Tek Bahadur Aire			

10	Mr. Dhan Raj Rai			
11	Dr. Manju Maharjan			
12	Dr Chanda Shrestha			
13	Dr Nabaraj Shrestha			
14	Dr. Luna Gongal			
15	Dr Suraj Subedi			
16	Dr. Binita Tamag			
17	Dr. L B Sahi			
18	Ms. Sushma Pokherel		1	1
19	Mr. Shiva Raj Khadka		1	1
20	Mr. Hari Lal Kandel	Animal Health Technician	8	8
21	Ms. Rita Dahal			
22	Mr Krishna Mani Kafle			
20	Mr. Sudip Kafle			
21	Ms. Sunita Adhakari			
22	Ms. Mina Kumari Tamang			
23	Ms. Bimala K.C			
24	Mr Deepak Ghimire			
25	Mr. Rikesh Yadav	Asst. Animal Health Technician	4	1
26	Ms. Anita Shrestha			
27	Mr. Jeevan Rai	Computer Operator (Contract)	1	1
29	Mr. Kumar Nagarkoti	Driver	2	1
30	Mr. Dipesh Rana Magar			
31	Ms. Devaki Rimal	Office Assistant	6	6
32	Ms. Yam Kumari Rai			
33	Ms. Bhima Acharya			
34	Mr. Chandra Bahadur Rana Magar			
35	Ms. Laxmi Adhikari			
36	Mr. Surendra Shrestha			
Total			**39**	**35**

CHAPTER II: ANNUAL PROGRESS

5. Annual progress

The detail of the annual program and progress of CVL for FY 2079-80 is given in table below.

Table 2: Annual program and progress report of CVL in the Fiscal Year 2079-80

S.N.	Activities	Unit	Target	Progress
1	**Parasitology**			
1.1	EPG counts of parasites	Number	400	732
1.2	Skin scrapping examination	Number	60	81
1.3	Larvae culture of parasite	Number	40	40
2	**Pathology**			
2.1	Clinical hematological examination	Number	800	1904
2.2	Bio-Chemical examination	Number	500	1741
2.3	Post-Mortem Examination	Number	2400	4811
2.4	Histopathological examination	Number	40	40
2.5	Test Protocol for Histopathological examination	Times	1	1
2.6	Preparation of SOP for Post-Mortem of Small ruminants and Poultry	Times	1	1
3	**Microbiology**			
3.1	**Bacteriology**			
3.1.1	Isolation and Identification of Bacteria	Number	800	1254
3.1.2	Sample collection, Isolation and Identification of Fungus	Number	40	40
	Participation in EQA, NEQA and dispatch of sample to Veterinary Laboratories and report compilation	Times	4	11
3.2	**Virology**			
3.2.1	Rabies test	Number	60	122
3.2.2	Investigation of PPR outbreak	Times	4	3
3.2.3	PPR Diagnosis by Pen-side Test	Number	80	85
4	**Serology**			
4.1	Seromonitoring of PPR (National PPR Disease Control Program)	Number	6000	6000
4.2	Poultry sample collection and examination for salmonella and Mycoplasma	Times	8	8

S.N.	Activities	Unit	Target	Progress
4.3	Screening of cattle and buffalo disease in government and private farm)M. bovis, CCHF,CBPP, blood parasites	Number	400	436
4.4	Screening of goat disease in government and private farm (CCPP, Brucellosis etc.)	Number	400	824
4.5	Testing of Salmonella and Mycoplasma (ELISA)	Number	200	242
5	**Molecular unit**			
5.1	Molecular Diagnostic examination for Bird-Flu	Number	400	536
5.2	Molecular diagnosis (LSD, Anthrax etc.)	Number	40	449
5.3	Molecular diagnosis (Salmonella, PRRS, Erysepalas etc)	Number	40	364
5.4	Molecular diagnosis (PPR, CCPP, Pasturella etc)	Number	40	256
5.5	Diagnosis of Glanders in Horse and Mules	Number	40	108
5.6	Dispatch of sample to international reference laboratories	Times	2	1
6	**Disease surveillance and investigation**			
6.1	Emergency disease investigation team (EDIT) deploying	Times	12	12
6.2	Surveillance of poultry disease (AI, ILT etc.)	Number	300	300
6.3	Avian Influenza Surveillance	Number	800	800
6.4	AMR surveillance in Poultry	Number	80	101
7	**Zoonotic disease Investigation**			
7.1	Sample collection and examination for Brucella.	Number	160	288
7.2	Surveillance of zoonotic diseases (Toxoplasmosis, Leptospirosis etc.)	Number	300	963
8	**Staff development**			
8.1	Laboratory Technology Transfer	Times	6	6
8.2	Training on Laboratory Technique (Non-Officer Level 14 days)	Times	2	2
8.3	Publishing laboratory result in international journal	Times	1	
9	**Public Health Program**			

S.N.	Activities	Unit	Target	Progress
9.1	Veterinary Drug Residue Testing	Number	800	805
9.2	Hormones detection in milk and meat	Number	240	240
9	**Laboratory Monitoring**			
9.1	Monitoring and inspection of veterinary laboratories at provincial and local level	Times	12	12
9.2	Monitoring managemental aspects of Veterinary Laboratories	Times	8	8
10	**Publications**			
10.1	Publication of annual technical bulletin	Times	1	1
10.2	Standard Operating Procedure preparation for Biosecurity and Biosaftey and waste management and water treatment	Number	2	2
11	**Laboratory Management**			
11.1	Health examination of staffs	Person	39	1
11.2	LIMS Management	Times	12	12
11.3	Teaching lab management	Times	12	12
11.4	Management of Serum Bank	Times	12	12

CHAPTER III: LABORATORY SERVICES

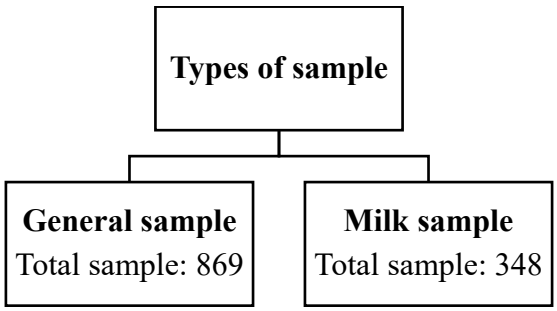
6. Microbiology section

Microbiology section at CVL has 2 units viz Bacteriology and mycology unit and Virology unit.

6.1 Bacteriology and Mycology Unit

6.1.1 Types of samples

In the fiscal year 2079/80, bacteriology unit received 1217 samples from various sources such as farms, Central Veterinary Referral Hospital, private clinics, veterinary laboratories, Veterinary Hospitals, and Livestock Service Expert Centers, and directly from the field- collected during the periodic sampling. The samples received were of two types:



The general samples include urine, blood, nasal swabs, ear swabs, skin scrapping, tissues from animals collected during postmortem examination, water and the animal feed.

6.1.2 Types of isolates

Out of the total 869 general samples and 348 milk samples, 758 (87.3%) and 313 (89.9%) showed bacterial growth respectively on culture. Among 758 general samples, 15 different types of isolates were identified by cultural characteristics, gram's staining, and biochemical tests. Also, few of the bacteria were identified via MALDI TOF, VITEK and BD PHOENIX. Similarly, of the 313 milk samples, 19 different types of bacteria were identified. The description of various bacteria isolated in general sample and milk sample are shown in tables below.

Table 3: Organism isolated from general samples.

S.N.	Organism	Nos.
1	*Bacillus spp.*	2
2	*Corynebacterium spp.*	1
3	*E coli*	646
4	*Enterobactor spp.*	3
5	*Enterococcus spp.*	1
6	*Klebseilla spp.*	11
7	*Listeria spp.*	1
8	*Mannheimia spp.*	1
9	*Mycoplasma spp.*	1

Table 4: Organism isolated from milk samples.

S.N.	Organism	Nos.
1	*Acinetobacter spp.*	2
2	*Aeromonas spp.*	1
3	*Bacillus spp.*	5
4	*Candida spp.*	6
5	*Citrobacter spp.*	2
6	*E coli*	128
7	*Enterobactor spp.*	1
8	*Enterococcus spp.*	7
9	*Haemophilus spp*	2

S.N.	Organism	Nos.
10	*Pasteurella multocida*	10
11	*Proteus spp.*	8
12	*Pseudomonas spp.*	3
13	*Salmonella spp.*	24
14	*Staphylococcus spp.*	44
15	*Streptococcus spp.*	2
Total		758
No Growth		111
Grand total		869

S.N.	Organism	Nos.
10	*Klebseilla spp.*	28
11	*Micrococcus spp.*	2
12	*Moraxella spp.*	1
13	*Pasteurella spp.*	1
14	*Proteus spp.*	6
15	*Pseudomonas spp.*	13
16	*Serratia spp.*	2
17	*Staphylococcus aureus*	88
18	*Streptococcus spp.*	16
19	*Yersinia spp.*	2
	Total	313
	No Growth	35
	Grand Total	348

Table 5: Month wise bacterial species isolated from general samples submitted to CVL.

Organism	**Shrawan**	**Bhadra**	**Ashoj**	**Kartik**	**Mangsir**	**Poush**	**Magh**	**Falgun**	**Chaitra**	**Baishakh**	**Jesth**	**Ashar**	**Total**
E.coli	47	46	31	38	79	49	59	58	58	70	70	41	646
Staphylococcus	7	10	10	2	3	2	3	2	1	2	1	1	44
Salmonella	4	2	1	0	2	1	1	0	1	3	3	6	24
Klebsiella	0	1	3	0	0	2	2	0	1	0	2	0	11
Proteus	1	2	0	2	1	1	0	0	0	0	0	1	8
Pasturella	1	2	1	2	2	1	0	0	0	0	0	1	10
Enterobacter	0	0	1	1	0	0	0	0	0	0	1	0	3
Pseudomonas	0	1	0	0	0	0	1	0	0	0	0	1	3
Bacillus	1	1	0	0	0	0	0	1	0	0	0	0	3
Streptococcus	0	0	0	0	2	0	0	0	0	0	0	0	2
Corynebacterium	0	0	1	0	0	0	0	0	0	0	0	0	1
Enterococcus	0	0	0	0	0	1	0	0	0	0	0	0	1
Manheimia	0	0	0	0	0	0	0	0	0	0	1	0	1
Listeria	0	0	0	0	0	0	0	0	0	0	0	0	1

Table 6: Month wise bacterial species isolated from milk samples submitted to CVL.

Organism	No. of isolates												
	Shrawan	**Bhadra**	**Ashoj**	**Kartik**	**Mangsir**	**Poush**	**Magh**	**Falgun**	**Chaitra**	**Baishakh**	**Jesth**	**Ashar**	**Total**
E.coli	27	12	3	1	1	7	10	8	5	15	14	19	128
Staphlococcus	14	16	12	6	6	4	10	3	9	2	2	4	88
Klebsiella	0	4	4	2	2	0	2	0	1	3	2	8	28
Streptococcus	0	1	0	2	2	1	6	0	0	0	1	3	16
Pseudomonas	1	1	0	0	0	1	0	1	2	1	1	5	13
Enterococcus	0	0	0	0	0	0	2	0	1	1	1	2	7
Candida	1	0	0	0	0	0	0	0	0	1	2	2	6
Proteus	0	0	0	0	0	0	0	0	0	1	3	2	6
Bacillus	0	0	0	0	0	0	0	1	0	1	0	3	5
Acinetobacter	0	1	1	0	0	0	0	0	0	0	0	0	2
Citrobacter	1	0	0	0	0	0	0	0	0	0	0	1	2
Haemophilus	0	0	1	0	0	0	0	0	0	0	0	1	2
Micrococcuus	0	2	0	0	0	0	0	0	0	0	0	0	2
Yersinia	0	0	0	0	2	0	0	0	0	0	0	0	2
Aeomonas	0	1	0	0	0	0	0	0	0	0	0	0	1
Enterobacter	0	0	0	0	0	0	1	0	0	0	0	0	1
Moraxella	0	0	0	0	0	0	0	0	0	1	0	0	1
Pasteurella	0	1	0	0	0	0	0	0	0	0	0	0	1
Serratia	0	0	0	0	0	1	0	1	0	0	0	0	2

6.1.3 Antimicrobial Susceptibility Testing (AST)

All the bacteria isolates were routinely tested for their antimicrobial susceptibility by disc diffusion method. The antibiotics against each isolate were matched according to Clinical and Laboratory Standard Institute (CLSI) guidelines, 2018.

6.1.3.1 General sample

All the bacterial isolates were tested for antimicrobial sensitivity for commercially available antimicrobials such as Gentamycin, Ciprofloxacin, Tetracycline, Amikacin, Amoxycillin, Ceftriaxone, Cefoxitin, Chloramphenicol, Sulphanomaide and Trimethroprim, Penicillin G, Imipenem etc. Details of the major five bacteria: *E. coli, Salmonella, Enterococcus, Moraxella* and *Klebsiella* is presented in the table and figure below.

Table 7: AST result on *Escherchia coli.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	7.02%	67.32%	25.66%
Ciprofloxacin	2.24%	92.44%	5.32%
Levofloxacin	2.82%	88.03%	9.15%
Amikacin	2.74%	87.67%	9.59%
Tetracyline	0.55%	93.39%	6.06%
Ceftriaxone	8.00%	26.40%	65.60%
Ampicillin	3.73%	83.68%	12.59%
Amikacin	10.64%	32.34%	57.02%
Chloramphenicol	1.99%	55.68%	42.33%
Cefoxitin	3.06%	25.51%	71.43%

Table 8: AST result on *Salmonella spp.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	0.00%	21.43%	78.57%
Ciprofloxacin	7.14%	50.00%	42.86%
Levofloxacin	0.00%	14.29%	85.71%
Tetracyline	5.88%	35.29%	58.82%
Ceftriaxone	11.11%	0.00%	88.89%
Cefoxitin	0.00%	11.11%	88.89%
Amikacin	0.00%	7.69%	92.31%
Chloramphenicol	0.00%	16.67%	83.33%

Table 9: AST result on *Klebsiella spp.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	0.00%	57.14%	42.86%
Ciprofloxacin	0.00%	80.00%	20.00%
Levofloxacin	0.00%	85.71%	14.29%
Tetracyline	12.50%	75.00%	12.50%
Ceftriaxone	66.67%	33.33%	0.00%
Chloramphenicol	0.00%	80.00%	20.00%
Amikacin	0.00%	75.00%	25.00%
Florfenicol	0.00%	50.00%	50.00%

6.1.3.2 Milk sample

Bacterial isolates from milk samples were subsequently subjected to antibiotic sensitivity test with different antibiotic panels. The detail description of major five bacteria; *E. coli, Staphylococcus, Streptococcus, Moraxella* and *Klebsiella* is depicted on table below.

Table 10: AST result on *Escherchia coli.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	6.59%	23.08%	70.33%
Ciprofloxacin	22.22%	37.04%	40.74%
Levofloxacin	6.82%	36.36%	56.82%
Ceftriaxone	14.71%	44.12%	41.18%
Cefoxitin	3.95%	42.11%	53.95%
Ampicillin	4.08%	75.51%	20.41%
Chloramphenicol	7.41%	29.63%	62.96%
Amoxcillin	0.00%	95.00%	5.00%

Table 11: AST result on *Staphylococcus spp.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	2.63%	22.37%	75.00%
Ciprofloxacin	16.67%	31.48%	51.85%
Levofloxacin	6.45%	32.26%	61.29%
Ampicillin	11.11%	33.33%	55.56%
Tetracyline	4.82%	31.33%	63.86%
Ceftriaxone	16.13%	35.48%	48.39%
Cefoxitin	0.00%	40.82%	59.18%
Ampicillin	7.41%	37.04%	55.56%
Chloramphenicol	0.00%	6.25%	93.75%

Table12: AST result on *Streptococcus spp.*

Drugs	Intermediate	Resistant	Sensitive
Gentamicin	0.00%	28.57%	71.43%
Ciprofloxacin	28.57%	42.86%	28.57%
Levofloxacin	16.67%	50.00%	33.33%
Tetracyline	10.00%	30.00%	0.60%
Ceftriaxone	50.00%	50.00%	0.00%
Cefoxitin	0.00%	75.00%	25.00%
Ampicillin	20.00%	80.00%	0.00%
Chloramphenicol	11.11%	55.56%	33.33%
Amoxycillin	0.00%	50.00%	50.00%

6.1.4 Antimicrobial Resistance Surveillance

Antimicrobial resistance (AMR) is one of the major global challenges for both human and animal health. CVL is the national reference laboratory for the AMR surveillance from Animal Health aspects. There has been collaboration with CVL and National Public Health Laboratory (NPHL) for the laboratory training. As a part of quality assurance, CVL takes part in External Quality Assessment (EQA), and quarterly receive unknown bacterial strains from NPHL for isolation, identification, and antimicrobial susceptibility test reporting. Along with this, CVL participates in EQA with Mahidol University, Thailand and Chulalongkorn university, Thailand in collaboration with Denmark technical university quarterly in year 2020 to 2021. CVL also provide EQA system among the veterinary laboratories within the country.

In recent years, the Fleming Fund Country Grant in Nepal (FFCGN) supported CVL in capacity building through human resources training, infrastructure development. FFCGN also assisted in the laboratory document preparation, such as surveillance plans and guidelines. As a part of this support, CVL has initiated a program for active AMR surveillance program in poultry fecal and cecal samples since 2020. The main targeted bacteria for the surveillance are E coli, salmonella, enterococcus, and campylobacter spp. CVL is leading the active surveillance activity, and three veterinary laboratories (VL) of Biratnagar, Pokhara, and the NADIL, Chitwan, are participating in the program during 2019-22. CVL is continuing the surveillance program through its annual program.

In addition, the microbiology unit of CVL is participating the WHO tricycle project on Extended Spectrum *E. coli* (ESBL)-Animal health component in collaboration with NPHL focusing the resistance patterns of ESBL *E. coli* in healthy chicken through One Health approach.

6.2 Virology unit

This unit is responsible for the diagnosis of viral diseases. Most of the samples are submitted from the postmortem unit of CVL followed by VL in different provinces, NADIL, Central Veterinary Referral Hospital, Veterinary Hospital and Livestock Services Expert Center (VHLSEC) and local level. Samples are also submitted by the quarantine check posts, private clinicians, farmers, and staffs of CVL during disease outbreak investigations. The unit has a facility for competitive ELISA, Fluorescent Antibody Test, Plate Agglutination Test and Lateral Flow Assay. Mainly, Lateral Flow Assay is used for the initial screening of Avian Influenza (AI), New Castle Disease (ND), Infectious Bursal Disease (IBD), Infectious Bronchitis (IB), PPR, ASF and Rabies. For the confirmative diagnosis of AI, ND, PPR and ASF. The samples are sent to the Molecular Section. Likewise, the confirmative diagnosis of rabies is done through Fluorescent Antibody Test (FAT).

6.2.1 Lateral Flow Assay

In the fiscal year 2079/80, a total of 992 samples were tested by lateral flow assay test kit method where 703 samples were positive for different diseases.

Table 13: Lateral flow assay diagnostic test record of different viral diseases.

Month	Samples	Lateral flow assay test result							Total
		AIV	NDV	IBDV	IBV	PPR	Rabies	ASF	
Shrawan	Total	8	0	1	0	0	9	0	18
	Positive	7	0	1	0	0	3	0	11
Bhadra	Total	52	2	14	2	3	10	2	85
	Positive	47	1	7	1	3	2	1	62
Ashoj	Total	33	0	3	0	3	3	1	43
	Positive	33	0	2	0	3	0	1	39
Kartik	Total	26	1	3	0	0	2	1	33
	Positive	22	0	1	0	0	2	1	26
Mangsir	Total	67	1	9	6	0	6	0	89
	Positive	62	1	5	2	0	1	0	71
Poush	Total	41	0	12	1	1	7	1	63
	Positive	37	0	4	1	0	2	1	45
Magh	Total	127	4	7	9	1	12	0	160
	Positive	105	0	1	1	1	1	0	109
Falgun	Total	78	8	7	4	0	10	0	107
	Positive	69	0	2	1	0	2	0	74
Chaitra	Total	73	6	14	3	1	17	1	115
	Positive	65	2	7	1	1	3	0	79
Baishakh	Total	64	8	21	6	1	8	0	108
	Positive	57	3	4	5	1	2	0	72
Jesth	Total	83	11	5	8	3	10	0	120
	Positive	64	6	1	3	2	4	0	80
Ashar	Total	31	2	5	2	0	11	0	51
	Positive	28	1	0	2	0	4	0	35
Grand Total	**Total**	**683**	**43**	**101**	**41**	**13**	**105**	**6**	**992**
	Positive	**596**	**14**	**35**	**17**	**11**	**26**	**4**	**703**

6.2.2 Rabies diagnosis

In the fiscal year 2079/80, 122 rabies suspected samples were tested out of which 104 (85.42%) were found to be positive. Most of the samples received were of canine species. Species wise details of rabies is presented in the Table.

Table 14: Species wise distribution of rabies cases

S.N.	Species	Positive	Negative
1.	Dog	67	16
2.	Cattle	14	0
3.	Buffalo	7	1
4.	Goat	14	1
5.	Jackle	2	0
	Total	104	18

Table 15: District wise distribution of rabies cases

S. N.	District	Total	Positive
1.	Bajhang	1	1
2.	Bhaktapur	2	2
3.	Chitwan	4	3
4.	Kailali	21	21
5.	Kanchanpur	19	17
6.	Kathmandu	55	41
7.	Kavrepalanchok	3	3
8.	Lalitpur	12	11
9.	Morang	2	2
10.	Sarlahi	1	1
11.	Sunsari	1	1
12.	Tanahun	1	1
	Total	**122**	**104 (85.24%)**

7. Pathology section

Pathology section consists of postmortem unit, histopathology, parasitology and clinical hematology and biochemistry unit. Samples are submitted either by VLs, NADIL, CVRH, VHLSEC and Local level or they are brought directly by the veterinary practitioners, livestock, and poultry farmers.

7.1 Postmortem unit

Post-mortem examination is the first step in disease diagnosis for morbid animals. Along with history, clinical findings, and epidemiological surveillance, it aids in making an accurate diagnosis, which is confirmed through various tests at CVL. In the fiscal year 2079/80 a total of 1,695 carcasses were brought to CVL for Post-mortem. The most common species were poultry with 1,581 cases, followed by dogs (48 cases), goats (48 cases), pigs (10 cases), rabbits (3 cases), wild animals (2 cases), wild birds (2 cases), and cats (1 case). The table below shows the species-wise distribution of pathological conditions.

Table 16: Reported Cases of Diseases by Animal Species at CVL

Species	Disease/Pathological condition	No. of reported cases
Cat	Non-specific disease	1
Dog	Gastritis	1
	Rabies	33
	Non-specific disease	14
Goat	Rabies	1
	Pneumonia	2
	PPR	2
	Monieza	1
	Abomastitis	1
	Coccidiosis	1
	Hemonchus infestation	10
	Enterotoxemia	13
	Posoning	1
	Bloat	1
	Non-specific disease	15
Pig	ASF	7
	Non-specific disease	3
Rabbit	Coccidiosis	1
	Non-specific disease	2
Wild animal	Injury	2
Wild birds	Non-specific disease	2
Poultry	Detail in table below	1581
Total		1695

A total of 1,581 poultry were brought to CVL for post-mortem examinations. The highest number of cases was reported in the month of Jestha with 223 cases, and the least number of cases were reported in the month of Asoj with 76 cases.

Table 17: Month-wise distribution of poultry disease diagnosed at CVL

Disease	Sharwan	Bhadra	Asoj	Kartik	Mangsir	Poush	Magh	Falgun	Chaitra	Baisakh	Jestha	Ashad	Total
Colibacillosis	28	19	31	29	32	21	55	43	45	75	53	38	469
Mycotoxicosis	14	15	20	16	14	9	11	5	13	17	34	19	187
CRD	8	7	7	3	11	10	24	7	22	29	38	13	179
IBD	9	5	3	6	10	17	11	4	4	14	23	11	117
Ascites	2	2	1	4	11	18	17	20	9	13	8	8	113
Avian Influenza	5	4	0	7	4	3	20	9	9	3	14	1	79
CCRD	2	7	2	5	12	5	6	5	11	0	6	5	66
IB	1	2	0	3	6	1	7	3	0	2	5	1	31
Coccidiosis	1	1	0	4	1	3	3	3	1	3	2	4	26
Gout	2	0	4	1	2	0	4	1	1	3	1	6	25
Salmonellosis	5	3	3	2	5	4							22
ND	0	0	0	0	0	0	3	8	3	1	6	0	21
Urolithiasis	0	0	0	0	0	0	3	3	1	7	3	1	18
Necrotic Enteritis	3	0	0	0	0	0	2	2	5	2	2	1	17
Immune Suppression	0	0	0	0	0	0	0	0	2	4	4	4	14
Nephritis	0	0	0	0	0	0	0	1	3	1	0	8	13
Ascariasis	3	0	0	2	1	1	2	1	1	0	1	1	13
Fatty Liver Syndrome	0	0	0	0	0	0	3	0	2	4	0	1	10
Fowl Typhoid	0	0	0	0	0	0	3	1	3	0	0	2	9
Marek's Disease	1	1	1	3	3	0	0	0	0	0	0	0	9
ALC	0	0	1	1	2	0	0	0	0	0	0	3	7
IBH	0	0	0	0	0	0	0	0	0	0	5	1	6
Stress	0	0	1	1	0	0	0	0	2	1	0	1	6
Pullorum Disease	0	0	0	0	0	0	0	0	0	2	3	0	5
Flu A Positive	0	0	0	0	0	0	0	0	0	0	1	3	4
Omphalitis	0	0	0	0	0	0	0	1	1	0	0	1	3
Pneumonia	0	0	0	0	0	0	0	0	0	0	1	1	2
Histooniasis	1	0	0	0	0	0	0	1	0	0	0	0	2
Fowl Pox	0	0	0	0	0	0	0	0	0	0	1	0	1
Aspergillosis	0	0	0	0	0	0	0	0	0	0	1	0	1
Rabies	0	0	0	0	0	0	0	0	0	0	0	1	1
Non specific	13	12	2	2	19	14	7	8	6	6	11	5	105
Total	**98**	**78**	**76**	**89**	**133**	**106**	**181**	**126**	**144**	**187**	**223**	**140**	**1581**

The table shows the distribution of various diseases among different types of poultry brought to CVL for post-mortem examinations. Broilers are the most affected, especially with Colibacillosis and CRD. Layers and local poultry have significant numbers of cases of Mycotoxicosis. Ducks and other less common poultry types have fewer cases overall.

Table 18: Distribution of poultry disease in different bird types based on Post mortem

Diseases	Broiler	Layers	Broiler Parent	Layer parent	Kuroiler	Kalij	Local	Giriraj	Duck	Pigeon	Others	Total
Colibacillosis	393	38	4	1	16	2	8	5	0	0	1	468
Mycotoxicosis	91	35	4	5	12	9	17	8	2	3	1	187
CRD	148	5	1	1	5	1	13	2	0	1	1	178
IBD	105	3	0	0	0	1	6	2	0	0	0	117
Ascites	107	0	2	0	1	1	0	2	0	0	0	113
AI	17	32	3	0	11	0	13	1	0	0	2	79
CCRD	55	4	0	0	1	0	3	3	0	0	0	66
Nephritis	26	10	0	0	2	3	8	4	0	3	0	56
IBH	17	8	0	0	2	0	6	1	0	1	0	35
IB	23	4	0	0	0	0	3	1	0	0	0	31
Coccidiosis	9	1	1	0	0	1	13	1	0	0	0	26
Salmonellosis	8	7	0	0	1	2	4	0	0	0	0	22
NE	11	0	1	1	2	0	1	0	0	1	0	17
Fowl Typhoid	1	2	0	0	2	1	2	1	0	0	0	9
MD	3	0	0	0	3	0	2	0	0	0	1	9
Asciariasis	0	1	0	0	2	0	3	0	0	3	0	9
ALC	0	0	0	1	1	0	2	1	0	0	0	5
Non-specific Disease	64	12	5	0	3	4	8	2	4	3	0	105
Others	37	3	1	0	4	1	3	0	0	0	0	49
Total	1115	165	22	9	68	26	115	34	6	15	6	1581

*Other diseases includes fatty liver disease, urolithiasis, nephritis, enteritis, stress, immunesupression, injury, sudden death, starvation etc.

Note: CRD-Chronic Respiratory Disease, CCRD-Complex Chronic Respiratory Disease, IBD-Infectious Bursal Disease, ND-Newcastle Disease, AI-Avian Influenza, IB-Infectious Bronchitis, MD-Mareks Disease, ALC-Avian Leukosis Complex

7.2 Hematology and biochemistry unit

A total of 1928 blood samples were tested in FY 2079/80. Blood samples were tested for hematological parameters and blood parasites.

7.2.1 Blood parasites

Out of 1928 blood samples, 305 samples (15.82%) were found to be positive for at least one blood parasites. *Babesia spp.* (76.39%) was the predominant hemoprotozoan found especially in the cattle followed by *Anaplasma* (14.75%). *Erlichia* (5.65%) was only observed in canine samples.

Table 19: Blood parasites diagnosed in different animal species in 2079/80

Species	CBC Test	Hemoprotozoan				
		Anaplasma	Babesia	Trypanosoma	Ehrlichia	Theliaria
Cattle	993	39	198	0	0	1
Buffalo	20	1	7	0	0	0
Goat	168	2	11	0	0	0
Sheep	49	0	0	0	0	0
Dog	606	3	17	6	20	0
pig	16	0	0	0	0	0
Cat	6	0	0	0	0	0
Yak	70	0	0	0	0	0
Total	1928	45	233	6	20	1

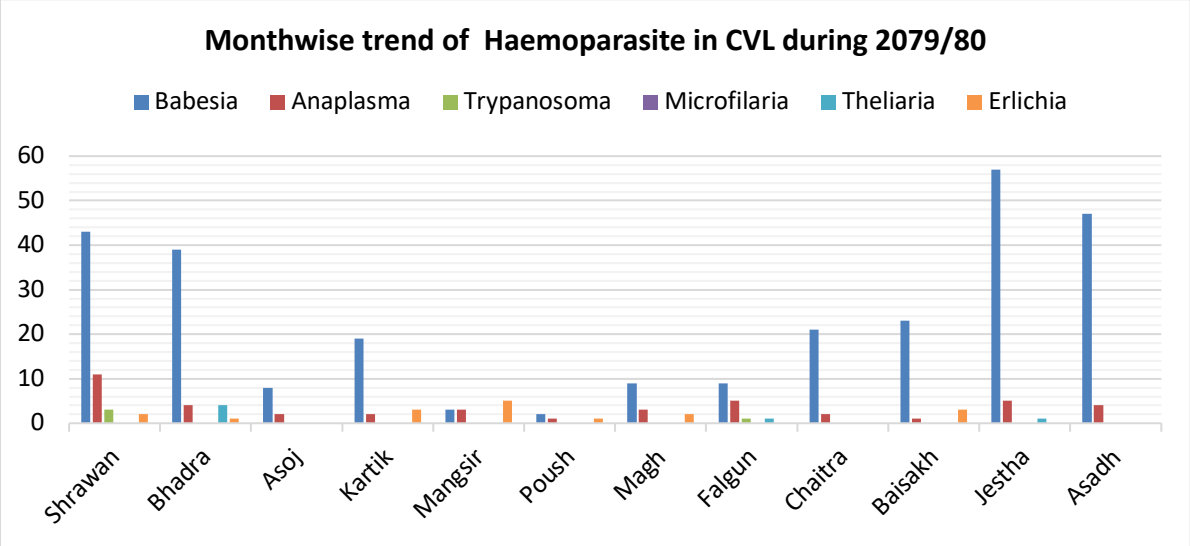


Figure 3: Monthwise trend of Haemoparasite in CVL during 2079/80

Table 20: Biochemical Analysis of blood samples collected from Government farms in 2079/80

Parameters	Cattle	Yak	Total
Calcium	88	66	154
Protein	60	66	126
Magnesium	88	66	154
Alkaline Phosphatase	60	66	126
Phosphorus	88	66	154
SGPT	60	66	126
SGOT	60	66	126
Total	504	462	966

7.3 Parasitology Unit

The parasitology unit is involved in routine examination of different types of internal and external parasites of animals and birds. For the investigation and diagnosis of parasites, fecal samples, and skin scrapings samples were collected and examined by adopting standard veterinary laboratory protocols.

7.3.1 Fecal examination

For a routine examination or diagnosis of internal parasites, direct smear method, sedimentation method and the floatation methods were used. During FY 2079/80 a total 1480 faecal samples were tested, out of which 465 samples (31.48 %) were found to be positive for at least one parasite.

Table 21: Fecal examination

Month	District	Species	Number of Sample	Positive	Parasite
Shrawan	Kathmandu	Cow	5	3	Liver fluke
	Kathmandu	Cow	5	1	Paramphistomum
	Kathmandu	Cow	5	2	Ascaris
	Dhading	Cow	2	1	Liver fluke
	Dhading	Cow	2	1	Paramphistomum
	Dhading	Cow	3	1	Ascaris
	Bhaktapur	Goat	3	1	Paramphistomum
	Bhaktapur	Goat	3	3	Ascaris
	Dhading	Goat	64	7	Paramphistomum
	Dhading	Goat	64	5	Liver fluke
	Dhading	Goat	64	8	Trichuris
	Dhading	Goat	138	5	Strongyles
	Dhading	Goat	64	6	Moneiza
	Dhading	Goat	64	6	Coccidia
Bhadra	Kavre	Goat	4	1	Strongyles
	Kathmandu	Cow	4	2	Liver fluke
	Kathmandu	Cow	4	1	Paramphistomum
	Nawalparasi	Goat	52	7	Trichuris
	Nawalparasi	Goat	52	11	Strongyles
	Nawalparasi	Goat	52	4	Capillaria
	Nawalparasi	Goat	52	9	Moneiza
	Nawalparasi	Goat	52	9	Strongyles
Ashwin	Nawalparasi	Goat	62	5	Liver fluke
	Nawalparasi	Goat	31	5	Trichuris
	Nawalparasi	Goat	31	1	Strongyles
	Nawalparasi	Goat	31	4	Capillaria
	Nawalparasi	Goat	31	4	Moneiza
	Chitwan	Elephant	2	2	Strongyles

	Chitwan	Elephant	2	2	Paramphistomum
	Kathmandu	Cow	4	1	Strongyles
	Kathmandu	Cow	4	3	Liver fluke
Kartik	Lalitpur	Goat	11	3	Strongyles
	Kathmandu	Cow	2	1	Liver fluke
	Dolakha	Chauri	5	5	Strongyles
	Dolakha	Chauri	4	4	Trichuris
	Rasuwa	Chauri	1	1	Moneiza+Trichstrongylus
	Rasuwa	Chauri	2	2	Moneiza
	Rasuwa	Chauri	1	1	Trichuris
	Rasuwa	Chauri	1	1	Strongyles
	Rasuwa	Chauri	1	1	Moneiza+Strongyles
	Rasuwa	Chauri	1	1	Strongyles
Mangsir	Dolkha	Cow	37	2	Paramphistomum
	Dolkha	Cow	37	1	Strongyles
Poush	Dolkha	Cow	30	6	Mixed
	Kailali	Sheep	40	20	Mixed
	Rasuwa	Yak	21	5	Mixed
	Rasuwa	Yak	21	4	Strongyles+Trichuris
	Nuwakot	Environment	10	10	Strongyles
	Kathmandu	Goat	8	8	Strongyles
Magh	Kathmandu	Cow	2	2	Paramphistomum
	Kathmandu	Cow	2	2	Strongyles
	Kailali	Goat	84	84	Mixed
Falgun	Kailali	Goat	89	86	Mixed
	Sindhuli	Goat	34	22	Mixed
Chaitra	Kathamandu	Cattle	4	4	Coccidia
	Nuwakot	Goat	3	3	Strongyles
	Nuwakot	Goat	3	3	Coccidia
	Kathamandu	Cattle	2	2	Strongyles
	Kathamandu	Goat	2	2	Strongyles
Baisakh	Bhaktapur	Cattle	2	2	Coccidia
	Kathamandu	Cattle	1	1	Mixed
	Kathamandu	Cattle	1	1	Strongyles
	Nuwakot	Goat	4	4	Strongyles
	Kathamandu	Poultry	6	3	Strongyles
	Kathamandu	Cattle	2	1	Strongyles
	Kathamandu	Buffalo/Cow/Goat	11	5	Mixed
	Kathamandu	Buffalo/Cow/Goat	11	5	Strongyles
	Kathamandu	Cattle	3	1	Strongyles
	Kathamandu	Cattle	3	3	Coccidia+Strongyles

	Kathamandu	Poultry	1	1	Coccidia
	Kathamandu	Cattle	3	3	Coccidia
	Kathamandu	Cattle	3	3	Coccidia
	Kathamandu	Buffalo	1	1	Paramphistomum
Jestha	Kathamandu	Cattle	2	2	Liver fluke
	Makawanpur	Buffalo	1	1	Liver fluke
	Kathamandu	Goat	4	4	Strongyles
Ashad	Kathamandu	Poultry	2	2	Coccidia
	Kathamandu	Sheep	1	1	Strongyles
	Sarlahi	Goat	7	7	Strongyles+Trichuris
	Sarlahi	Goat	7	7	Mixed
Total			1490	465	

7.3.2 Skin Scrapping examination

In the FY 2079/80, 62 skin scrapping samples from different species were tested for the presence of external parasites. 54.83% of the samples were found to be positive.

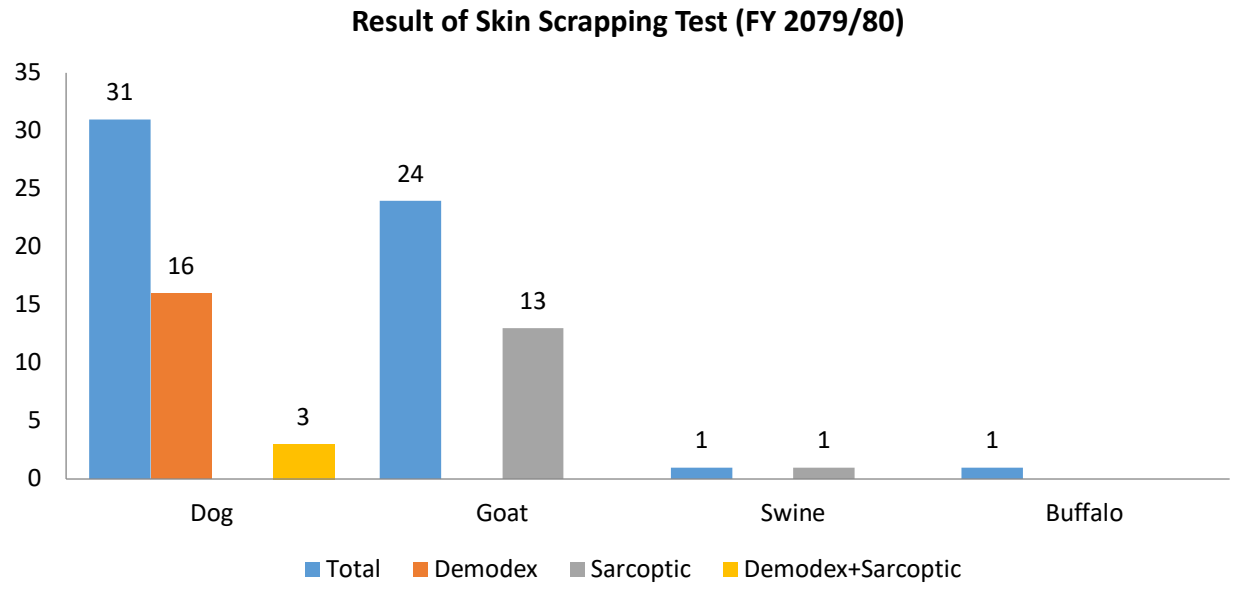


Figure 4: Results of Skin Scrapping Test (FY 2079/80)

8. Molecular Biology Section

8.1 Molecular Unit

Molecular biology unit was established in CVL in 2003 A.D. Earlier this laboratory started to diagnose avian influenza by using RT-PCR technique. Later, from 2010, CVL started real time PCR for diagnosis of avian diseases like AI, ND, and IBD. Since 2016, CVL also started multiplex for respiratory disease of small ruminant (PPR, MCCP, Capripox, Pasteurella) and swine diseases (African swine fever virus, Classical swine fever virus, Salmonella and Erysipela*)* and other diseases like Porcine Reproductive and Respiratory Syndrome (PRRS-NA and PRRS-EU). Later on, the technologies has been expanded for diagnosis of other zoonotic and economically

importance diseases like Glanders, Lumpy skin Diseases (LSD) and Enterotoxaemia. Molecular unit is also participating in proficiency testing (PT) for AI, PPR and Swine diseases since 2016.

In the Fiscal year 2079/80, a total of 411 swab samples of avian species suspected for avian influenza were received from twenty-five districts. The samples were tested by using Real Time Reverse Transcriptase Polymerase Chain Reaction (rRT PCR). Out of those samples, 69.58% samples were found to be positive for subtype H9N2. During this period, HPAI outbreak also occurred in five (Kathmandu, Lalitpur, Bhaktapur, Kabrepalanchowk and Dhading) districts. 34 samples were found to be positive for HPAI (H5N1). Among them, few samples were sent to WOAH Reference laboratory, Australia. According to WOAH Report, (HPAI) H5N1 associated with the current outbreak belonged to 2.3.2.1a clade.

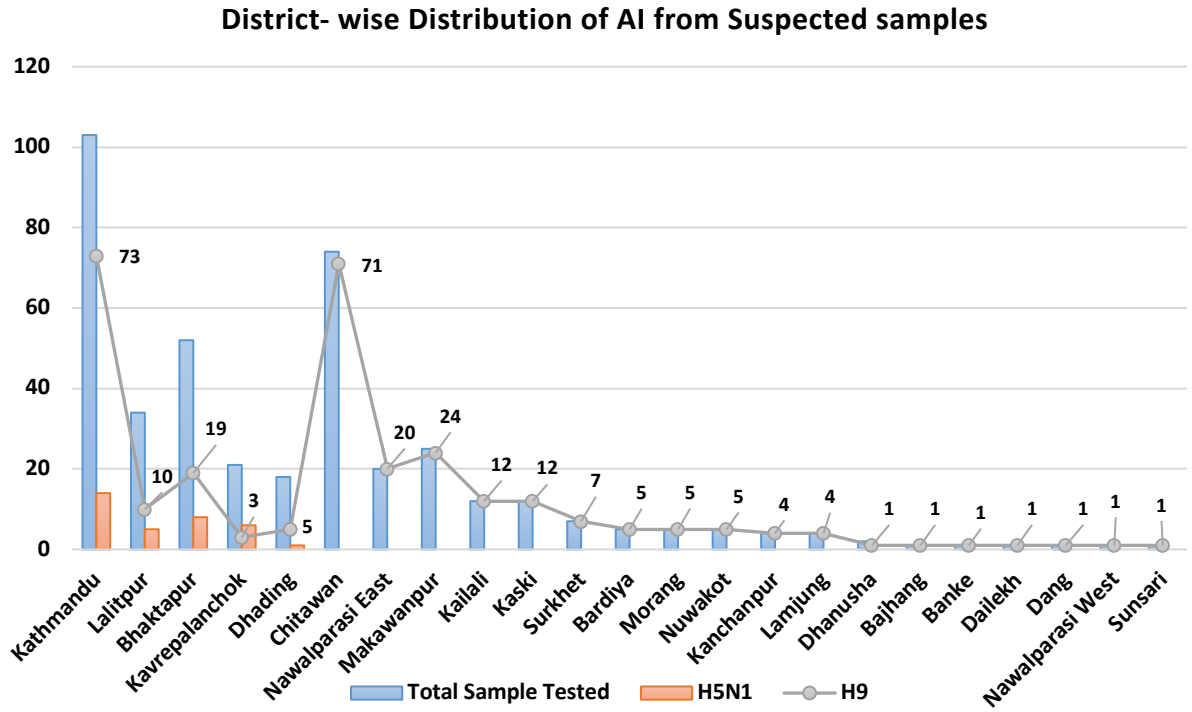


Figure 5: District wise H9 and H5 outbreak of AI.

Similarly, CVL received 100 samples for the diagnosis of PPR from 29 districts, out of which 19% were positive from 7 districts (Dhanusha, Dolakha, Kailali, Kathmandu, Morang, Ramechhap and Rupandehi).

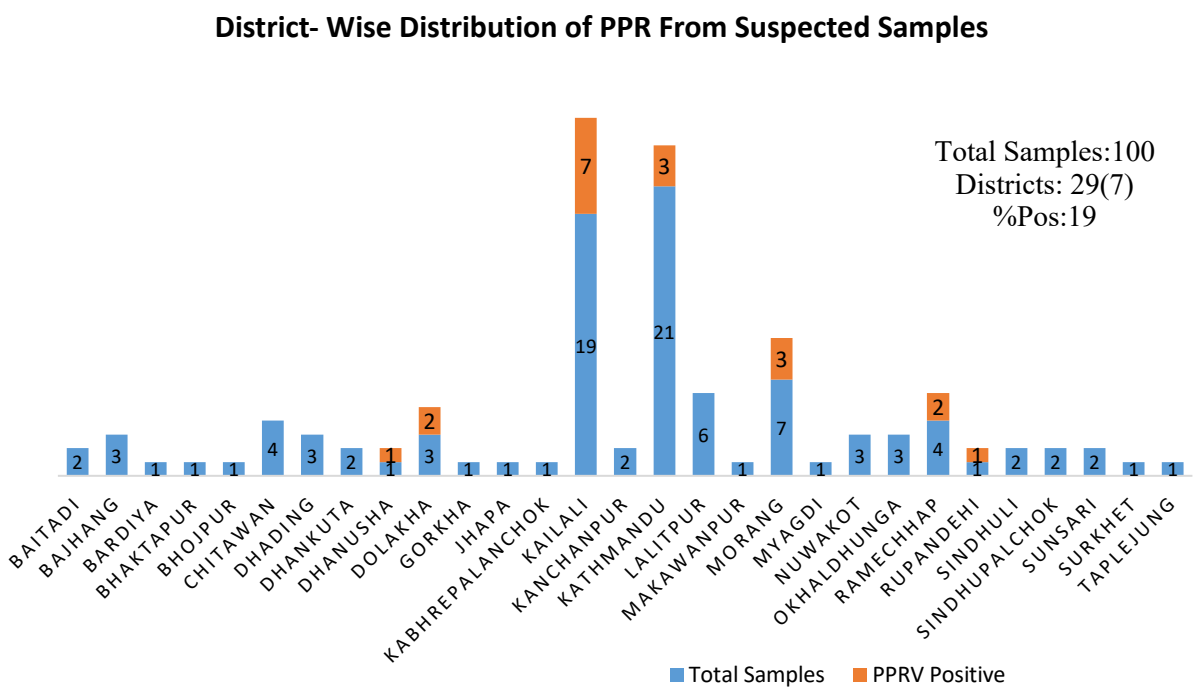


Figure 6: District wise distribution of PPRV in goats.

Also 107 samples from 36 districts were received for the diagnosis of respiratory multiplex. 8 Samples from Surkhet, Okhaldhunga, Baitadi, Bardiya and Lalitpur were positive for Pasteurellosis.

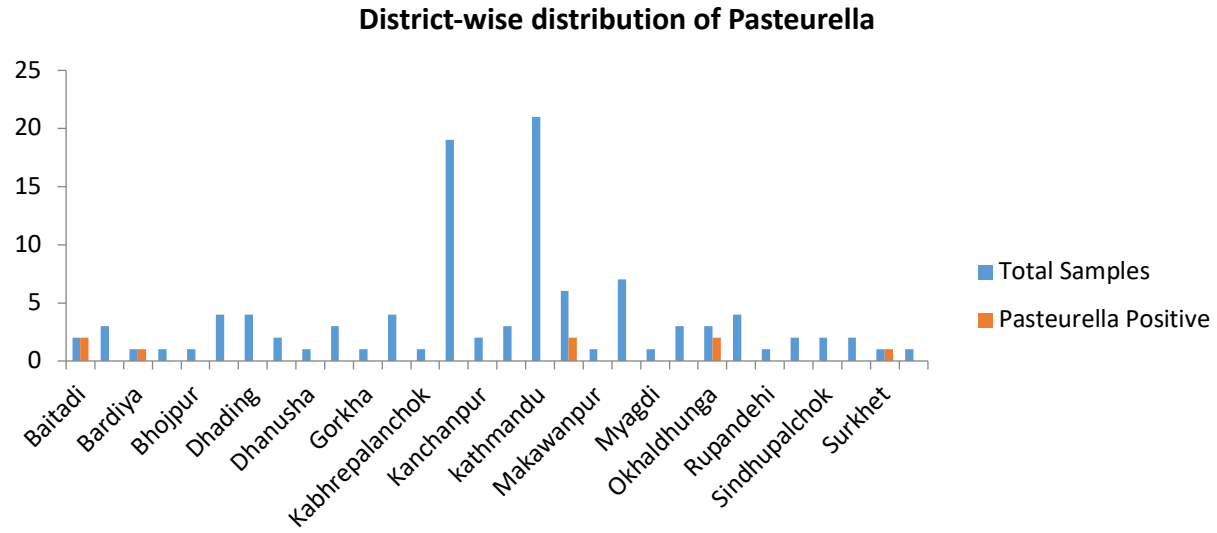


Figure 7: District wise distribution of Pasteurellosis in goats.

Likewise, CVL received 169 samples (Tissues/blood) from thirty-two districts for Swine diseases. The samples were initially tested for ASF followed by CSF, PRRS-NA, PRRS-EU, Salmonella and Erysipelas. Out of those samples, 75.14% samples were found to be positive for ASF.

According to OIE Report, phylogenetic analysis based on partial p72 nucleotide gene sequences of ASFV belonging to Genotype II. None of the samples were positive for both PRRS-NA and PRRS-EU.

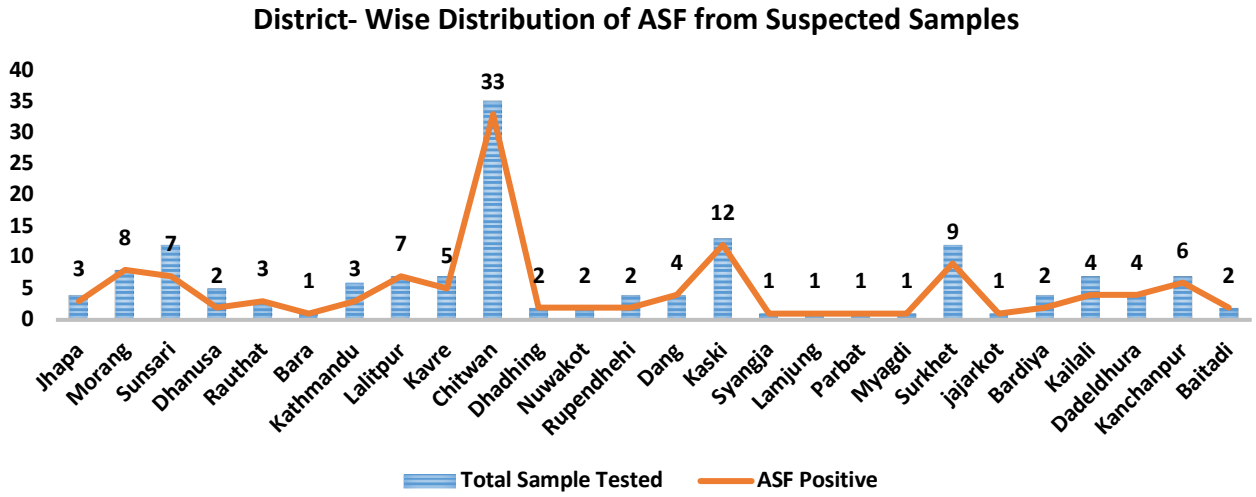


Figure 8: District wise distribution of ASF

CVL also received bovine samples (Swab/pus/scar) from forty-eight districts for Lumpy Skin Disease (LSD). A total of 449 samples were tested using PCR. 88.2% of the samples were positive for LSD.

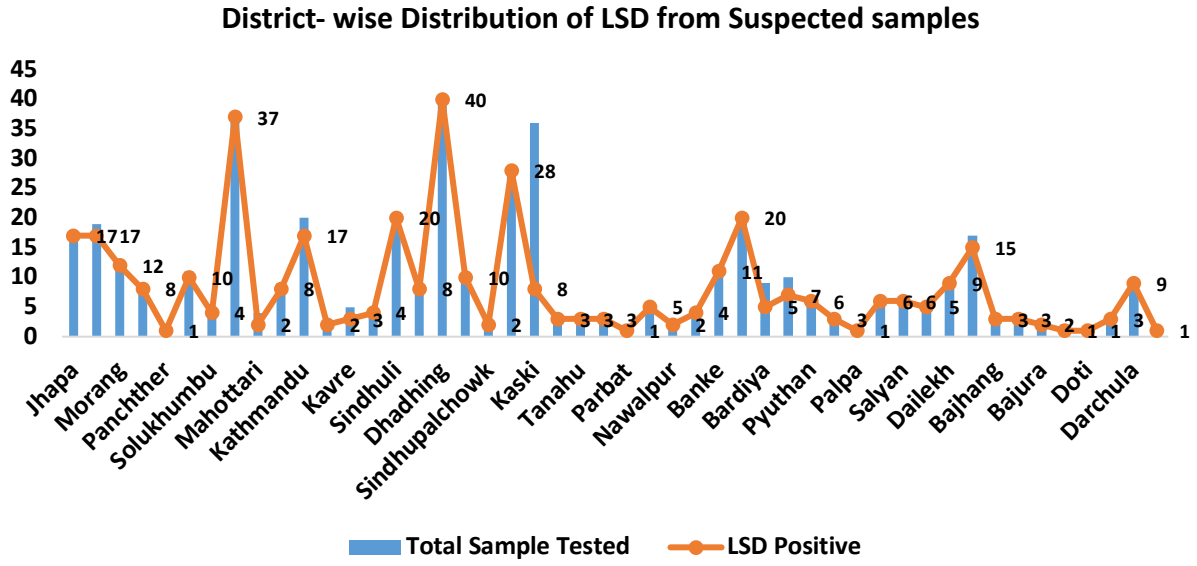


Figure 9: District wise distribution of LSD in bovine

CVL also received 108 equine pus swab samples from seven districts. 14.14% samples from Banke, Bardiya, Bhaktapur, Lalitpur and Rautahat were found to be positive.

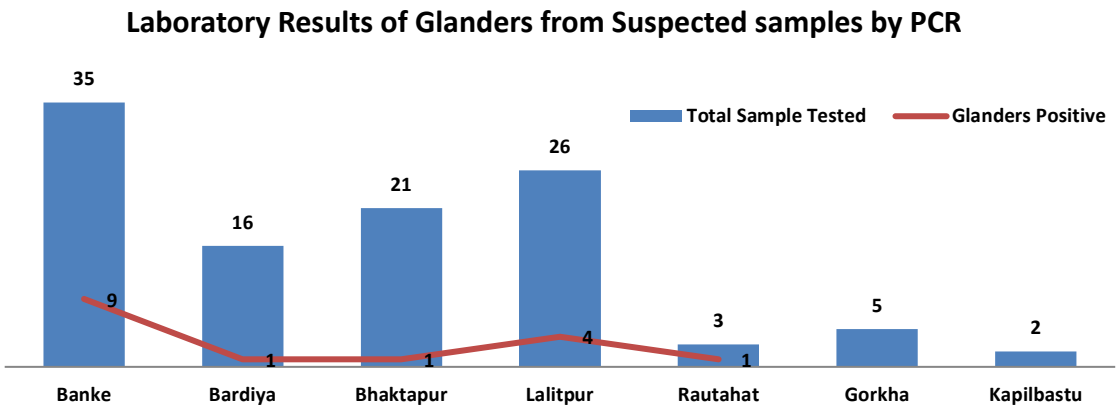


Figure 10: District-wise distribution of glanders from suspected samples

A total of 471 swab samples were collected from 10 districts for the surveillance of Avian Influenza.Two samples from Chitwan district were found to be positive for Low Pathogenic Avian Influenza (H9).

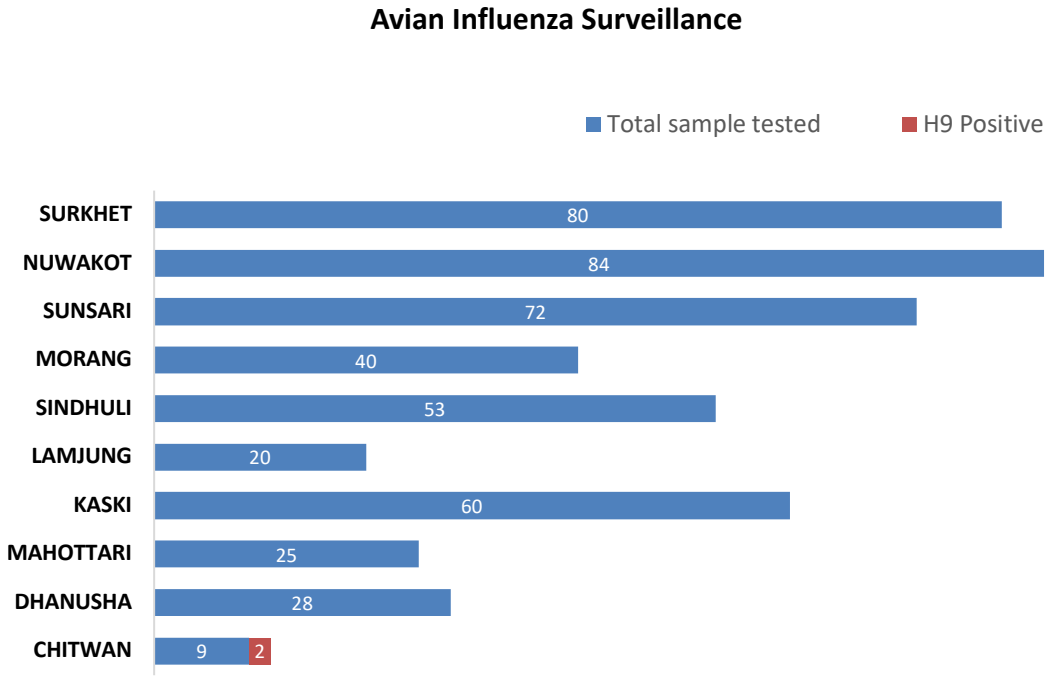


Figure 11: Avian Influenza Surveillance

8.2 Serology Unit

Serology unit of CVL performs different serological tests for the diagnosis, monitoring and surveillance of animal diseases. Most of the samples are submitted to this unit by VL, NADIL, VHLSEC, Quarantine Check-posts, private practitioner, farmers and staff of CVL during disease

outbreak investigations, routine diagnosis well as sero-monitoring. This section possesses facility of Competitive Enzyme Linked Immunosorbent Assay (ELISA), Immuno-capture ELISA, Indirect ELISA, Tube agglutination Test, Agar-Gel Immuno-Diffusion (AGID) test, Plate agglutination test and rapid tests.

Serology unit also participates in proficiency testing (PT) for PPR diagnosis by ELISA and Brucellosis by PAT since 2016. Progress report of Serological investigation of various diseases in animals and birds during 2079/80 is as follows.

8.2.1 PPR Seromonitoring

For PPR sero-monitoring, sample collected during the end of Ashad 2078/79 to 2079/80 were tested in 2079/80. CVL received serum samples from 49 districts. During 2079-80, a total 8214 serum samples were tested. Out of those samples, 66.84% were found positive for PPR antibody. The result shows that the PPR antibody positive percentage was found highest 98.(more than 90%) in four districts, Morang, Sunsari, Bhojpur and Sankhuwasava respectively. Low antibody titre was found in the serum samples of Dhanusha, Kavre, Dolakha, Rasuwa, Sindhupalchowk, Kaski, Baglung, Dailekh and Banke (below 50%). The antibody positive percentage ranged between 50-89 in remaining 36 districts.

The low sero-conversion might be due to sampling error or problem in cold chain maintenance of the vaccine. Furthermore, PPR vaccination program have been affected due to massive LSD outbreaks during vaccination season.

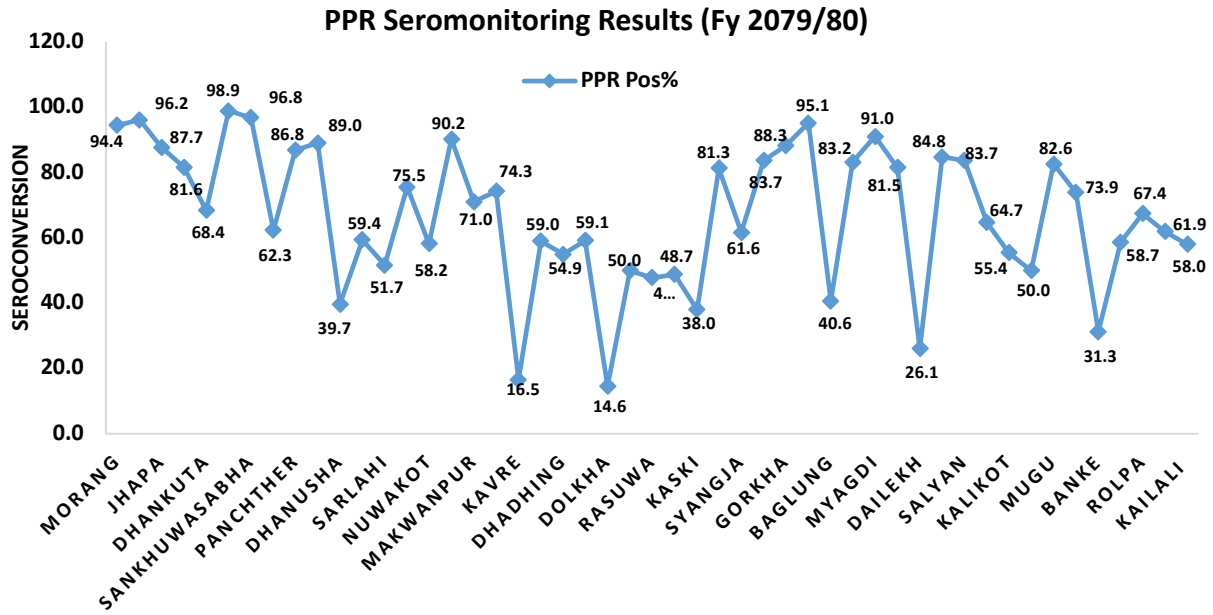


Figure 12: Peste-des Petitis Ruminant (PPR) Seromonitoring C-ELISA antibody test results

8.2.2 Brucellosis

A total of 873 serum samples from bovine, caprine, swine, ovine and feline and were tested for *Brucellosis* antibody by ELISA and PAT method and among them 35 (4.01%) samples were found positive for brucella antibody.

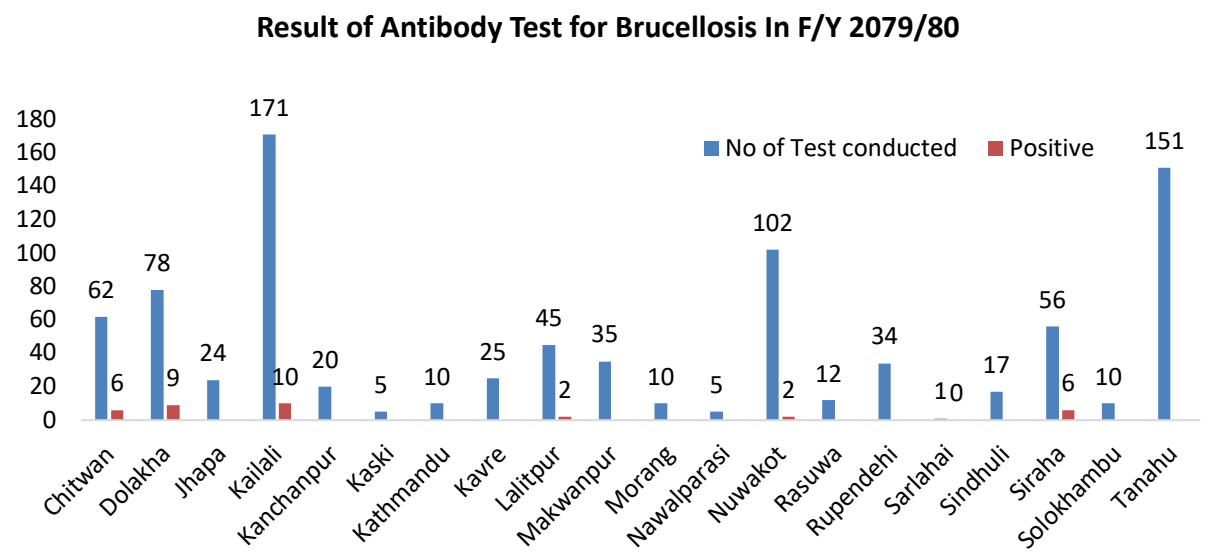


Figure 13: Result of Antibody test for Brucella by ELISA

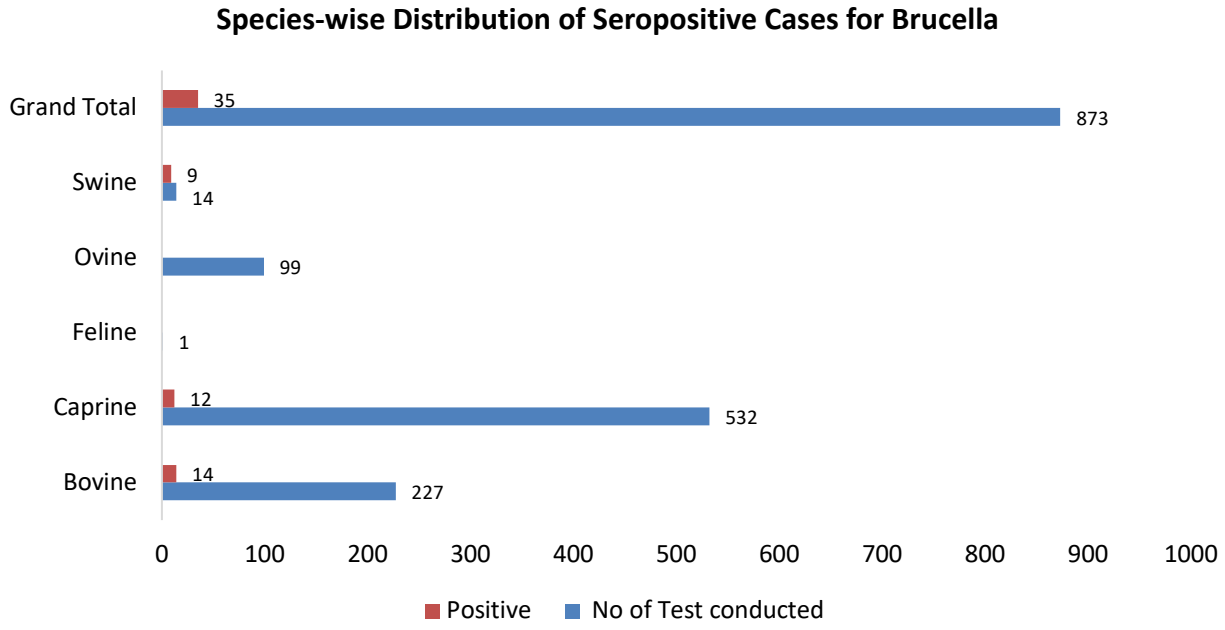


Figure 14: Species-wise Distribution of Seropostive cases for Brucells by ELISA /PAT Test

8.2.3 Toxoplasmosis

A total of 482 caprine, bovine, ovine and feline serum samples were tested from 18 different districts in terai, hilly and mountain regions for antibody of Toxoplasma by ELISA method. Among them, 21% samples were found to be positive.

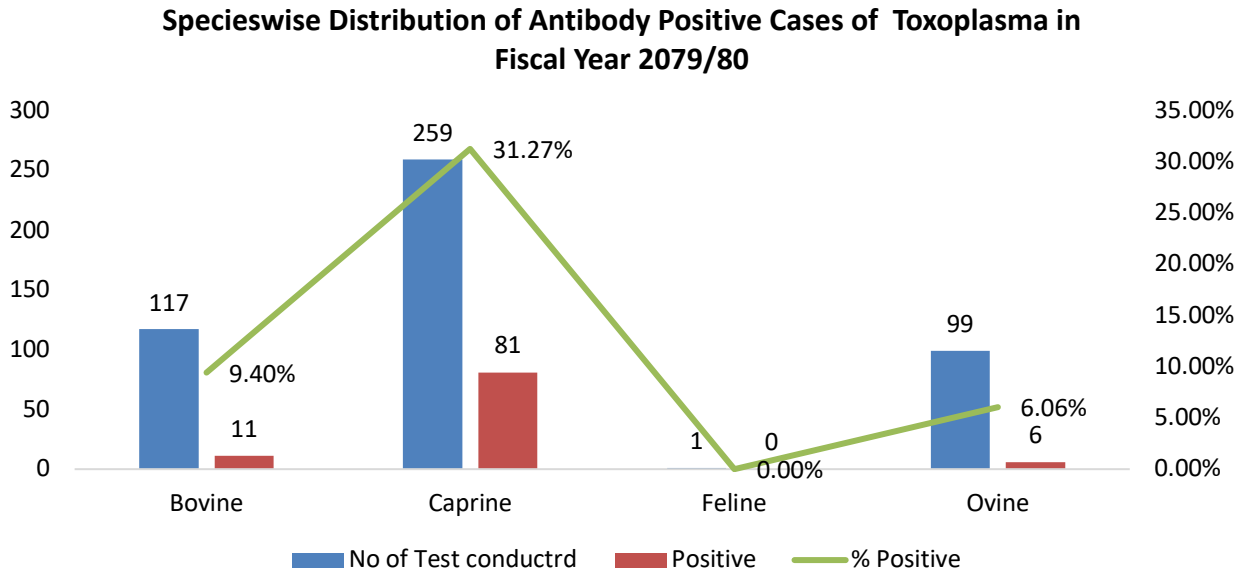


Figure 15: Species-wise Distribution of Antibody Positive cases of Toxoplasma by ELISA

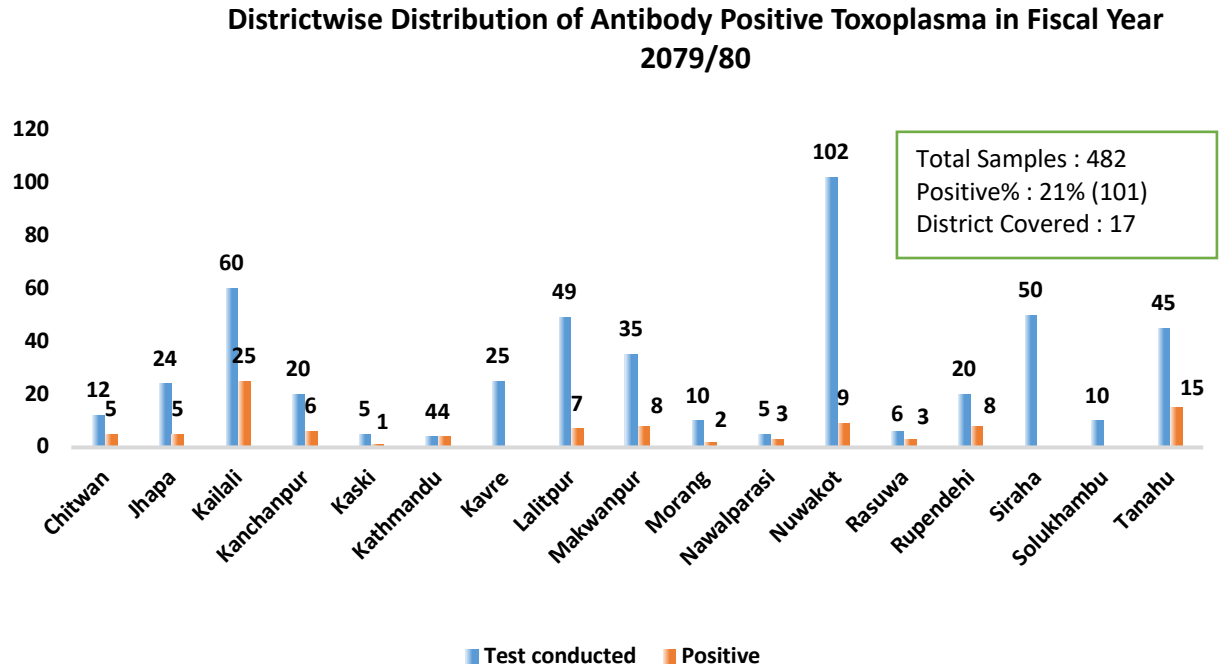


Figure 16: District-wise distribution of Antibody Positive Cases of Toxoplasma

8.2.5 Enterotoxemia

A total of 48 Caprine sample (Intestinal Content) from 48 districts were tested for different toxin for enterotoxemia from *Clostridiun perfringens* toxin detection multiscreen antigen ELISA method. Among them, 56.25% of suspected samples from 11 districts were found positive.

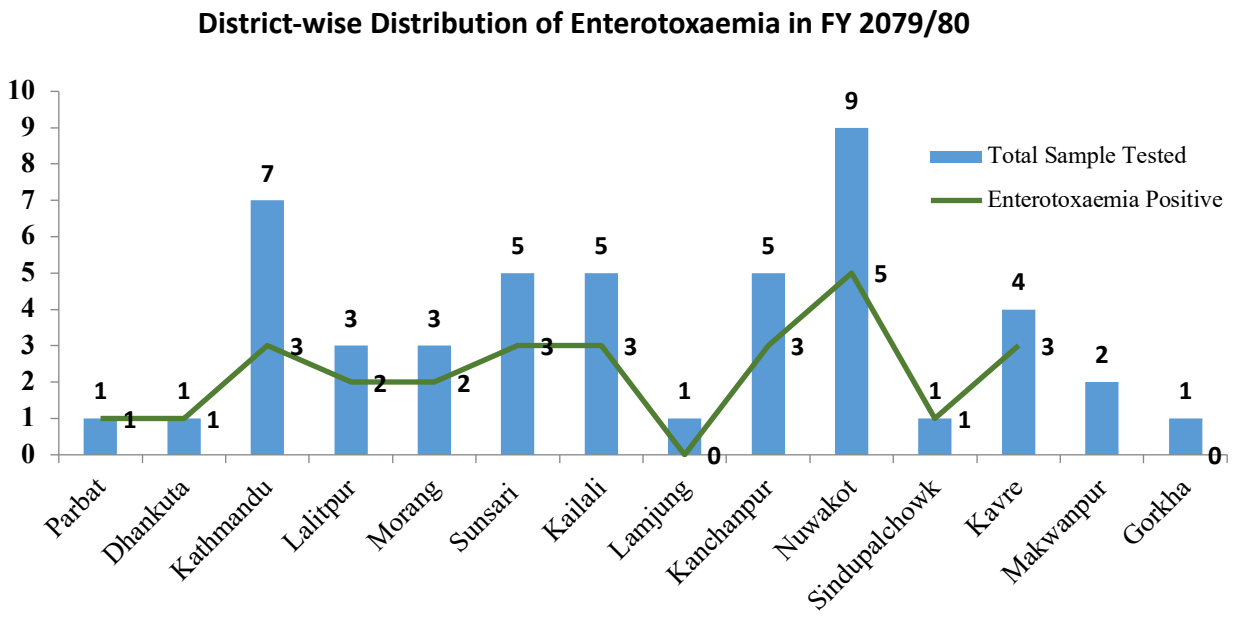


Figure 17: District- wise distribution of Enterotoxaemia in Fiscal Year 2079/80

8.2.6 Glanders Surveillance

Glanders Surveillance was conducted in different districs of Nepal where population of equines as well as risk of disease is high. 11 districts were covered in surveillance including Banke, Bardiya, Gorkha, Bhaktapur, Lalitpur, Kapilbastu, Chitwan, Kathmandu, Bara, Rautahat and Mustang. A total of 411 samples were collected among which 84 samples were positive from Banke, Bardiya, Gorkha, Bhaktapur, Lalitpur and Kapilbastu districts.

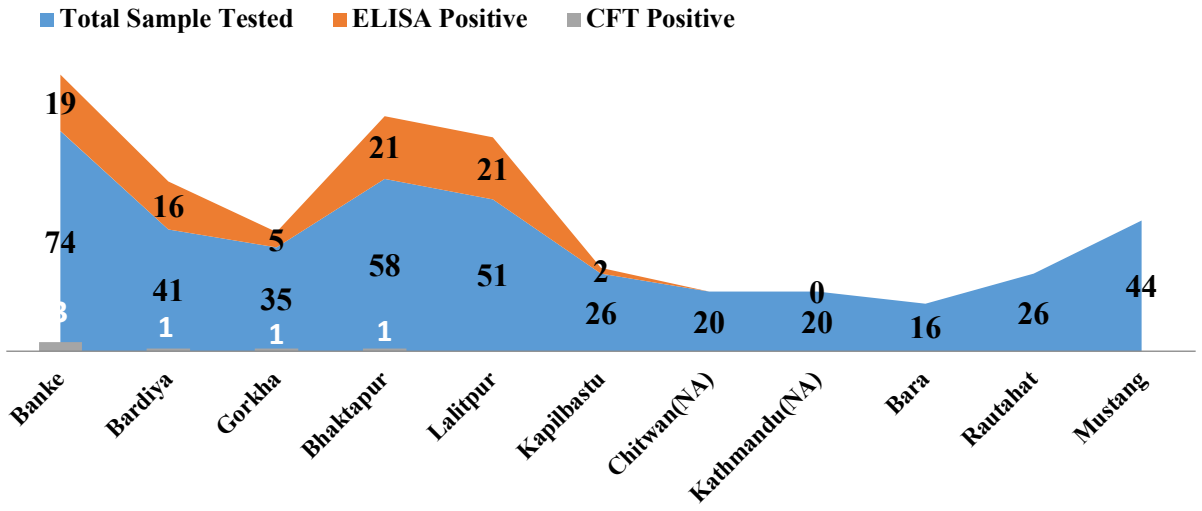


Figure 18: District- wise distribution of Glanders from Surveillance samples (NA-Nepal Army)

8.2.7 Leptospirosis

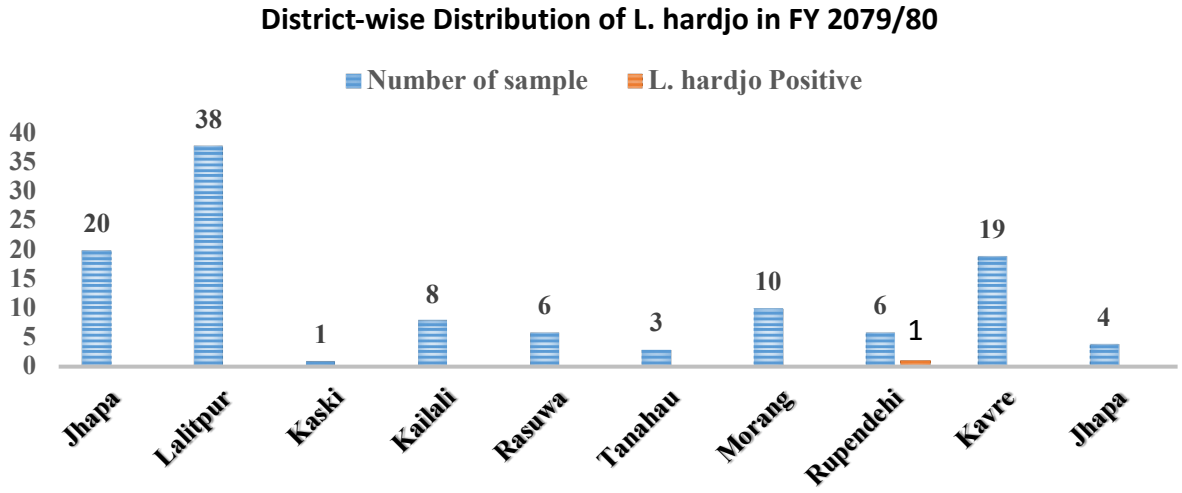


Figure 19: District- wise Distribution of *L. hardjo* from suspected samples

The graph presents data on the testing of samples for the presence of Leptospira hardjo across various districts. A total of 115 samples were collected, with the distribution as follows: Jhapa (24 samples), Lalitpur (38 samples), Kaski (1 sample), Kailali (8 samples), Rasuwa (6 samples), Tanahau (3 samples), Morang (10 samples), Rupendehi (6 samples), and Kavre (19 samples). Out of these samples, only one tested positive for L. hardjo, which was from Rupendehi. The remaining districts, including Jhapa, Lalitpur, Kaski, Kailali, Rasuwa, Tanahau, Morang, and Kavre, had no positive cases. This indicates a very low prevalence of L. hardjo in the tested regions, with only Rupendehi showing any presence of the bacteria.

8.2.7 Plate agglutinaiton test

A total 194 number of poultry serum samples were collected from Sindhuli, and Kaski districts and tested for *Salmonella pullorum* and *Mycoplasma gallisepticum* antibody respectively by PAT method. 4 samples were found positive for *Salmonella pullorum* and 58 samples for *Mycoplasma gallisepticum* antibody.

Table 22: Antibody Test Results of *Salmonella pullorum* and *Mycoplasma gallisepticum* in poultry serum by using Plate Agglutination Test (PAT)

S.N.	Districts	*Salmonella pullorum* (PAT)			*Mycoplasma gallisepticum* (PAT)		
		Total Tested sample	Positive	Negative	Total Tested sample	Positive	Negative
1	Kaski	51	0	51	51	41	10
2	Sindhuli	46	4	42	46	17	29
Total		**97**	**4**	**93**	**97**	**58**	**39**

8.2.8 SARS-CoV2 Survey

A total of 346 samples from companion animals (dog and cat) from the owners with COVID infection were collected for SARS-CoV2 Survey. 29 samples were found to be positive from dog. whereas none of the samples were positive from cats.

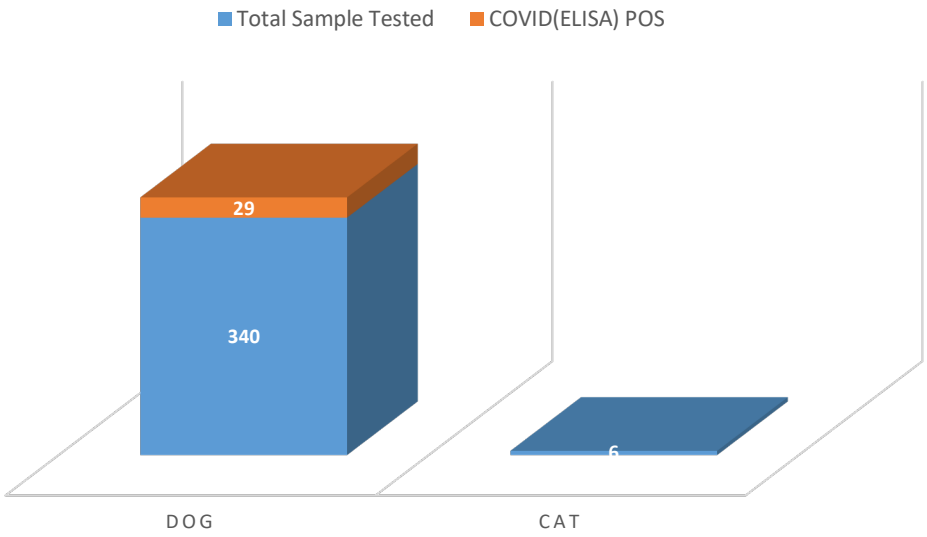


Figure 20: SARS-CoV2 Survey

9. Veterinary public health section

Veterinary Public Health section is responsible to assure the safety of public with consumption of animal products. This section conducts various programs to address public health issues such as veterinary drugs, insecticides/hormones/pesticides residue testing in milk, egg, meat and fish. This section is conducting antibiotic residue testing in certain group of antibiotics like as Streptomycin, Gentamycin, Sulphonamides and Tetracycline. It conducts hormone test in the milk and meat that are harmful to the health of people. It is conducting the ractopomine and progesterone hormone residue test in milk and meat sample. It is responsible to conduct zoonotic disease surveillance that are prevalent and at high risk in Nepal and monitoring of meat shop.

9.1 Antibiotic residue test in milk and meat

Table 23: Antibiotic residue test in milk and meat

Sample	Drug/Toxin	District	Negative	Positive	Total
Gizzard content from suspected bird	Total Aflatoxin	Kathmandu	0	8	8
Fish	Malachite Green	Chitwan	45	0	45
Liver	Benzylpenicillin	Dhadhing	17	2	19
Meat		Dhadhing	24	2	26
Meat	Colistin	Dang	59	1	60
Meat		Dolakha	9	0	9
Meat		Kathmandu	14	1	15
Meat	Fluoroquinolone	Kathmandu	21	0	21
Meat	Tetracycline	Dang	60	0	60
Meat		Dolakha	5	0	5

Sample	Drug/Toxin	District	Negative	Positive	Total
Meat		Kathmandu	15	0	15
Meat	Tylosin	Dang	56	4	60
Meat		Dolakha	13	1	14
Meat		Kathmandu	10	0	10
Milk	Aflatoxin M1	Bhaktapur	24	4	28
Milk		Kathmandu	102	10	112
Milk		Lalitpur	27	1	28
Milk	Gentamicin	Dhadhing	84	0	84
Milk		Kathmandu	82	2	84
Milk		Tanahun	32	0	32
Milk	Oxytocin	Kathmandu	82	0	82
Milk	Streptomycin	Chitwan	26	0	26
Milk		Lamjung	8	0	8
Milk	Sulfonamide	Tanahun	32	0	32

In the FY 2079/80, 314 meat and 266 milk samples were tested for the presence of antibiotic residue of the drugs that are commonly used in the field. The list included benzyle penicillin, fluoroquinolones, colistin, tetracycline and tylosin for the meat samples whereas for the milk samples it included gentamicin, sulphonamide and streptomycin. In 1.8% of the milk samples and 9.8% of the meat samples, there were presence of the tested antibiotics residue.

9.3. **Toxoplasmosis surveillance**

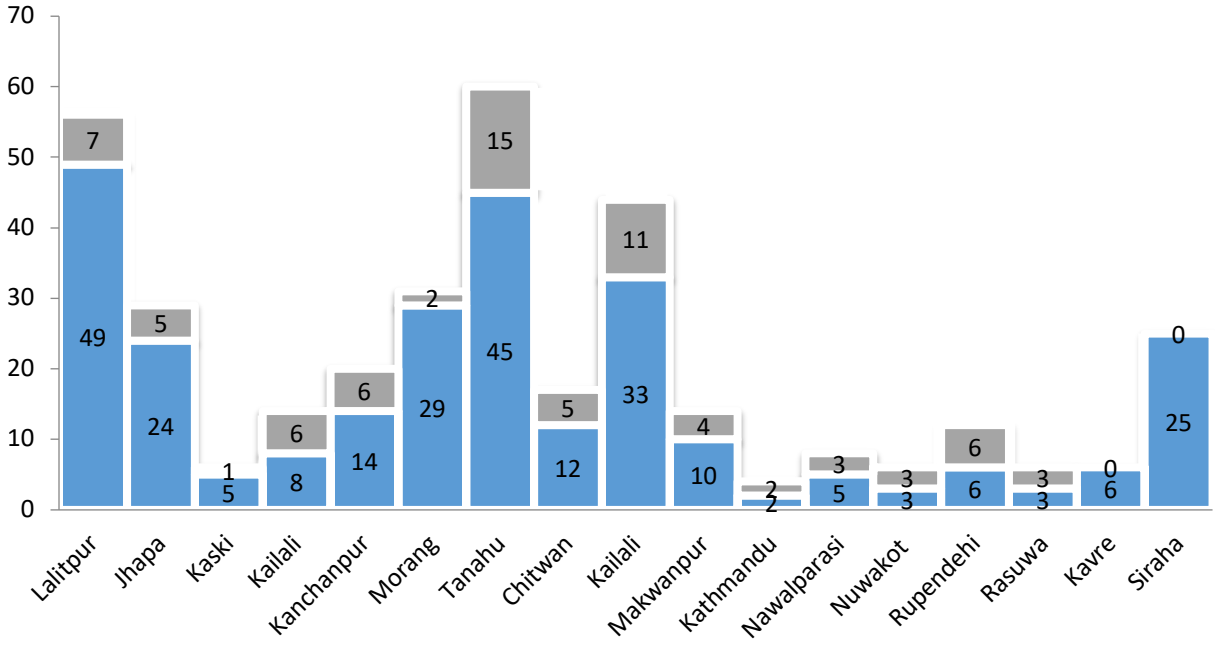


Figure 21: Toxoplasmosis surveillance (FY 2079/80)

A total of 279 serum samples from 17 districts were collected out of which 28.31% were found to be positive.

10. Disease status in Government Farm

10.1 Sheep and goat

Table 24: Sheep and goat

Government Farm	Brucellosis		Toxoplasmosis		*C. abortus*		MCCP	
	Total Sample Tested	Positive	Total Sample Tested	Positive	Total Sample Tested	Positive	Total Sample Tested	Positive
Sheep Genetic Resource Centre, Nuwakot	99	0	20	6	_	_	58	1
Goat Genetic Resource Center, Kailali	101	0	_	_	101	0	_	_
Bakhra Bikash Farm, Makwanpur	25	0	25	4	_	_	_	_

10.2 Cattle and yak

Table 25: Cattle and yak

Government Farm	Brucellosis		Leptospirosis		Tuberculosis		CCHF		Toxoplasmosis	
	Total Sample Tested	Positive	Total Sample Tested	Positive	Total Sample Tested	Positive	Total Sample Tested	Positive	Total Sample Tested	Positive
Yak Genetic Resource Centre, Solukhambu	69	0	46	1	_	_	46	0	69	1
Cattle Genetic Resource Center, Dolakha	_	_	23	0	44	0	23	0	_	_

VETERINARY LABORATORY

1. Introduction

To provide diagnostic facilities throughout the country, CVL works through its five Veterinary Laboratories (VLs) located in different provinces of the nation; Koshi Province Veterinary Laboratory (Biratnagar), Madhesh Province Veterinary Laboratory (Janakpur), Gandaki Province Veterinary Laboratory (Pokhara), Karnali Province Veterinary Laboratory (Surkhet) and Sudurpashim Province Veterinary Laboratory (Dhangadhi). Specimens that cannot be processed in the aforementioned laboratories due to insufficient facilities and expertise or needed to be further tested for confirmation are referred to the CVL.

2. Objectives

The objectives of veterinary laboratories as follows;

- To provide prompt and efficient disease diagnostic services to the farmers of respective province/working areas.
- To investigate and diagnose the epidemics in the province.
- To assist and support VHLSEC and local level governments in sample collection, disease diagnosis and epidemic control.
- To supervise and assist in diagnostic services to basic and primary laboratories situated in VHLSEC in the province.
- To collect, analyze and predict the animal diseases prevailing in the province.
- To develop human resources for the field level veterinary services.
- To co-ordinate and support national livestock disease control and eradication program.
- To support and facilitate the national veterinary regulatory services.
- To participate actively in collaborative and coordinated research program in animal health and production in the region.

3. Laboratory services at VL

To meet the above-mentioned objectives, the VLs have been providing services under four sections.

3.1 Pathology section

Post-mortem examination, hematology and biochemistry are the major areas under the pathology section. Mostly the section receives specimens from all over the province either directly or through the respective VHLSEC or local level government. Besides this, veterinary practitioners, livestock and poultry farms as well as farmers deliver specimens for the purpose of disease diagnosis.

3.2 Microbiology section

This section is responsible for isolation and identification of bacteria and fungus, which receives samples from various sources such as farmers, local level governments, referral samples from private clinics, VHLSEC and directly from the field during the outbreaks. Various samples like milk, urine, tissues, water, nasal swabs, ear swabs and skin scrapping are received in this unit for isolation, identification and antibiotic sensitivity testing of the organism. Also, this section is responsible for the diagnosis of viral disease through different tests. Lateral flow assay (antigen

based) is used for the initial diagnosis of ND, AI, IB, ASF and Rabies. For the further confirmative diagnosis of disease, the samples are sent to the CVL.

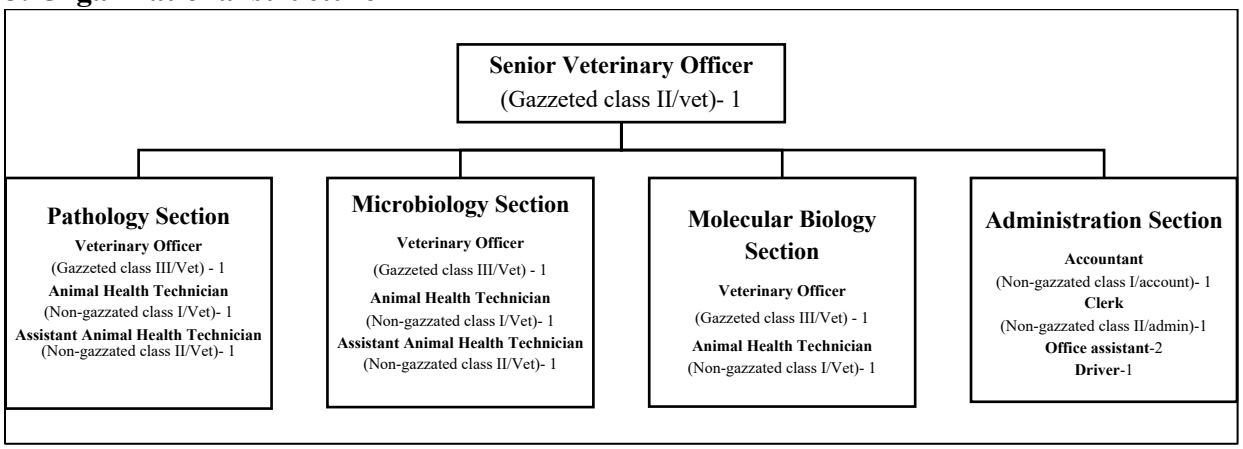
3.3 Molecular biology/Serology section

Molecular tools are increasingly important in modern animal disease investigation. Molecular biology section is functioning in some labs and are planning to make functional in other labs facilities for handling infectious agents. This section also the facility for ELISA and other serological tests. Most of the samples are submitted to this unit by the post mortem unit and field leve for the further confirmative diagnosis of disease, the samples are sent to the CVL.

3.4 Administration Section

This section governs overall financial, logistic and administrative management of the laboratory. The section is responsible for revenue collection, maintenance of reagents and supplies, financial transaction, vehicle management etc.

3. Organizational structure



VETERINARY LABORATORY BIRATNAGAR

1. Introduction

The Veterinary Laboratory, Biratnagar is located in Biratnagar Metropolitan City in Koshi Province. It was established in F.Y.1988/1989 as regional veterinary laboratory, Biratnagar and covered all the 16 districts of the Eastern Development region. This laboratory was restructured during 2074/75 with the scope to work in all 14 districts of Koshi Province. In Veterinary Laboratory, Biratnagar, following diagnostic services are available: feacal test, blood test, skin scraping and postmortem examination, bacterial culture etc.

2. Human resource

Table 26: Staffing of Veterinary Laboratory, Biratnagar, Morang

S. N	Name of Staff	Designation	Class
1	Dr. Sanjay Kumar Yadav	Senior Veterinary Officer	Gazetted II
2	Dr. Ananta Koirala	Veterinary Officer	Gazetted III
3	Dr. Khila Bogati	Veterinary Officer	Gazetted III
4	Dr. Roshan Dahal	Veterinary Officer	Gazetted III
5	Mr. Balaram Pokharel	Accountant	Non- gazetted I
6	Mr. Yogendra lal Yadav	Veterinary Technician	Non- gazetted I
7	Ms. Babita Rai	Veterinary Technician	Non- gazetted I
8	Mr. Bikash Adhikari	Asst. Veterinary Technician	Non- gazetted II
9	Mr. Bishnu Prasad Dulal	Asst. Veterinary Technician	Non- gazetted II
10	Mr. Ramesh Kumar Khadka	Kharidar	Non- gazetted II
11	Mr. Ram Sewak Mandal	Office Assistant	Classless
12	Mr. Gajanand Thakur	Office Assistant	Classless
13	Mr. Ram Narayan Yadav	Light Vehicle Driver	Classless

3. Laboratory Service

3.1 Pathology section

3.1.1Parasitological examination:

Parasitology unit examine the faecal samples of various livestock species by direct smear, sedimentation and flotation method. Parasitological unit identify the parasites and quantify the parasitic burden of the nematodes, trematodes and cestodes by Mc-Master method. In this fiscal year 2079/80, a total of 127 faecal samples were examined and were found positive for various species. Major internal parasites include *Haemonchus, Strongylus, Trochoostrongylus, Fasciola Paramphistomum, Trichuris*.

Result of the faecal examination is presented below.

Table 27: Fecal examination result.

District	Species	Total sample	Positive	Negative	Positive %
Dhankuta	Goat	50	30	20	60%
Bhojpur	Goat	25	7	18	28%
Morang	Goat	52	17	35	32%
Total		**127**	**54**	**73**	**42.5%**

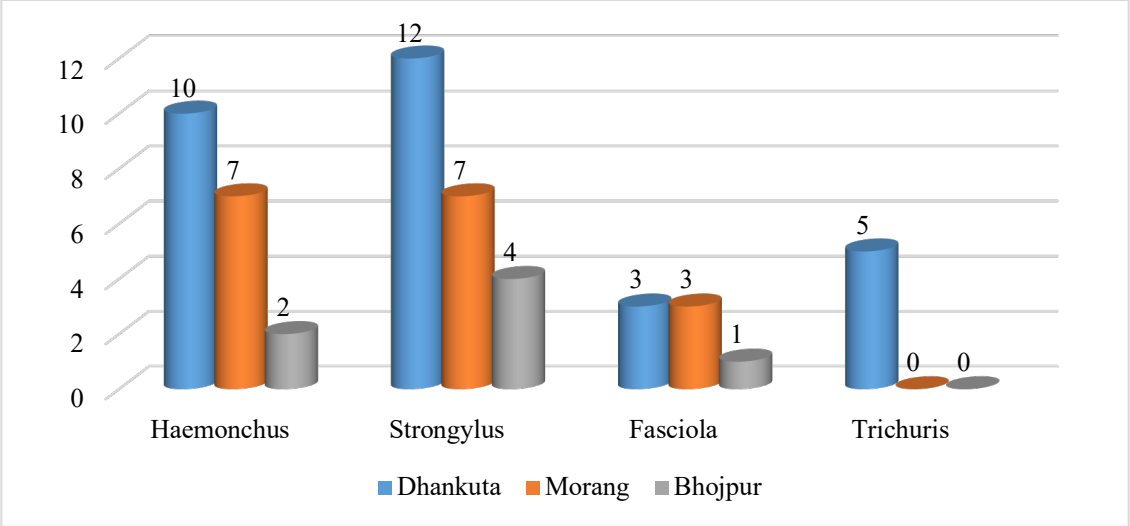


Figure 22: District wise distribution of helminthes parasites

3.1.2 Hematological test:

Hematology unit examine the blood samples of various species of animals in Koshi province. Hematological test includes the Hb, PCV, DLC, ESR, PLT estimation and identification of blood protozoa. This test is performed by the hematoanalyzer and microscopic examination of blood. In this fiscal year total of 642 blood samples were tested in which 130 samples were found positive for blood protozoa. Blood protozoa like; *Trypanosoma sps., Babesia sps., Anaplasma sps., and Theileria sps.* were identified. Result of the blood examination is presented in graphical representation in below.

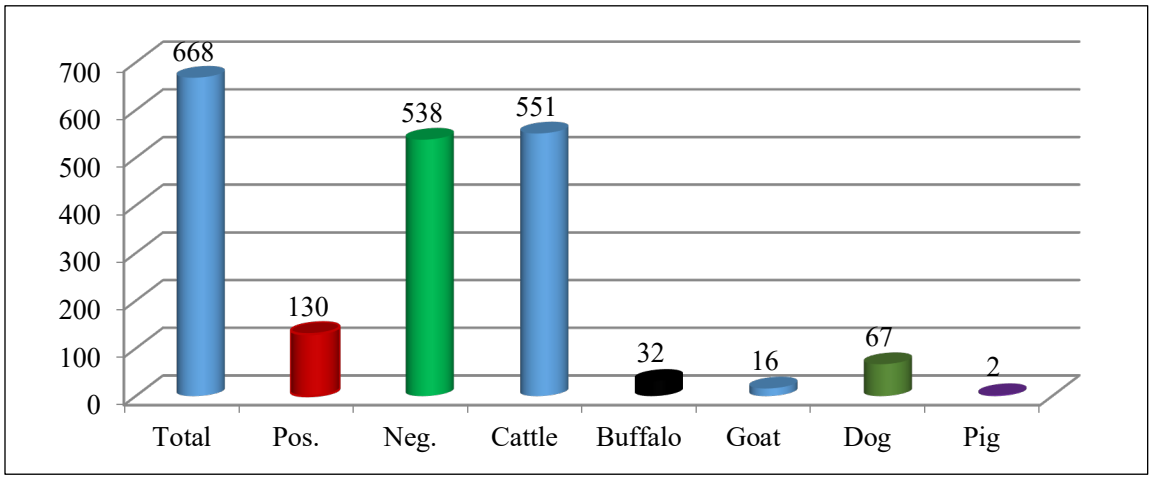


Figure 23: Species wise blood samples received at the laboratory.

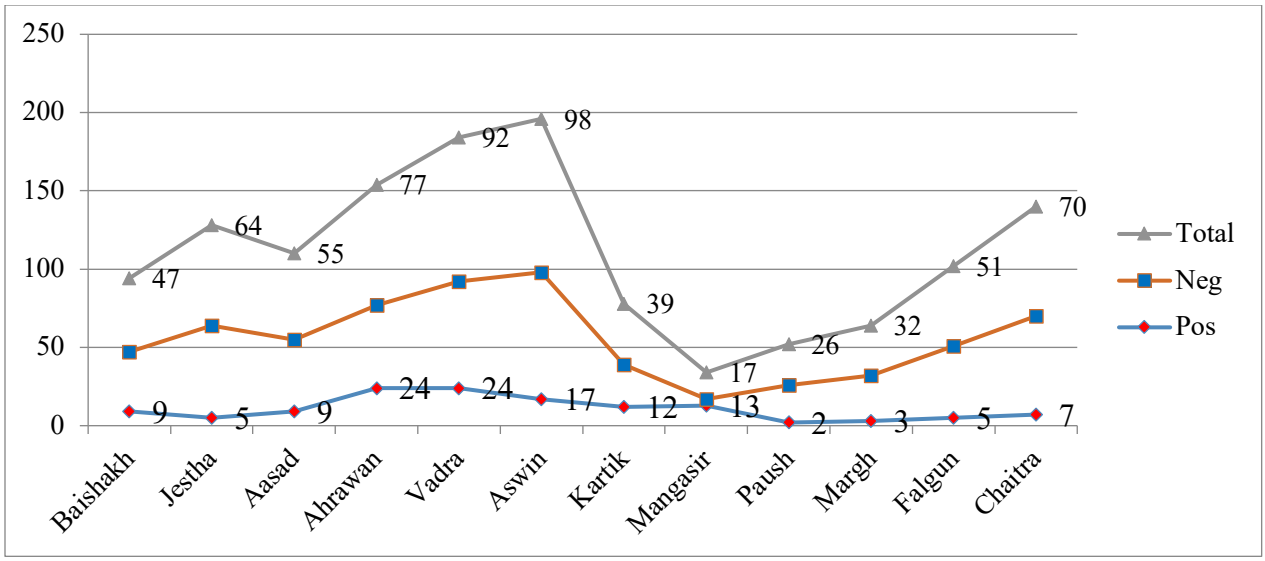


Figure 24: Month wise trend of positive cases of blood parasites.

3.1.3 Skin scraping test

In this year, out of a total 71 samples tested 7 samples were of cow and 64 samples were of dogs. All the samples from cattle were negative for mange mites, while 11 samples from dogs were positive (8 psoroptes and 3 demodex).

Figure 25: Psoroptes in dog.

Figure 26: Demodex in dog

3.1.4 Urine test

Out of 57 urine sample, 12 samples were found abnormal (3 with traces of blood, 7 with increased urobilinogen and two with pus).

3.1.5 Post-mortem examination

A total of 1171 samples (1104 samples chickens, 60 goats and 7 pigs) were brought at VL Biratnagar for postmortem examination. Samples were received from Sunsari, Morang, Jhapa, Dhankuta, Saptari and Udayapur.

3.1.5.1 Postmortem of poultry

Diseases of poultry is shown in below figure.

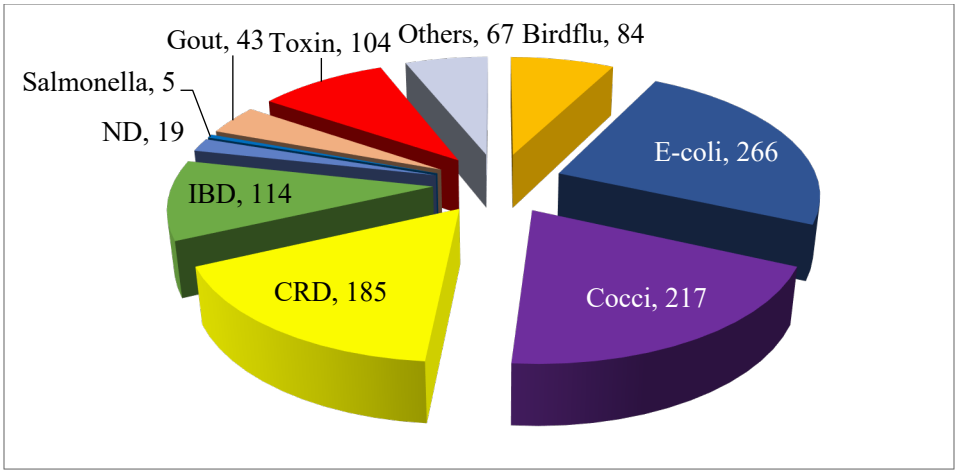


Figure 27: PM examination of poultry

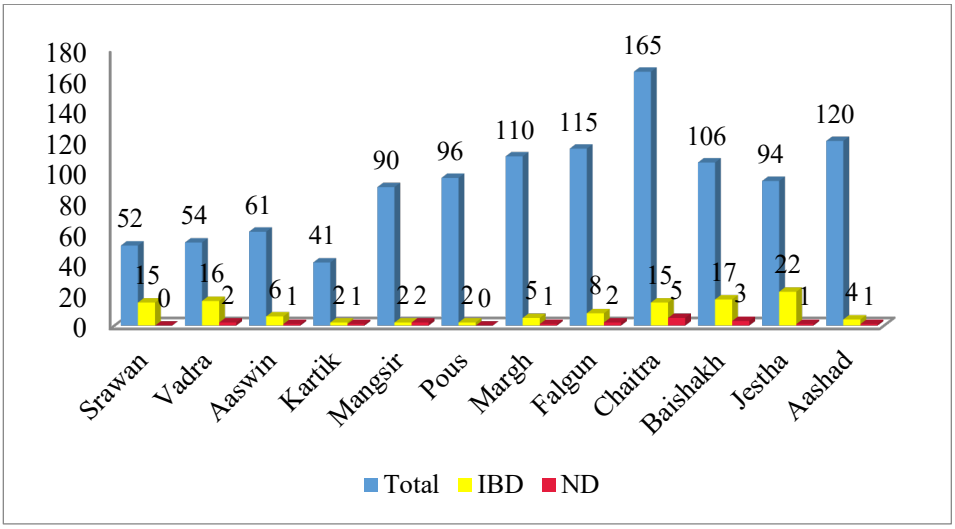


Figure 28: Month wise comparison of positive case IBD and ND

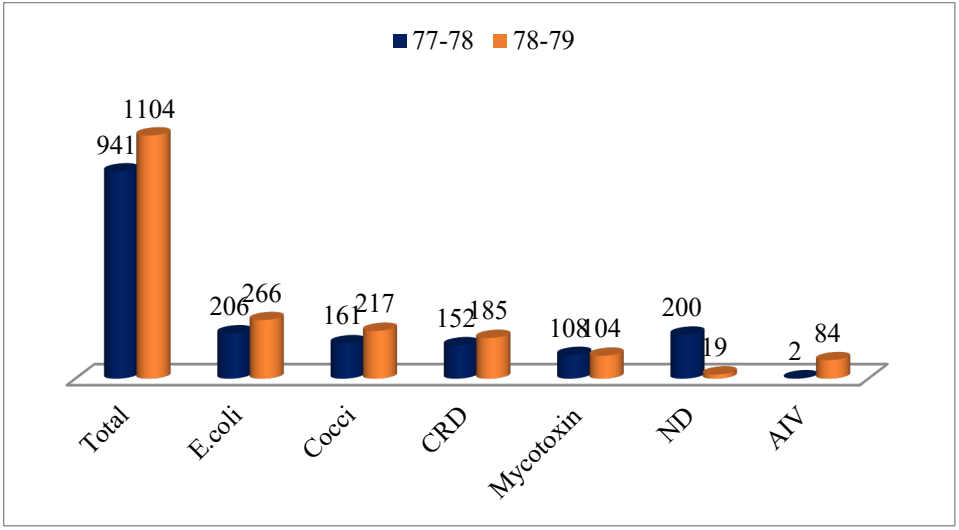


Figure 29: Comparison of the poultry diseases.

In this fiscal year 2079/80 HPAI was detected in Morang, Sunsari and Udayapur. The outbreak was managed with stamping out operation as follows.

Sunsari	Morang	Udayapur
• Outbreak-3 • Total bird-11000	• 10 outbreak • Total birds-111259	• 1 outbreak • Total birds-1625

Figure 30: Bird flu outbreak in Koshi Province.

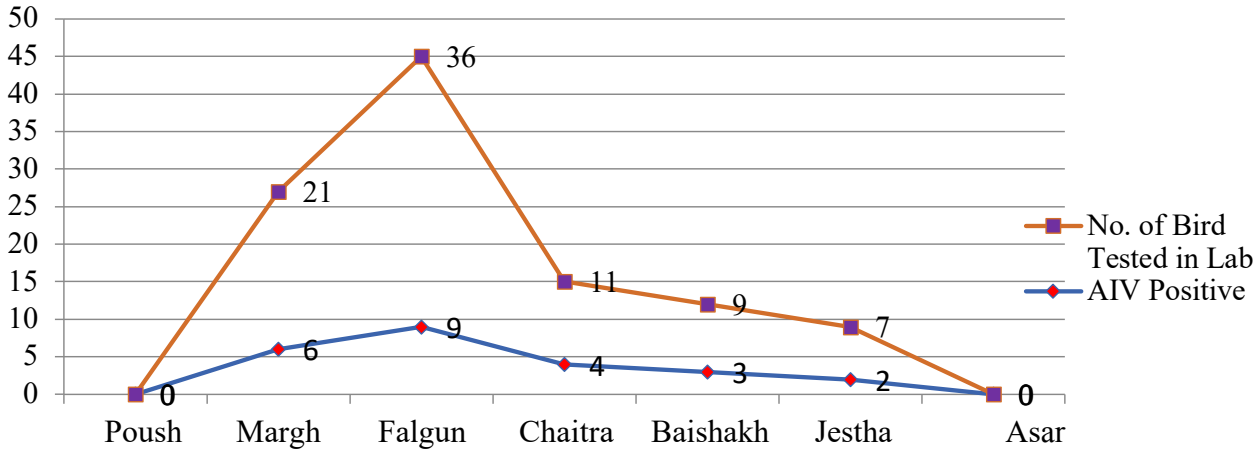


Figure 31: Month wise trend of Avian Influenza.

3.1.5.2 Animal disease

A total of 60 dead goats were brought at VL for post mortem examination. Enterotoxemia was found to be the major cause of death in goats.

Table 28: District wise goat brought at VL for post mortem.

S.N.	District	Species	No. of samples	Test Method	Result
1	Sunsari	Goat	17	PM, Clinical symptoms	
2	Morang	Goat	26	"	
3	Jhapa	Goat	7	"	
4	Udayapur	Goat	5	"	
5	Dhankuta	Goat	5	"	
	Total		**60**		

Similarly, seven pigs were brought for post mortem examination, out of which all were diagnosed with CSF by rapid diagnostic test.

Table 29: District wise pig brought at VL for post mortem.

S.N.	District	Species	No. of samples	Test Method	Result
1	Morang	Pig	4	PM, Clinical symptoms	CSF Ag +ve
2	Sunsari	Pig	3	PM, Clinical symptoms	CSF Ag +ve

3.2 Microbiology section

3.2.1 California mastitis test (CMT)

A total of 777 milk samples were tested by CMT, out of which 332 samples were found positive. Positive milk samples were tested for the bacteria culture, identification and AST.

3.2.2 Bacteriology

Bacteriological culture, bacteria identification and AST was conducted for the microbiological samples. The samples included milk, blood and tissue. Out of total 355 samples for culture, 332 were milk and 23 were of blood and tissue.

Result of the microbiological examination is presented in figure below.

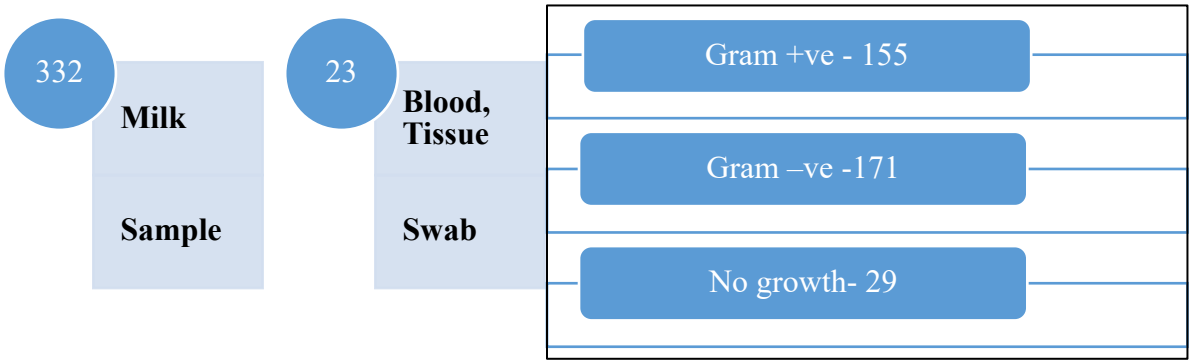


Figure 32: Microbiological examination result.

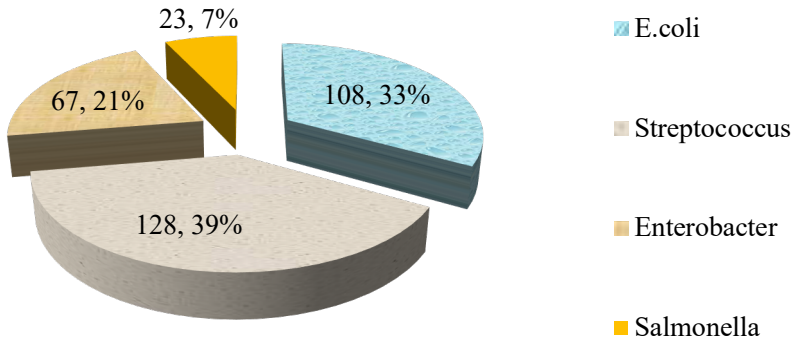


Figure 33: Types of bacteria found in culture.

3.2.3 Antibiotic sensitivity test (AST)

AST was performed for the bacteria identified in culture using disc diffusion method. Trend of susceptibility for different classes of antibiotic including Chloramphenicol, ceftriaxone, gentamicin, ciprofloxacin, enrofloxacin, ampicilline and tetracycline is shown in below figure.

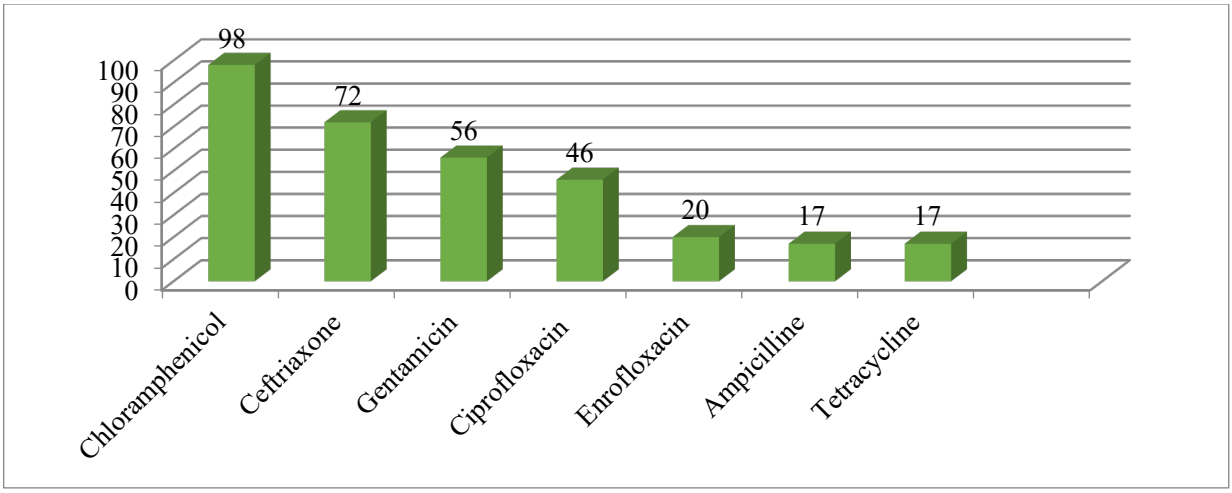


Figure 34: AST result.

In this fiscal year, under the active AMR surveillance program VL Biratnagar tested 121 bird samples from Sunsari (31 samples) and Morang (90 samples) district. VL Biratnagar followed the CLSI guideline for the bacterial culture. Among the common isolates 75.20% was *E. coli*, 51.2% were *Enterococcu*s and 11.57% were *Salmonella*.

3.2.4 Virological test

Virological unit of VL Biratnagar examined different types of viral diseases of an animal. Disease diagnosis was done by the using of rapid test kits and ELISA. The test detects Antigen or Antibody of the virus and also detect the serotype of the virus of some disease like; FMD. List of the virological test conducted by VL, Biratnagar were:

i. Animal disease: - FMD, PPR, ASF, CSF, Rabies, Canine distemper, PRRS, Canine parvo, Canine corona, Rota viral, BVD, etc.
ii. Avian disease: - ND, IBD, IBV, AIV, etc.
iii. Out of 55 samples tested for Rabies 2 were found positive.

3.3 Molecular biology section

3.3.1 Serological test

Serological unit of VL Biratnagar conduct the different types of test like PAT, Rapid Kit Test, and ELISA. Details of the tests are shown in below table and Figure.

Table 30: Types of tests conducted at VL.

S.N.	Test	Disease
1	PAT	Salmonella, Mycoplasma, Brucella
2	Rapid kit test	Bovine TB, B. Brucellosis, *E. canis*, etc

In a total, 665 samples were tested and the details of the result is presented below.

Table 31: Result of different serological tests.

Test	Annual Target	Total sample	Positive	Negative
RBPT	150	217	4	213
***Salmonella* PAT**	150	245	4	241
***Mycoplasma* PAT**	150	245	34	211
Total	**450**	**707**	**42**	**665**

Similarly, 227 samples were collected from Sunsari, Morang, Jhapa, Saptari and Udayapur and were tested for Bovine TB and 183 samples were tested for Bovine Brucellosis and the result is presented below.

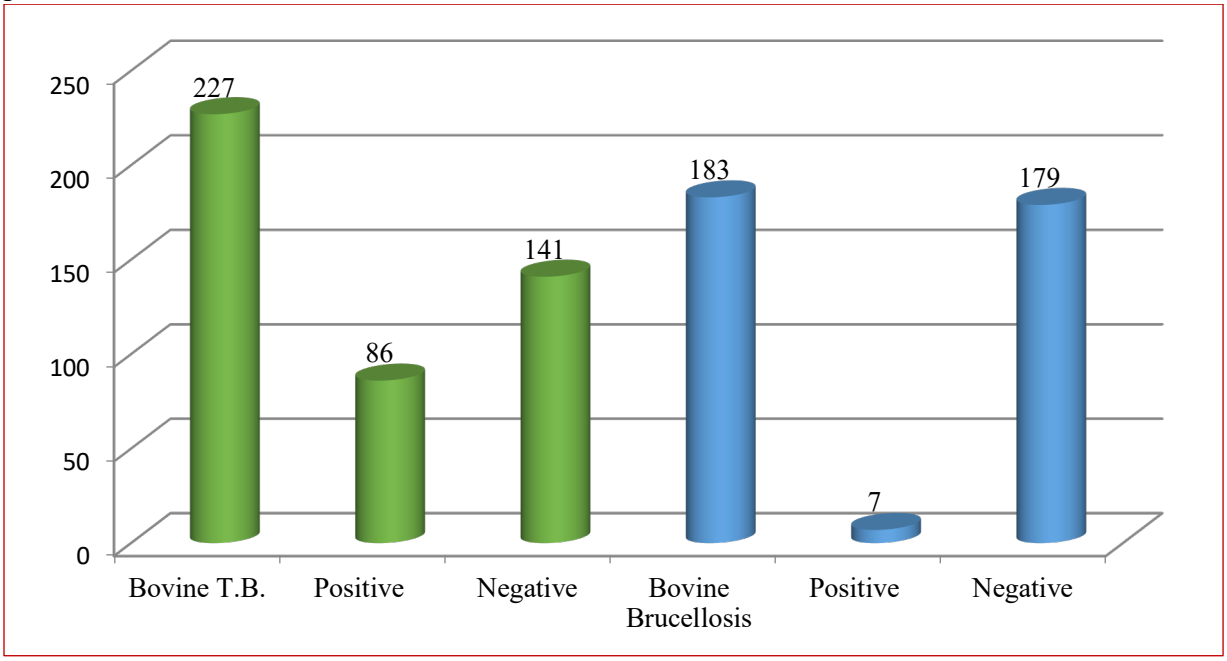


Figure 35: Serological test of the cattle diseases.

Note:

- Bovine Tuberculosis Positive sample from Sunsari, Morang and Jhapa
- Bovine Brucellosis Positive sample from Sunsari and Jhapa

4. Vaccine bank

In this fiscal year, under the national disease control program vaccine bank was managed at VL, Biratnagar for Koshi province. For National Disease Control Program, vaccines were deployed for different diseases.

5. Seromonitoring

In this fiscal year a total of 1500 serum samples were collected to monitor the efficacy of National Animal Disease Control Program from different district of the Koshi province. Out of total sample 618 were for FMD seromonitoring, 1024 for PPR seromonitoring and 29 CSF seromonitoring.

VETERINARY LABORATORY JANAKPUR

1. Introduction

Veterinary Laboratory is situated in Janakpurdham city of Madhesh province. The laboratory is providing diagnostic services eight districts of Madhesh Province and occasionally to adjoinging districts of Bagamati province. The working areas of this Laboratory include Dhanusha, Mahottari, Sarlahi, Rauthat, Bara, Parsa, Siraha, Saptari, and Sindhuli. The laboratory has various units viz pathology, parasitology, microbiology, hematology, biochemistry, sterilization and serology. Histopathological and molecular laboratory test results are obtained by dispatching the relevant specimens to CVL, as these diagnostic facilities are not available in VL, Janakpur at present.

2. Human resources

Table 32: Staffing of VL, Jankpur, Dhanusha

S.N.	Name of staff	Post	Responsibilities
1.	Dr. Rakesh Mahoan Singh	S.V.O.	Chief
2.	Dr. Mukesh Nayak	V.O.	• Laboratory quality management and Planning • Microbiology, Postmortem, Serology and Pathology
3.	Mr. Rakesh Kumar Sah	V.O.	Parasitology and Hematology
4.	Mr. Anirudh Sah	J.T.	Microbiology, Postmortem Serology and Pathology
5.	Mr. Laxmi Mandal	J.T.	• Administration, Store • Parasitology and Hematology
6.	Mr. Satyanarayan Sah	J.T.	Parasitology and Hematology
7.	Mr. Sudhir Kumar Sah	Accountant	Financial
8.	Mr. Kula Nand Jha	Driver	Driving
9.	Mr. Surndra Mishra	Office assistant	Office Attendant
10.	Mr. Binod Kapad	Office assistant	Office Attendant

3. Laboratory service

The routine work of VL, Janakpur mainly involves examination of fecal sample, postmortem examination, sero-surveillance, CMT test of milk samples and bacterial culture and sensitivity testing. Milk samples were tested to isolate and identify the bacteria responsible for Mastitis. Blood samples was brought from different districts of Madhesh Province for HB, PCV, TC, DLC and blood protozoa identification. Serum samples were examined for total protein, calcium, phosphorus, glucose, magnesium, brucellosis, etc. Examination of skin scraping and urine was frequently conducted in VL Janakpur. AST is conducted in the culture positive samples in regular basis.

3.1 Pathology section

3.1.1 Parasitological examination

Parasitological examination from faeces of different animals has been done routinely. The fecal sample are received mainly from farmers directly or referred by VHLSEC of Dhanusha, Mahottari, Sarlahi, Sinnduli, Rauthat and Bara. For the gastro intestinal parasites, Mc'master technique is followed to count eggs per gram (EPG) in feces.

In the F/Y 2079/80 total 1778 fecal samples were received among them 1712 from cattle and 66 from goat were examined. Among these samples 1682 samples (95%) were found positive and 96 samples (5%) were found negative. The results of fecal test revealed that *Fasciola* 670 (38%), *Paramphistomum* 572 (32%), *Ascaris* 261 (14%), *Strongylus* 137 (8%), *Trichuris* 124(7%) and *Toxocara* 14(1%).

Table 33: Average number of eggs found in EPG

Nematodes			
Species	**Mild infection**	**Moderate infection**	**Severe infection**
Cattle/Buffalo	100-300	300-600	600-800
	3	13	6
Sheep/Goat	300-500	1000-1500	2000-3000
	2	7	1
Trematodes			
Cattle/ Buffalo	50-100	100-200	200-400
	23	73	94
Sheep/Goat	50-100	100-200	300-600
	4	9	2

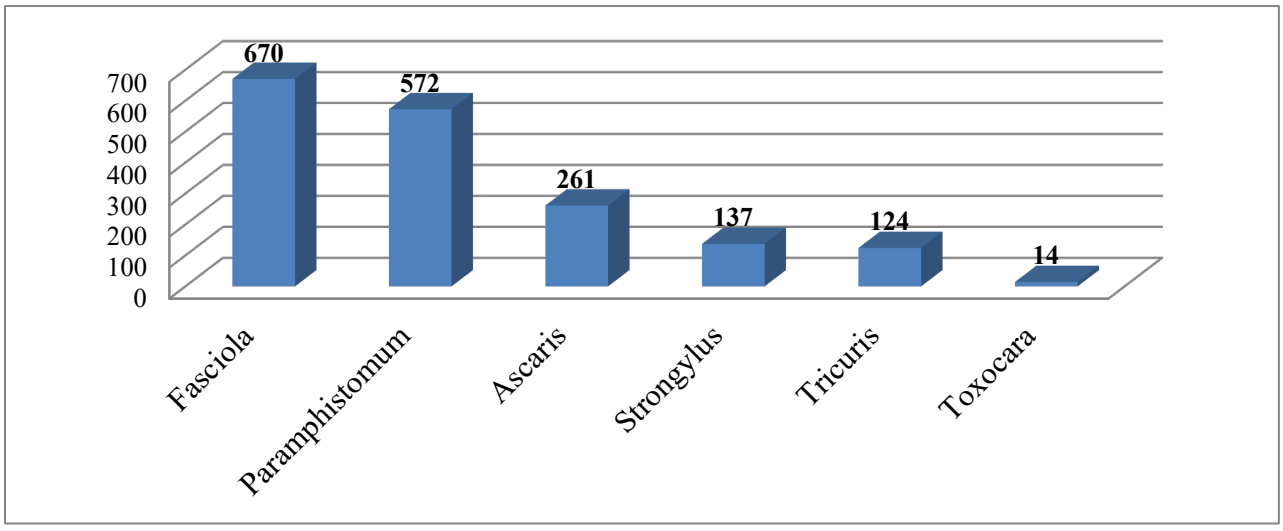


Figure 36: Showing species of parasite found during feacal examination

3.1.2 Haematological examination

Haematological examination TLC, TEC, DLC, PCV and, HB estimation is done routinely in this laboratory.

A total of 120 Blood sample were examined for different blood parameters as well as for blood parasites. Among them 78 samples were found negative for any blood parasites and rest 42 were found positive for different blood parasites.

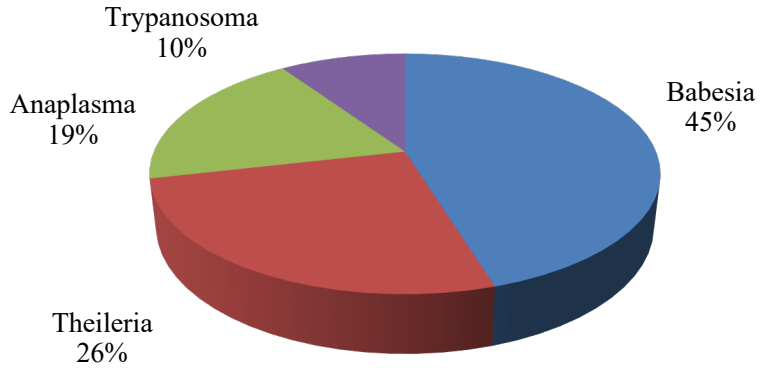


Figure 37: Hemoprotozoans species identified at VL.

3.1.3 Pathological Examination

The pathological examination includes mostly post-mortem examination of the dead birds received from commercial poultry farms Most of the cases were from Dhanusha and Mahottari district and sometimes from Sindhuli, Sarlahi, Rauthat and Sirha district. A total 1032 cases of post mortem examination were presented during the F/Y 2079/80. All the case received were of birds. The status of poultry disease in the area is shown in Table.

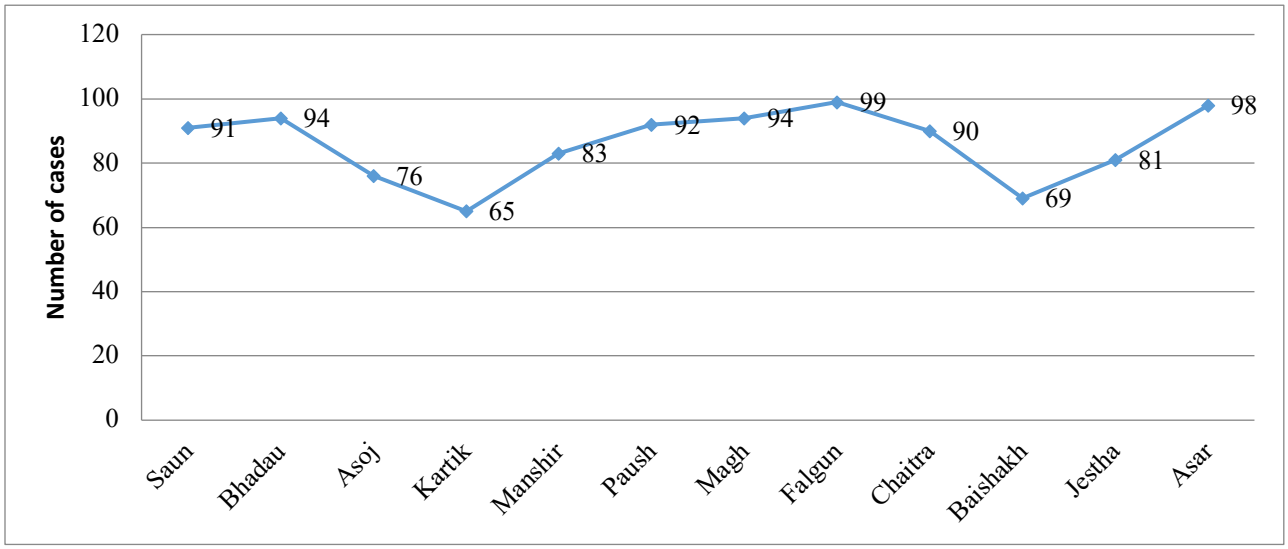


Figure 38: Month wise postmortem case flow rate at veterinary laboratory Janakpur.

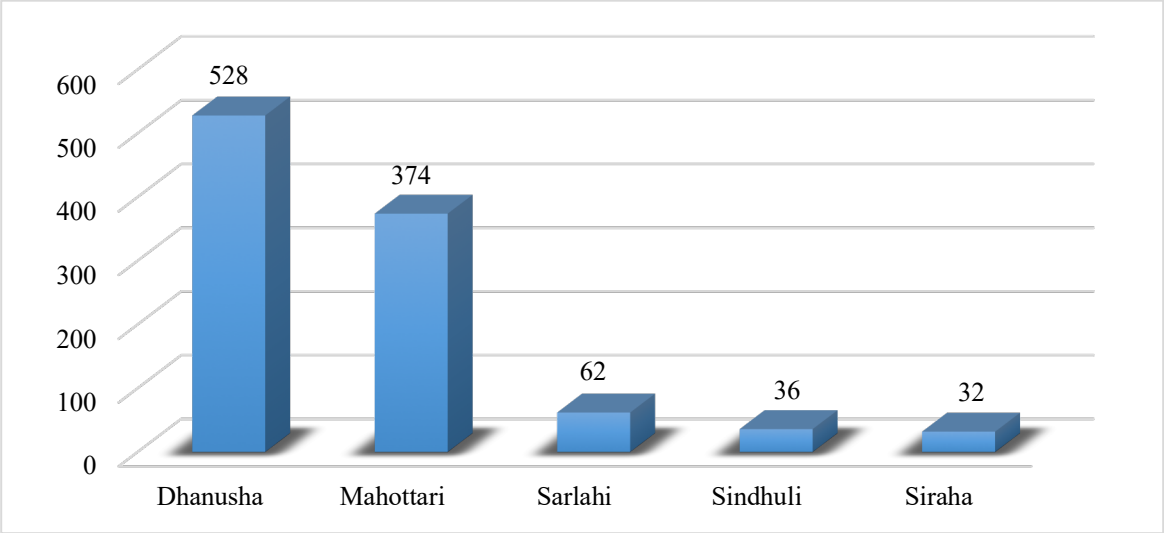


Figure 39: District wise case flow at VL.

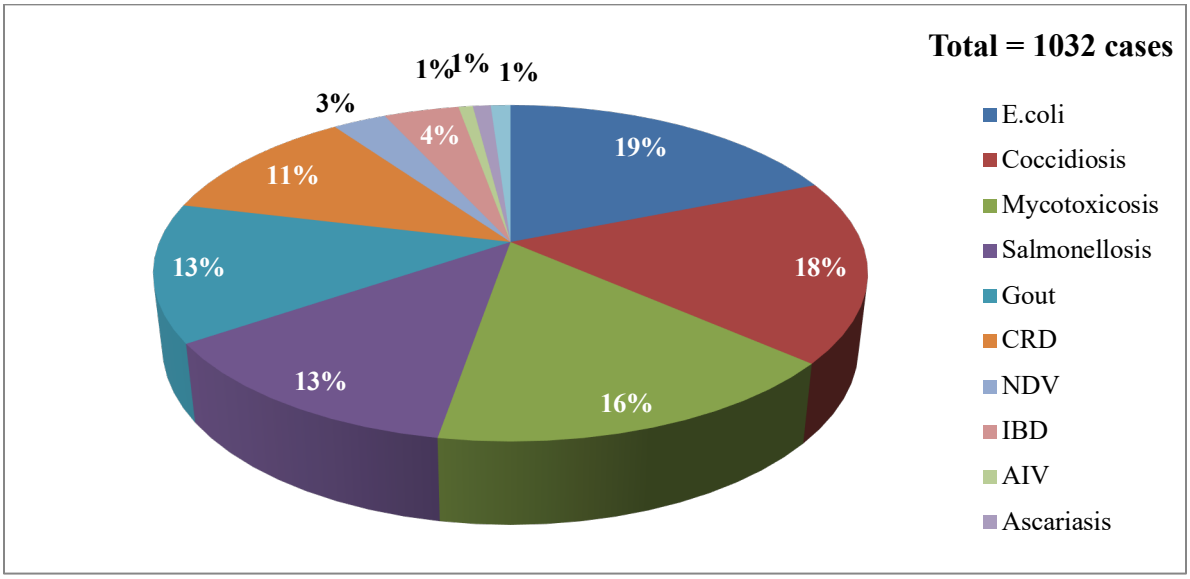


Figure 40: Disease diagnosed based on postmortem examination

3.1.4 Biochemical examination

Table 34: Result of biochemical tests.

Type of animal	Number of Sample	Calcium gm/100ml Normal Value	Calcium gm/100ml Normal Result	Phosphors gm/100ml Normal Value	Phosphors gm/100ml Normal Result
Cow	152	9-12	7-9	4-7	3-6
Buffalo	127	9-12	7-10	4-7	3-6
Goat	21	10-11	9-10	3-11	4-8
Poultry	1	9-12	10-12	4-8	4-7

3.2 Microbiology section

Milk samples were received from farmers at veterinary laboratory Janakpur for the bacterial culture and identification. A total of 356 (Cattle-202 and Buffalo-154) milk sample collected from the following district on which microbiological culture and AST was performed. The most prevalent bacteria isolated show *Staphylococcus, Streptococcus and E. coli* etc.

Table 35: Bacterial species isolated at VL

S.N.	District	No. of samples	Organism
1.	Dhanusha	181	*Staphylococcus, Streptococcus and E. coli*
2.	Mohattari	115	*Staphylococcus, Streptococcus and E. coli*
3.	Sindhuli	9	*Staphylococcus, Streptococcus and E. coli*
4.	Sarlahi	12	*Staphylococcus, Streptococcus and E. coli*
5.	Bara	2	*Staphylococcus, Streptococcus and E. coli*
6.	Parsa	1	*Staphylococcus, Streptococcus and E. coli*

3.3 Molecular biology and serological examination

3.2.1 Serological examination

A total of 204 samples were tested for PPR out of which 47 samples were found positive. Likewise, 172 samples were tested for Avian Influenza Virus (AIV) in which 8 samples were positive which was further confirmed H5N1 by CVL. The 84 samples from the infertile and repeat breeder cattle/buffalo were tested for Brucellosis in which none of the samples were found positive. There was an outbreak of Foot and Mouth Disease in the working area of VL Janakpur from where 184 samples were collected and tested in which 32 samples were found positive for FMD. 207 samples of mycoplasma were tested in which 137 samples were found positive. The 188 samples from suspected Salmonellosis was tested in which 132 samples were found positive and similarly 106 and 108 samples of ND and IBD were tested in which 30 and 42 samples were found positive respectively.

3.4 Sample sent to CVL for further investigation

137 Samples were dispatched to CVL for further testing.

VETERINARY LABORATORY POKHARA

Veterinary Laboratory (VL), Pokhara was established in 2049 B.S. as a Regional Veterinary Laboratory with the objective of disease diagnosis and outbreak investigation in sixteen districts of western development region at that time. After the organization reform as per the constitution the Veterinary Laboratory, Pokhara belongs to the federal government and provides services to 11 districts of Gandaki province and 6 districts of Lumbini Province. The mission of the Veterinary Laboratory, Pokhara is to promote the health of livestock, poultry and companion animals and to ensure safe animal products for the consumer by assisting Veterinary Hospital and Livestock Service Expert Centers (VHLSEC) of these provinces and Livestock Service Sections (AHS) of local levels, veterinarians, clients, and others responsible for animal health in the detection and prevention of disease by conducting responsible investigation on animal diseases and providing accessible, accountable, timely, and accurate diagnostic services. The laboratory is responsible for food safety, outbreak investigation, formulating disease control strategy, prepare epidemiological profile of livestock and poultry diseases and maintain and disseminate the regional epidemiological information database on animal health in the regional as well as in the national networks.

In the present context, commercialization in Livestock and poultry production has increased the challenge for precise and prompt diagnosis of different animal and poultry diseases. For this

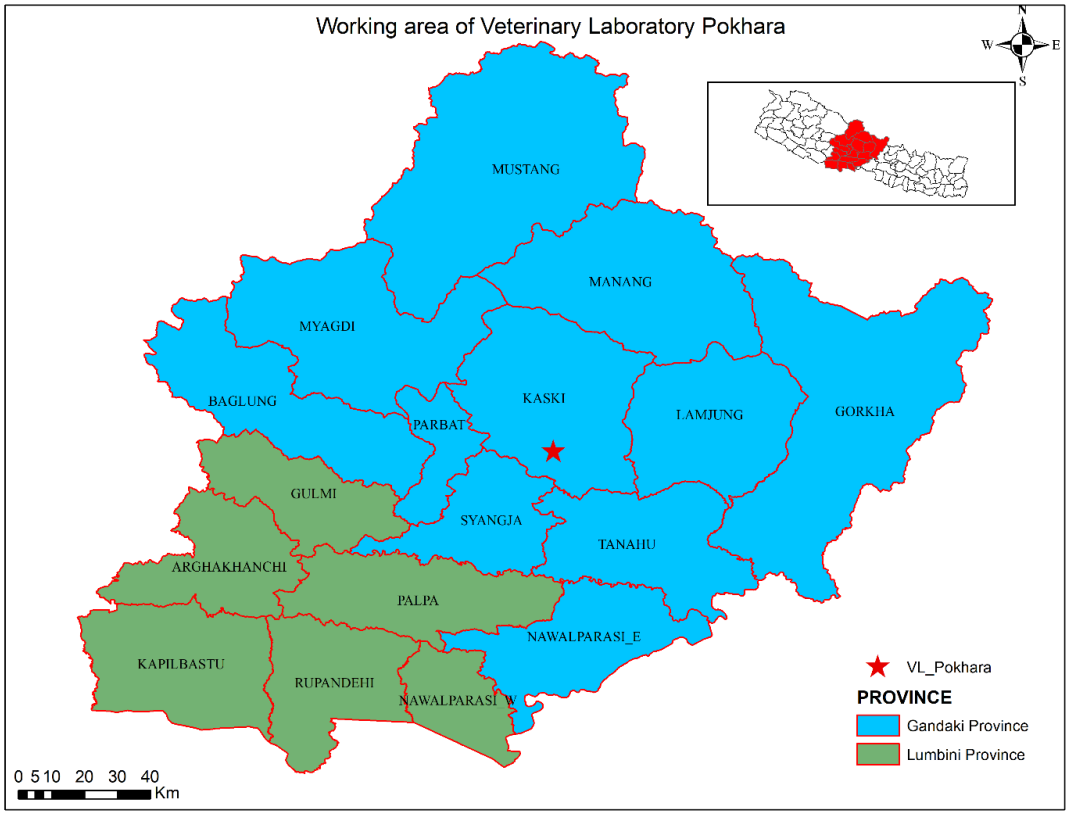


Figure 41: Working areas of veterinary Laboratory, Pokhara

specific reason the molecular basis of disease diagnosis has been set up at Veterinary Laboratory. Currently lab has both Conventional and Real-time PCR facilities.

1. Major Laboratory Facilities of VL, Pokhara

VL, Pokhara, located at Ramghat-12, the center of Pokhara city, provides diversified veterinary laboratory test facilities for the farmers, private veterinary practitioners and VHLSEC of this region. It mainly tests the following categories of the samples under mainly 3- sections

1.1 Pathology

A. **Parasitological Unit:** Parasitological unit tests for internal parasites through microscopic examination of faecal samples. Skin scrappings are also tested for the presence of external parasitic infection e.g., Mange, Sarcopties. It also conducts blood parasite test using thick and thin blood smear examination e.g., *E. canis*, *B. canis*, Trypanosomiasis etc.

B. **Post-mortem Unit**: Pathology unit mainly performs PME on various species of animals and collects appropriate samples for the microbiological, parasitological and molecular biological examination. The unit performs necropsy of morbid and dead birds and animals.

C. **Biochemistry Unit**: Biochemistry unit analyzes mainly serum for the estimation of Ca, P, Mg, TP, Fe, Albumin etc. It also performs urine tests by estimating Albumin, Bilirubin, ketone bodies, urobilinogen etc. using a dipstick test kit.

D. **Hematology Unit**: This unit provides routine hematological examinations of all animals and birds using an automated hemato-analyzer.

1.2 Microbiology

Microbiological unit tests diversified samples like milk, tissues, blood, aspirated fluids, and feces etc. Both aerobic and anaerobic culture facilities are available.

A. Bacteriology and Mycology: It also performs identification of the bacterial and fungal organisms using various biochemical tests, staining, morphology etc. The microbiology unit also performs antibiotic susceptibility test and advice for the appropriate antibiotic for the treatments.

B. Virology: In the virology unit, the laboratory is capable of HA/HI tests.

1.3 Molecular Biology

A. Molecular unit: The Molecular Biology unit is performing PCR test for FMD and PPR disease of ruminants using conventional PCR machine.

B. Serological Unit: Serological unit of VL, Pokhara mainly performs RBPT for *Brucella*, Penside test for PPR, PAT test for *Mycoplasma* and *Salmonella* as well as ELISA for various viral and bacterial diseases of livestock and poultry.

2. Human resource

Table 36: Staffing of VL, Pokhara

S. N	Name of Staff	Designation	Class
1	Dr. Kedar Raj Pandey	Senior Veterinary Officer	Gazetted II
2	Dr. Ganesh KC	Veterinary Officer	Gazetted III
3	Dr. Dilip Kumar Upadhyaya	Veterinary Officer	Gazetted III
4	Dr. Anil Regmi	Veterinary Officer	Gazetted III
5	Mr. Santosh Adhikari	Accountant	Non- gazetted I
6	Ms. Bishnu Kumari Basnet	Veterinary Technician	Non- gazetted I
7	Mr. Bishnu Prasad Kafle	Veterinary Technician	Non- gazetted I
8	Mr. Khim Lal Adhikari	Veterinary Technician	Non- gazetted I
9	Mr. Radheshyam Malla	Asst. Veterinary Technician	Non- gazetted II
10	Ms. Laxmi Sharma	Asst. Veterinary Technician	Non- gazetted II
11	Mr. Gautam GC	Kharidar	Non- gazetted II
12	Mr. Surya Prasad Sapkota	Office Assistant	Classless
13	Mr. Jhalak Bahadur Chhetri	Office Assistant	Classless
14	Mr. Bal Ram Acharya	Light Vehicle Driver	Classless

3. Details of Sample flow in Veterinary laboratory Pokhara 2079/80

In Fiscal Year 2079–80, a total of 8332 samples were received from different districts. Out of the

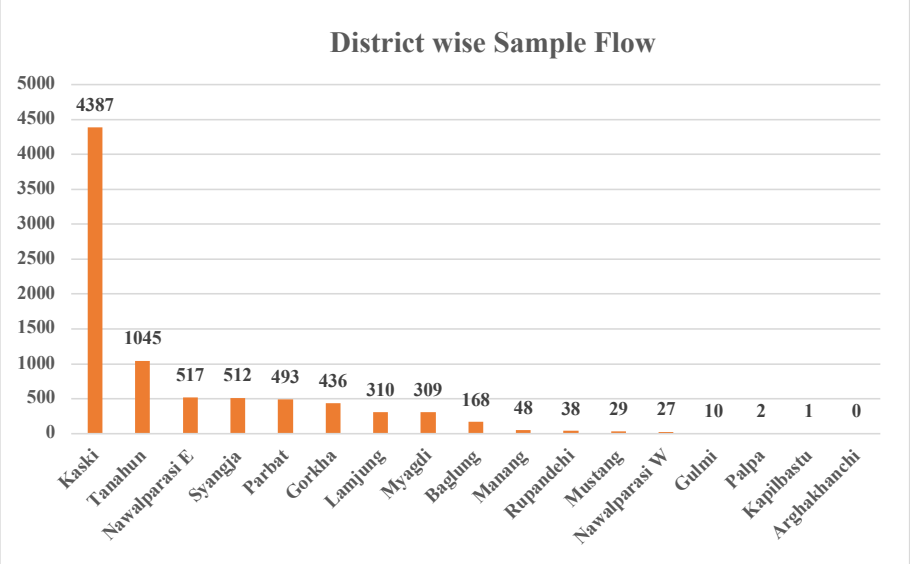


Figure 42: District wise sample flow

total samples received, most (52.65%) were from Kaski district, followed by Tanahun (12.54%), Nawalparasi E (6.20%), Syangja (6.14%), Parbat (5.92%), Gorkha (5.23%), Lamjung (3.72%), Myagdi (3.71%), Baglung (2.02%), Manang (0.58%), Rupandehi (0.46%), Mustang (0.35%), Nawalparasi

W (0.32%), Gulmi (0.12%), Palpa (0.02%), and Kapilvastu (0.01%), as shown in Figure. No samples were received from Arghakhanchi district this fiscal year.

The majority of the 8332 samples collected in this FY were from poultry (3287), followed by cattle and buffalo (2402), goat (2199), dog (237), swine (192), sheep (9), equine (4), rat (1), and wild animal (1) as shown in Figure. This demonstrates the degree to which Farmers in our working region are aware that their animals should only be treated following an accurate test diagnosis.

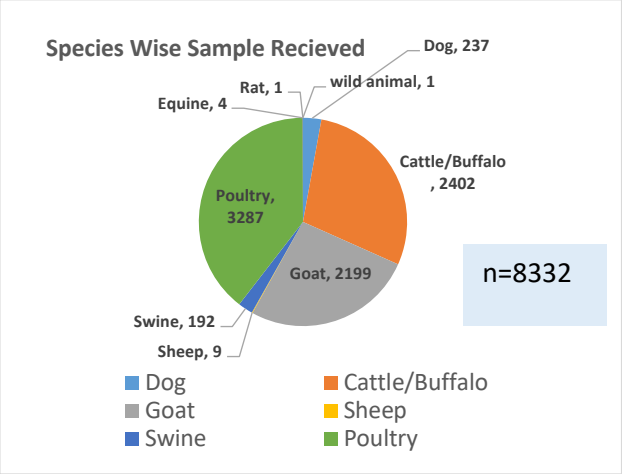


Figure 43: Species wise sample received

As seen in the figure, the highest number of sample flow was in the month of Jestha followed by Asar, Chaitra, and Baishakh. The Sero-monitoring programs for various diseases in cattle, buffalo, goats, sheep, pigs, and dogs accounted for higher number of cases. Except for Ashoj and Falgun due to public holidays, other months show a steady number of sample flow.

Dead bodies sample for necropsy examination, made up most of the samples, followed by milk,

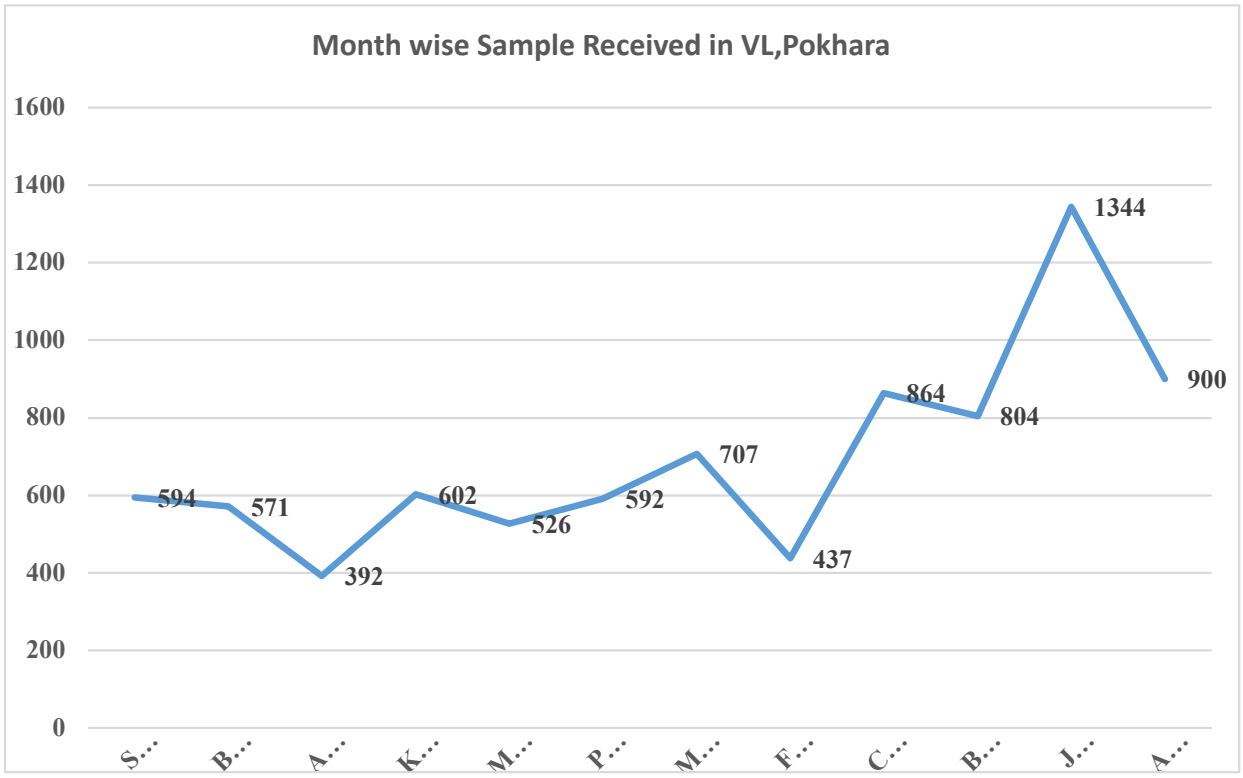


Figure 44: Month wise Sample Received in VL, Pokhara

serum, whole blood, and feces. A nasal swab, urine sample, skin scab, brain sample, tracheal sample, and fresh tissue were also received as shown in the figure.

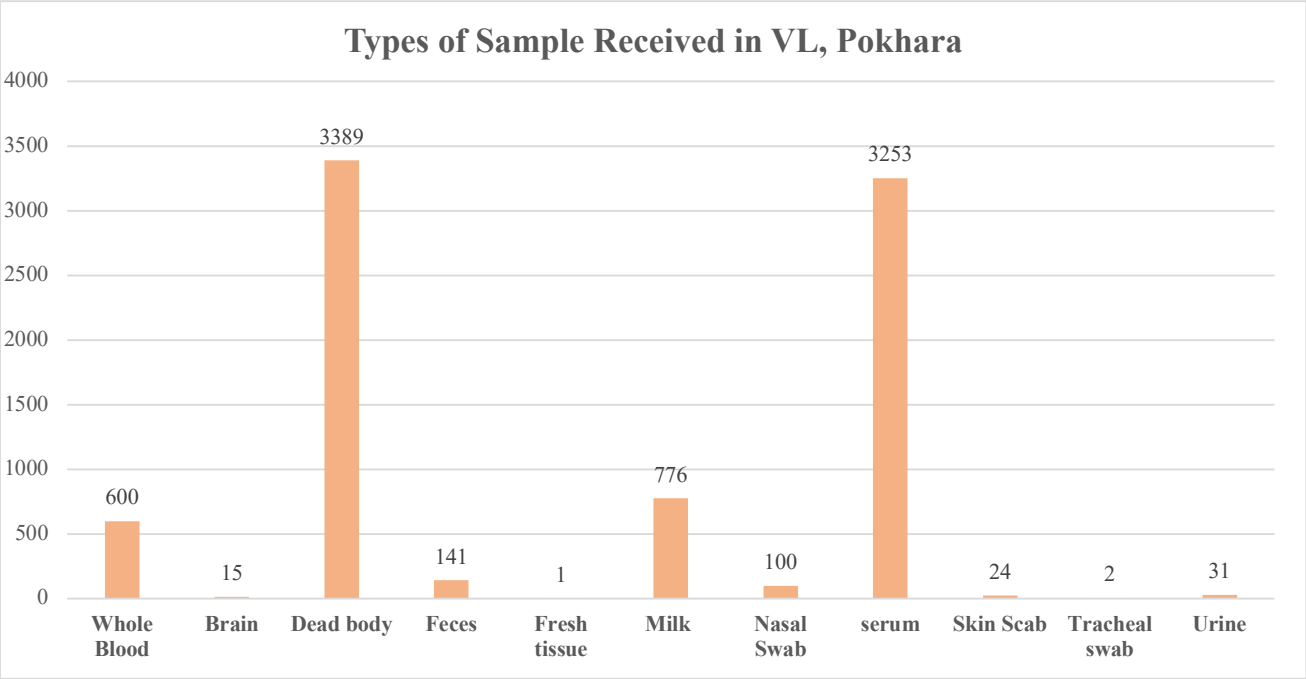


Figure 45: Types of Sample Received in VL, Pokhara

4. Details of Post-mortem in FY 2079/80

Table 37: Details of Post-mortem in FY 2079/80

S.N	Species	Total no of sample	Major PM findings	Remarks
1	Avian	3267		
1.1	Commercial Broilers	2805	Coccidiosis, IBD, Colibacilosis, Mycotoxicity, Salmonellosis, CRD, ND, Omphalitis, Enteritis, Ascites, CCRD	Diagnosed on the basis of history, Post-Mortem Examination, rapid diagnostic test (RDT), culture and ELISA
1.2	Commercial Layers	184	ND, Mycotoxicity, IBH, IB, ALC, Salmonellosis, Colibacilosis,	
1.3	Backyard Chicken	278	Enteritis, Mycotoxicity, Colibacilosis, Histomoniasis, Ascariasis	
2.	Caprine	47	Parasitic infestation, Pneumonia, Urolithiasis, Rabies	
3.	Swine	63	Mycotoxicity, HE, Pneumonitis, Helminth Infestation, ASF	
4.	Canine	12	Rabies	RDT
Total		**3389**		

A total of 3389 postmortem examinations were performed in this fiscal year 2079/80. Out of 3389 PM, 3267 were of poultry birds followed by swine (63), caprine (47) and canine (12). Out of the total poultry cases presented, 85.85 % were broilers, 5.63 % were layers and the remaining 8.5 % were local/backyard chickens as shown in figure. Disease diagnosis was done based on history, PM examination, Rapid test, Culture and ELISA. Most of the cases recorded were from small scale farmers, followed by medium scale farmers and large-scale farmers/entrepreneurs.

5. Poultry Diseases Pattern based on PM

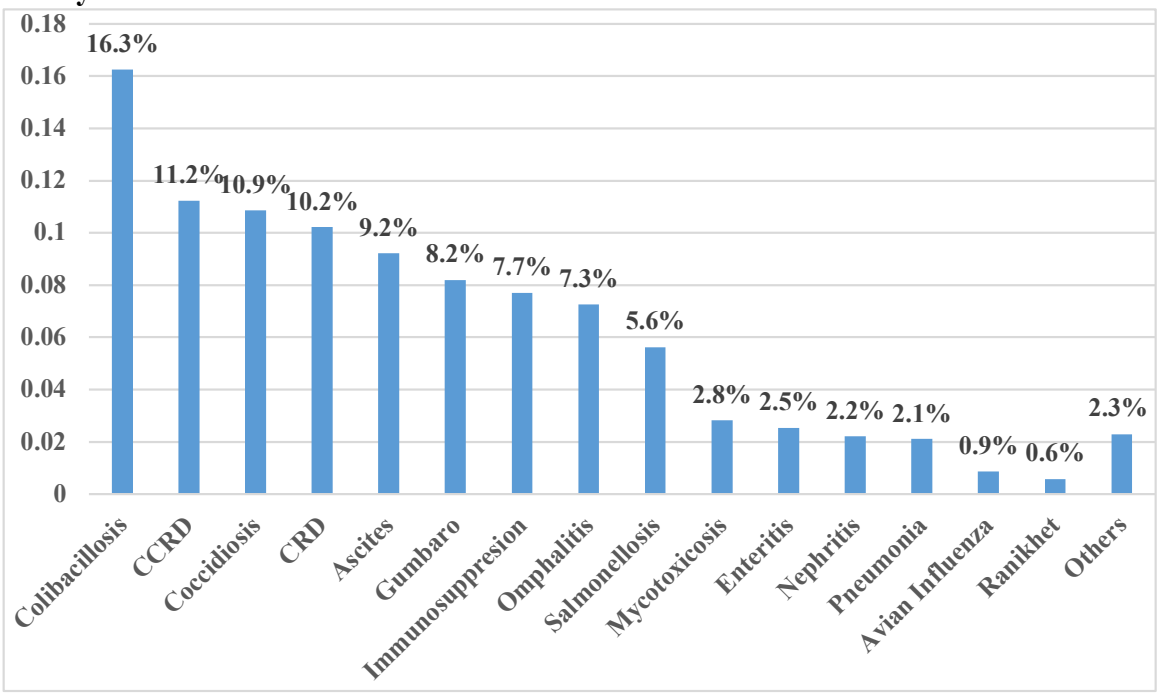


Figure 46: Poultry Diseases Pattern based on PM

On PM examination of Poultry, different bacterial, viral, fungal, protozoal and managemental disease were diagnosed. Out of 3267 dead birds necropsied, bacterial disease was found to be most prominent among the poultry farmers which account for about 50.6% followed by viral, protozoal, fungal and managemental disease. Colibacillosis (16.3%) was found to be most prominent bacterial disease among poultry followed by CCRD, CRD, salmonellosis and omphalitis. Among the viral diseases, IBD was diagnosed in 8.2% of cases followed by avian influenza, and ND. Other diseases diagnosed were Ascites, Toxicity, Enteritis, Nephritis and Pneumonia which is shown in figure.

6. Hematological Test: FY 2079/80

Table 38: Hematological Test: FY 2079/80

S.N.	Species	Total Hematological Test	Total Blood Smear Test	Remarks
1	Buffalo	27	14	
2	Cow	62	42	
3	Dog	181	147	
4	Goat	23	11	
5	Pig	22	13	
Total		**315**	**227**	

6.1. Summary of Blood Smear Test

Table 39: Summary of Blood Smear Test

Species → / Blood Parasites ↓	Dog	Cow	Buffalo	Pig	Goat
Anaplasma spp.	12	1	2	1	
Bacillus anthracis		2		7	
Trypanosoma spp.	1	1			
Babesia spp.	1	2			
Theleria spp.		1			

7. Mastitis Test

Table 40: Mastitis Test Results

SN	Species	SLST Test			Sample received condition		Culture	Organism identified
		Total test	Positive	Negative	nonsterile bottle	sterile bottle		
1	Cattle	441	310	131	376	379	353	Staphylococcus (115), E. coli (80), Streptococcus (51), Enterococcus (18), Pseudomonas (5), No growth (84)
2	Buffalo	314	194	120				
3	Goat	11	5	6	0	2	2	*E. coli* (2)
Total		**766**	**509**	**257**			**355**	

In FY 2079/80, a total of 766 samples were tested by CMT. Out of 766 samples received, 441 were from cattle, 314 were from buffalo, and 11 were from goats. 65% (509) of the milk samples tested were found to be positive for mastitis. Almost half of the samples received were in non-sterile bottles. This means that most of the farmers are still unaware of the milk sample collection and dispatch techniques for CMT, Culture, and AST. Out of 355 samples cultured, no growth was found in 84 samples, and bacterial growth was found in 271 samples. The predominant bacteria isolated in the milk sample were Staphylococcus (115), followed by E. coli (82), Streptococcus (51), Enterococcus (18), and Pseudomonas (5).

8. Major Infectious disease outbreaks in Fiscal year 2079/80

8.1 Anthrax

Blood samples were collected from the suspected swine and cattle with a history of high fever, sudden death, unclotted blood discharge from natural orifices, and absence of rigor mortis for the diagnosis of anthrax. A total of 19 samples were collected from swine and cattle, and blood smears were prepared for observing *Bacillus anthracis* (Causative agent of Anthrax). Out of 19 samples, 8 (1 cattle and 7 swine) samples were positive for anthrax. Cattle from Tanahun (Bandipur), swine from Kaski (Pokhara, Madi, Annapurna), and Tanahun (Suklagandaki) were positive for anthrax

Table 41: Results of samples tested for Anthrax

Species	Area	Number	Positive
Cattle	Tanahu (Bandipur)	1	1
Swine	Kaski (Pokhara, Madi, Annapurna), Tanahu(Sukla Gandaki)	18	7

8.2 African Swine Fever (ASF)

Rapid diagnostic techniques and PCR method were used for ASF Diagnosis. A whole blood sample from orbital sinus was collected from affected pigs. Rapid test followed by PCR was done at CVL for confirmation of ASF. A total of 121 blood samples were collected from Kaski, Myagdi, Baglung, Tanahun, Syangja, and Parbat districts. 69 (57.02%) samples were positive for ASF, with Kaski having the greatest number of positives (52) followed by Myagdi (7), Baglung (6), Tanahun (2), Syangja (1), and Parbat (1). The first case was reported in 2079 (Shrawan 29) from Pokhara-14, Kaski.

Table 42: Results of samples tested for ASF

Area	Sample tested	Positive
Kaski	93	52
Syanga	6	1
Tanahun	3	2
Baglung	10	6
Myagdi	7	7
Parbat	2	1

8.3 Lumpy Skin Disease (LSD)

A total of 126 serum, skin scab and whole blood samples were tested for LSD diagnosis. Serum samples were analyzed for presence of LSD antibodies by antibody ELISA whereas skin scab and whole blood were analyzed using RT PCR at CVL for LSD viral antigen. Out of 126 samples tested, 73 samples (57.93%) were positive for LSD.

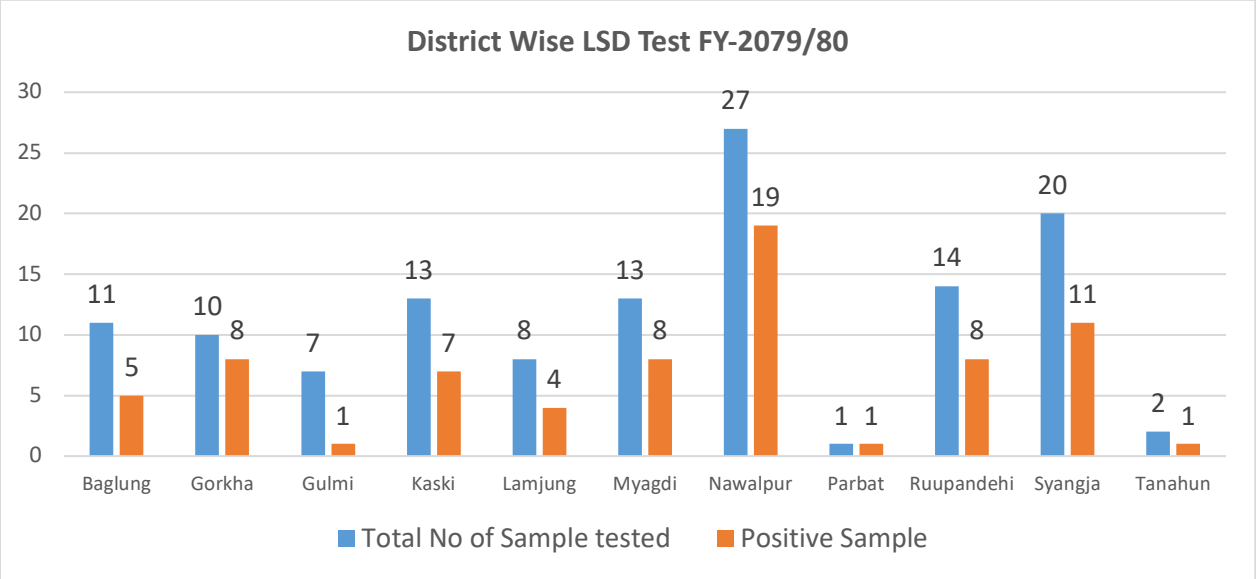


Figure 47: District Wise LSD cases FY-2079/80

District wise sample received and test result is shown in above chart. Out of 126 samples, highest number of samples were received from Nawalpur, 27 samples (19 positive) followed by Syagnja, 20 samples (11 positive), Rupandehi (14 samples, 8 positive), Myagdi (13 samples, 8 Positive), Kaski (13 samples, 7 positive), Gorkha (10 samples, 8 positive), Baglung (11 samples, 5 positive) and one sample positive from each of Gulmi, Parbat and Tanahun.

8.4 Haemorrhagic Septicaemia (HS)

Table 43 : Haemorrhagic Septicaemia (HS)

Species	Area	Tested Samples Number	Positive
Yak	Manang (ward 3,5,8)	7	1

In FY 2079/80, whole blood and bone marrow samples were collected from 7 suspected Yaks for HS from ward 3, 5 and 8 of Manang district. Out of 7 samples tested for HS, 1 bone marrow sample was positive for HS with the remaining 6 samples negative.

8.5 Peste des Petits Ruminants (PPR)

In the FY 2079/80, 131 Nasal swabs were received from goat with a sign of nasal discharge, depression, diarrhea, and death for PPR test. PPR was tested at VL Pokhara by using Rapid test. Out of 131 samples tested, 14 samples were positive for PPR. Details of the PPR outbreak are shown in the figure below.

Table 44 : Peste des Petits Ruminants (PPR)

District	Local level	Ward	Species	Breed
Kaski	Pokhara Metropolitian	32	Goat	Boer Cross
Kaski	Pokhara Metropolitian	22	Goat	Khari
Syangja	Biruwa RM	3	Goat	Khari
Kaski	Pokhara Metropolitian	4	Goat	Boer Cross
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Kaski	Pokhara Metropolitian	14	Goat	
Mustang	Baragung Muktichhetra RM	2	Goat	Chyangra
Mustang	Baragung Muktichhetra RM	2	Goat	Chyangra

8.6 Rabies

41 suspected Brain samples were received in FY 2079/80 from different districts of our working areas for Rabies testing. All the samples were tested by Rapid test methods. Out of 41 samples, 16 samples were tested positive for Rabies.

District and specise wise Rabies outbreak in FY 2079/80 is shown in below Charts. Out of 16 samples positive, 6 samples were positive from Tanahun District followed by 3 from Kaski, 2 from each of Syangja, Baglung and Gulmi district and 1 from Kapilvastu District. Out of 16 Positive

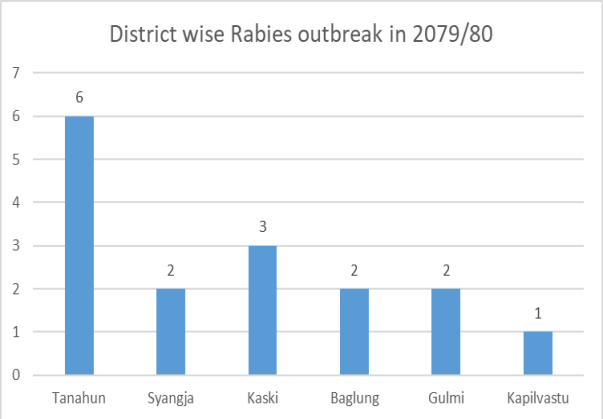


Figure 48: District wise Rabies outbreak in 2079/80

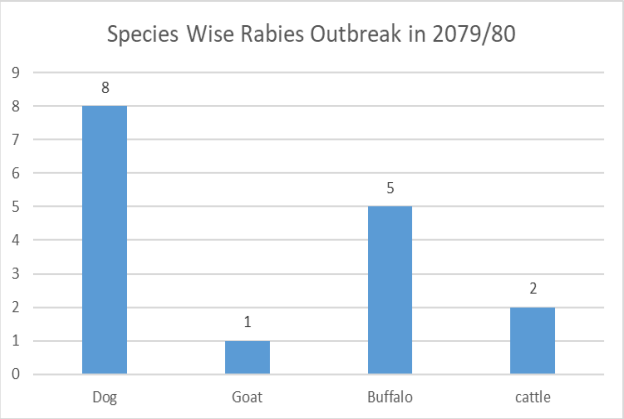


Figure 49: Species wise Rabies outbreak in 2079/80

samples, 8 samples were positive from Dog, 5 samples positive from Buffalo, 2 Positive from cattle and 1 positive from Goat.

8.7 Foot and Mouth Disease(FMD)

In the fiscal year 2079/80, a single outbreak of FMD occurred in cattle of Machhapuchchre-04, Kaski in the Month of Jestha.

8.8 Classical Swine Fever (CSF)

19 samples were tested for CSF in FY 2079/80 with only 2 samples positive. The outbreak area was Pokhara-14 Kaski.

8.9 Avian Influenza Virus (Type A H9)

In FY 2079/80, tracheal samples were collected from 399 poultry birds with a sign of anorexia, respiratory distress, high mortality and decreased in egg production. All the 399 samples were tested by using Rapid test methods. Out of 399 samples tested, 28 samples (7%) were positive in Rapid test kit. All the 28 samples positive in Rapid test kit were dispatched to CVL for differentiation of H5 or H9. All the 28 samples were positive for H9 in PCR examinations. Out of 28 samples positive for H9, 21 samples were from Broiler, 6 samples were from Layers and 1 from Backyard poultry. Out of 28 samples positive, the highest positive samples were from Kaski district i.e. 23 followed by 1 sample positive from each of the Syangja, Tanahun, Baglung and Parbat.

Table 45 : Avian Influenza Virus (Type A H9)

Area	Total Samples	Positive
Kaski	308	23
Syanga	28	1
Tanahun	32	1
Baglung	5	1

Myagdi	12	-
Parbat	11	1
Total	399	28

8.10 Brucellosis:

63 samples were tested by Rose Bengal Plate Agglutination Test (RBPT) and ELISA for Brucellosis. All the samples were negative for brucellosis.

Table 46: Brucellosis test result

District	Samples tested	Positive
Baglung	4	-
Kaski	48	-
Nawalparasi E	2	-
Syangja	1	-
Tanahun	8	-

Table 47: Brucellosis test result in species

Species	Samples Tested	Positive
Buffalo	8	-
Cattle	19	-
Goat	34	-
Pig	2	-

9. Summary of Rapid Test Report: FY 2079/80

VL, Pokhara provides rapid diagnostic tests for various animal and poultry diseases. The results obtained from RDTs are confirmed by other tests such as culture, ELISA, PCR and other suitable tests. The samples that could not be tested in the laboratory due to lack of resources or that are to be confirmed as per standard are dispatched to CVL and tested there.

Table 48 : In fiscal year 2079/80, VL, Pokhara performed the RDTs of following diseases.

Name of Disease/ Virus/ Bacteria/ Protozoa	Total number of samples tested	Positive	Negative	Positive (%)
Avian Influenza virus (AIV)	399	28	371	7.02
Infectious Bursal Disease virus (IBDV)	144	82	62	56.94
Infectious Bronchitis virus (IBV)	30	2	28	6.67
Rabies Virus	41	16	25	39.02
PPR Virus	131	14	117	10.69
African Swine Fever virus (ASFV)	121	69	52	57.02
Classical Swine Fever virus (CSFV)	19	2	17	10.53
Salmonella pullorum	513	194	319	37.82
Mycoplasma gallisepticum	512	174	338	33.98
Rose Bengal Plate Test (RBPT)	63	0	63	0
Ehrlichia canis	19	11	8	57.89
Anaplasma phagocytophilum/Anaplasma platys	11	4	7	36.36

10. Microbiological test

In Microbiology Unit, All the samples brought were subjected to cultural examination on Nutrient Agar, Mac Conkey Agar and Blood Agar media. They were incubated at 37º C for 24 hours.

Cultural isolates were identified based on colony characteristics, Gram's staining, and different biochemical tests; IMViC, Oxidase, Catalase and Motility.

10.1 Bacterial isolates and AMR patterns

Table 49: Bacterial isolates and AMR patterns

Type of sample	No. of sample tested	Organism isolated	No growth	Type of Bacteria isolated
Milk	353	269	84	*E. coli, Streptococcus spp, staphylococcus spp,Pseudomonas*
Poultry liver/caeca/spleen	49	26	23	*E coli, Salmonella spp, Enterococcus spp*
Bone marrow	3	1	2	*Pasteurella hemolytica*
Whole blood	6	1	5	*Staphylococcus spp*
Lungs	5	1	4	*Enterococcus spp*
Pus	4	4	0	*Staphylococcus spp, Enterococcus spp*
Nasal swab	3	0	3	
Total	**423**	**302**	**121**	

10.2 Different Bacterial isolates from Milk and Poultry Sample

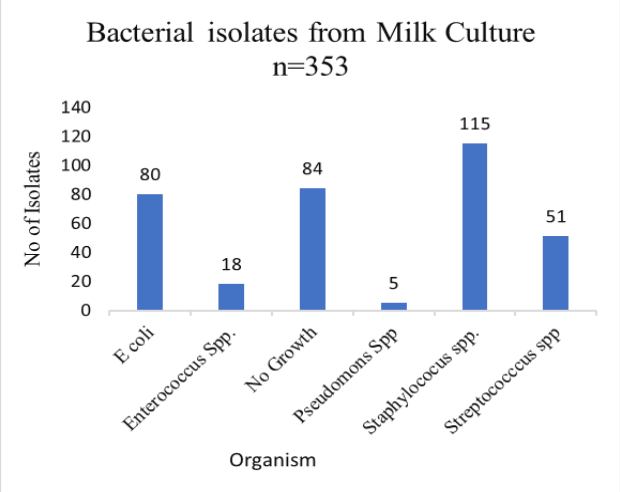


Figure 50: Bacterial isolates from Milk Culture

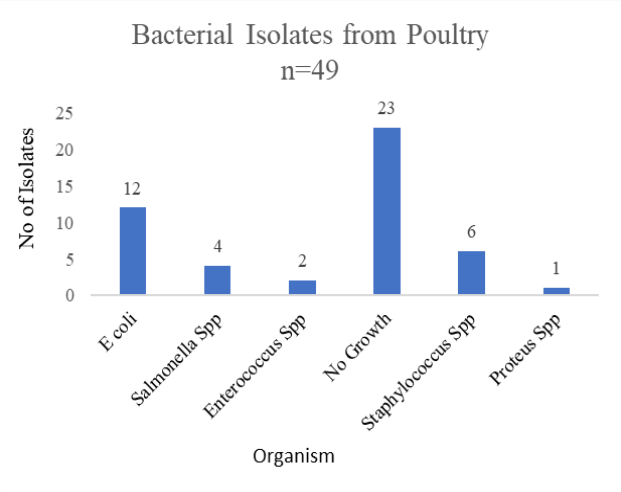


Figure 51: Bacterial isolates from Poultry

Table 50: Anti-microbial resistant patterns for E. coli organism from milk and poultry liver sample

SN	Antibiotic Name	Sensitivity %	Intermediate %	Resistance %
1	Ceftriaxone	58	7	35
2	Ciprofloxacin	24	44	32
3	Enrofloxacin	24	41	35
4	Amikacin	62	25	13
5	Gentamicin	59	14	27
6	Oxytetracycline	25	63	12

11. Sero-monitoring of Fiscal Year: 2079/80

A cross sectional study was carried out from April 2023 to July 2023 in 9 districts of Gandaki Province. A multistage sampling technique was used. One local level from each district was selected based on history of at least 3 successive years of vaccination. Wards and livestock farms were randomly selected from respective local levels. Based on number of animals vaccinated 1.5% sheep and goat were sampled. Similarly, 0.5% of vaccinated cattle and buffalo were sampled for this study purpose.

While selecting study area the reference was taken from secondary data of VHLSEC of respective districts and Livestock Service Section (LSS) of respective local levels and other details were obtained from farmers of study area from where samples were collected.

12. Samples and data collection

About 5 ml blood was aseptically collected by venipuncture in a sterile 10 ml syringes and transferred to clot activator tube. Then the blood samples were left to clot at room temperature overnight, and serum was obtained. Serum samples were then stored at -20^{0}C until testing. The serum samples were taken after 3 weeks of vaccination in animals. The questionnaire was developed to gather information related to the status of farms and animals during sample collection and is included in appendix-I.

13. Laboratory Investigation

Laboratory work was performed at Veterinary Laboratory, Pokhara. ID-Screen® FMD Type O Competition ELISA kit manufactured by "ID.vet Innovative Diagnostics, France" was used for the detection of antibodies in individual serum for sero-monitoring of FMD and ID-Screen® PPR Competition ELISA kit manufactured by "ID.vet Innovative Diagnostics, France" was used for the detection of antibodies in individual serum for sero-surveillance of PPR. They both result within 4 hours. The test procedure was done in accordance with the procedure detailed in the manufacturer's protocol.

A total of 2590 serum samples were collected (980 samples for FMD sero-surveillance from cattle and buffaloes, 1290 samples for PPR sero-surveillance from sheep and goats, for the test from vaccinated animals.

13.1. Test result and Seropositivity of FMD Vaccine

The ID-Screen® FMD Type O Competition ELISA kit was used for the detection of antibodies in individual serum. Out of 980 samples tested for antibodies against the Foot and Mouth Disease Virus (FMDV) serotype O by competitive ELISA, 833 samples (85.00%) samples were found to be positive while 147 samples (15.00%) tested were found to be negative (Table 3).

Table 51: Test result and Seropositivity of FMD Vaccine

Total number of samples	**Test result**		**Seropositivity (%)**
	Number of positive samples	Number of negative samples	
980	833	147	85.00 %

Table 52: Test result of sero-positivity of FMD Vaccine among different districts

District	Total number of samples	Number of positive samples	Seropositivity (%) 2079.80	Seropositivity (%) 2078.79
Kaski	96	52	54.17	76.50
Tanahun	202	185	91.58	98.39
Nawalpur	191	185	96.86	97.00
Gorkha	65	54	83.08	37.50
Lamjung	92	80	86.96	96.15
Myagdi	84	65	87.84	98.00
Parbat	104	100	96.15	94.44
Baglung	74	65	77.38	96.15
Syangja	72	47	65.28	99.00
Total	**980**	**833**	**85**	**88.36**

The overall seropositivity of FMD vaccine was found to be 85%. About (96.86%) animals were found seropositive in Nawalpur district followed by 96.15% in Prabat district. The least number of seropositive cattles and buffaloes (54.17%) were found in Kaski followed by Syangja (65.28) district.

13.2. Test result and Seropositivity of PPR Vaccine:

The ID-Screen® PPR Competition ELISA kit was used for the detection of antibodies in individual serum. Out of 1290 samples tested for antibodies against the Peste des Petits Ruminants (PPR) virus by competitive ELISA, 1020 samples (79.06%) samples were found to be positive while 270 samples (20.94%) tested were found to be negative (Table 5).

Table 53: Test result and Seropositivity of PPR Vaccine

Total number of samples	Test result		Seropositivity (%)
	Number of positive samples	Number of negative samples	
1290	**1020**	270	79.06 %

Table 54: Test result of seropositivity of PPR Vaccine among different districts

District	Total number of samples	Number of positive samples	Seropositivity (%) 79.80	Seropositivity (%) 78.79
Kaski	92	35	38.04	67.50
Tanahun	221	180	81.39	91.94
Nawalpur	303	290	95.56	75.31
Gorkha	162	143	88.27	87.78
Lamjung	104	87	83.65	83.70
Myagdi	100	91	91	86.96
Parbat	113	94	83.18	81.70
Baglung	92	37	40.21	80.77

District	Total number of samples	Number of positive samples	Seropositivity (%) 79.80	Seropositivity (%) 78.79
Syangja	103	63	61.16	78.01
Total	**1290**	**1020**	**79.06**	**80.30**

The overall sero-positivity of PPR vaccine was found to be 79.06%. 95.56 % were found seropositive in Nawalpur district followed by 91.00% in Myagdi district and 88.27% in Gorkha district. The least number of seropositive goats and sheeps (38.04%) were found in Kaski district followed by Baglung 40.20% and Syangja 61.16 %.

The sero-conversion both FMD and PPR vaccine is satisfactory in Gandaki Province. In comparison to fiscal year 2078.79 the overall sero-conversion has decreased for both FMD and PPR vaccine by 3.36% and 1.24% respectively. The Seroconversion of FMD and PPR vaccine is low in Kaski and Syangja district whereas seroconversion of PPR is also low in Baglung. Vaccine cold chain, vaccine transportation, high ratio of animal replacement in a herd and difficulty in identification of vaccinated animals due to lack of animal traceability and poor recording system could be the possible reason for low sero-conversion.

VETERINARY LABORATORY SURKHET

1. Introduction

Veterinary Laboratory (VL), Surkhet is located at Birendranagar Municipality-7, of the Karnali Province. Since its establishment in F/Y 1988/1989 AD, it has been providing diagnostic services in 10 districts of Karnali Province and six districts of Lumbini province.

The mission of the VL, Surkhet is to promote the health of livestock, poultry and ensure safe animal products for consumer by assisting VHLSEC, local level veterinarians, animal health workers, and others who are responsible for animal health in detection and prevention of animal diseases.

2. Staffing of Veterinary Laboratory, Birendranagar, Surkhet

Table 55: Staffing of Veterinary Laboratory, Birendranagar, Surkhet

S.N.	Designation	Class	Number of Post	Fulfilled	Remarks
1	Senior Veterinary Officer	Gaz.2	1	1	
2	Veterinary Officer	Gaz.3	3	3	
3	Animal Health Technician	Non.Gaz.1	3	3	
4	Junior Animal Health Technician	Non.Gaz.2	2	2	
5	Accountant	Non.Gaz.1	1	1	
6	Kharidar	Non.Gaz.2	1	1	
7	Driver	Class less	2	1	1 contract
8	Office Helper	Class less	2	0	2 contract
Total			**15**	**12**	

3. Laboratory Services

3.1 Pathology section

3.1.1 Parasitological examination

Parasitological unit examines the fecal samples of animal species using different methods to estimate the parasitic burden of the nematode, trematode and cestode by various methods. A total of 1126 faecal samples were examined and 831 were found positive for various internal parasites. Among them *Fasciola, Coccidia*, *Haemonchus, Strongylus, Paramphistomum, Trichuris,* were found major internal parasites identified. Reports of faecal examination are presented in table below.

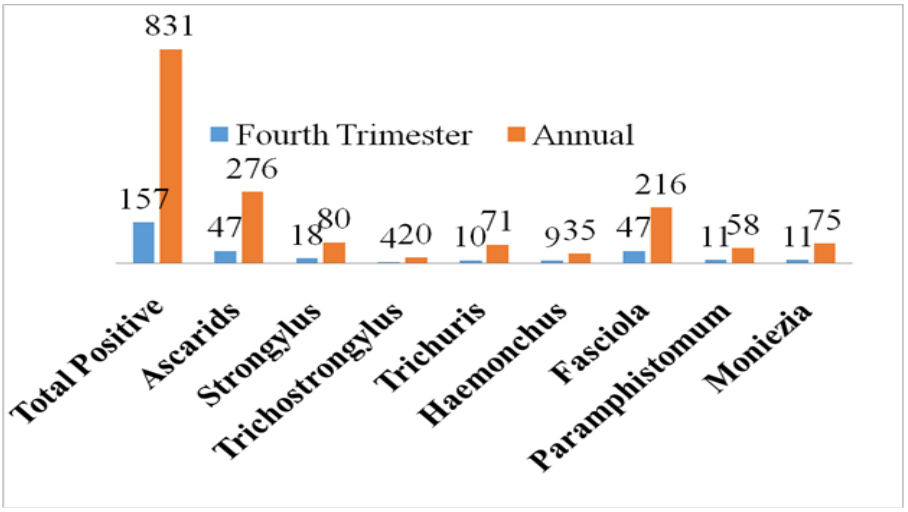


Figure 52: Fecal examination of different animal conducted at VL, Surkhet

3.1.2 Blood Parasite Examination

Out of 132 samples, 15 sample were positive. Among positive sample, 7 cases of Babesia spp were found in 3 sample from cattle and 4 sample from buffalo. In blood sample of horse 10 sample were found positive for trypanosoma (8 sample of Banke and 2 sample of Bardiya).

3.1.3 Postmortem examination

A total of 577 birds were brought at VL for postmortem examination. The pathological examination includes mostly post-mortem examination of poultry received from commercial poultry farms. Most of the cases were brought from Birendranagar Municipality and surrounding Rural Municipalities (Lekbesi, Simta, Bheriganga, Gurbakot, Barahatal) and some cases involved from Municipality and rural Municipality of Banke, Bardiya and Dang District. The details of the disease diagnosed are presented in the figure below.

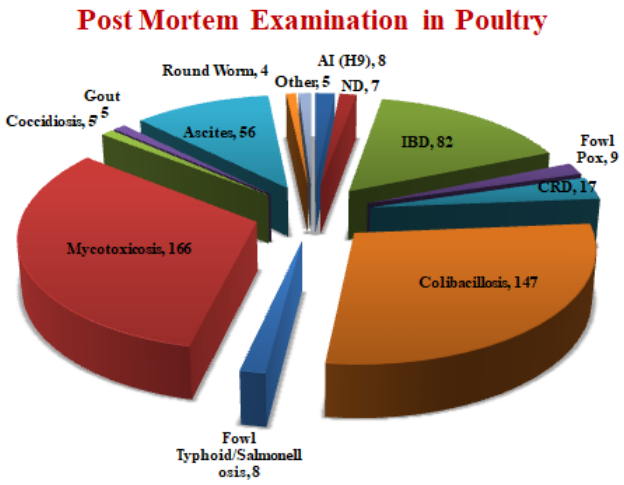


Figure 53: Postmortem examination in Poultry

Table 56: Animal species brought at VL for PM examination

S.N.	Species	Total samples (Fourth trimester)	Total samples (Annual)	Major diseases identified
1	Commercial Broiler	123	377	Colibacillosis, ND, AI, IBD, IB, Fowl Pox, CRD, Fowl Typhoid/Salmonellosis, Mycotoxicosis, Coccidiosis, Ascites, Gout Round Worm, Tape Worm, Stress
2	Commercial Layers	27	95	
3	Commercial Parents	2	12	
4	Local Birds	16	28	
5	Duck	2	5	
6	Pheasent	1	2	
7	Goat	2	17	Endoparasite
8	Pig	3	28	ASF, CSF, Pneumonia, Endoparasite
9	Dog	3	13	Parvo-enteritis, Rabies
Total		**179**	**577**	

3.1.4 Skin Scraping Test

Out of 30 samples of skin scrapings, 9 samples were found positive (4 Sarcoptic mange mite in goat, 4 Demodex mite in dog and 1 fungal infection in Buffalo).

3.1.5 Clinical-Hematological Test

The Hematology unit is providing routine hematological testing of all the animals and poultry. Hematological parameters include Total Erythrocyte Count (TEC), Total Leukocyte Count (TLC), Differential Leukocyte Count (DLC), Hemoglobin estimation (Hb), Packed Cell Volume (PCV), and staining of blood smears for blood protozoa. Anaplasma and Babesia species was found in blood samples collected from improved breed of cattle from Bardiya and Surkhet districts. Altogether 63 sample were tested in clinico-hematological unit.

3.1.6 Urine Test

Out of 71 urine samples tested 5 samples were found with abnormal result.

4. Microbiological examination

In a total 574 samples were received at VL for bacterial culture and AST. Similarly, 52 samples were received for fungal culture.

Out of 574 samples submitted at microbiology, 508 and 21 samples produced bacteria and fungus respectively. The distributions of major bacterial isolates are described in table below.

Table 57: distributions of major bacterial isolates

SN	Name of Bacteria Isolated.	No of Isolates
1	E. coli	342
2	Salmonella	5
3	Streptococcus	5
4	Staphylococcus	49
5	Pseudomonas	4
6	Enterococcus	5
7	Bacillus	6
8	Klebsiella	10
9	Campylobacter	18
10	Fungus	21
11	No Growth	99
Total (isolates in mixed infection)		**374**

4.1 California Mastitis Test (CMT)

CMT was used to diagnose clinical and sub clinical mastitis of animal in laboratory as well as in field level. In total 304 milk sample were tested, among which 93 sample were found positive.

Table 58: California Mastitis Test Result

CMT Test	Positive	Negative	Total
No of Sample tested	93	211	304

4.2 Surveillance on mastitis

Table 59: Test results on samples collected during active and passive surveillance

Address	Species	CMT Total	CMT Positive	CMT Negative	Gram Positive	Gram Negative	Isolated Spp
Surkhet	Cow, Buffalo	234	73	161	38	23	*E.coli*, Staph, Strepto, Pseudomonas, Klebsiella
Bardiya	Cow	2	2	0	0	1	*E.coli*
Banke (khajura, Nepalgunj)	Cow	26	12	12	14	0	
Salyan	Buffalo	42	6	36			
Total		304	93	209	52	24	

4.3 AMR Active Surveillance in poultry

Since 2021, VL Surkhet is participating in AMR surveillance under the Central Veterinary Laboratory. Fleming Fund Fellowship (AMR Lab Fellow) was provided to Bacteriology Unit staff.

Table 60: Result of AMR Surveillance

S.no.	Location	Gentamicin		Ciprofloxacin		Tetracycline		Cotrimoxazole		Ceftriaxone		Ampicllin	
		S	R	S	R	S	R	S	R	S	R	S	R
1.	Bheriganga	1	10	7	4	0	11	0	11	1	10	0	11
2.	Gurvakot	1	12	7	6	2	11	0	13	0	13	0	13
3.	Melkuna	0	11	5	6	0	11	0	11	1	10	0	11
4.	Birendranagar	0	15	6	9	0	15	0	15	8	7	0	15
Total		**2**	**48**	**25**	**25**	**2**	**48**	**0**	**50**	**10**	**40**	**0**	**50**

4.5 Biorepository Management in Laboratory

In Lab more then 200 bacteria isolate from passive and active surveillance program has been preserved. Some of the samples are sent for the biobank of CVL. Isolates of even anaerobic bacteria has also been isolated. Some of the bacterial isolates was sent to the Australian Doherty Laboratory for whole genome sequencing.

5. Virological examination

A total of 650 samples of different domestic animal were collected at field level for virological testing using lateral flow assay tests kits. The results of the test are presented in the table below.

Table 61: Result of Virological testing

S. N	Rapid Test	Target	Positive	Negative	Total
1	AI	200	10	577	587
2	IBD	70	27	48	75
3	ND	150	8	577	585
4	IB	70	0	70	70
5	Rabies	20	34	9	43
6	PPR	100	2	100	102
7	ASF	20	27	3	30
8	Parvoenteritis	10	2	8	10
9	Canine Distemper	10	2	8	10
Total		**650**	**112**	**1400**	**1512**

6. Serological examination

A total of 504 serum samples of different domestic animal were collected and tested at VL, Surkhet. The results of the test are presented in the table below.

Table 62: Result of plate agglutination test (FY 2079/080)

S.N.	Conducted Test	Total Sample Tested	No of Positive Sample	Positive %
1	Brucella	202	6	2.97
2	Salmonella	100	24	24
3	Mycoplasma	100	21	21
4	PPR	102	2	1.96
5	**Total**	**504**	**53**	**10.51**

6.1 PPR Seromonitoring Program

Under the National PPR Control program, Department of Livestock Services provided 1636000 doses of PPR vaccine to be used in 16 districts of this region. VL, Surkhet supported the program by sero-monitoring from the vaccinated animals. District-wise collection of serum sample and laboratory test result is presented in the table below.

Table 63: PPR seromonitoring test result

SN	Name of District	Target of Sample	Collected Sample	Test Sample	Positive	Negative	Positive Percent
1	Banke	200	204	204	62	142	30.39
2	Bardiya	100	92	92	72	21	78.26
3	Dailekh	100	92	92	24	68	26.08
4	Jajarkot	100	92	92	79	13	85.86
5	Jumla	150	122	122	99	7	81.14
6	Kalikot	100	62	62	55	6	88.70
7	Pyuthan	100	92	92	54	38	58.69
8	Mugu	200	184	184	155	13	84.23
9	RukumPaschim	200	184	184	123	14	66.84
10	Salyan	100	92	92	79	13	85.86
11	Surkhet	100	92	92	76	16	82.60
12	Rolpa	100	92	92	61	31	66.30
	Total	**1550**	**1400**	**1400**	**939**	**461**	**67.07**

6.2 CSF Sero-monitoring

Department of Livestock services provided 150000 dose of CSF vaccine to be used under national swine fever control program. VL, Surkhet supported the program by sero-monitoring.

Table 64: District wise collection of serum sample.

S. N.	District	Serum collection		Swine Fever AbElisa Test	Positive Sample	Percentage
		Target	Progress	No of Sample		
1	Rolpa	60	39	39	37	94.87

2	Rukum Paschim	60	53	53	30	56.66
3	Dang	60	92	92	69	75
	Total	**180**	**184**	**184**	**136**	**73.91**

6.3 FMD Sero-monitoring Program

Department of Livestock services distributed 150000 doses of FMD vaccine under National FMD control program to be used in Banke, Bardiya and Dang. VL, Surkhet supported the program by sero-monitoring. The table below shows district wise collection of serum sample.

Table 65: District wise sample collection and test result.

S. N.	District	Serum collection		FMD O type Elisa Test	Positive Sample	Percentage
		Target	Progress	No of Sample		
1	Banke	100	92	92	82	89.13
2	Bardiya	100	92	92	86	93.47
3	Dang	200	184	184	143	77.71
	Total	**400**	**368**	**368**	**311**	**84.51**

6.4 ND Sero-monitoring Program

Department of Livestock services distributed 195000 doses of ND vaccine under National ND control program for 9 districts of this region. VL, Surkhet supported the program by sero-monitoring. The table below shows district wise collection of serum sample.

Table 66: District wise collectionand test result of serum sample.

S. N.	District	Test Result ND Serosurvillance		
		Sample collected and tested	No of Positive	Positive %
1	Dailekh	144	101	70.13
2	Pyuthan	44	26	59.09
3	Rukum paschim	48	15	31.25
4	Jajarkot	31	31	100
5	Dang	65	20	30.76
	Total	**332**	**193**	**58.13**

7. Regional vaccine bank

VL, Surkhet maintains regional vaccine bank for major livestock and poultry disease. In FY 2079/80, vaccines of PPR and ND were distributed in different districts as the part of National Animal Disease Control Program. A detail of the distribution is mentioned in table below.

Table 67: List of Vaccine distribution of PPR and ND

S. N	District	PPR	ND	Rabies	FMD	CSF	HSBQ
1	Surkhet	300000	-	2320	0		27000
2	Dailekh	157000	42000	1250	8000	2080	
3	Salyan	150000	40000	800	3500	1000	
4	Jajarkot	90000	-	-	-	100	
5	Jumla	130000	25000	200	1000		11200
6	Kalikot	53000	30000	-	20000		
7	Humla	101000	25000	-	0		9800
8	Dolpa	60000	40000	-	0		
9	Mugu	60000	40000	-	53000		
10	West Rukum	150000	25000	750	25000	2000	
11	Dang	200000	-		-		
12	Banke	180000	-		14600		60000
13	Bardiya	200000	-		3000		3000
14	Rolpa	150000	25000			100	
15	Pyuthan	140000	25000				
16	East Rukum	50000	25000				
Total		**2171000**	**342000**	**5320**	**128100**	**5280**	**111000**

8. Epidemic Investigation:

Various disease outbreaks of animal and poultry were investigated during F/Y 2079/80. Whenever requested for investigation of an outbreak was received from the district to the VL, a veterinarian or a technician or a team of technicians with necessary sampling kit visited to the site, collected epidemiological information and appropriate samples. In the laboratory, samples collected from the field were processed to identify the etiology of the event. Epidemiological information gathered from the site was used to decide the test to be performed in the laboratory and to assist in the confirmation of disease. Samples which were not possible in this laboratory were referred to CVL, Kathmandu for further testing.

Table 68: Epidemic investigation

SN	Program	Target	Progress	Remarks
1	Epidemic disease investigation	4	3	Surkhet, Jajarkot, Banke, Bardiya
2	Province level emergency disease investigation team management	4	4	Surkhet, Jajarkot, Banke, Bardiya
3	Avian Influenza Disease Survillance and Test	4	5	Surkhet, Dailekh, Salyan, Dang, Banke, Bardiya

8.1 Status of PPR outbrehaks in 2079/80

SN	Date of Outbreak	Location	Remarks
1	2079/06/05	Gurash 4, Dailekh	PPR Rapid Test and PCR (CVL)

8.2 Status of FMD outbreaks 2079/80

SN	Date of Outbreak	Location	Remarks
1	2079/5/11	Raskot 1, Kalikot	FMD and TADs Lab

8.3 Status of LSD outbreaks 2079/80

SN	Date	Location	Type of Sample	Total Sample	Result (PCR)		Remarks
					Positive	Negative	
1	2079/12/02	Chaurjhari, Rukumpaschim	Blood	9	9	0	
2	2079/12/06	Bangachili, Dang	Blood	1	1	0	
3	2079/12/07	Bangachili, Dang	Blood	2	2	0	
4	2079/12/08	Bangachili, Dang	Blood	3	5	0	
5	2079/12/09	Bangachili, Dang	Blood	2	2	0	
6	2079/12/09	Bheri-12, Jajarkot	Blood	5	5	0	
7	2079/12/10	Bangachili, Dang	Blood	5	4	0	
8	2079/12/19	Surumarani-1, Pyuthan	Blood	1	1	0	
9	2080/01/04	Birendranagar-8, Surkhet	Blood	1	1	0	
10	2080/01/04	Chingad-3, Surkhet	Blood	5	1	0	
11	2080/01/04	Rolpa	Blood	3	5	0	
12	2080/01/04	Madhuban-5, Bardiya	Blood	2	3	0	
13	2080/01/04	Bheriganga-7, Surkhet	Blood	2	2	0	
14	2080/01/04	Babai-7, Dang	Blood	8	2	0	
15	2080/01/04	Kalimati, Salyan	Blood	2	8	0	
16	2080/01/15	Sharadanagar, Salyan	Blood	4	2	0	
17	2080/01/16	Lekhbeshi, Surkhet	Blood	1	1	0	
18	2080/01/28	Shantinagar, Dang	Blood	7	1	0	
19	2080/01/29	Sanibheri, Rukumpaschim	Blood	6	6	0	
20	2080/02/09	Bhairabi, Dailekh	Blood	5	6	0	

SN	Date	Location	Type of Sample	Total Sample	Result (PCR)		Remarks
					Positive	Negative	
21	2080/02/10	Chamundabindasini-8, Dailekh	Blood	4	4	0	
22	2080/02/16	Simta, Surkhet	Blood, Swab	5	5	0	
23	2080/02/30	Chamundabindasini, Dailekh	Blood	4	4	0	
24	2080/02/31	Simta, Surkhet	Serum	5	4	0	
25	2080/03/19	Khajura-3, Banke	Blood	29	29	0	
26	2080/03/24	Raptisonari, Banke	Blood, Swab	66	66	0	

8.4 Status of Classical Swine Fever outbreaks 2079/80

SN	Date of Outbreak	Location	Remarks
1	2079/4/15	**Birendranagar 1, Surkhet**	**FMD and TADs Lab**

8.5 Status of Low Pathogenic Avian Influenza outbreaks 2079/80

SN	Date of Outbreak	Location	Remarks
1	2079/04/31	Basgadhi-4, Bardiya	Rapid Test and qPCR H9 (CVL)
2	2079/05/16	Birendranagar-12, Surkhet	
3	2079/09/10	Dullu-12, Dailekh	
4	2079/10/16	Birendranagar-13, Surkhet	
5	2079/04/31	Birendranagar-4, Surkhet	
6	2080/01/11	Birendranagar-12, Surkhet	
7	2080/01/25	Basgadhi-4, Bardiya	
8	2080/01/31	Basgadhi-4, Bardiya	
9	2080/02/04	Birendranagar-9, Surkhet	
10	2080/02/12	Gularia-3, Bardiya	
11	2080/02/25	Basgadhi-10, Bardiya	
12	2080/03/03	Birendranagar-11, Surkhet	

8.6 Status of Rabies outbreaks.

SN	Date	Location	Animal Infected
1	2079/04/16	Chaukuna-4, Surkhet	Buffalo
2	2079/05/23	Bheriganaga-4, Surkhet	Dog
3	2079/06/05	Bheriganga-3, Surkhet	Dog
4	2079/06/14	Musikot-10, Rukumpashchim	Buffalo
5	2079/06/25	Darma-5, Salyan	Buffalo
6	2079/07/23	Bheriganga-10, Surkhet	Goat
7	2079/07/27	Bheriganaga-4, |Surkhet	Dog
8	2079/08/14	Bheriganga-2, Surkhet	Goat
9	2079/08/29	Kohalpur, Banke	Dog
10	2079/09/11	Gurvhakot, Surkhet	Goat
11	2079/09/12	Birendranagar 13, Surkhet	Goat
12	2079/09/12	Bashgadhi-1, Bardiya	Buffalo
13	2079/09/20	Birendranagar-4, Surkhet	Dog
14	2079/09/22	Chingard Surkhet	Buffalo
15	2079/09/28	Lekhbasi-4, Surkhet	Buffalo
16	2079/10/03	Birendranagar-9, Surkhet	Dog
17	2079/10/05	Gurvhakot-13, Surkhet	Cow
18	2079/10/10	Chingard-1, Surkhet	Buffalo
19	2079/10/16	Musikot-5, Rukumpashchim	Dog
20	2079/10/17	Chingard-1, Surkhet	Dog
21	2079/10/23	Chingard-1, Surkhet	Dog
22	2079/10/25	Birendranagar-4, Surkhet	Dog
23	2079/10/26	Chingard, Surkhet	Dog
24	2079/10/27	Birendranagar-8, Surkhet	Dog
25	2079/11/03	Birendranagar-9, Surkhet	Dog
26	2079/11/05	Birendranagar-10, Surkhet	Dog
27	2079/12/01	Bheriganga-9, Surkhet	Goat
28	2079/12/01	Bheriganga-9, Surkhet	Buffalo
29	2079/12/26	Birendranagar-11, Surkhet	Goat
30	2079/12/26	Chingard-4, Surkhet	Sheep
31	2080/02/08	Bheriganaga-7, Surkhet	Goat
32	2080/02/14	Bheriganaga-9, Surkhet	Buffalo
33	2080/02/24	Chaukuna, Surkhet	Dog
34	2080/03/12	Birendranagar-2, Surkhet	Dog

8.7 Status of African Swine fever outbreaks 2079/80

SN	Date	Location	Remarks
1	2079/05/12	Birendranagar-13, Surkhet	**Rapid Test And PCR(CVL)**
2	2079/05/16	Birendranagar-9, Surkhet	

3	2079/07/02	Birendranagar-12, Surkhet	
4	2079/07/09	Birendranagar-14, Surkhet	
5	2079/07/16	Birendranagar-14, Surkhet	
6	2079/07/21	Birendranagar-1, Surkhet	
7	2079/07/30	Birendranagar-9, Surkhet	
8	2079/08/16	Birendranagar-9, Surkhet	
9	2079/08/16	Birendranagar-9, Surkhet	
10	2079/08/20	Birendranagar-9, Surkhet	
11	2079/08/21	Birendranagar-10, Surkhet	
12	2079/08/23	Birendranagar-10, Surkhet	
13	2079/08/29	Bheriganga-9, Surkhet	
14	2079/09/04	Birendranagar-1, Surkhet	
15	2079/09/18	Birendranagar-13, Surkhet	
16	2079/09/20	Birendranagar-1, Surkhet	
17	2079/10/18	Birendranagar-5, Surkhet	
18	2079/10/23	Birendranagar-9, Surkhet	
19	2079/10/26	Birendranagar-9, Surkhet	
20	2079/11/18	Bashgadhi-06, Bardiya	
21	2079/12/01	Birendranagar-1, Surkhet	
22	2079/12/09	Gorahi-17, Dang	
23	2079/12/09	Gorahi-15, Dang	
24	2079/12/17	Birendranagar-10, Surkhet	
25	2079/12/28	Birendranagar-9, Surkhet	
26	2080/01/21	Birendranagar-12, Surkhet	

VETERINARY LABORATORY DHANGADI

1. Introduction

Veterinary Laboratory is situated in Dhangadhi Sub Metropolitan city of Far-Western Province of Nepal. This laboratory was established as the designated laboratory of the Far-Western development region with its service area covering nine districts. Veterinary Laboratory, Dhangadhi was established in 2049/050 as with the name of Regional Veterinary Laboratory, Dhangadhi. With the administrative reform of Nepal during the year 2074/75, the laboratory was renamed as Veterinary laboratory, Dhangadhi, Kailali with working areas as before. Veterinary laboratory aims to protect the livestock with the provision of prompt diagnosis of diseases.

Table 69: Staffing of Veterinary Laboratory, Dhangadi, Kailali

S.N.	Type of post	Class	Number	Fulfilled	Vacant	Remark
1	Senior veterinary officer	G.II	1	1	0	
2	Veterinary officer	G.III	3	2	1	
3	Animal Health Technician	NG.I	3	3	0	
4	Assistant Animal Health Technician	NGII	2	1	1	
5	Accountant	NGI	1	1	0	
6	Clerk	NG2	1	1		
7	Driver	No class	1	1	0	Contract
8	Office helper	No class	2	2	0	

2. Progress of FY 2079/80

Table 70: Summary of Achievements in fiscal year 2079/80

S.N.	Programs and Activities	Unit	Annual Progress		Percentage	Remarks
			Target	Progress		
A.	**Fixed cost activities**					
1	Printer purchase	No	2	2	100%	
2	DSLR camera purchase for Lab Purpose	No	1	1	100%	
3	CCTV Purchase and installation	No	1	1	100%	
4	Laptop purchase for Lab Purpose	No	2	2	100%	
5	Washing Machine, Solar Geaser etc Purchase for Lab Purpose	No	1	1	100%	
6	A.C(1.5ton) purchase and installation in Laboratory	No	1	1	100%	
7	Book Shelf purchase	No	2	2	100%	
8	Locker, Bench, Tool etc purchase for Office and Laboratory Purpose	No	1	1	100%	
9	Construction of Post-mortem Biosafety Pit	No	1	1	100%	
10	Repairment of Sample Registration Room	No	1	1	100%	

S.N.	Programs and Activities	Unit	Annual Progress		Percentage	Remarks
			Target	Progress		
11	Ambulance remodelid and equipment, tool jointed	No	1	1	100%	
12	Laboratory Premise Improvement and Making BSL 2 Lab Woodless	No	1	1	100%	
13	Scooter purchase	No	1	1	100%	
B	**Laboratory Service programme**					
1	Identification of parasites and EPG Count	No.	300	301	100%	Budget withhold
2	Skin scraping Test	No.	60	0	0%	Budget withhold
3	Clinical hematological examination	No.	400	0	0%	
4	Biochemical examination	No.	200	0	0%	
5	Urine examination	No.	60	0	100%	Budget withhold
6	Postmortem examination	No.	1200	1458	100%	
7	Bacteria culture and antibiotic sensitivity test	No.	400	230	58.75%	
8	Fungus culture and Identification	No.	50	50	0%	Budget withhold
9	AI Test	No.	300	282	94%	Budget withhold
10	IBD Test	No.	400	197	49.25%	Budget withhold
11	ND Test	No.	200	121	60.5%	Budget withhold
12	PPR disease diagnosis (Penside)	No.	200	28	14%	
13	Salmonellosis examination (PAT)	No.	400	38	9.5%	Budget withhold
14	Mycoplasma examination (PAT)	No.	400	0	0%	Budget withhold
14	Sample collection, test and dispatch to CVL	Time	8	8	100%	
16	Viral disease examination of Dog (Distemper/parvovirus/corona) rapid Test examination	No.	30	10	33.33%	Budget withhold
C	**Investigation, Study and Treatment program**					
1	Provincial level Investigation of Emergency Diseases Team management	Time	4	4	100%	
2	Pandemic Disease Investigation in Livestock and Poultry	Time	4	4	100%	
3	Investigation of Kumri disease in goats (Kailali/Kanchanpur)	Time	1	1	100%	Budget withhold

S.N.	Programs and Activities	Unit	Annual Progress		Percentage	Remarks
			Target	Progress		
4	Khari disease investigation (Baitadi/Darchula)	Time	1	1	100%	Budget withhold
5	Sub clinical mastitis investigation in milking cattle and Buffalo	Time	4	4	100%	
6	Fish Disease Investigation	Time	1	0	0%	Budget withhold
7	Lumpy Skin Disease Control Program	Time	1	1	100%	Budget withhold
D	**Zoonotic disease investigation programs**					
1	Avian Influenza Rapid Test	No.	300	282	94%	
2	Brucella examination	No.	200	129	64.5%	
3	Rabies antigen test	No.	20	41	100%	
E	**Workshops**					
1	Orientation on Sub-Clinical Mastitis to Local level Technicians and Farmers	Time	1	1	100%	Budget withhold
2	Interaction about Epidemiological Reporting to local and provincial level techinicains	Time	1	0	0%	Budget withhold
3	District level interaction on sample collection, storage and dispatch with	Time	2	0	0%	Budget withhold
F	**Disease prevention and control program**					
1	Provincial Vaccine Bank Management PPR, FMD and Rabies	Time	4	4	100%	
2	PPR Sero-monitoring- Sample collection and Dispatch	No.	1200	1381	100%	Budget withhold
3	CSF Sero-monitoring- Sample collection and Dispatch	No.	300	50	16.66%	ASF outbreak
4	FMD Sero-monitoring- Sample collection and Dispatch.	No.	300	52	17.33%	Budget withhold
3	FMD surveillance	Time	4	4	100%	
G.	**Publication and others Programs**					
1	Annual technical book material preparation	Time	1	1	100%	
2	Participation in national level conference, symposium and workshop	Time	4	0	0%	Budget withhold
3	Province level laboratories Monitoring	Time	5	5	100%	
	Total progress				83.83%	

3. Laboratory activities conducted offered during the fiscal year 2079/80

3.1 Parasitology:

Altogether 301 fecal sample (295 goats,6 cattle) samples were tested for identification of parasitic conditions of livestock populations. Samples for examination mainly consisted of the regular fecal samples submitted to the livestock service section, Dhangadhi Submetropolitan, Kailali. Apart

from this samples were also collected from field during outbreak investigation and the other investigation programme as per annual workplan.

Out of 301 fecal samples tested for EPG, 245 (81.39%) samples were positive. The most common helminthes identified during faecal examination were Heamonchus sp., Tricostronglyus sp., Fasciola sp., Paramphistomum sp., Strongylus sp., Hemonchus sp., Trichuris sps. It was found that most of the fecal samples examined at the VL were positive for more than one parasite.

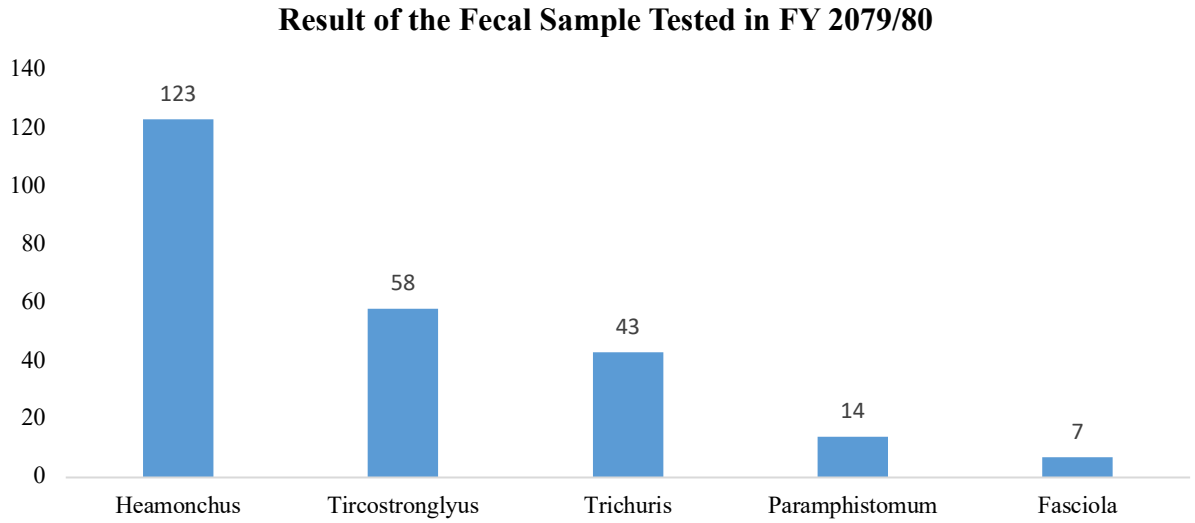


Figure 54: Result of the Fecal Sample Tested in FY 2079/80

3.2 Serology:

During the fiscal year 2079/80 most of the serum samples collected were from goats and cattle for various diseases like Toxoplasmosis, Brucellosis, PPR (Pen side Test),), Rabies rapid antigen test, Salmonella and Mycoplasma (Plate Agglutination Test-PAT) for poultry disease was conducted in the laboratory. Due to withholding of budget and budget restrictions we were unable to perform many pre planned tests. Some of the samples were forwarded to CVL Tripureswor Kathmandu for further testing.

Table 71: Results of serological examination performed at the VL, Dhangadhi, Kailali

S.N.	Disease Name	Test method	Sample Tested	Result	Remarks
1.	Salmonellosis	PAT	38	38 -ve	
2.	Brucellosis	PAT	38	38-ve	(Sample collected by VL Dhangadhi and Test was performed by CVL, Kathmandu)
		ELISA	91	91-ve	
3.	Peste des petits ruminants (PPR)	Penside test	28	7+ve	
4.	Rabies	Antigen test	41	39 +ve	RDT at VL, Dhangadhi and FAT at CVL
5.	Avian Influenza type A		282	28+ve	H9 in PCR

6.	New Castle Disease	Antigen test (Lateral flow asssy)	121	120-ve	
7.	Infectious bursal disease		197	84 +ve	
8.	Toxoplasmosis	ELISA	91	41+ve	(Sample collected by VL Dhangadhi and Test was performed by CVL, Kathmandu)
9.	African Swine Fever	Rapid Test/PCR	31	24+ve	PCR performed by CVL, Kathmandu
10.	Lumpy Skin Disease	ELISA	38	36+ve	(Sample collected by VL Dhangadhi and Test performed by CVL, Kathmandu)
11	FMD	ELISA	26	26+ve	ELISA performed by FMD and TADs Laboratory
12	CSF	PCR	24	1+ve	PCR performed by CVL, Kathmandu
13	Enterotoxemia	ELISA	7	2+ve	Sample collected by VL Dhangadhi and Test performed by CVL, Kathmandu)

3.3 Pathology:

This section of the laboratory mostly receives poultry, goat and pig carcasses for necropsy examination and disease diagnosis. However, dead bodies of other animal species are also received

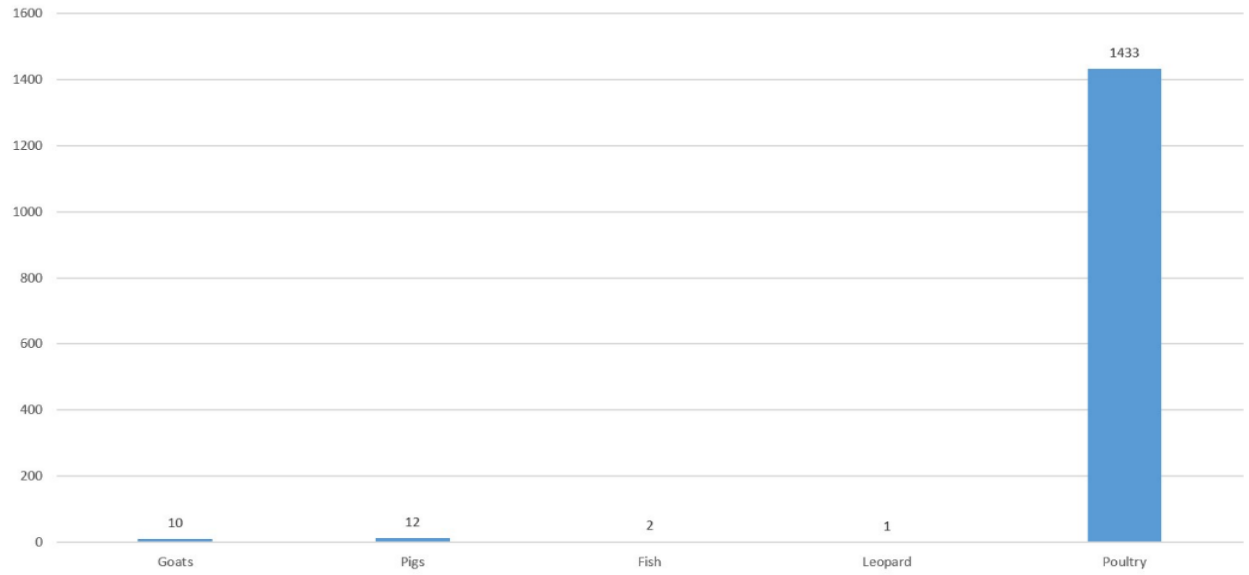


Figure 55: Post-Mortem Record

occasionally, especially during disease outbreak. Most of the pathological samples comprised of poultry. During the fiscal year 079/80, altogether 1458 carcasses belonging to 729 farms were registered at VL, Dhangadhi for postmortem examination. The major conditions diagnosed in the pathology section are presented below.

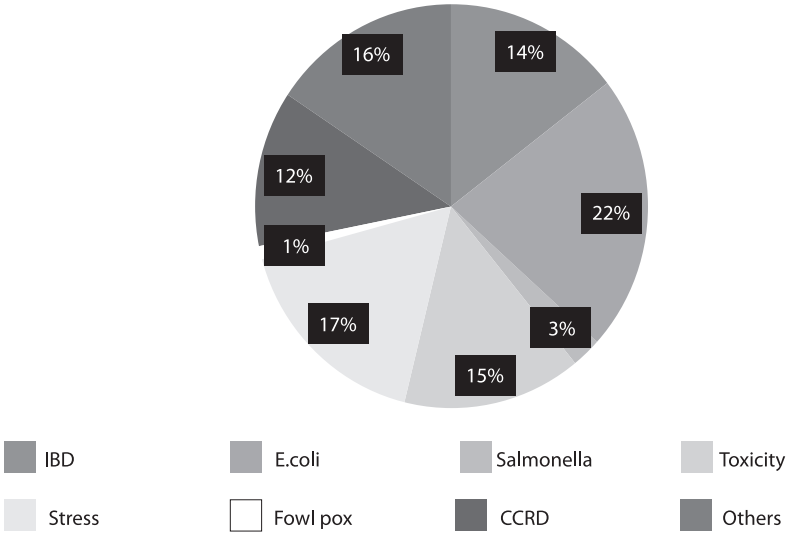


Figure 56: Local Level wise Post-Mortem Record

Table 72: Post mortem findings in Caprine and Swine

S. N	Animal species	Disease/condition	Number of farms	Remarks
1	Swine	TGE(Suspected)	4	
		ASF	8	
2	Caprine	Non-specific death	2	
		Pneumonia	5	
		Plant poisoning	1	
		Parasitic gastroenteritis	2	

3.4 Microbiology:

The samples received to microbiological examination at the VL, Dhangadhi constitutes of milk, and swab from visceral organs like liver, lungs, intestine etc. of various animal species. The media used for microbiological culture were Nutrient agar, MacConkey agar, Blood agar, EMB Agar, XLD Agar, and MHA etc. Due to withholding of budget, lab was unable to perform fungus culture as planned. Bacteria were identified on the basis of colony characteristics, gram's staining and the biochemical test. Due to limitation of trained technical resource person and budget restrictions the lab was unable to meet the target for the fiscal years. The result of microbiological test is presented in the table as below.

Table 73: Microbiological test result

Animal type	Sample type	Number of samples tested	Results	Major bacteria identified
Cattle/Buffalo/ Goat	Milk	45	35+ve	*Staphylococcus, E.coli,*
Poultry	Liver, lungs, heart	186	139 +ve	*E.coli, Salmonella, Klebsiella, Streptococcus, Enterococcus, Staphylococcus*
Total		231	174 +ve	

Table 74: Summary of Major Bacteria seen in Microbiology (079/80)

S. N	Major Bacteria	Number	Remarks
1	*E.coli*	78	
2	*Staphlyococcus*	15	
3	*Klebsiella*	24	
4	*Salmonella*	14	
5	*Enterobacter*	4	
6	No growth	57	
7	Total	192	

3.4.1 Antibiotic Sensitivity Profile of Enterobacteriaceae

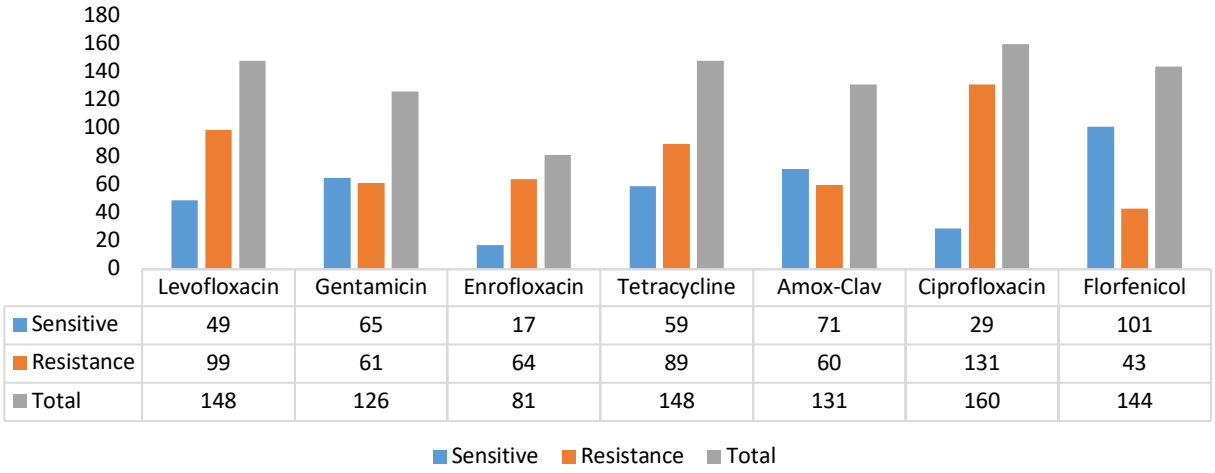


Figure 57: Antibiotic Sensitivity Profile of Enterobacteriaceae

4. Investigation of Kumri (*Seteria sp.*) in Goats

A part from above activities VL, Dhangadhi has performed investigation of a Kumri in Goat in Kailali and Kanchanpur district. The main objective of the study was to find the prevalence of Kumri in goat, especially in Hilly and Terai area of Kailali and Kanchanpur district. Active

surveillance was performed through questionnaire. Serum and faeces samples were collected from infected goats.

This investigation was conducted in Chure Rural Municipality and Mohanyal Rural Muncipality of Kailali district. Summary of investigation and finding are given below.

Method of Investigation

Active surveillance through administration of standard questionnaire, clinical examination and laboratory testing.

Major findings

1. None of the goat's wear affected by Kumari (*Setaria sp.*).
2. Prevalence of Kumri disease was found to be 0% in Kailali district.

5. Investigation of sub clinical mastitis in dairy animals

This program was conducted to know the prevalence of sub clinical mastitis in milking cattles and buffaloes of Kailali and Kanchanpur districts. Fresh milk samples were taken from farmers directly and tested by CMT immediately.

Investigation Site: Kailali and Kanchanpur

Objectives: To find the prevalence of sub clinical Mastitis in dairy animals of Kailali and Kanchanpur.

Method – Active surveillance and questionnaire

- Milk sample were directly taken from animals.
- Test Method: CMT

Table 75: Summary of Sub clinical mastitis Investigation program

District	Number of farms	Number of cattle milk samples	Number of buffalo milk samples	Number of goats milk samples	Total samples tested
Kailali	12	33	18	6	57
Kanchanpur	5	20	25	5	50
Total	17	53	43	11	107

Major findings

- Total number of samples positive for mastitis: 94 (87.85%).
- There was low prevalence of sub clinical mastitis in local breeds of cattle as compared to the exotic breeds.

Major Disease Outbreak Status

Table 76: PPR Outbreak status (F/Y 2079/80)

District	No. of Outbreaks	Location	Animal type	No. of affected animals	No. of deaths	No. of animals at risk
Dadeldhura	1	Parsuram municipality	Goat	35	6	300
Kailali	1	Mohanyal Rural Muncipality	Goat	50	10	150

Table 77: FMD Outbreak Status

District	No. of Outbreaks	Location	Animal type	No. of affected animals	No. of deaths	No. of animals at risk
Darchula	2	Marma Rural Municipality, Sailyashikar Rural Municipality	Cattle, Buffalo	750	0	2000
Baitadi	1	Dilasani Rural Municipality	Cattle, Buffalo	43	0	500
Dadeldhura	1	Navdurga Rural Municipality	Cattle, Buffalo	35	0	350

Table 78: Summary of PPR Sero-monitoring

S. N	Districts	Sample collected	Test Result		Remarks
			Positive	Negative	
1	Bajhang	160	99	54	7 doubtful
2	Kailali	418 (317 tested)	184	133	Samples not processed due to lack of test kit and to be tested in next FY
3	Kanchanpur	376			
4	Baitadi	133			
5	Dadeldhura	124			
6	Doti	170			
7	Total	1381	283	187	

Table 79: Summary of FMD Sero-monitoring

S. N	Districts	Sample collected	Test Result		Remarks
			Positive	Negative	
1	Kailali	52			Samples are not processed due to lack of Test Kit and to be tested in next FY

Table 80: Summary of CSF Sero-monitoring

S. N	Districts	Sample collected	Test Result		Remarks
			Positive	Negative	
1	Kailali	52			Samples are not processed due to lack of Test Kit and to be tested in next FY

Table 81: Vaccine distribution as per national animal health program in sudurpaschim province (F/Y 2079/80).

District	**PPR**	**FMD**	**Anti-Rabies**	**ND I-2**	**HS+BQ**	**CSF**
Darchula	55000	10000	250	0	0	0
Baitadi	102000	0	200	51000	0	0
Dadeldhura	100000	20000	550	51000	0	0
Kanchanpur	113100	28250	2760	0	0	13500
Bajhang	71000	0	150	38000	3000	0
Bajura	83000	0	350	25000	0	0
Doti	69000	0	510	63000	0	0
Achham	70000	0	250	60000	0	0
Kailali	138500	83500	1550	76000	28000	14000
Total	**801600**	**141750**	**6570**	**364000**	**31000**	**27500**

ANNEX: PHOTOGRAPHS OF LABORATORY SERVICES

Avian Leukosis Complex in Poultry

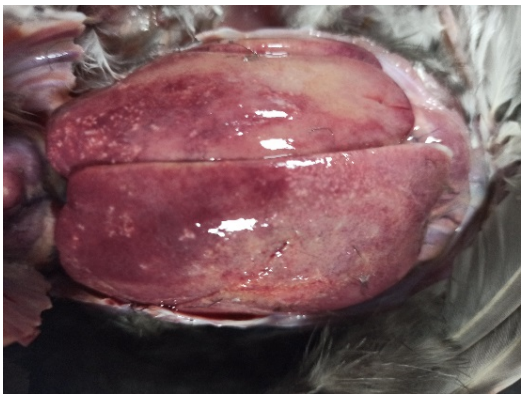


Figure 1. Hepatomegaly and diffused necrosis in liver.

Figure 2. Multiple white nodules, diffused necrosis in liver and hepatomogaly.

Marek's Disease in Poultry

Figure 3. Multiple tumors in proventriculus.

Figure 4. Multiple tumors in liver.

Figure 5. Tumor on heart.

Figure 6. Tumors on mesentery.

Newcastle Disease in Poultry

Figure 7 and 8. Caecal tonsil and payer's patches hemorrhages in small intestine.

Figure 9. Pin point hemorrhage in proventriculus.

Figure 10. Hemorrhages in trachea.

Fowl Typhoid in Poultry

Figure 11. Necrotic foci with bronze colored liver.

Figure 12. Pedunculated ovary.

African Swine Fever in Pig

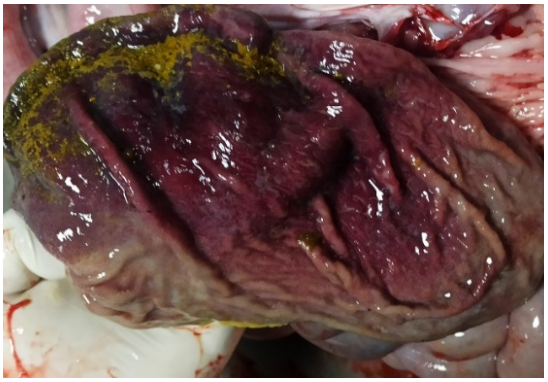


Figure 13: Hemorrhages in stomach.

Figure 14. Hemorrhagic spleen.

Figure 15. Hemorrhagic and necrosis of kidney.

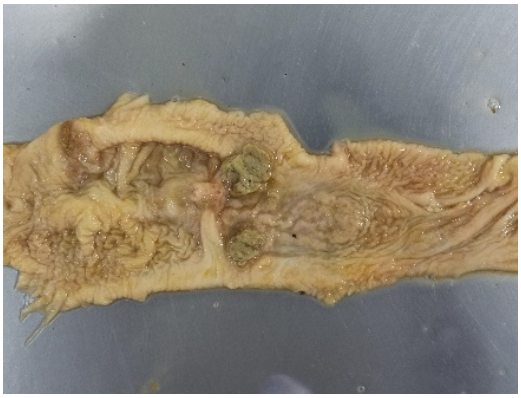


Figure 16. Button like ulcer in caecum.

Figure 17. Congested lungs.

Figure 18. Hemorrhagic mesenteric lymphnodes.

Figure 17. LSD infected animals

Figure 17. Anthrax suspected pigs

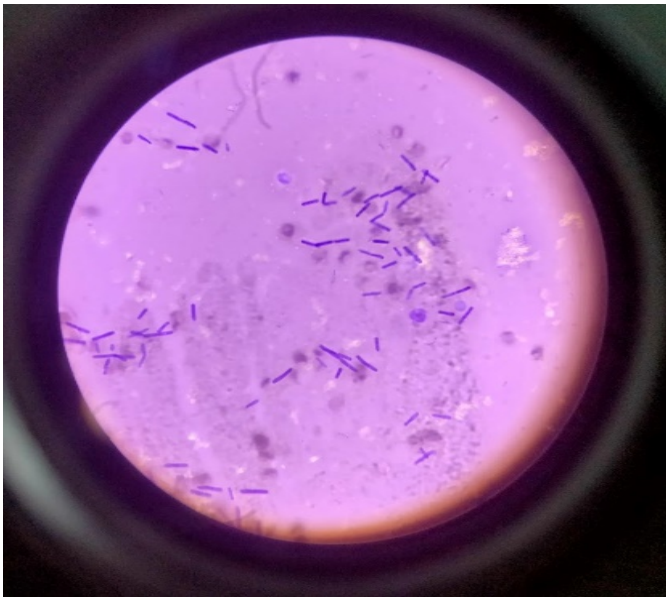


Figure 18. Anthrax Bacilli on blooed smear

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