

Annual Technical Report

2070/071 (2013-2014)



Government of Nepal
Ministry of Agriculture Development
Department of Livestock Services

Directorate of Animal Health
Central Veterinary Laboratory

Tripureshwor, Kathmandu, Nepal

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Foreword

I am very much pleased to present this annual technical report for the fiscal year 2070/071 (2013-2014). This issue includes activities and progress report of Central Veterinary Laboratory (CVL), five Regional Veterinary Laboratories (RVLs) and National Avian Disease Investigation Laboratory (NADIL).

Recognition of disease is the foundation of disease control and prevention. The rapid and accurate diagnosis of diseases can only be assured in fully equipped laboratories that have range of standardized diagnostic reagents and trained manpower.

In the context of Nepal as a WTO Member, for the implementation of SPS measures among the scientific basis, laboratory based diagnosis is of paramount importance. Therefore, the role of national and regional veterinary laboratories including basic labs in the districts is very important in animal disease diagnosis. And also there is still a scope to uplift the standards and quality assurance of the laboratory diagnosis conducted by these labs. With the establishment of molecular diagnostic techniques at the CVL, routine molecular diagnosis of Avian Influenza, ND, and IBD is performed. Similarly, the ELISA, IFAT, HA/HI, AGPT, Virus isolation and characterization and other routine diagnostic tests are used for the diagnosis of various livestock and poultry diseases.

We are putting our efforts to upgrade CVL, RVLs and NADIL diagnostic laboratories to provide reliable and prompt diagnostic services all over the country. We already have good co-ordination between regional labs and basic labs and the sample flow in the national reference laboratories is routinely being done.

I would like to extend my sincere thanks to all RVLs and NADIL staffs for providing information required for this publication. I express my personal appreciation and sincere thanks to all the staff of CVL who worked hard to shape the annual technical report in this form.

I would be grateful if you provide any suggestions for the improvement of the future issue of this publication.

Dr. Sudarsan Prasad Gautama
Act. Chief Veterinary Officer
Central Veterinary Laboratory
Tripureshwar, Kathmandu

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CENTRAL VETERINARY LABORATORY, KATHMANDU

1. Introduction

Central Veterinary Laboratory (CVL) focuses programme with the objective of rapid and accurate diagnosis of diseases and securing healthy national flocks of animals and birds throughout the nation by controlling the livestock and poultry diseases at the earliest. CVL also works on epidemic investigation as well as surveillance and investigation on various diseases in its approved annual programme. 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 lab and Molecular Biology with a considerable progress in the later. At present the molecular based diagnosis of avian influenza is in routine use.

CVL has standard Operating Procedures, test protocols and quality guideline manual. CVL is gradually practicing the bio-safety/ bio-security measures so that good laboratory practice is followed in our all the diagnostic laboratories. CVL has already been adopting test verification system through international reference laboratories which will help in the accreditation of CVL for international certification as well.

To provide diagnostic facilities throughout the country, CVL works through its five Regional Veterinary Laboratories (RVLs) located one in each of the development regions of the nation; eastern (Biratnagar), central (Janakpur), western (Pokhara), mid-western (Surkhet) and far-western (Dhangadhi) as well as t National Avian Disease Investigation Laboratory (NADIL) located in Chitwan. To provide the diagnostic services smoothly throughout the nation, 15 basic laboratories have been established in 15 District Livestock Service Offices (DLSOs) namely, Illam, Jhapa, Saptari, Sarlahi, Rautahat, Parsa, Makawanpur, Kabhrepalanchowk, Chitwan, Rupandehi, Dang, Banke, Jumla, Dadeldhura and Kanchanpur, and 60 primary laboratories one in each DLSOs of other districts. The basic laboratories are capable to perform parasitological examinations, microbial culture and antibiotic sensitivity test. Specimens that cannot be processed in the aforementioned laboratories due to insufficient facilities and expertise are referred to the CVL. In this way, CVL works as a reference veterinary laboratory in Nepal.

2. Objectives

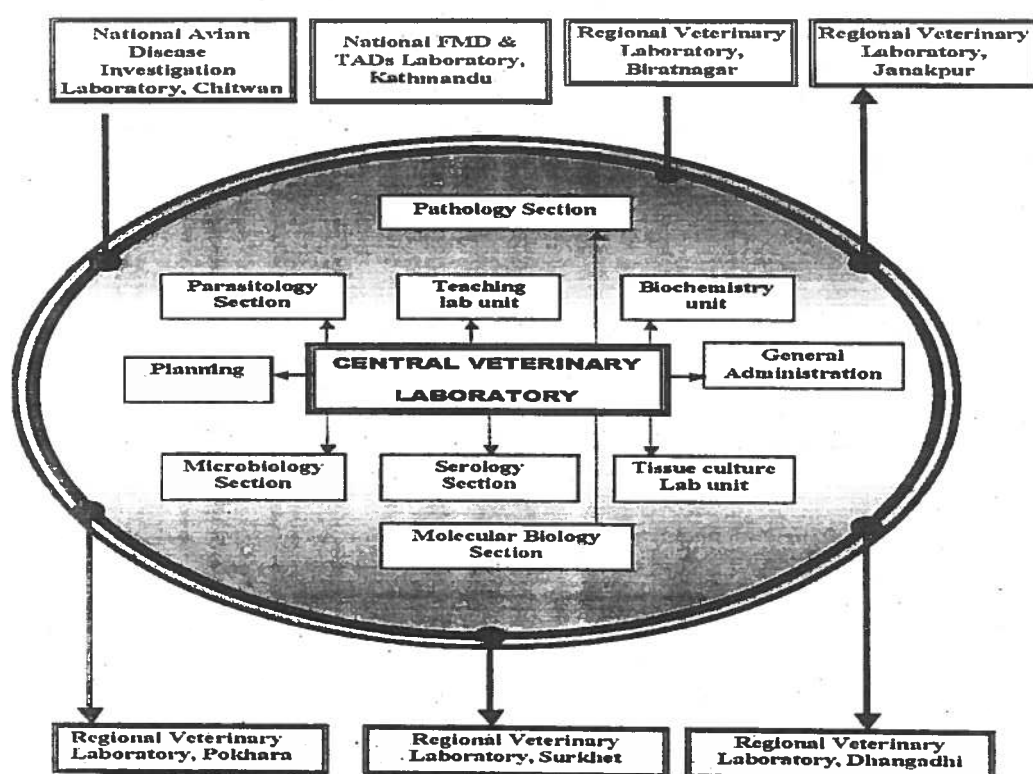
The role of veterinary laboratory system has become dynamic in the advent of food safety issues, economic liberalization and trade globalization. Nepal joined as a WTO member in 2004. Therefore, Nepal follows the guidelines provided by Office International des Epizootics (OIE) for the provision of Sanitary and Phytosanitary (SPS) agreement under WTO that seeks scientific procedures and evidences 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 in the country in the area of animal health and veterinary
- ❖ public health.
- ❖ Acts as a national veterinary reference laboratory.

- ❖ Conduct epidemiological disease investigation & laboratory diagnosis of livestock and poultry disease.
- ❖ Support the national disease control and surveillance programmes.
- ❖ Acquire, adopt, upgrade and disseminate different laboratory diagnostic test methodologies for Live stock and poultry diseases.
- ❖ Assist Animal Health Directorate (AHD) in the animal health policy development and formulation of animal disease control and eradication programmes.
- ❖ Extend & disseminate information concerning livestock and poultry disease controls.
- ❖ Collaborate with international reference laboratories & institutions on veterinary laboratory diagnosis.
- ❖ Assist in implementation of national epidemic control strategies.
- ❖ Conduct laboratory diagnosis techniques training for the veterinarians and veterinary paraprofessionals.
- ❖ Strengthen and coordinate regional and district level veterinary laboratories.

To achieve the aforementioned objectives, there are a series of approved annual activities carried out by different laboratory sections of the CVL, five RVLs and NADIL.

3.Organization Chart



Annual Technical Report- 2070/071 (2013-2014)

Table: Staff working at Present at CVL

S.N	Name or the staff	Designated Post	Total Number	Fulfilled	Vacant
1		Chief Veterinary Officer	1		1
2	Dr. Suderson Prasad Gautam	Senior Veterinary Officer	2	2	
3	Dr. Pragya Koirala	Senior Veterinary Officer			
4	Dr. Indresh jha	Veterinary Officer	7	7	
5	Mrs. Gouri Devi Thapa	Veterinary Officer			
6	Mr. Purn B. Buda	Veterinary Officer			
7	Mr. Bal B. Kuwar	Veterinary Officer			
8	Mr. Prakash Devkota	Veterinary Officer			
9	Mr. Tek B. Air	Veterinary Officer			
10	Dr. Chanda Sherestha	Veterinary Officer			
11	Dr. Manju Maharjan	Veterinary Officer			
12	Mr. Bakta B. Katuwal	Animal Health Technician	6	5	1
13	Mr. Indal sha	Animal Health Technician			
14	Mr. Shyam Sundar Ray yadav	Animal Health Technician			
15	Mr. Mithilesh Kumar Karan	Animal Health Technician			
16	Mr. Tul B. Rai	Animal Health Technician			
17	Mr. Bimsen Adhikari	Assistant Animal Health Technician	8	3	5
18	Mr. Purn Maharjan	Assistant Animal Health Technician			
19	Mr. Bal Kumar Rai	Assistant Animal Health Technician			
20		Senior clerk (Typist)	1		1
21	Mr. Budhi B. Lama	Accountant	1	1	
22	Mrs. Prem Kumari Sherpa	Clerk (Kharidar)	1	1	
23	Mr. Macha kaji Maharjan	Driver	1	1	
24	Mr. Dipes B. Rana Magar	Driver		1	Contract
25	Mr. Rajan Adhikari	Office Helper	3	3	
26	Mrs. Bhima Acharya	Office Helper			
27	Mr. Chandra B. Rana Magar	Office Helper			
28	Mr. Jiwan Rai	Office Helper	1	1	Contract
Total			32	25	8

Table: Annual programme, Target & progress of CVL in fiscal Year 2070/071

S.N.	Activities	Unit	Target	Achievement
1	Parasitology			
1.1	EPG and Larva Culture	Number	300	833
1.2	Investigation of Blood Protozoa Out brake and diagnosis	Times	3	3
2	Pathology			
2.1	Clinical hematological examination	Number	300	1031
2.2	Bio-Chemical examination	Number	250	592
2.3	Post-Mortem Examination	Number	2000	4142
2.4	Histopathological examination	Number	100	104
3	Micro-Biology			
3.1	Bacteriology			
3.1.1	Isolation and Identification of Bacteria	Number	1000	1609
3.1.2	Diagnosis of micro-plasma in goats	Times	4	4
3.1.3	Sample collection, Isolation and Identification of Fungus	Number	100	114
3.2	virology			
3.2.1	Sample collection of virological examination	Number	1000	1904
3.2.2	Investigation of PPR out-brake	Times	5	6
3.2.3	PPR Diagnosis by ELISA Method	Number	300	444
3.2.4	PPR Diagnosis by Pen site Test	Number	300	359
3.2.5	Bacteria and Virus Preservation for Vaccine Production.	Times	3	3
4	Serology			
4.1	PPR sero monitoring	Number	4000	4277
4.2	Serum Collection and examination for PPR control.	Times	1	4
4.3	Poultry sample collection and examination for salmonella and micro-plasma	Number	400	621
5	Molecular Diagnosis			
5.1	Molecular Diagnostic examination for Bird-Flu	Number	500	911
5.2	Gene sequencer Maintenance.	Times	3	4
5.3	Dispatch of sample to reference laboratory.	Number	200	303
6	Disease surveillance and investigation			
6.1	Emergency disease investigation team	Times	6	6
6.2	Sample collection and examination of PRRS disease in pig	Times	3	4
6.3	Disease Investigation in Animal Health and Anoestrus camping	Times	3	3
7	Zoonotic disease Investigation			

7.1	Sample collection and examination for Rabies	Times	3	3
7.2	Sample collection and examination for Brucella.	Number	200	1983
7.3	Disease surveillance and Investigation of H7N9	Times	2	2
7.4	Investigation of Japanese Encephalitis in Pig	Times	2	2
8	Food security			
8.1	Clinical mastitis Investigation in Dairy pocket Farm	Times	3	4
9	Staff Motivation			
9.1	Training on Laboratory Technology(30 Days duration) for mid Laval Technician	Times	1	1
10	Interaction programs			
10.1	Participator on Regional workshop	Times	5	5
10.2	Laboratory Program Planning	Times	1	1
10.3	Participator on Animal and Quarry workshop in RD office	Times	5 -	5

4. Laboratory Services

4.1 Parasitology section

Introduction : The parasitology section is involved in routine examination of different types of internal and external Parasites of animals and birds, besides of this, it is also involved in the investigations of different types of internal and external parasite causing adverse effect on animals and poultry health and production. For the investigation and diagnosis of parasites, fecal samples, blood samples and skin scrapings samples are collected and examined by adopting standard veterinary laboratory protocols.

As a routine examination or diagnosis of parasites, Direct smear method, Sedimentation method and The floatation methods and EPG count are commonly used. These methods are adopted for both quantitative and qualitative detection of parasites. The quantitative method to detect the gastrointestinal parasites in a gram of faces is performed using Mc-Master's slide by counting numbers of eggs.

The main objectives of Parasitological section is to, provide day to day diagnostic service to the farmers who come to this Laboratory with samples for parasitic diagnosis. And the investigation programmes are also conducted as a regular programmes.of CVL.

Parasitic burden during the fiscal year 2070/71

During the fiscal year 2070/71, A total of 822 faecal samples of cattle, buffaloes, sheep, goats, dogs poultry and other species of animals were tested in this section. Out of which 325 samples were found positive for different species of parasites. The month wise test result of fecal examination is given in table 1.

Table: Month wise distribution of Parasites found in the faecal sample examination

Month	No. of Sample tested	Species of Parasites					
		Fasciola	Param Phistomum	Strongylus	Trichuris	Ascaris	Coccidia
Shrawan	44	14(31.81%)	6(13.63%)	4(9.09%)	2(4.45%)	3(6.81%)	Cocci.
Bhadra	59	3(5.08%)	2(4.54%)	8(13.55%)	1(1.69%)	-	-
Aswin	41	9(21.95%)	6(14.63%)	1(2.43%)	1(2.43%)	-	-
Kartik	25	-	-	-	-	-	1(4%)
Mansir	28	3(10.71%)	3(10.71%)	-	-	-	1(3.57%)
Push	114	17(14.91%)	11(9.64%)	8(7.01%)	-	-	-
Magh	119	27(22.68%)	22(18.48%)	6(5.04%)	2(1.68%)	1(0.8%)	-
Fagun	157	24(15.28%)	26(16.56%)	5(3.18%)	2(1.25%)	2(1.25%)	-
Chaitra	95	10(10.52 %)	20(21.05%)	5(5.25%)	-	-	1(1.05%)
Baishak	69	9(13.04%)	10(14.49%)	14(20.28%)	3(4.34%)	-	-
Jeth	41	7(17.07%)	6(14.63%)	6(14.63%)	1(2.43%)	-	-
Ashad	30	4(13.33%)	4(13.33%)	3(10%)	-	-	1(3.33%)
Total	822	127(15.45%)	116(14.11%)	60(7.29%)	12(1.45%)	6(0.72%)	4(0.48%)

Sample tested under zoonosis programme

An investigation programme was carried out for the determination of parasitic Zoonosis in cats and dogs of Kathmandu Valley which was financially supported by the Zoonosis project, Budhanikanth Kathmandu. A total of 1000 samples were Collected from dog of Kathmandu and Lalitpur Districts but , the samples from cats were not available during the investigation period. The samples were tested by direct smear method for the detection of the Zoonotic species of parasites in dogs.

Table: Zoonotic species of parasites found in faecal samples of dogs of Kathmandu and Lalitpur district

S.N.	Type of parasite	Details of Results
1	Ascaris	70
2	Capillaria	61
3	Taenia	67
4	Trichuris	72
5	Negative samples	730
6	Total Samples	1000

4.2 Pathology section

Postmortem examination, hematology, histopathology and clinical biochemistry are major areas under the pathology section in CVL. Mostly the section receives specimens from all over the country either directly or through the the respective RVLs and NADIL. Beside this, the District Livestock Services Offices (DLSOs), veterinary practitioners and livestock and poultry farms as well as farmers deliver specimens for the purpose of disease diagnosis. Necropsy examination plays very important role in the diagnosis of diseases in different species of animals. Necropsy examination is the first step of disease diagnosis in case of dead animals. The history, clinical findings, epidemiological surveillance information is also helpful to necropsy examination for diagnosis of diseases.

Postmortem examination

During the fiscal year 2070/71, all together 4142 animals/poultry were brought for postmortem at CVL. The detail monthly and species wise distribution is shown in the table below.

Table: Month-wise postmortem examination of different species conducted at CVL during 070/71

S.No.	Type of animals/birds	Shrawan	Bhadra	Asoj	Kartik	Mangsir	Poush	Magh	Falgun	Chaitra	Baisakh	Jestha	Asadh	Total
1	Poultry Broilers	438	384	66	84	112	238	96	147	169	396	257	274	2661
2	Poultry Layers	119	84	27	29	21	47	22	34	32	28	33	24	500
3	Poultry Parents	46	36	0	0	26	0	7	8	24	37	32	43	259
4	Backyard poultry	53	97	22	34	28	45	31	29	41	33	36	29	478
5	Ducks	9	18	4	6	1	34	6	13	12	18	6	0	127
6	Pigeons	2	0	1	2	0	0	2	4	0	0	0	0	11
7	Wild Birds	3	4	1	0	0	0	0	0	0	0	0	0	8
8	Other birds	1	1	0	1	1	3	0	0	0	0	0	0	7
9	Goats	3	2	4	3	2	2	0	4	3	2	2	0	27
10	Pigs	5	1	2	4	5	7	3	3	4	4	1	1	40
11	Dogs	1	0	0	0	1	0	2	0	1	1	0	0	6
12	Rabbit	0	1	1	3	0	0	0	2	0	5	3	2	17
13	Cow	0	0	0	0	1	0	0	0	0	0	0	0	1
	Total	680	628	128	166	198	376	169	244	286	524	370	373	4142

The above table shows the number of postmortem was highest in the Shrawan followed by Bhadra and lowest in Asoj.

Table: Species-wise number of postmortem conducted at CVL during 070/71

S.No.	Species	Number	Percentage
1	Avian	4051	97.80%
2	Swine	40	0.96%
3	Caprine	27	0.65%
4	Rabbit	17	0.41%
5	Canine	6	0.14%
6	Bovine	1	0.02%
	Total	4142	100%

It can be seen in the above table that among the different species of animals brought for postmortem at CVL, avian species is the major one 97.80% followed by swine 0.96%.

Table: Monthly distribution of postmortem examination and diseases/pathological conditions diagnosed other than avian at CVL during year 070/71

Species	Diseases/ Pathological conditions	Shrawan	Bhadra	Asoj	Kartik	Mangsir	Poush	Magh	Falgun	Chaitra	Baisakh	Jestha	Asadh	Total
Caprine	PPR	2	2	3	2	2	2	0	3	2	2	2	0	22
	Tympany	0	0	1	0	0	0	0	0	0	0	0	0	1
	Pneumonia	1	0	0	0	0	0	0	1	0	0	0	0	2
	Toxicosis	0	0	0	0	0	0	0	0	1	0	0	0	1
	Enterotoxaemia	0	0	0	1	0	0	0	0	0	0	0	0	1
Swine	Classical Swine fever	3	1	2	2	3	3	3	3	4	4	1	1	30
	Toxicosis	0	0	0	0	1	1	0	0	0	0	0	0	2
	PRRS	0	0	0	1	0	1	0	0	0	0	0	0	2
	Pneumonia	2	0	0	1	1	2	0	0	0	0	0	0	6
Canine	Pneumonia	1	0	0	0	1	0	1	0	1	1	0	0	5
	Parvovirus infection	0	0	0	0	0	0	1	0	0	0	0	0	1
Rabbit	Pneumonia	0	1	1	3	0	0	0	2	0	5	3	2	17
	Hepatitis	0	0	0	0	0	0	0	0	0	0	0	0	0
	Coccidiosis	0	0	0	0	0	0	0	0	0	0	0	0	0
	Mycotoxycosis	0	0	0	0	0	0	0	0	0	0	0	0	0
Cow	Road accident	0	0	0	0	1	0	0	0	0	0	0	0	1
Total		9	4	7	10	9	9	5	9	8	12	6	3	91

Table: Monthly distribution of postmortem examination and diseases/pathological conditions diagnosed in avian species at CVL during year 070/71

S.N.	Diseases/Pathological conditions	Shrawan	Bhadra	Asoj	Kartik	Mangsir	Poush	Magh	Falgun	Chaitra	Baisakh	Jestha	Asadh	Total
1	Colibacillosis	47	41	21	36	43	31	34	47	58	13	48	37	582
2	Colibacillosis	58	63	13	17	21	53	49	68	73	56	33	41	545
3	Colibacillosis +Mycotoxicity	23	26	9	13	12	13	5	8	12	24	23	19	187
4	Colibacillosis	64	44	6	18	18	67	19	29	27	19	28	47	386
5	Egg peritonitis/ Salpingitis	11	9	0	0	1	2	0	0	0	1	2	0	26
6	Omphalitis	26	22	13	9	11	17	6	9	12	78	56	48	307
7	Salmonellosis	5	2	0	0	0	0	0	1	0	1	0	2	11
8	Nephritis	17	13	0	1	0	3	1	2	2	7	8	4	58
9	Enteritis	12	7	1	2	2	5	0	2	4	4	3	8	50
10	Pneumonia	27	31	5	6	3	33	3	5	13	21	17	14	178
11	Chronic Respiratory	89	72	14	18	17	42	15	22	28	26	24	39	406
12	Complicated CRD	78	65	4	7	8	23	9	14	13	21	19	27	288
13	CRD+Ascites	13	11	1	2	5	14	2	3	5	13	11	17	97
14	Avian Influenza	96	93	24	0	0	0	0	0	0	0	0	0	213
15	Ranikhet Disease	6	2	0	0	0	1	0	0	2	2	1	0	14
16	Ranikhet Disease+ Colibacillosis	2	0	0	0	0	1	0	0	0	0	1	0	4
17	Gumboro Disease(IBD)	22	17	2	8	6	13	4	6	9	40	24	31	182
18	IBD+Coccidiosis	2	1	1	2	0	1	1	1	1	15	8	2	35
19	IBD+Colibacillosis	1	1	1	1	0	1	1	1	1	2	1	1	12
20	IB/IB complex	1	0	0	0	0	0	0	0	0	0	1	0	2
21	Litchi Heart	5	2	0	0	0	1	0	1	1	2	1	2	15
22	LHD+ Coccidiosis	0	1	0	0	0	1	0	1	0	0	1	0	4
23	Marek's Disease	1	1	0	1	1	0	0	0	1	0	0	1	6
24	Lymphoid	1	1	0	0	1	0	0	0	1	0	0	1	5
25	Avian Encephalomyelitis	0	0	0	0	0	0	0	0	1	0	0	1	2
26	Coccidiosis	5	12	1	3	9	7	4	0	3	14	16	11	85
27	Coccidiosis+ Colibacillosis	3	14	1	2	2	2	1	0	1	8	2	1	37
28	Coccidiosis+CRD	2	12	1	4	2	2	2	0	0	5	2	1	33
29	Aspergillosis	0	1	0	0	0	0	1	0	0	2	0	0	4
30	Mycotoxicosis	17	19	1	4	11	11	3	2	2	7	4	3	84
31	Mycotoxicosis+CR	21	18	1	2	7	9	1	2	1	3	2	2	69
32	Mycotoxicosis+ Coccidiosis	3	5	0	0	0	2	0	0	0	0	0	0	10
	Gout	3	2	0	0	1	1	0	0	0	2	7	0	16
	Ascites	2	5	0	0	3	3	3	11	2	0	5	7	41
	Mineral	0	1	0	0	1	0	0	0	1	0	2	0	5

Vitamin	0	1	0	0	1	0	0	0	1	0	3	0	6
Sudden Death Syndrome	0	1	0	0	0	0	0	0	0	0	7	1	9
Immunosuppression	5	2	1	0	1	3	0	0	0	0	2	1	15
Stress Condition	1	5	0	0	1	5	0	0	2	0	1	0	15
Fowl Pox	1	1	0	0	1	0	0	0	1	0	1	1	6
Inclusion Body Hepatitis (IBH)	0	0	0	0	0	0	0	0	0	0	0	0	0
Putrified	1	0	0	0	0	0	0	0	0	0	0	0	1
Total	67	62	12	15	18	36	16	23	27	51	36	37	405

4.3 Hematology and Biochemistry Unit

A total of 1031 Blood samples were tested in FY 2070/071 for blood analysis. Out of 801 cattle blood samples examined 74 were positive for Babesia spp., 59 for Anaplasma spp. and rest were negative for any Blood Parasite. Similarly out of 16 buffalo blood samples, 4 were positive for Babesia spp., 3 for Anaplasma spp. and rest were found Negative. Out of 54 Dog blood samples, 7 were positive for Babesia spp., 6 Anaplasma spp. and rest were negative for any blood parasites. Similarly out of 6 Horses blood samples examined, 1 was positive for Trypanosoma spp. and 2 for Babesia spp. Out of 150 Goat and Sheep blood samples, 2 Micro flora spp and rest were Negative. Out of 4 Pig blood samples tasted all were negative for any blood parasites.

Table: Blood samples were tested in FY 2070/071

Species	PCV	HB	TLC	DLC	Blood protozoa positive	Total Sample	Remarks
Cattle	801	801	801	801	94 Babesia, 59 Anaplasma sps	801	
Buffaloes	16	16	16	16	4 Babesia, 3 Anaplasma sps	16	
Dog	54	54	54	54	7 babesia, 6 Anaplasma sps	54	
Horse	6	6	6	6	1 Tripanosoma sps, 2 Babesia	6	
Goat/Sheep	150	150	150	150	2 Microflaria sps	150	
Pig	4	4	4	4	negative	4	
Total Sample	1031	1031	1031	1031		1031	

Table: Blood samples tested for biochemical parameters in FY 2070/071

Type of Sample tasted	Dog	Cattle	Buffaloes	Goat	Sheep	Total
Urine	4	12	3			19
Calcium	3	14	4	120	56	197
Phosphorous		5	2	120	56	183
Magnesium		5		120	56	181
Gulcouse	12					12
Total	19	36	9	360	168	592

4.4 Histopathological Section

Postmortem examination provides a tentative diagnosis of a disease and the histopathology technique always lays a presumptive diagnosis. In this way, it is essential to circulate necropsy and sometimes biopsy samples through various laboratory disciplines. Histopathology section processes the samples and provides the result within 7-10 days period by wax technique method.

Histopathology unit received a total of 104 samples from different animal species during the fiscal year 2070/71. Out of 104 samples examined, 33 samples were found to be positive for various histopathological conditions. Out of them Infectious Bursal Disease (10), Marek's Disease (5), Nephritis (5), Hepatitis (3) and coccidiosis (10) were major disease condition.

Table : The month-wise sample flow from different species

S.No.	Months	Species			
		Layers	Broiler	Goat	Total
1	Shrawan	10	5	0	15
2	Bhadra	2	5	0	7
3	Ashoj	5	4	0	9
4	Kartik	2	0	3	5
5	Mangshir	5	4	5	14
6	Poush	0	5	2	7
7	Magh	5	5	0	10
8	Falgun	5	3	0	8
9	Chaitra	3	3	0	6
10	Baishak	10	5	0	15
11	Jestha	0	2	0	2
12	Ashar	3	3	0	6
Total		50	44	10	104

4.5 Microbiology section

4.5.1 Bacteriology and mycology unit

The bacteriology unit receive samples from various sources such as farmers, Central veterinary hospital, referral samples from private clinics, Regional veterinary laboratories and direct from the field as well DLSO's. In total 2797 samples were received in this unit in fiscal year 2070/71. The sample received was milk, urine, tissues, water, nasal swabs, ear swabs, and skin scrapings. Different bacterial species were isolated from specimens (milk, urine, tissues, water, nasal swabs, ear swabs, and skin scrapings) in the FY 2070/71.

Table : The Bacterial species isolated in the laboratory in the FY 2070/71

S.N.	Bacterial species	Number of isolated
1	<i>Escherichia coli</i>	509
2	<i>Staphylococcus spp.</i>	213
3	<i>Streptococcus spp.</i>	54
4	<i>Salmonella spp.</i>	52
5	<i>Klebsiella spp.</i>	6
6	<i>Enterobacter spp.</i>	22
7	<i>Bacillus spp.</i>	71
8	<i>Mycobacterium spp.</i>	1
9	<i>Micricoccus</i>	3
10	<i>Pasteurella multocida</i>	7
11	<i>Clostridium chauvoei</i>	1
12	<i>Proteus</i>	2

A total of 240 fungal samples were received in this unit for mycological culture and among these 90 samples were found negative for fungal growth. The most common fungus isolated from pathological lesion of PM sample and poultry feed are *Penicillium* and *Aspergillus Spp.*

Table : The Bacterial species isolated in the laboratory in the FY 2070/71

S.N.	Fungal species	Number of isolated
1	<i>Penicillium spp.</i>	98
2	<i>Aspergillus spp.</i>	52

CMT and Bacteriological test for milk samples:

A total of 540 bovine samples were received in this unit for CMT and bacteriological culture and among these 98 sample were found negative for CMT test and no growth on bacteriological culture.

Table : The Bacterial species isolated in the milk samples of CMT positive cases in the FY 2070/71

S. N.	Bacterial species	Number of isolated
1	<i>Escherichia coli</i>	137
2	<i>Bacillus spp.</i>	25
3	<i>Klebsiella spp.</i>	35
4	<i>Staphylococcus spp.</i>	207
5	<i>Streptococcus spp.</i>	33
6	<i>Proteus</i>	2
7	<i>Micricoccus</i>	2
8	<i>Candida spp.</i>	1

Antibiotic sensitivity Test :

The bacterial isolates from the milk sample received from mastitis cases were subjected to antibiotic sensitivity test. Among the antibiotic Gentamicin, tetracycline, ciprofloxacin and Azithromycin are sensitive for the treatment of cattle in case of mastitis disease for the given organism.

Table : Antibiotic Sensitivity test Result in mastitis positive cases in the FY 2070/71

S. N.	Bacterial specimen isolated	Number of isolates tested	Antibiotic sensitivity percentage													
			G	T	Cip	Am	A	S	Azm	CP	Cl	N	Li	E	Ak	Do
1	<i>E.coli</i>	137	86	81	87	44	42	22	97	42	55	10	3	6	61	32
2	<i>Staphylococcus spp.</i>	207	85	84	87	44	43	21	95	43	45	9	4	6	60	30
3	<i>Streptococcus Spp.</i>	33	85	84	86	40	39	20	91	41	42	8	4	5	59	29
4	<i>Bacillus spp.</i>	25	56	48	34	40	8	19	52	18	-	-	-	-	-	38
5	<i>Klebsiella spp.</i>	35	36	36	28	33	32	-	-	33	-	21	-	-	54	-
6	<i>Proteus spp.</i>	2	100	100	100	-	-	-	100	100	-	-	-	-	-	-
7	<i>Micrococcus spp.</i>	7	98	98	95	-	-	-	90	90	-	-	-	-	-	-
8	<i>Candida spp.</i>	3	-	-	-	-	-	-	-	-	-	-	-	-	-	-

G= Gentamicin, T= Tetracycline, Cip= Ciprofloxacin, Am=Amoxicillin, A= Ampicillin, S= Streptomycin, Azm= Azithromycin, Cp= Cephalaxin, Cl= Colistin, N= Neomycin, Li= Livomycin, E= Erythromycin, Ak= Amikacin, Do= Doxycycline, - = not use

4.5.2 Virology unit

This unit is responsible for diagnosis of various viral diseases of birds and animals by using various laboratory diagnostic methods. It receives the samples from postmortem unit of CVL itself and from the field, RVL and DLSOs. The unit has great contribution in the diagnosis of important zoonotic diseases like Avian Influenza and Rabies. Rapid test is used for the initial test of Avian Influenza followed by PCR in the molecular Biology section. Newcastle Disease and Infectious Bursal Disease are diagnosed using rapid test kit and HA/HI is used for ND as well.

During the fiscal year 2070/71, a total of 2058 samples from chicks, ducks, wild birds, goats, dogs and other animal species were examined for different viral diseases and by various methods. Out of this only 428 samples were found positive for various viral diseases.

Table: Month-wise distribution of different diseases in virology unit.

S.N.	Months	AI			ND			IBD			PPR			Rabies		
		Total	Pos	Neg	Total	Pos	Neg	Total	Pos	Neg	Total	Pos	Neg	Total	Pos	Neg
1	Shrawan	189	133	20	55	1	54	4	0	4	1	0	1	1	1	0
2	Bhadra	172	48	20	12	0	12	8	1	7	0	0	0	3	1	2
3	Ashoj	29	6	23	31	1	30	16	1	15	0	0	0	1	1	0
4	Kartik	5	0	5	4	0	4	12	8	4	50	0	50	1	1	0
5	Mangshir	15	6	9	8	0	8	11	3	8	2	0	2	2	1	1
6	Poush	12	3	9	2	0	2	1	0	1	148	1	147	4	2	2
7	Magh	9	1	8	11	1	10	9	4	5	0	0	0	4	0	4
8	Falgun	33	4	29	26	1	25	27	3	24	58	0	58	1	1	0
9	Chaitra	21	2	19	30	5	25	33	15	18	8	8	0	4	2	2
10	Baishak	11	2	9	8	0	8	9	1	8	94	22	72	8	4	4
11	Jestha	12	2	10	13	3	10	20	16	4	0	0	0	3	2	1
12	Ashar	20	5	15	20	2	18	18	11	7	50	0	50	2	0	2
Total		528	212	176	220	14	206	168	63	105	411	31	380	34	16	18

In the fiscal year 2070/71, a total of 528 tracheal, cloacal swab and brain samples were received from CVL, NAL, RVLs and DLSOs for Avian Influenza (AI) diagnosis. Out of these, 212 samples were found to be positive for Flu A in rapid test kit method. All positive samples and 10% of negative samples were sent to molecular unit for identification of subtype H5 and H9.

Similarly, a total of 220 tracheal swab samples from poultry were received from different districts of Nepal which were tested for Newcastle Disease (ND) through antigen test method and further by HA/HI. Out of these 14 samples were found to be positive for ND.

A total of 168 samples suspected for Infectious Bursal Disease (IBD) from poultry were received from different districts of Nepal which were tested through rapid antigen test method. Out of these, 63 samples were found to be positive for IBD.

Likewise, a total of 411 nasal and ocular swab samples received for PPR test and 34 samples for rabies test, 31 and 16 samples were found to be positive for PPR and Rabies respectively when PPR was tested through penside test and Rabies by rapid antigen test method.

Table: Result of PPR and ALV Ag ELISA Test.

S.No.	Months	PPR Ag ELISA			ALV Ag ELISA		
		Total	Pos	Neg	Total	Pos	Neg
1	Shrawan	0	0	0	0	0	0
2	Bhadra	0	0	0	0	0	0
3	Ashoj	0	0	0	0	0	0
4	Kartik	0	0	0	0	0	0
5	Mangshir	0	0	0	0	0	0
6	Poush	4	4	0	0	0	0
7	Magh	0	0	0	145	5	140
8	Falgun	40	0	40	0	0	0
9	Chaitra	0	0	0	0	0	0
10	Baishak	0	0	0	0	0	0
11	Jestha	28	12	16	324	21	303
12	Ashar	60	11	49	96	39	57
Total		132	27	105	565	65	500

Out of 132 and 565 samples tested through PPR and ALV Ag ELISA test, 27 and 65 samples were positive for PPR and ALV respectively.

4.6 Serology section

Serology section of CVL performs different serological tests for the diagnosis, monitoring and surveillance of animal diseases mainly associated with viral and bacterial infection. Most of the samples are submitted to this unit by Regional Veterinary Laboratories, District Livestock Service Offices, Quarantine Check-posts, private practitioner, farmers and staff of CVL during disease outbreak investigations well as routine diagnosis. This section possess capacity and facility of Competitive Enzyme Linked Immunosorbent Assay (ELISA), IgM capture ELISA, Indirect ELISA, Tube agglutination Test, Agar-Gel Immuno-Diffusion (AGID) test, Plate agglutination test, A solid phase immune assay (Immuno- comb) and rapid tests. Progress report of Serological investigation of various diseases in animals and birds during 069/70 is as follows

Table :Peste-des Petitis Ruminant (PPR), C-ELISA Antibody Test Results (Outbreak Samples)

S.N.	Districts	No. of sample tested	Test Results		Percentage Positive (%)
			Positive	Negative	
1	Sunsari	4	1	3	25.00
2	Saptari	17	7	10	41.18
3	Rauthat	3	3	0	100.00
4	Makwanpur	11	7	4	63.64
5	Chitwan	12	5	7	41.67
6	Dhadhing	26	13	13	50.00
7	Kathmandu	2	0	2	0.00
8	Lalitpur	4	4	0	100
9	Nuwakot	7	0	7	0.00
10	Sindhupalchowk	16	0	16	0.00
11	Ramechhap	12	12	0	100
12	Nawalparasi	22	5	17	22.73
13	Jumla	8	2	6	25.00
Total		144	59	85	40.97

A total of 144 serum samples of goats from outbreaks areas were tested, 40.97 % were positive for PPR antibody. The result shows that the PPR antibody positive percentage was found highest in two districts (100%) and lowest (0%) in three districts.

Table : PPR Sero-monitoring C-ELISA Antibody Test Results

S.N.	Districts	No. of sample tested	Test Results		Percentage Positive (%)
			Positive	Negative	
1	Morang	308	233	75	75.65
2	Sunsari	200	138	62	69.00
3	Saptari	260	164	96	63.08
4	Siraha	136	79	57	58.09
5	Jhapa	373	157	216	42.09
6	Nuwakot	122	59	63	48.36
7	Dhadhing	72	46	26	63.89
8	Rammechhap	53	28	25	52.83
9	Chitwan	43	19	24	44.19
10	Sarlahi	143	79	64	55.24
11	Dhanusha	242	158	84	65.29
12	Mahottari	50	25	25	50.00

13	Rauthat	50	18	32	36.00
14	Makwanpur	71	27	44	38.03
15	Kapilbastu	199	39	160	19.59
16	Rupendhehi	224	125	99	55.80
17	Nawalparasi	27	9	18	33.33
18	Kaski	148	81	67	54.73
19	Gorkha	157	109	48	69.43
20	Baglung	99	25	74	25.25
21	Magdi	120	59	61	49.17
22	Dang	232	119	113	51.29
23	Surkhet	276	227	49	82.25
24	Banke	167	76	91	45.51
25	Bardia	112	46	66	41.07
	Total	3884	2145	1739	55.23

A total 3884 serum samples of vaccinated goats were tested. Out of those samples 55.23% were found positive for PPR antibody. The result shows that the PPR antibody positive percentage was found highest in Surkhet (82.25%). The antibody positive percentage found in the serum samples of Rauthat, Makwanpur, Kapilbastu, Nawalparasi and Baglung were not satisfactory. The low antibody positive percentage might be due to either sampling error or cold chain problem or the samples were collected earlier.

Table : Test Result Of Brucellosis, Toxoplasmosis and Leptospirosis

S.N.	Diseases	Test Method	Total No. of Sample tested	Results	
				Positive	Negative
1	Brucellosis	ELISA/PAT	3077	16	3061
2	Toxoplasmosis	ELISA	2760	419	2341
3	Leptospirosis	ELISA	2112	201	1911

A total of 3077 serum samples from cattle, buffalo, goat, sheep and pig were tested for *Brucellosis* antibody in CVL by ELISA method and plate agglutination test (PAT) method. Only 16 samples were found positive. Likewise a total of 2760 serum samples from cattle, Buffalo, sheep, goat, pig and dog were tested for *Toxoplasma gongii* antibody in CVL by indirect ELISA method. Only 419 samples were found positive. Similarly a total of 2112 serum samples from cattle and Buffalo were tested for *Leptospira hardjo* antibody in CVL by ELISA method. Only 201 samples were found positive.

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

S.N.	Districts	<i>Salmonella pullorum</i> (PAT)			<i>Mycoplasma gallisepticum</i> (PAT)		
		Total Tested sample	Positive	Negative	Total Tested sample	Positive	Negative
1	Kaski	81	37	44	81	24	57
2	Dhading	72	11	61	72	27	45
3	Jhapa	56	16	40	50	17	33
4	Lalitpur	52	20	32	52	14	38
5	Kathmandu	85	9	76	85	7	78
6	Chitwan	0	0	0	11	0	11
Total		- 346	93	253	351	89	262

A total 346 number of poultry serum samples were tested for *Salmonella pullorum* antibody by PAT method and 93 were found positive. Similarly, 351 number of poultry serum samples were tested for *Mycoplasma gallisepticum* antibody by PAT method and 89 were found positive.

Table: Antibody Test Results of Porcine Reproductive and Respiratory Syndrome Virus (PRRS) in swine serum by using ELISA

S.N.	Districts	No. of sample tested	Test Results	
			Positive	Negative
1	Jhapa	58	0	58
2	Morang	121	0	121
3	Sunsari	100	0	100
4	Kaski	30	0	30
5	Kailai	41	1	40
6	Kavre	48	0	48
7	Bardia	44	0	44
8	Makwanpur	5	0	5
Total		447	0	446

A total of 447 serum samples of swine were tested for Porcine Reproductive and Respiratory Syndrome (PRRS) Virus Antibody by ELISA method at CVL. The samples were received from Jhapa, Kavre, Morang, Sunsari, Kaski, Kailai, Bardia and Makwanpur district. None of them were found positive.

Table: Japanese Encephalitis Virus Antibody test result in swine serum using ELISA test

S.N.	Districts	No. of sample tested	Test Results	
			Positive	Negative
1	Narc	182	141	141
2	Kathmandu	62	0	62
3	Jhapa	44	2	42
4	Surkhet	36	0	36
5	Rupendhehi	16	0	16
6	Kanchanpur	17	0	17
	Total	357	143	314

A total of 357 serum sample of swine was tested for Japanese Encephalitis Virus Antibody by ELISA method at CVL. Out of those samples only 143 samples were found positive.

Table: Avian Luekosis Virus Antigen test result in Cloacal Swab by using ELISA test

S.N.	Districts	No. of sample tested	Test Results	
			Positive	Negative
1	Kaski	175	4	171
2	Kathmandu	5	4	1
3	Lalitpur	5	2	2
4	Bhaktapur	6	3	3
5	Nuwakot	14	13	1
	Total	205	26	179

A total 205 cloacal swab samples of poultry were tested for Avian Leukosis Virus Antigen by ELISA method at CVL. The samples were received from Kathmandu, Bhaktapur, Lalitpur, Kaski and Nuwakot districts. Out of those samples 26 samples were found positive.

Table: Avian Influenza Flu A Virus Antibody Test Results in poultry serum using ELISA

S.N.	Districts	No. of sample tested	Test Results	
			Positive	Negative
1	Morang	57	0	57
2	Dhading	36	8	18
3	Saptari	35	0	36
4	Jhapa	56	8	48
	Total	184	16	159

A total 184 serum samples of poultry (duck/chicken) were tested for Avian Influenza Flu A Virus Antibody by ELISA method at CVL. The samples were received from Morang, Dhading, Saptari and Jhapa districts. Out of those samples 16 samples were found positive.

Table: MS/MG Virus Antibody Test Results in poultry serum by using ELISA

S.N.	Districts	No. of sample tested	Test Results	
			Positive	Negative
1	Dhadhing	38	1	37
2	Kaski	51	0	51
	Total	89	1	88

A total 89 serum samples of poultry were tested for *Mycoplasma synovae* and *Mycoplasma gallisepticum* Virus Antibody by ELISA method at CVL. The samples were received from Dhadhing and Kaski districts. Out of those samples only 1 sample was found positive.

4.7 Molecular Biology section

Molecular tools are increasingly important in modern animal disease research not only in other country but also in our country Nepal. In view of this Central Veterinary Laboratory (CVL) has established molecular biology unit in 2003. This laboratory has started to diagnose disease especially avian influenza by using RT-PCR technique. This technique is also useful for the epidemiological study of diseases and understanding of the genetic relationships between and within species of infectious organisms.

Table: Month-wise samples flow and test results of Molecular diagnosis of avian influenza virus (subtype H5 and H9) from outbreak samples by PCR Method

S.No.	Month	Total Sample tested	Results		
			H5	H9	Negative
1	Shrawan	218	126	4	88
2	Bhadra	144	48	3	93
3	Ashwin	10	0	2	8
4	Kartik	6	0	0	6
5	Mansir	5	0	4	1
6	Poush	6	0	3	3
7	Magh	0	0	0	0
8	Falgun	4	2	2	0
9	Chaitra	16	0	5	11
10	Baishakh	11	0	8	3
11	Jestha	29	0	0	29
12	Ashad	7	0	0	3
	Total	456	176	34	246

In the Fiscal year 2070/ 2071, a total of 456 tracheal and cloacal swab samples different avian species from the outbreak areas were tested by molecular diagnosis methods in CVL. The samples were tested for Flu A by Rapid test. For H5, N1 and H9, Conventional and Real Time PCR (rRT-PCR) method was used. Out of those samples, 38.59% and 7.45% were found positive for avian influenza sub type H5 and H9 respectively.

During this period samples were received from 19 districts out of which outbreaks were occurred in Bhaktapur, Lalitpur, Kathmandu, Dhadhing Kaski, Makwanpur, Rupendhehi, Nawalparasi Sindhuli, Tanahu, Sindupalchowk, Kavre and Sunsari districts. The highest numbers of samples were received in the month of Shrawan and Bhadra. None of the samples received in month of Magh.

Sub-clinical Mastitis in Cattle of Morang, Jhapa, Dhanusha and Saptary District in Nepal

Dr. S. P. Gautam and Mr. M.K. Karna

Abstract

To find the incidence of sub clinical mastitis within the dairy pocket of Morang, Jhapa, Dhanusha and Saptary districts. A total of 65 milk Samples were submitted to Central Veterinary laboratory. These samples were subjected for the Sodium Laurel Sulphate screening test for detecting mastitis in the animals. In screening test 33 milk samples of animals were found to be positive. These positive samples were subjected for the microbiological culture to acquaint with the bacterial spectrum responsible for sub clinical mastitis and the drugs sensitive to them. Study showed that the incidence of the sub-clinical mastitis were less in first or second lactation and were increased with the increase in lactation. This increment in the percentage may be due to entry of the bacteria in the higher yielding animal and older animal through the relaxed teat sphincters. The percentage of major pathogen contributing to the subclinical mastitis was *Staphylococcus* Spp. (38.10), *E-coli* (26.20), *Streptococcus* Spp. (21.42), *Bacillus* Spp. (11.90), *Candida* Spp. (2.38). From the result obtained in the CVL Gentamycin is the most potent antibiotic to treat the sub clinical mastitis affected animals.

Introduction

Sub-clinical mastitis is a herd problem, where as clinical mastitis is an individual problem of the Lactating animals. Intra mammary infection with organisms leads to substantial losses from both clinical and sub clinical mastitis. Sub-clinical mastitis may cause milk loss up to 80% in a herd causing enormous economic loss. Bovine mastitis is universally recognized as one of the most economic diseases confronting the dairy industry. The loss result from sub clinical mastitis is more than the clinical mastitis. The magnitude of the losses due to the disease warrants the need for continued work on different aspect of mastitis. Clinical mastitis is easy to diagnose while sub clinical mastitis is difficult to diagnose as it does not exhibit any clinical symptoms. Not much attention has been paid on sub clinical mastitis and only limited study has been carried out in our country. Four terai districts Morang, Jhapa, Dhanusha and Saptary of Eastern and Central Development region are highly potential districts for milk production. Therefore, an attempt has been made to know the status of sub-clinical mastitis and suggest an appropriate measure to combat the disease.

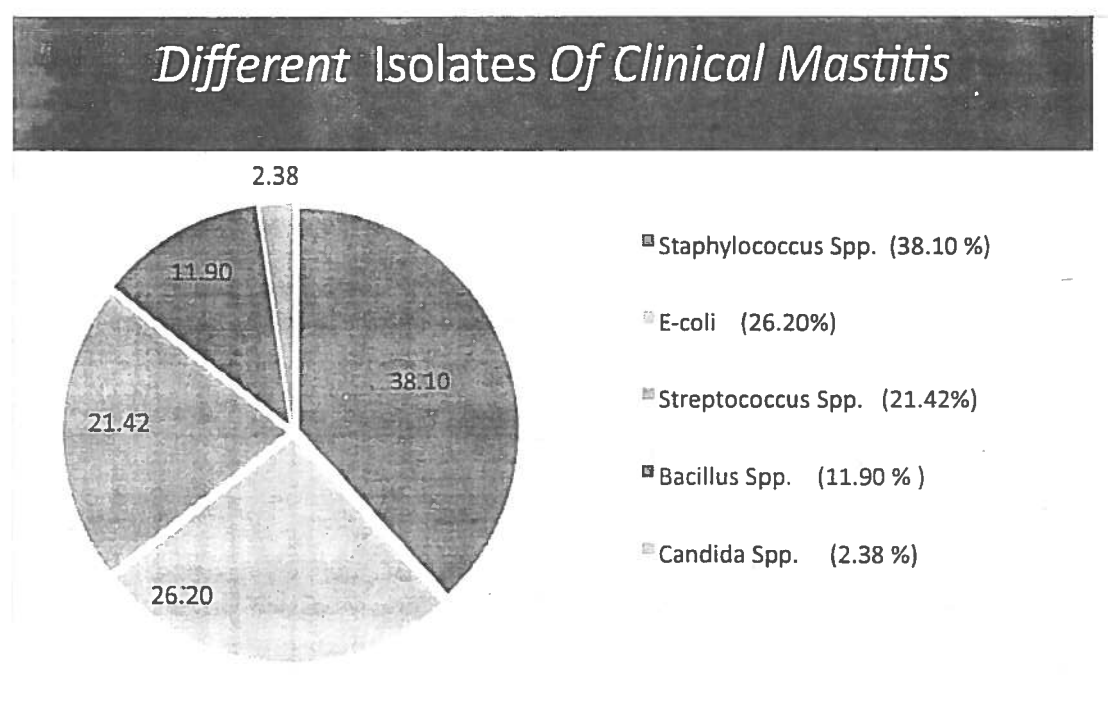
Materials and methods

The dairy pocket of Morang, Jhapa, Dhanusha and Saptary districts were selected for the study and a total of 65 milk Samples were collected and brought to Central Veterinary laboratory these were subjected for the Sodium Laurel Sulphate screening test. History and relevant information such as age of animal, lactation number, date of calving, previous mastitis history, date of collection of samples were duly collected and recorded before sample collection. In the process of collecting samples the udders of the animals were thoroughly cleaned with water and allowed to dry, the teats were mopped with sterile gauze several times and finally with gauze soaked in 70 per cent alcohol, nearly 20 ml of milk sample from each quarter was collected in two separate sterilized screw-capped bottles. The collection tubes were marked as right front (RF), right hind (RH), left front (LF) and left hind (LH). Samples were kept in icebox at 4 °C. Isolation of bacteria was done by streaking the sample on Mc Con key Agar, Nutrient agar, and blood agar These were incubated aerobically at 37 °C for 24

hours. The isolates were identified on the basis of cultural, morphological and biochemical characteristics.

Result and discussion

In screening test 33 milk samples of animals were found to be positive. These positive samples were subjected for the microbiological culture to acquaint with the bacterial spectrum responsible for sub clinical mastitis and the drugs sensitive to them. Study showed that the incidence of the sub-clinical mastitis were less in first or second lactation and were increased with the increase in lactation. This increment in the percentage may be due to entry of the bacteria in the higher yielding animal and older animal through the relaxed teat sphincters. The percentage of major pathogen contributing to the subclinical mastitis was *Staphylococcus* Spp. (38.10), *E-coli* (26.20), *Streptococcus* Spp. (21.42), *Bacillus* Spp. (11.90), *Candida* Spp. (2.38). From the result obtained in the CVL Gentamycin is the most potent antibiotic to treat the sub clinical mastitis affected animals.



Conclusion

Bovine mastitis is universally recognized as one of the most economic diseases confronting the dairy industry. The loss result from sub clinical mastitis is more than the clinical mastitis. The magnitude of sub clinical mastitis is more than 50% in the present study in the dairy pockets of eastern development region. Hygienic practices during milking and early diagnosis as well as treatment of subclinical mastitis will reduce the loss incurred by the farmers. Teat dipping practice using Glycerin and povidin Iodine in equal volume showed good result in the past in controlling the mastitis in the country.

REGIONAL ANIMAL DISEASE INVESTIGATION LABORATORY, BIRATNAGAR (EASTERN REGION)

Regional Veterinary Laboratory (RVL) has been situated in sub-metropolitan city, Biratnagar-17, of eastern Nepal and was established in the fiscal year 1988/1989 AD. But until 1990/1991, the laboratory was not functional and could not perform its activities as per objectives due to lack of manpower, necessary equipments and frequent changes in organizational structure. From fiscal year 1991/1992, the RVL has its separate identity. There was provision of manpower and other logistics. The program was launched as per objectives.

The working area of this RVL is all districts of Eastern Development Region (EDR). In this eastern region, there are three zones (Mechi, Koshi and Sagarmatha) and 16 districts. Geographically, the region is divided into three eco-zones (high hills, mid hills and terai).

High hills

This eco- zones lies in the northern part of the region covering Taplejung district of Mechi zone, Sankhuwasabha district of Koshi zone and Solukhumbu district of Sagarmatha zone. Livestock rearing is the main occupation of the farmers in this region. Yak/Nak, chauri, sheep and goat are being reared in this region.

Mid-hills

This region falls between high hills on its north and tarai at the south. Panchthar, Illam, Dhankuta, Terathum, Bhojpur, Okhaladhunga, Khotang, and Udaypur districts are under this eco-zone. Farmers follow mixed farming system and agro-based livestock industries are their main occupation. Cattle, buffalo, swine, goat are being reared in this region. Poultry and rabbit farming are also popular among the farmers

Terai

Jhapa, Morang, Sunsari, Saptari and Siraha districts of Nepal are under this eco-zone. Though traditional system of livestock rearing is followed in this region, in recent years, poultry, dairy industries and pigery are being commercialized especially in east-west highway corridor.

To provide proper laboratory diagnosis and improve in the quality of veterinary services, the government has established five regional laboratories, one in each development region of the country. Primary laboratories in 55 and basic laboratories in 15 districts have been established to improve and upgrade existing disease diagnosis system.

Objectives of Regional Veterinary Laboratory

- To provide prompt and efficient disease diagnostic services to the farmers of the region.
- To investigate and diagnose the epidemics in the region.
- To assist and support DLSOs in disease diagnosis and epidemic control.
- To supervise and assist in diagnostic services to basic and primary laboratories situated in DLSOs of the region.
- To collect, analyze and predict the animal diseases prevailing in the region.
- To develop human resources for the field level veterinary services.
- To co-ordinate and support national animal 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.
- To support animal health and infertility camps in the region.

Annual Progress Report (2070/2071)

तेश्रो चौमासिक तथा वार्षिक प्रगति आ.व. २०७०।०७१										
सि.न	विवरण	इकाई	वार्षिक लक्ष	वार्षिक बजेट	तेश्रो चौ. लक्ष	तेश्रो चौ. प्रगति	तेश्रो चौ. बजेट	भार	वार्षिक प्र.	वार्षिक प्रगति प्रतिशत
१	Parasitological examination									
	परजिबि ई.पि.जि. गणना लाभ कल्चर	संख्या	४००	७३	१३०	१३०	२३	०।५३	४०६	१००
	रगत परीजीवी परीक्षण	संख्या	३००	८५	१००	१०३	२५	०।५८	३३१	१००
	क्लिनिकल हिमाटोलोजिकल परीक्षण	संख्या	४००	७७	१३०	१०३	२५	०।५८	३३१	८२।७५
२	वायोकेमिकल परीक्षण									
	क्यलसियम, फस्फोरस, प्रोटी, ग्लूकोज	संख्या	४००	८०	१३०	२२०	२५	०।५८	४९६	१००
	पिसाप परीक्षण	संख्या	१००	४१	३३	३३	१४	०।३२	१०१	१००
३	प्याथोलोजिकल परीक्षण									
	शव परीक्षण	संख्या	३५०	६१	११६	१८४	२१	०।४६	५६३	१००
४	माईक्रोवायोलोजिकल परीक्षण जिवाणु कल्चर तथा आईसोलसन	संख्या	८००	१३०	२६६	४०५	४३	०।९९	१२४०	१००
५	एन्टिवायोटिक सेन्सीटीभिटी परीक्षण	संख्या	४५०	७४	१५०	२५९	२५	०।५८	६५४	१००
६	ट्रुसि कल्चर तथा आईसोलसन	संख्या	१००	६३	३३	५२	२१	०।४६	१५५	१००
७	माईरोबिज परीक्षण									
	रनिखेत रोग परीक्षण	संख्या	७५	६७	२५	६१	२३	०।५३	१२८	१००
	गम्बारो रोग परीक्षण	संख्या	७५	६६	२५	५०	२१	०।४६	१००	१००
	पि.पि.आर. पेन्साईट परीक्षण	पटक	३	५०	१	१	२१	०।४६	११	१००
८	सिरोलोजिकल परीक्षण									
	माईक्रोप्लाजमा परीक्षण	संख्या	२००	८०	६६	११०	२७	०।६२	२८३	१००
	सालमोनेला परीक्षण	संख्या	२००	७३	६६	११५	२३	०।५३	३२१	१००
९	नमूना परीक्षणको लागी अन्य प्रयोगशालामा पठाउने	संख्या	४००	७५	१००	१८४	२५	०।५८	५५४	१००
१०	गाईमा सवलिकनिकल मासटार्डिटिस अन्वेषण	पटक	३	१२६	१	१	४५	१।०४	३	१००
११	एन्टिवायोटिक रेसिटेन्स परीक्षण	पटक	३	१५०	१	१	५०	१।१५	३	१००
१२	दूध तथा मासमा एन्टिवायोटिक रेसिड्यू परीक्षण	पटक	३	५०	१	१	१७	०।३९	३	१००
१३	EDIT व्यवस्थापन	पटक	३	६४	१	२	२१	०।४८	५	१००

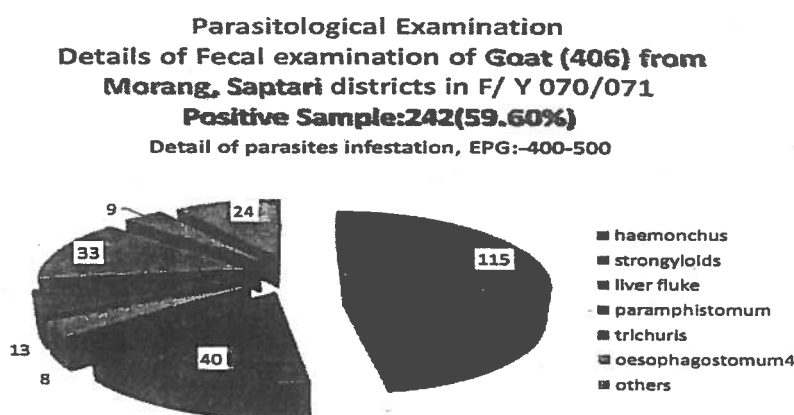
१४	ईपिडेमीक अन्वेषण	पटक	३	१७७	१	२	५७	११३१	५	१००
१५	क्षेत्रीयस्तरमा प्राथमिकता रोग पहिचान, अभिलेखिकरण तथा रिपोर्टिङ	पटक	१	३९	१	१	३९	०१९०	१	१००
१६	जूनोर्टिकरग निदान प्रयोगशाला कार्यक्रम									
	एभियन इन्फ्लून्जा परिक्षण	संख्या	१५०	६७	५०	१२८	२२	०१५१	२३१८	१००
	रेविज रोग परिक्षण	संख्या	५०	१५६	१५	५	५०	१११५	२०	४०
	बूमेला परीक्षण	संख्या	२००	८५	६७	११६	२८	०१६५	२५७	१००
	क्षय रोग परिक्षण	संख्या	५०	१३४	१५	१५	४४	११०१	५०	१००
१७	क्ष.स्तरमापि.पि.आर. खोप वैक व्यावस्थापन	पटक	३	५५	१	१	१५	०१३५	३	१००
१८	क्ष.स्तरमा रेविज खोप वैक व्यावस्थापन	पटक	३	५५	१	१	१५	०१३५	३	१००
१९	पि.पि.आर. सेरो मनिटरिङको लागि नमूना संकलन तथा प्रेषण	पटक	१	२३१	१	१	२३१	५१३३	१	१००
२०	खोरेत सिरो मनिटरिङको लागि नमूना संकलन तथा प्रेषण	पटक	२	२२७	१	१	११७	२१७०	१	१००
२१	रानिखेत सिरो मनिटरिङको लागि नमूना संकलन तथा प्रेषण	पटक	१	१४५	१	१	१४५	३१३४	१	१००
२२	जिल्ला प्रयोगशाला अनुगमन गर्ने	पटक	३	१०४	१	१	३५	०१८१	३	१००
२३	पशूपक्षी रोग अन्वेषण सम्बन्धि प्राविधिक अन्तरक्रिया गोष्ठी (क्षेत्रियस्तरिय)	पटक	१	१०१	०	०	०	२.३३	१	१००
२४	जिल्ला स्तरीय स्तलगत प्रयोगशाला क्ष.स्तर तालिम	पटक	१	११८	०	०	०	२१३३	१	१००
२५	बजेट तर्जुमा प्रगति, समिक्ष अन्य गोष्ठी भाग लिने	पटक	३	४८	१	१	१६	०१३७	३	१००
२६	अर्ध वार्षिक इपिडेमीयोलोजि बुलेटिन प्रकाशनको सामाग्री तयार गर्ने	पटक	२	२८	१	१	१४	०१३२	२	१००
२७	वार्षिक प्राविधिक पुस्तिका सामाग्री तयार गर्ने	पटक	१	४२	०	०	३१	०१७२	१	१००
२८	पुस्तक तथा जर्नल खरिद	पटक	१	१५	१	१	१५	०१३५	१	१००
२९	ल्याव एनिमल व्यवस्थापन	पटक	१२	४३	४	४	१४	०१३२	१२	१००
३०	मेशिनरी सामान प्रयोगशाला सामाग्री सरक्षण गर्ने	पटक	१	२८	१	१	२८	०१६५	१	१००
३१	कम्पटर अपरेटर, माली, स्वीपर तथा अन्य	पटक	१	४७	१	१	४७	११०८	१	१००
३२	कार्यालय सरचना संभार	पटक	१	११०	१	१	११०	२१५४	१	१००
३३	एलाइजा परिक्षण (खोरेत रोग)	पटक	६	३४३	२	२	११५	२१६५	६	१००

Laboratory Services

The routine laboratory works of RVL, Biratnagar, involve examination of fecal samples, CMT and MWT tests of milk samples. Cultural examination of mastitis positive milk samples are done to isolate and identify the bacteria responsible for this disease. Blood samples are received here, particularly for Hb, PCV, TC, DLC tests, total protein and blood protozoa identification. Serum samples are used to estimate Ca and P level in the blood of animal. Similarly, serological test is done to screen brucella affected animal and salmonella and mycoplasma affected poultry in this region. Hypersensitivity test (tuberculin test) is done to isolate tuberculosis affected animal. Examination of skin scraping and urine samples is frequently done in RVL, Biratnagar.

Parasitological examination

In this examination, both internal and external parasites are identified from the samples. For internal parasites, fecal examination of Goat is done routinely. The fecal samples are received mainly from farmers, DLSOs and also collected from field during investigation programs.



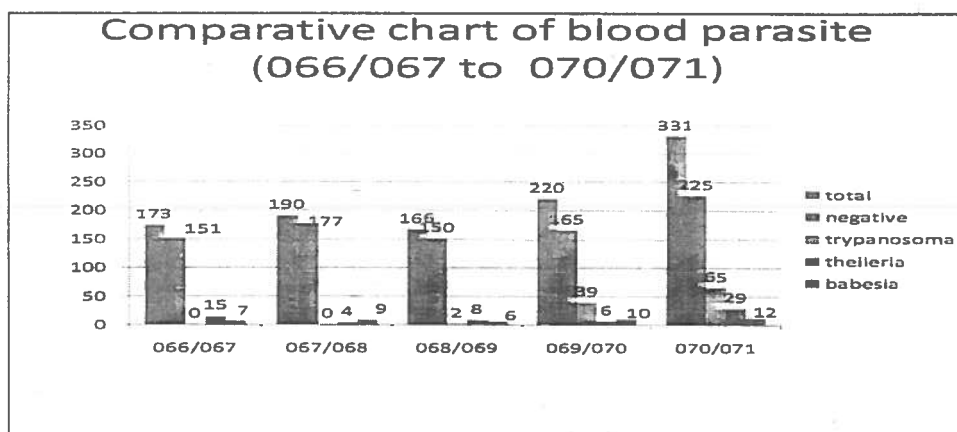
Most frequently the fecal examination is done by sedimentation and floatation techniques to identify the gastro-intestinal parasites. However, in certain cases, Mc Master Technique is followed to quantify the eggs per gram (EPG) in feces.

In the fiscal year 2070/071, altogether 406 fecal samples from different species of animal were received and examined. Among 406 samples, 242 samples (59.60%) were positive. The result of fecal test revealed that Hemoncus (28.32% %) is the most prevalent parasitic infestation followed by Strongylus (9.86 %) and Trichuris (8.13 %) and others (40.39%).

Haematological examination:

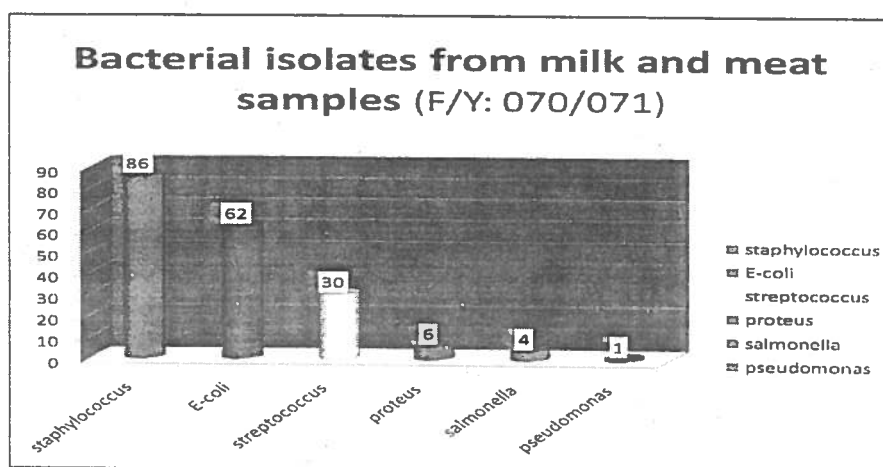
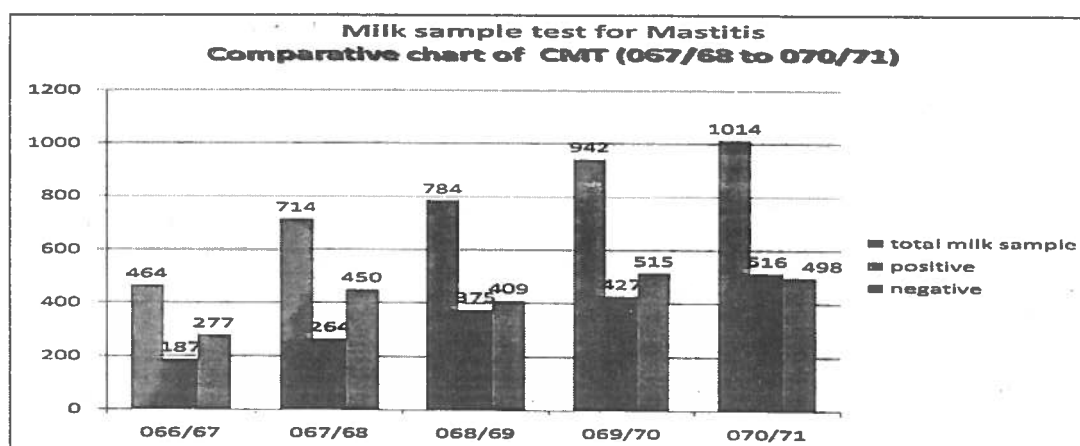
Under haematological examination, TLC, TEC, DLC, PCV and Hb tests are done in this lab. Hb estimation is done by Sahli's haemoglobinometer, PCV by microhaematocrit method, total count of RBC and WBC by haemocytometer. For DLC, blood samples are stained with Gajemsa.

Blood samples received from different districts of eastern region were examined for blood parasites. A total number of 331 samples were examined for blood parasites. Out of them, 106 samples were positive in which Trypanosoma was dominant having 65 positive samples. Rest 29 samples were of theileriosis & 12 positive sample were of babesiosis brought here in this fiscal year.



Microbiological examination

Altogether 1014 milk samples were registered in this laboratory in the fiscal year 2070/071. Out of them 516 samples were positive for CMT and MWT tests. The most prevalent bacteria isolated from these positive milk samples were *E. coli*, *staphylococcus*, *Streptococcus*, *Klebsiella*, *Pseudomonas*, *Enterobacter* etc.



Pathological Examination

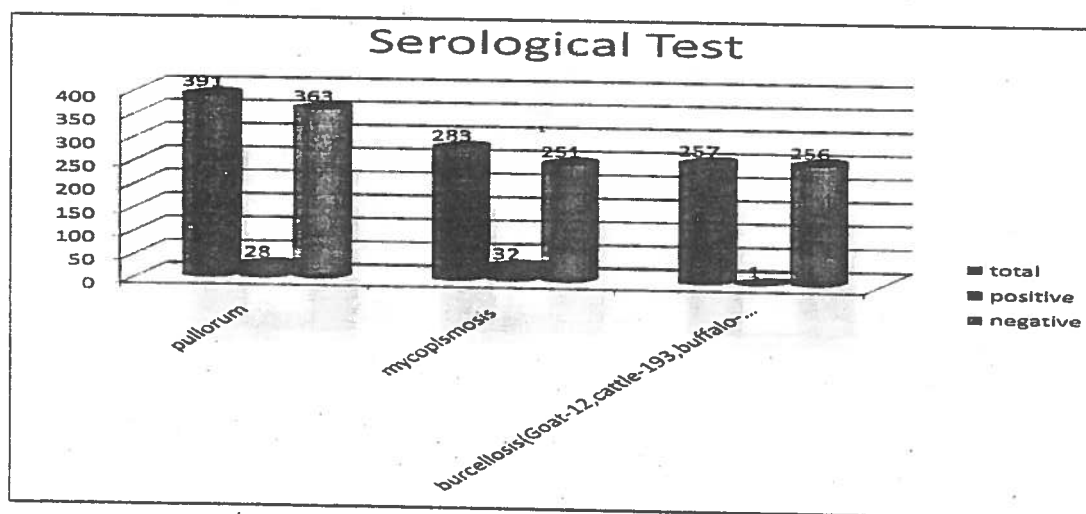
Mostly postmortem examinations of dead birds and occasionally of dead animals are done in the laboratory. During PM examination impression smears, swab, tissues are collected for required tests. Altogether 632 dead birds were received to the lab. On the basis of PM examination and lab tests, diagnosis is done. Out of 632 samples, colibacillosis had higher incidence followed by IBD.

Biochemical examination

Examination of urine and analysis of blood is routinely done to assess the different conditions of urine and blood constituents. Serum samples are collected from farmers, sites of investigation program, etc. Altogether 496 serum samples were collected and analyzed in the fiscal year 070/071 for the estimation of Calcium, Phosphorus, Total protein, glucose, using specific kits. 101 Urine samples were tested by using dipsticks (multisticks) as well as biochemical methods. Examination of urine was done for specific gravity, PH, sugar, albumin, ketone bodies, urobilinogen etc. Mostly Rothera's test and Robert's test were done to detect ketone bodies and protein respectively.

Serological Examination

Serological examination is done mainly for three diseases (salmonellosis, mycoplasmosis and brucellosis) in this laboratory. In fiscal year 2070/071 altogether 391 samples were tested for pullorum disease by Plate agglutination method (PAT). 28 samples were found to be positive. Similarly, Rose Bengal plate test (RBPT) is done for screening the brucella positive animals. Total 257 samples were tested for brucella, out of them 1 sample was found to be positive. Similarly out of 283 sample, 32 were found to be positive for mycoplasma.



Sample sent to CVL, Kathmandu in F/Y 2070/071

As the laboratory is not well equipped with the modern equipments, the samples are sent to CVL. Sometimes, the samples have to be sent to CVL for reconfirmation of the diagnosis.

Zoonosis sample sent to CVL for diagnosis of Brucel, Lepto & Toxo							
	cattle	buffalo	goat	sheep	pig	dog	total
Sunsari	65	15	15				95
Ilam	55						55
Jhapa	60	15	15		64	11	165
Panchthar	20	15	10		5	5	55
Dhankutta	20		10				30
Terrahthum	17	5	20				42
Solu	17	20	10				47
Taplejung	15	4	5	5			29
Khotang			9				9
Udaypur	25	21	15				61
Saptari	81	30	15	10	6	4	146
Siraha	60	20	10	30			120
Sankhuwasabha	15	5	26				46
total	450	150	160	45	75	20	900

Detail of FMD outbreak in EDR						
sn	districts	No. of outbreak	Total affected pop.	No. of infected animals	No. of death	Mortality rate
1	Morang	5	2500	133	8	6.01%
2	Ilam	3	4500	75	5	6.66%
3	Panchthar	2	6600	194	17	8.76%
4	Khotang	1	500	25	0	0
5	Saptari	1	2000	50	0	0
6	Sunsari					
7	Siraha	1	7511	90	6	6.66%
8	Udaypur					
9	Okhaldhunga					
10	Jhapa	16	18000	529	4	0.75%
total						

Confirmation report of PPR from CVL

sn	districts	Total sample	positive	negative	Positive %
1.	saptari	13	4	9	31%
2.	sunsari	5	1	4	20%

REGIONAL VETERINARY LABORATORY, JANAKPUR

Introduction

Regional Veterinary Laboratory of the central development region is situated in Janakpur. It provides diagnostic services to all the 19 districts of central development region. Various diseases diagnosed at the laboratory units i.e. pathology, Parasitology, Microbiology, Haematology & Biochemistry. Serological & Histopathological Laboratory test results are obtained by dispatching the relevant specimens to CVL, as these diagnostic facilities are not available in RVL Janakpur at present.

The working areas of this RVL are all districts of central region but mainly focused on those districts that are Dhanusha, Mohottary, Bara, Parsa, Rautahat and Sinduli. In this Central Region there are 3 zones and 19 districts geographically the region is divided into 3 Eco-zones, Mountain, Hill & Terai.

Mountain

This ecozone lies in the northern part of the region covering Rasua, Sindhupalchok of Bagmati zone. Livestock rearing is the main occupation of the farmers in this region. Yak, Nak, Chauri, sheep & goat are being reared in this region.

Hills

Kathmandu, Bhaktapur, Lalitpur, Dolkha, Makawanpur, Sindhuli are under this region. Farmers follow mixed farming system and agro-based livestock industries are their main occupation. Cattle, Buffalo, Pig, Poultry & Goat are being reared in this region. Poultry farming is also popular among the farmers.

Terai

Dhanusha, Mohottary, Sarlahi, Bara, Parsa, Rautahat & Chitwan are under this Terai region. Through traditional system of livestock farming is followed in this region. Poultry, dairy & piggy are being commercialized especially in east-west highway and its vicinity.

Objectives of RVL Janakpur

- To provide disease diagnostic to the Farmers.
- To investigate & diagnose the epidemics in the region.
- To assist & support DLSOs in disease diagnosis and epidemic control.
- To support animal health and infertility camps in the region.
- To supervise basic laboratories run in DLSOs of the region.

Staff of RVL Janakpur (At the end of F/Y 2069/070)

S.N.	Name of staff	Post	Work of Responsible
1.	Dr. Surya Deo Sah	S.V.O.	Chief
2.	Mr. Ram Ashish Sah	V.O.	Planning
3.	Mr. Anirudra Sah	J.T.	Microbiology, Serology
4.	Mr. Ram Adhar Tiwari	J.T.	Parasitology
5.	Mr. Umeash Sah	J.T.	Biochemistry, Help desk
6.	Mr. Ram Ratan Ray Yadav	J.T.	Sterilization
7.	Mr. Ram Bahadur Chudhari	Na.Su.	Administration, Store
8.	Mr. Hari Narayan Balmpaki	Accountant	Financial
9.	Mr. Kula Nand Jha	Driver	Driving
10.	Mr. Surindra Mishra	Office assistant	Office guard
11.	Mr. Dinesh Datta	Office assistant	Office Attendant

Table: Annual Progress Report (2068/069)

S.N.	Programmers	Unit	Annual Target	Annual Progress	Annual Weightage	Progress %
1.	Laboratory Service					
1.1	Parasitological Examination					
1.1.1	E.P.G. Counts & Larva Culture	Nos.	200	200	2.15	100%
1.1.2	Blood protozoa Identification	Nos.	300	302	2.10	100%
1.2	Pathological Examination					
1.2.1	Clinical hematological Examination	Nos.	200	200	2.35	100%
1.2.2	Calcium, Phosphorus, Protein & Glucose Estimation	Nos.	200	200	2.20	100%
1.2.3	Urine Test	Nos.	60	71	1.25	100%
1.2.4	Postmortem Examination	Nos.	250	250	2.52	100%
1.3	Microbiological Examination					
1.3.1	Bacterial Culture & Identification	Nos.	200	200	3.89	100%
1.3.2	Antibiotic Sensitivity test	Nos.	200	203	2.17	100%
1.3.3	Fungus Culture & Identification	Nos.	200	200	2.92	100%
1.4	Virological Examination					
1.4.1	Virological Examination for Birds (Ranikhet & Gambaro)	Nos.	200	200	3.32	100%
1.4.2	HAHI Examination	Nos.	81	104	2.50	100%
1.4.3	PPR Pen side Examination	Nos.	81	81	1.25	100%
1.5	Serological Examination					
1.5.1	Micoplasma Identification	Nos.	600	600	2.70	100%
1.5.2	Salmonella Identification	Nos.	200	203	2.37	100%
1.5.3	Disease Investigation by Elisa	Nos.	300	640	4.39	100%
1.7.1	Sample Collation & dispatch	Nos.	750	750	3.84	100%
1.8	Disease Investigation & Surveillance Programme					
1.8.1	Clinical Sub Mastitis Test	Times	3	3	1.62	100%
1.8.2	Antibiotic Resistant Bacteria Serve & Scaring	Times	3	3	3.00	100%
1.8.7	Antibiotic Residue Test milk & meat	Times	3	3	2.87	100%
1.8.8	Regional Emergency district investigation team management	Times	3	3	0.75	100%
1.8.9	Investigation of Epidemic Disease	Times	3	3	5.69	100%
1.8.10	Regional Priority for disease Identification, recording &	Times	1	1	2.25	100%

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	Reporting					
1.9	<i>Identification of Zoonosis Disease</i>					
1.9.1	Avian Influenza examination	Nos.	600	600	3.99	100%
1.9.2	Brucella examination	Nos.	400	454	3.79	100%
1.9.2	Rabies examination	Nos.	50	50	2.75	100%
2	<i>Vaccine Bank management</i>					
2.1	Regional PPR Vaccine bank management	Times	3	3	0.75	100%
2.1	Regional Rabies Vaccine bank management	Times	3	3	0.75	100%
3	<i>Seromonitoring</i>					
3.1	Seromonitoring for PPR sample collection & dispatch	Times	1	1	2.25	100%
3.4	Seromonitoring for Ranikhat sample collection & dispatch	Times	1	1	1.45	100%
4	<i>Supervision</i>					
4.1	Follow up District Level Labortory	Times	3	3	2.22	100%
5	<i>Workshop</i>				1.00	100%
5.1	Veterinary disease investigation workshop	Times	1	1	1.25	100%
5.2	District level lab Training 1 day	Times	1	1	1.52	100%
5.1	Planning, Budget & Progress report Workshop	Times	3	3	0.62	100%
7	<i>Publication</i>					
7.1	Publication of Half yearly Epidemiological Bulletin	Times	2	2	0.62	100%
7.2	Publication of Yearly Epidemiological Bulletin	Times	1	1	0.50	100%
7.3	Preparation of Records for yearly publication	Times	1	1	0.75	100%
8	<i>Purchasing of books & Journals</i>	Times	1	1	0.75	100%
9	<i>Lab animal management</i>	Month	12	12	3.02	100%
10	<i>Contract Service</i>					
10.1	Purchasing of machinery equipment	Times	1	1	4.57	100%
10.2	Computer operator, Sweeper & mali	Times	1	1	4.49	100%
11	Office Structure Management	Times	1	1	5.62	100%

Laboratory Services

The routine Laboratory Works of RVL Janakpur , mainly involves examination of faecal Sample CMT test of milk samples and culture examination of mastitis. Positive milk samples are done to isolate and identify the bacteria responsible for this disease. Blood samples brought here through DLSOs particularly for HB, PCV, TC, DLC. Total protein and blood protozoa identification. Examination of skin scraping & Urine test frequently has been done in RVL Janakpur.

Parasitological Examination

Parasitological examination (Internal & external), fecal examination of different animals has been done routinely. The fecal sample are received mainly from farmers referred by DLSOs and also collected from Dhanusha, Mahottary, Sarlahi, Sinnduli, Rauthat and Bara, field area during surveillance and investigation program me. The gastro intestinal parasites. Mc'master technique is followed to quantify the eggs per gram (EPG) in faeces. In the F/Y 2070/71 total 200 faecal sample from different species of animals were received & examined. Among these sample 138 sample (69%) were positive result and 62 sample (31%) showed negative results. The results of faecal test revealed that fasciola (25%), Paramphistomum (48%), Trichuris (11%) Mixed & Other parasites(7%).

Figures of faecal sample examined and types of parasites in this test are folled

Month	No. of Sample	Fasciola	Param Phistomum	Stronggles	Trichuris	Other	-VE
Srawan	20	3				2	14
Bhadra	18		7		1		9
Asoj	19		6				12
Kartik	26	2					24
Mensik	18		5	2	3	1	17
Poush	18	2					17
Magh	20		8				13
Falgun	17	3		2	2	1	11
Chaitra	10	5					9
Baisakh	12		4			1	4
Jestha	10			1	1		8
Asad	12						0
Total	200	15	30	5	7	5	138

Haematological examination :

Haematological examination TLC, TEC, DLC, PCV, & HB test are done in this laboratory. Total count of RBC, WBC by haemocytometer for DLC blood sample are stained with Giemsa blood samples received from different district of central region.

Total 302 Blood sample were examined for different blood parameters as well as for blood parasites. Out of 78 samples 224 samples were found negative for any blood parasites & rest 86 were found positive for different blood parasites.

Details of blood sample examination

Month	No. of Sample	Anaplasma	Babesia	Theileria	Tryps	Other	-VE
Srawan	23	10	3		2	2	6
Bhadra	24	4	8		3	7	2
Asoj	26	5	6	3	3	8	1
Kartik	15	2	3			4	6
Mensik	20	3	2		5	2	8
Poush	26	6	5	4		3	8
Magh	22	2	3			5	12
Falgun	23	8	2		1	6	6
Chaitra	27	10	4	2	2	9	0
Baisakh	20	14	1		1	3	1
Jestha	36	17	2	1	3	4	9
Asad	40	10	1	3		7	19
Total	302	91	40	13	20	60	78

Pathological Examination :

The pathological examination includes mostly post mortem examination of the dead birds received from commercial poultry farms 99% cases bought from janakpur municipality area & periphery the rural area of the Dhanusha district for pathological test, A total 250 cases of post mortem examination were presented during the F/Y 2070/071 all the case received was birds. No cases of large & small ruminants and other species of animals were received. The status of poultry disease in the area is shown below

Trend of disease occurrence in poultry

S.N.	Tentative Diagnosis	Total Cases	
		Number	Percent
1.	Coccidiosis	51	20
2.	IBD	12	4
3.	CRD	24	9
4.	Ranikhet	27	10
5.	Aflatoxin	54	21
6.	Colibacillosis	18	10
7.	Enteritis	6	2
8.	Mixed infection	58	24
	Total	250	100

Microbiological Examination :

Bacterial identification in buffalo milk. Total 200 Buffalo milk sample collected from the following district & Sample were test for the presence of Bacteria in milk. The most prevalent bacterial isolated show staphylococcus, Streptococcus & E. coli etc.

S.N.	District	Tested No. of Sample	Result
1.	Dhanusha	80	staphylococcus, Streptococcus & E. coli
2.	Mohattari	40	staphylococcus, Streptococcus & E. coli
3.	Sarlahi	30	staphylococcus, Streptococcus & E. coli
4.	Rauthat	50	staphylococcus, Streptococcus & E. coli

Antibiotic Sensitivity Test Results :

Antibiotics Used	Percent Efficacy
Enrofloxacin	H. Sensitive
Ciproflaxacin	H. Sensitive
Gentamycin	H. Sensitive
Ampicloxacillin	Sensitive
Teracylin	Low Sensitive

Serological Examination :

All together micoplasma test 600 serum sample tested among them 84 were +ve & rest found negative, for salmonella 203 sample tested 119 sample showed positive & rest showed negative, for Brucella 454 sample tested & rest showed negative.

Biochemical Examination :

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	58	9-12	8-10	4-7	6-8
Buffalo	61	9-12	9-11	4-7	5-8
Goat	49	10-11	11-13	3-11	4-12
Poultry	32	9-12	10-14	4-8	5-7
Total	200				

Sample send to CVL for further Investigation in F/Y 2070/071 :

Regional veterinary laboratory, Janakpur is not well equipped with the modern equipments. The sample are sent to CVL the samples have to be sent to CVL for diagnosis of the disease & Sometimes

reconfirmation of the different types of disease diagnosis . FMD suspected samples sent to FMD laboratory Budhanilkanth, Kathmandu In total 750 Samples dispatched to CVL.

S.N.	Types of Samples	Number
1.	Bird serum	156
2.	Bird swab	185
3.	Serum for PPR	195
4.	FMD	214
	Total	750

National PPR Programme Sero-Surveillance F/Y 2070/071

S.N.	Name of DLSOs	Serum to be collected (No.)
1.	Dhanusha	550
2.	Mahottary	550
3.	Sarlahi	375
4.	Chitwan	550
6.	Sindhuli	200
7.	Makwanpur	400
8.	Routahat	250
9.	Parsha	550
10.	Bara	550
	Total	3975

Bird flu Surveillance in Cental Development Region, RVL Janakpur (F/Y 2070/071)

S.N.	District	Types of birds	Type of farming	Types of sample collected		Total	Test Result
				TS	CS		
1.	Dhanusha	Poultry	CB	20	24	44	-ve
		Duck	CB	18	16	34	-ve
2.	Mahottary	Poultry	CB	28	25	53	-ve
3.	Sarlahi	Poultry	CB	87	92	179	-ve
4.	Sindhuli	Poultry	CB	65	70	135	-ve
5.	Bara	Poultry	CB	26	29	55	-ve
6.	Parsa	Poultry	CB	30	25	55	-ve
7.	Rauthat	Poultry	CB	22	23	45	-ve
	Total			296	304	600	-ve

NATIONAL AVIAN LABORATORY, CHITWAN

Introduction

National Avian Disease Investigation Laboratory (NADIL) was established in 2004 under the Department of Livestock Service. This is the first commodity (Poultry) specific in country's Livestock Department. It works with direct supervision of Central Veterinary Laboratory, Tripureshwor. The Laboratory was proposed during ninth five year plan to address problem related to privately evolving poultry industries in the nation. So that it could diagnosed earlier problems regarding poultry and protect the farmer from the losses. Therefore it's major objective is to provide essential laboratory diagnostic services for the avian diseases so it can facilitate the fast growing commercial poultry industry in the country. More than 60 percent of the total population and the activities concerned with poultry are concentrated in Chitwan. Poultry sector has about 4 percent contribution in National economy through poultry and its product. Currently there are 61 hatcheries' and 42 feed industries supporting commercial broilers and layer farming business in Chitwan and major cities of the country. This laboratory provides diagnostic service through several units, such as post mortem, sample collection, histopathology, parasitology, bacteriology, virology, serology and molecular biology unit.

Objectives

- To diagnosed the major poultry diseases by adopting standard laboratory techniques.
- Isolation, characterization and preservation of different bacteria, virus, fungi, and protozoa causing disease in poultry.
- To develop the laboratory as a centre for the avian disease diagnosis.
- To assist in national poultry disease surveillance and control programs.
- To assess the immunological status of major viral diseases in commercial flock of Nepal through test like ELISA and HA/HI.
- Conduct HPAI laboratory diagnosis using real time PCR technique.

Technical Progress

Different activities of laboratory were performed by various units. The details of the unit wise technical progress are presented in:

Parasitological Unit

Total sample received :-278

Total sample examination :-278

ne	Positive test
	68
	7

All total 278 samples for parasitological study were prepared during F/Y 70/71. Among them 75 sample were positive for endoparasites. Examined sample revealed mostly the Ascariasis followed by intestinal tapeworms; caecal cocci and intestinal cocci were also found during examination.

Bacteriology Unit

Total sample received :- 412

Positive case :- 106

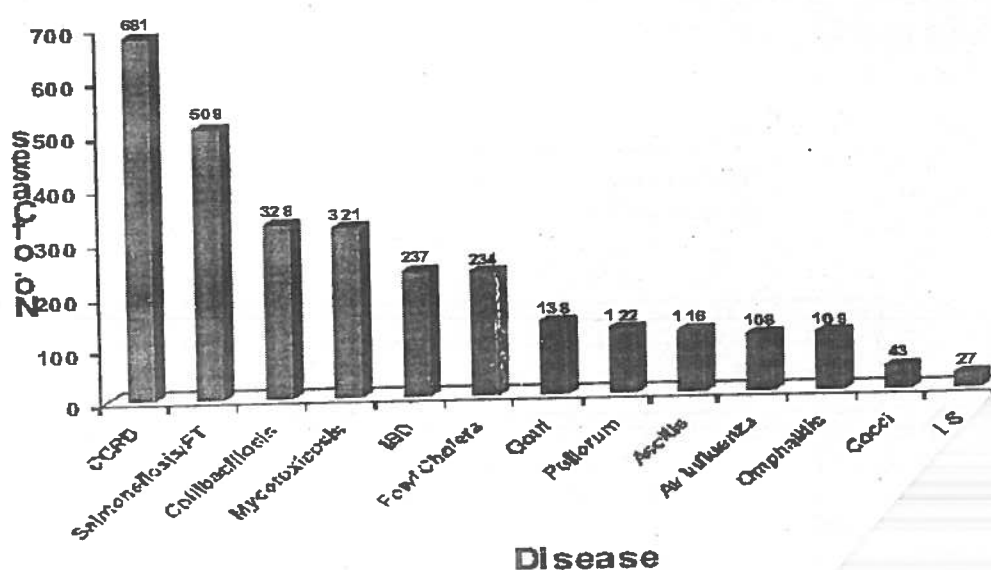
Negative case :- 306

S.N.	Name of micro-organisms	Positive
1.	E. coli	48
2.	Salmonella sps.	37
3.	Enterobacter	21
	Total positive test	106

Suspected sample collected from post mortem examination were brought to bacteriology unit for bacterial culture. Total 412 samples were collected and processed, out of which 106 sample were positive and 306 were negative. Micro-organism were identified on the basis of their colony structure and biochemical characteristics

Post mortem Clinical and post mortem examination of sick and dead birds is the major worked of this unit. From the suspected cases tracheal and cloacae swabs were taken and tested for Avian influenza, Ranikhet, and infectious Bursal Diseases rapid test kits. Blood sample were taken and sent to serology unit for further investigation. Diagnosis was also made by observing pathognomic and other lesion during postmortem.

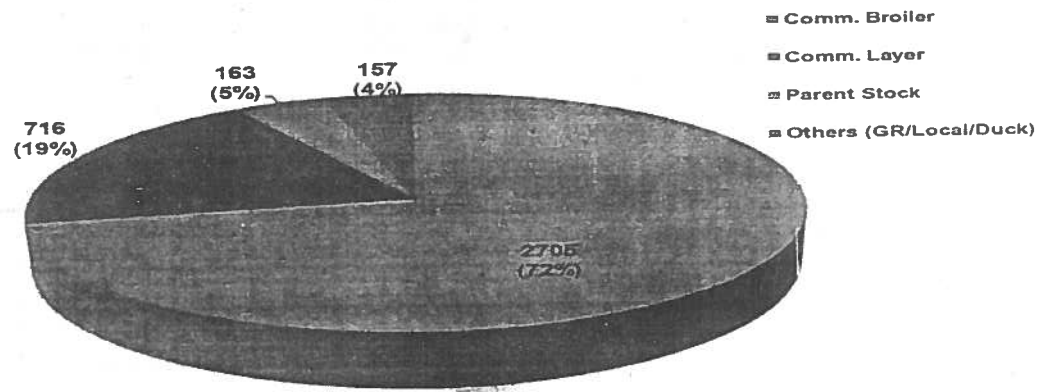
Disease Profile 2070/71



Among the diseases the CCRD dominate of all the diseases during this fiscal year, salmonellosis, colibacillosis, Mycotoxicosis, IBD and FC were other major diseases, beyond that, gout, BWD, ascites, avian influenza omphalitis, cocci, LS and worms were recorded according to their number of case during this year.

Similarly, the total 3740 cases during this year were enrolled, the sample were come from eastern Nepal Panchthar to western Rupendehi. The largest numbers were come from Chitwan (1937), Nawalparasi (976), Makawanpur (576), Tanahun (73), Gorkha (57), Dhading (48), Lamjung (39), Rupendehi (13), Bara (9), Palpa (7), Udayapur (3), Panchthar (2) respectively.

Type of Bird



Districtwise distribution of farmers F/Y 2070/71



Likewise, highest number of birds were enrolled from commercial Broilers (72%) followed by commercial Layers (19%), others from parent stock and 4% from Giriraja, Backyard and Ducks.

Flock category scenario Poultry farms (55%) having less than 1000 birds had brought their sick birds in this lab followed by (42%) less than 3000 birds. Only (3%) had used hospital facilities. This may due to large farmers have their own specialist and a sort of hospital. This

1.3.2	PAT For Salmonella	„	300	80	1.87	310	1.87	
1.4	Molecular Diagnosis							
1.4.1	Flu A PCR	„	50	325	7.61	79	7.61	
1.5	Sample Collection , Examin & Dispatch	„	200	102	2.39	529	2.39	
1.6	Seromonitoring for RD Diagnostic	Time	1	135	3.16	1	3.16	
2	Disease Investigation & Surveillance							
2.1	Migratory birds (Surveill, Monitoring)	„	3	150	3.51	3	3.51	
2.2	Poultry Disease Diagnosis By ELISA	Nos.	180	225	5.27	2090	5.27	
2.3	Immunocomb ELISA(ND,IBD, Mycoplasma)	„	180	210	4.92	193	4.92	
2.4	Bacterial AST Searing & Surveillance	Time	3	195	4.57	3	4.57	
2.5	Epidemic Inves, & Surveil, monitoring	„	3	280	6.56	3	6.56	
3	Epidemiological Reporting							
3.1	Monthly Epidemic Reporting	„	12	15	0.35	12	0.35	
3.2	Computerized data base preparation	„	3	10	0.23	3	0.23	
4	Monitoring & Supervision	„						
4.1	District Lab. Monitoring	„	3	110	2.58	3	2.58	
5	Training & Workshops							
5.1	Participation in Central & Other Workshops	„	6	148	3.47	6	3.47	
5.2	Workshops In Poultry Entrepreneurships	„	1	45	1.05	1	1.05	
5.3	Assistant Level (3 days Sample	„	3	135	3.16	3	3.16	

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	Col.) Training							
6	Publication							
6.1	Preparation Annual Technical Paper	„	1	10	0.23	1	0.23	
6.2	Brochures Publication	„	1	50	1.17	1	1.17	
7	Lab Animal Management	„	3	63	1.48	3	1.48	
8	Contract Services							
8.1	Driver/ Sweeper/ Maali	„	1	150	3.51	1	3.51	
8.2	Lab Equipment & Computer	„	1	75	1.76	1	1.76	
8.3	Others Contracts	„	1	75	1.76	1	1.76	
9	Book & Journal Purchase	„	1	50	1.17	1	1.17	

5. Man power Situation during F/Y 2070/071

S. N.	Type of the post	Class	Approved post	Fulfilled	Vacant	Remarks
A	Technical (Officer)					
1	Senior Veterinary Officer	Gaz.II	1	1	0	
2	Veterinary Officer	Gaz.III	3	3	0	
B	Technical (Non-Officer)					
1	Veterinary Technician	Non.Gaz I	2	2	0	
2	Junior Vet. Technician	Non.Gaz.II	1	1	0	
Total Tecnical Staffs			7	7	0	
C	Non-Technical (Non-Officers)					
1	Account	Non.Gaz.I	1	1	0	
2	Kharidar	Non.Gaz. II	1	1	0	
3	Light Vehicle Driver	No Class	1	1	0	
4	Office Helper	No Class	2	2	0	
Total Administrative staffs			5	5	0	
Grand Total			12	12	0	

REGIONAL VETERINARY LABORATORY, POKHARA

Introduction

Western Development Region (WDR) is geographically divided into Himalayan, Hilly and Terai main domains: Himalayan region is located in the northern part of the WDR, covering Mustang, Manang and upper

belt of Gorkha districts. Yak/Nak, sheep, alpine goats (Chyangra) and mule rearing formed the way of life of

the people in this region. Hilly region lies in between the Himalayan and Terai regions. This region is comprised of Arghakhanchi, Gulmi, Palpa, Shyanga, Kaski, Tanahu, Lamjung, and lower belt of Gorkha, Parwat, Baglung and Myagdi districts. People of divergent ethnic groups, casts and cultures share their common way of living. Agro-based livestock industry in this region is the main source of income of the people. Poultry farming, goat rearing and dairy industries are becoming familiar near the cities/towns and in the areas where market is accessible. Terai region covers Nawalparasi, Rupandehi and Kapilbastu districts.

The mission of the Regional 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 District Livestock Development Offices, 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.

Objectives

- To provide accessible, timely and accurate diagnostic services to the livestock and poultry farmers and to
- veterinarians, veterinary technicians and their owners in the region.
- To conduct diagnostic examinations, record results, report information, and assist in the interpretation of
- results to submitting DLSOs, Veterinarians, and veterinary technicians.
- To investigate the animal disease epidemic in the region and assist, advice and support DLSOs to control them.
- To 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.
- To investigate relatively important livestock diseases in the region and assist in the formulation of disease
- control measures
- To supervise and assist in diagnostic services to basic and primary laboratories based at DLSO's of the region.
- To conduct and support the laboratory and animal health related training programs for the veterinary paraprofessionals in the region.
- To assist the national disease control and eradication programs in the region.

Table : Annual work Program and summary of achievements of Regional Veterinary Laboratory, Pokhara (for fiscal year 2070/71 & Budget No:312125-3/4)

S.N.	Programmes and Activities	Annual Targets			Annual Progress	Weightage (%)	Remark
		Unit	Targets	Weightage (%)			
sA	Capital Expenditure						
1	Construction meeting hall	Nos	1	9.63	1	9.63	
2	Purchase of microcentrifuge machine	Times	1	1.35	1	1.35	
3	Purchase of furniture	Times	1	3.85	1	3.85	
B	Running Expenditure						
1	Laboratory Services						
1.1	Parasitological Examinations						
1.1.1	EPG Count and Larva Culture	Nos	300	2.45	305	2.45	
1.1.2	Examination of Blood Parasites	Nos	60	1.23	100	1.23	
1.2	Pathological Examinations						
1.2.1	Clinical Hematological Tests	Nos.	60	0.96	100	0.96	
1.2.2	Calcium, Phosphorus, T. Protein, Glucose, Urine Test	Nos	180	1.12	188	1.12	
1.2.3	Urine Analysis	Nos	50	0.48	67	0.48	
1.2.4	Necropsy Examination and Recording	Nos.	1800	4.21	2507	4.21	
1.3	Microbiological Examinations						
1.3.1	Isolation and Identification of Bacteria	Nos	200	5.83	480	5.83	
1.3.2	Antibiotic Sensitivity Test	Nos	180	2.06	183	2.06	
1.3.3	Isolation and Identification of Fungus	Nos	150	1.21	157	1.21	
1.4	Virological Tests						
1.4.1	Virological test in birds	Nos	45	1.48	200	1.48	
1.4.2	ND diagnosis using HA HI	Nos	50	1.93	79	1.93	
1.4.3	PPR testing using penside test	Nos.	200	2.04	270	2.04	
1.5	Serological Tests						
1.5.1	Mycoplasma tests in Poultry using PAT	Nos	45	0.67	255	0.67	
1.5.2	Salmonella tests in Poultry using PAT	Nos	120	1.69	283	1.69	
1.5.3	Disease Diagnosis using	Nos.	300	2.64	300	2.64	

	ELISA Tests(swine fever)						
1.6	Molecular diagnostic test	Times	1	0.96	1	0.96	
1.7	Sample collection and dispatch	Nos	300	1.64	442	1.64	
1.8	Disease Investigation and Surveillance Program						
1.8.1	Investigation of subclinical mastitis in cattle	Nos	300	2.79	306	2.79	
1.8.2	Finding out the Basic production and Fertility Profile in Cattle	Times	3	0.96	3	0.96	
1.8.3	Investigation of Parasitic Diseases in Migratory Sheep and Goats	Times	3	4.91	3	4.91	
1.8.4	Antibiotic resistant bacteria surveillance and sharing	Nos	120	3.02	120	3.02	
1.8.5	Test of antibiotic residue in meat and milk	Times	3	0.96	3	0.96	
1.8.6	Management of Regional EDIT	Times	3	2.54	3	2.54	
1.8.7	Investigation of Epidemic	Times	3	3.95	3	3.95	
1.8.8	Reporting of priority disease in western region	Timres	1	0.48	1	0.48	
1.9	Laboratory Diagnosis of Zoonotic Diseases						
1.9.1	Avian Influenza Diagnosis	Nos	450	4.12	1101	4.12	
1.9.2	Brucella Disease Diagnosis using RBPT	Nos	100	2.04	665	2.04	
1.9.3	Rabies Diagnosis	Nos	100	3.87	113	3.87	
1.9.4	Tuberculosis Testing using Tuberculin	Nos	100	2.52	104	2.52	
1.9.5	Campylobacter and Salmonella isolation in chicken	Nos	150	4.14	150	4.14	
2	Management of vaccine bank						
2.1	Management of PPR vaccine	Nos	3	0.48	3	0.48	
2.2	Management of Rabies vaccine	Nos	3	0.48	3	0.48	
3	Seromonitoring						
3.1	PPR	Times	1	1.73	1	1.73	
3.2	FMD	Times	-	-	-	-	
3.3	Swine Fever	Times.	1	0.87	1	0.87	
3.4	Ranikhet	Times	1	0.87	1	0.87	

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4	Monitoring and Supervision					
4.1	Follow up tour of district Laboratories management	Times	6	0.87	6	0.87
5	Workshops and seminars					
5.1	Animal Disease Investigation Interaction Workshop	Times	1	0.96	1	0.96
5.2	District level laboratory training	Times	1	0.96	1	0.96
5.3	Participation in Budget and Programme formulation workshops at Central Level	Times	3	0.62	3	0.62
7	Publication Program					
7.1	Half Yearly Epidemiologic Bulletin publication	Times	2	0.77	2	0.77
7.2	Annual Epidemiological Bulletin Publication	Times	1	0.39	1	0.39
8	Material Preparation for Annual Technical Report	Times	1	0.77	1	0.77
9	Lab Animal Management	Month	12	1.56	12	1.56
10	Contract Service					
10.1	Laboratory Equipment and Machineries Security Service	Times	1	0.96	1	0.96
10.2	Gardener/Office Assistant/Sweeper/Driver	Times	3	3.95	3	3.95
11	Security of Office buildings					

REGIONAL VETERINARY LABORATORY, SURKHET

1. Introduction

Regional Veterinary Laboratory (RVL) of the Mid-western Development Region (MDR) is situated in Birendranagar Municipality-7, Surkhet and was established in FY 1988/1989 AD. It provides laboratory diagnostic services to all the 15 districts of MDR. In this region, there are 3 zones (Rapti, Bhari and Karnali) and 15 districts. Geographically, the region is divided into 3 eco-zones (high hills, mid hills, and terai). To provide the diagnostic livestock services smoothly, three districts (Banke, Dang and Jumla) have established basic laboratories. They perform parasitological examinations, microbiological culture and antibiotic sensitivity test.

The mission of the RVL, Surkhet is to promote the health of livestock, poultry and ensure safe animal products for consumer by assisting DLSO's offices, veterinarians, clients, and others who are responsible for animal health in detection and prevention of animal diseases.

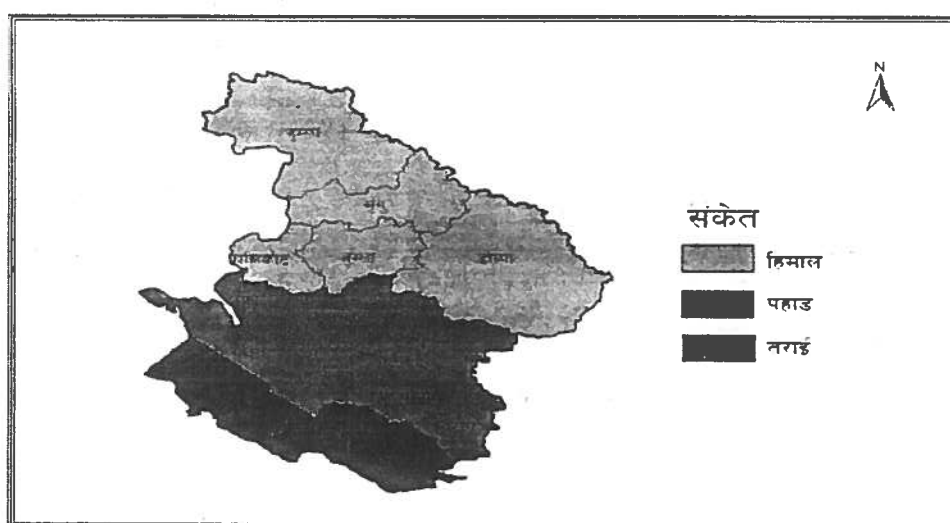


Figure. 1 Map of Mid-western region showing the three eco zones.

2. Objectives

- To provide animal disease laboratory diagnostic services to the farmers of the region.
- To investigate the animal disease epidemics in the region.
- To support DLSOs in disease diagnosis and epidemic control.
- To investigate important livestock diseases in the region & assist to formulate disease control measures.
- To monitor and report the incidence and threat of animal disease as well as zoonotic disease
- To supervise diagnostic service at DLSO's of the region and onsite coaching.
- To conduct and support the laboratory and animal health related training activities/programs for the veterinary paraprofessionals in the region.
- To assist in the national disease control and eradication programs in the region.

Progress Report of FY 070/71

The annual program and the summary of the progress report is presented in the table below.

Table : Annual work program and summary of achievements (FY 2070/071)

S.N.	Activities	Unit	Annual		Progress %
			Target	Progress	
1.	Furniture & Fixtures				
1.1	Sofa set	Set	1	1	100
1.2	Steel dares	„	1	1	100
2.	Purchase of equipment				
2.1	Laptop	No.	1	1	100
1	Laboratory services				
1.1	Parasitological Examination, identification and recording	No.			
1.1.1	EPG Count, Larvae Culture Identification and Treatment	No.	450	485	100
1.1.2	Blood Parasite Examination	No.	150	187	100
1.2	Pathological Examination				
1.2.1	Clinical Hematological Examination	No.	150	192	100
1.2.2	Biochemical Examination	No.	90	97	100
1.2.3	Urine Examination	No.	50	82	100
1.2.4	Post Mortem	No.	450	494	100
1.3	Microbiological Examination				
1.3.1	Isolation, Identification and Recording of Bacteria	No.	150	267	100
1.3.2	Antibiotic Sensitivity Examination	No.	150	235	100
1.3.3	Culture and Identification of Fungus	No.	60	102	100
1.4	Virological Tests				
1.4.1	Viral disease Examination in Poultry (IBD, ND)	No.	50	192	100
1.4.2	HA, HI Examination	No.	50	112	100
1.4.3	PPR testing using Pen site Test	No.	50	54	100
1.5	Serological Examination				
1.5.1	Mycoplasma Examination in Poultry	No.	50	101	100
1.5.2	Salmonella Examination in poultry	No.	300	484	100
1.5.3	Eliza Examination (FMD)	No.	100	120	100
1.7	Sample collection and dispatch				
1.7.1	Sample collection and dispatch for further examination in other Laboratories	Times	3	3	100
1.8	Epidemic Investigation	Times	3	3	100
1.8.1	Subclinical mastitis investigation and reporting in Cattle and Buffalo	Times	3	3	100
1.8.6	Antibiotic Resistance Bacteria Surveillance and shearing	Times	3	3	100

1.8.7	Antibiotic Residue examination (Milk, Meat)	Times	3	3	100
1.8.8	Regional label emergency Disease investigation team management	Times	3	3	
1.8.9	Epidemic investigation	Times	3	3	100
1.8.10	Disease investigation, filing, reporting in regional label	Times	1	0	
1.9	Zoonotic Disease investigation laboratory Program				
1.9.1	Avian influenza examination	No.	300	477	100
1.9.2	Brucella examination	No.	100	165	100
1.9.3	Rabies examination	No.	75	64	100
1.9.4	Tuberculosis examination	No.	50	55	100
2	Vaccine Bank Management				
2.1	Vaccine Bank Management in Regional label for PPR	Times	3	3	100
2.2	Rabies Vaccine Bank Management	Times	3	3	100
3	Sero monitoring				
3.1	Sample collection and dispatch of PPR for sero monitoring	Times	1	1	100
3.2	Sample collection and dispatch of FMD for sero monitoring	Times	0	0	100
3.3	Sample collection and dispatch of Swine fever for sero monitoring	Times	1	1	100
3.4	Sample collection and dispatch of ND for sero monitoring	Times	0	0	100
4	Monitoring and supervision				
4.1	Supervision and Monitoring of District Laboratories	Times	3	3	100
5	Work Shop program				
5.1	Animal Disease Investigation interaction workshop	Times	1	1	100
5.2	District label Laboratories training program (1 Days)	Times	1	1	100
5.3	Budget program ..	Times	3	3	100
7	Publication program				100
7.1	Half yearly epidemiological bulletin publication (preparation).	Times	2	2	100
7.2	yearly epidemiological bulletin publication (preparation).	Times	1	1	100
7.3	Annual Technical Bulletin Publication	Times	1	1	100
8	Purchase of Books and Journal	Times	1	1	100
9	Management of Lab Animal	Month	12	12	100
10	Contract Service				
10.1	Laboratory Equipment Preservation Services	Times	3	3	100
10.2	Computer Operator, Driver, Others	Times	3	3	100

4. Laboratory Services

4.1 Parasitological examination

Parasitological unit examines fecal samples of various species using different methods as direct smear, sedimentation, and floatation methods. Parasitological unit not only identify the parasites but also quantify the parasitic burden of the nematode, trematode and cestodes by McMaster method. In this fiscal year 2070/071, total 964 faecal samples were examined and 880 were found positive for various internal parasites. Among them *Fasciola*, *Coccidia*, *Haemonchus*, *Strongylus*, *Paramphistomum* were found major internal parasites identified. Result of faecal examination is presented in table and figure below.

Table : Fecal examination result different animal conducted at RVL, Surkhet (Y70/071)

Parasites	2070/071	Percentage
<i>Fasciola</i>	299	31
<i>Paramphistomum</i>	89	9
<i>Strongylus</i>	78	8
<i>Trichuris</i>	54	6
<i>Haemonchus</i>	128	13
<i>Coccidia</i>	232	24
Nad	84	9
Total	964	100

4.2 Microbiological examination

Microbiological examinations include the isolation and identification of organisms from the pathological samples. Bacteriological culture and antibiotic sensitivity test is conducted for microbiological investigation. The samples were milk, urine, tissue, skin scrapings etc. The organisms shown in the culture were subjected to sensitivity test. Antibiotic sensitivity test conducted at RVL Surkhet, during 2070/071 the test result is given below.

Table : Antibiotic sensitivity test result of RVL, Surkhet (FY 2070/071)

Antibiotic Disc	Sensitivity	Intermediate	Resistance
Gentamicin	11	61	31
Ciprofloxacin	35	48	20
Cefalexin	2	52	49
Choramphenicol	-	75	28
Amoxycilline	4	65	3
Tetracycline	9	71	23
Cloxacilline	8	67	18

4.3 Pathological 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 VDCs; Utterganga, Latikoili, Chhinchu, Jurbutta and some VDCs of Bardiya and Dang District for pathological examination. The status of poultry diseases according to post-mortem examination is shown in table and figure below.

Table: Postmortem result of poultry conducted at RVL, Surkhet (FY 2070/071)

Parasites	2070/071	percentage
Colibacillosis	149	20
IBD	141	19
Coccidiosis	61	8
Ascities	65	9
Mycotoxycosis	51	7
Salmonellosis	80	11
Omphalitis	66	9
Others	129	17
Total	742	100

1.4 Serological examination

A total of 720 serum samples of different domestic animals were sent to Central Veterinary Laboratory for serological analysis of zoonotic disease. Among them, 496 samples were tested. The results of the test are presented in the table below.

Table : Result of Zoonotic Diseases (FY 2070/71, Ref no. 29, Serology section CVL)

S. N.	District	Leptospirosis		Toxoplasmosis		Brucellosis	
		Total sample tested	Positive	Total sample tested	Positive	Total sample tested	Positive
1	Banke	102	11	92	11	92	2
2	Bardiya	74	10	73	7	63	1
3	Surkhet	0	0	71	26	121	0
	Total	176	21	236	44	276	2

2. National PPR Program

Under National PPR Control program, Directorate of Animal Health had provided 6,15,000 doses of PPR vaccine for 11 districts of this mid western region. RVL Surkhet supported the program by sero-monitoring. District-wise collection of serum sample is presented in the table below.

Table: District wise collection of serum sample.

S. N.	District	Vaccination	Serum collection	
			Target	Progress
1	Jumla	15000	75	75
2	Jajarkot	20000	100	0
3	Pyuthan	40000	200	200
4	Rolpa	40000	200	0
5	Rukum	40000	200	200
6	Dailakh	40000	200	200
7	Salyan	40000	200	200
8	Bardiya	70000	350	350
9	Banke	100000	500	500
10	Surkhet	100000	500	600
11	Dang	110000	500	500
	total	615000	3025	2825

3. Swine Fever Sero-monitoring Program

The total number of swine fever Vaccines distributed from Directorate of Animal Health was 40000 doses under national swine fever vaccine program for 2 districts for this region. RVL, Surkhet supported the program by sero-monitoring. The table below shows district -wise collection of serum sample.

Table : District wise collection of serum sample.

S. N.	District	Vaccination	Serum collection	
			Target	Progress
1	Banke	20000	200	90
2	Bardiya	20000	200	40
3	Surkhet	-	-	100
4	Dang	-	-	40
	Total	-	400	270

4. List of staff of RVL Surkhet

Table: List of staff of Regional Veterinary Laboratory, Surkhet (FY 2070/071).

S.N.	Designation	Class	Number of Post	Fulfilled	Vacant
1	Senior Veterinary Officer	Gaz.2	1	-	1
2	Veterinary Officer	Gaz.3	2	1	1
3	Animal Health Technician	Non.Gaz.1	2	2	1(Study leave)
4	Assistant Animal Health Technician	Non.Gaz.2	2	2	
6	Typist	Non.Gaz.1	1	1	
7	Accountant	Non.Gaz.1	1	1	
8	khaddar	Non.Gaz.2	1	1	
9	Driver	Class less	1	0	1 (1 contact)
10	Office Helper	Class less	2	0	2 (1contact)
	Total		13	8	5

5. Problems of RVL

- Limited budgets on logistics and fuel.
- Lack of staff (especially technical staffs) and laboratory equipments to provide the emergency services.
- Inadequate rooms to perform post-mortem of small and large animals.
- Lack of disposal pit to dispose biological and other wastes.
- Not fulfillment of vacant posts.
- Lack of refreshment training.

REGIONAL VETERINARY LABORATORY, DHANGADHI,

Introduction:

Regional Veterinary Laboratory, is situated in Dhangadhi ,municipality of far western Nepal. This laboratory is established as the reference laboratory of the region with its service area covering the nine district & two zones. It is the smallest region among the five development regions. The region shares borders with the Tibetan China to the north and with the Indian states of Uttranchal and Uttar Pradesh to the south west.

Geographically the region is divided into three parts namely, Mountains, Hills and Terai. The mountainous districts of the region comprises of Bajura, Bhajhang, and Darchula, Like wise Hilly districts of the region are Baitadi, Dadeldhura, Acchamm, & Doti. Kailali and Kanchanpur are the Terai districts of the Far western region. All districts have a high potentiality for the development of livestock industry. Almost all kinds of livestock viz cattle, buffalo, sheep, goat, pig, poultry, duck, etc..

On the other Goat and poultry farming is inclined towards commercialization, but particularly poultry farming is rapidly gaining popularity among the farming communities.

There are a number of infectious diseases which are constrain to the development of the livestock sector of the region, apart from managemental, nutritional and other factors. The major being the FMD, PPR, Swine Fever, HS,poultry diseases and parasitic diseases.

Now Layer and broiler are reared in commercial scale in Kailali and Kanchanpur districts. Recently some farmers of Dadeldhura, Baitadi and Doti are showing interest in poultry farming, and they have started to keep poultry in number of few hundreds. A few economically important diseases of poultry in this region are New Castle, Infectious Bursal Disease, Coccidiosis, Chronic Respiratory Disease (CRD), Inclusion Body Hepatitis (IBH) ,Leechi Heart Disease, Collibacillosis,Ecoli etc. Regional Veterinary Laboratory, Dhangadhi was established in 2049/050,

Objective:

The main objective of RVL Dhangadi, are as follows

- To conduct the Livestock and Poultry disease investigation work.
- To investigate animal disease epidemics in the region and assist DLSO's to control animal and poultry diseases.
- Prepare Epidemiological profile of livestock and poultry diseases.
- Supervise & assist in diagnosis to basic and primary laboratories of DLSO's.
- Support to conduct the Laboratory trainings for vet .Technicians.
- Coordinate and assist in national disease control and eradication programs.
- Monitor and report of public health importance diseases.

Staffing of RVL Dhangadi

S.N.	Type of post	Class/post	number	fulfilled	vacant	Remark
1	Senior vet officer	G.II,SVO	1	0	1	
2	Vet officer	G.III,VO	2	2	0	
3	AH Technician	NG.I	2	2	0	
4	AAH Tech	NGII	2	1	1	
5	Accountant	NGI	1	1	0	
6	Nayab Subba	NGI	1	1	0	
7	Driver		1	1	0	contract
8	Office helper	No class	2	2	0	

Summary of Achievement in f/y 070/071 is presented in Table below

S.N.	Programs and Activities	Unit	Annual Progress		Percentage
			Target	Progress	
A1	Constraction of upper floor in Duplex Quater	Time	100%	100%	100%
A2	Garez Constraction	Nos	1	1	100%
B1	Laboratory Service programme-				
1.	Parasitological Examination	Nos			
2.	Identification of parasites and EPG Count	Nos	300	324	100%
3.	Identification of blood protozoa	Nos	150	208	100%
4.	Clinical hematological examination	Nos	300	360	100%
5	Biochemici examination	Nos	300	328	100%
B2	Pathological examination				
1	Postmortem examination	Nos	500	548	100%
2	Microbiological culture & Identification	Nos	250	260	100%
3	Antibiotic sensitivity test	Nos	200	210	100%
4.	fungus culture& Identification	Nos	150	151	100%
5	Viral diseases examination of Birds(ND/IBD) examination	Nos	150	157	100%
6.	HAHI(ND/IBD)	Nos	50	68	100%
7	Salmonellosis examination (PAT)	Nos	500	554	100%
8	ELISA(FMD)	Nos	3	3	100%
9	Sample collectin test & Dispatch	Time	3	5	100%
10	Sub clinical mastitis investigation in cow	Nos	500	591	100%
11	Investigation of Kumri in goat	Time	3	3	100%
12	Khari disease preventatio & control.	Time	3	3	100%

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13	Antibiotic Resistant surveillance & Sharing	Time	3	3	100%
14	Antibiotic Residue test in milk & meat	Time	3	3	100%
15	Regional level Investigation of Emergency Diseases	Time	3	3	100%
16	Epidemic investigation	Time	3	3	100%
B3	Zoonotic disease investigation programmes				
1	Avian influenza test	Nos	200	645	100%
2	Brucella test	Nos	100	359	100%
3	Rabies test	Nos	50	50	100%
B4/1	Vaccine Bank Management PPR	Nos	3	3	100%
2	Vaccine Bank Management Rabies	Nos	3	3	100%
3	PPR Seromonitoring & Sample Dispatch	Nos	1	1	100%
4	FMD Seromonitoring & Sample Dispatch	Nos	2	2	100%
5	Swine fever Seromonitoring & Sample Dispatch	Nos	1	1	100%
6	Inspection and Supervision of District Labs.	Time	3	3	100%
7	Technical Interaction of animal disease	Time	1	1	100%
8	District level lab Technique training 1 day	Time	1	1	100%
9	Participation in budget, program, & workshop	Time	3	3	100%
B5	Publication Programme:				
1	Half Yearly Epidemiological bulletin publication	Time	2	2	100%
2	Yearly Epidemiological Bulletin publication	Time	1	1	100%
3	Annual Technical Book material Preparation	Time	1	1	100%
4	Purchase of scientific Books and Journals	Time	1	1	100%
B6	Management of Lab. animal	Time	12	12	100%
7	Protection of Lab. equipments & Machineries	Time	3	3	100%
8	Office assistant, computer operator & Driver for contact	Time	3	3	100%
	Total progress				100%

Zoonosis Disease Control Program Sample tested by RVL Dhangadhi 070/071

S.N.	Programs and Activities	Unit	Annual Progress		Percentage	Remarks
			Target	Progress		
1.	Nos	Nos	200	645	100%	7 Sample H9+ve according to cvl
2	Brucella	Nos	100	359	100%	28+ve suspected
3	Rabies	Nos	50	50	100%	All-ve

Zoonosis Disease Control Program Sample from RVL Dhangadhi tested by CVL Tripureshwar, 070/071

S.N.	District	Leptospirosis		Toxoplasmosis		Brucellosis	
		Total Sample tested	Positive	Total Sample tested	Positive	Total Sample tested	Positive
1.	Kanchanpur	154	23	185	48	251	0
2.	Kailali	14	5	63	8	92	0
3	Doti	8	0	28	1	25	0
	Total	176	28	276	57	368	0

Above samples were collected from Kailali, Kanchanpur, Doti & Bhanjhang districts. Most of the samples were negative in rapid test flu A type. But 7 samples from, Kailali Kanchanpur & Baitadi districts were found positive for H9 strain according to CVL. Total 359 sample of cattle, buffalo RBPT for Brucella, 28 samples were positive (suspected) & 820 sample test for Leptospirosis, Toxoplasmosis & Brucellosis by CVL.

1. Parasitological Examination :

Altogether 324 samples were tested for different parasitic conditions of livestock populations. Samples for examination mainly consisted of the regular fecal samples submitted to the district livestock service office, Kailali. Apart from this samples were also collected from field during epidemic disease investigation and the investigation programme in our set annual programme. The most common helminthic parasite identified during faecal examination was Fasciola followed by other internal parasites of nematode group, viz. Strongylus, Trichuris, Coccidiosis, etc. it is found that most of the fecal samples examined at the RVL are positive for one or the other internal parasites. Out of 324 samples tested 172 (53%) were positive and A total of 324 samples were tested for EPG count to see the prevalence of internal parasite in Goat.

In EPG counting the prevalence rate was 53% in goats. Strongyles, strongyloids, trichuris and monezia were the most parasite found in samples.

2. Serology:

A total of 646 serological tests of different types were performed against the target during the fiscal year. Most of the serum samples collected were from goats, cattle, buffalo for various diagnostic tests Brucellosis, Cattle, Buffalo serum for ELISA test and salmonella for poultry disease. 810 serum samples were forwarded to CVL..

Serum samples collected from bovine, caprine & poultry population were mainly obtained from Baitadi, Kailali, Kanchanpur, Doti, Dadeldhura Bajhang and Darchula. Performing the

regular screening test of Brucellosis using Rose Bengal Plate Agglutination Test & salmonella PAT. Test detail is given below Result of Serological examination performed at the RVL, Dhangadhi

S.N.	Disease Name	Target	Sample Tested	Result	Remarks
1.	Salmonella	500	554	83Pos. 471Neg	s.pullorum
2.	ELISA(FMD)	3 Time	92	6Pos. 86Neg	

3. Haematology:

A total of 360 samples were examined for Haematology at the RVL, Dhangadhi included TLC, DLC, PCV, Hb examination for presence of blood parasites. Blood samples were mainly collected from live stocks during outbreak of epidemic, cases referred by the DLSO's

On our examination of hematological parameters of parasitic infestation animals they invariably showed low Hb. concentration. Hb. value in these animals ranged from low of 7 to a high of 16g./dl. & PCV ranges from 11-38dl of blood, 208 blood samples were examined for blood protozoan were negative for babesia.

4. Pathology:

This unit of the laboratory mostly receives poultry & pig carcass for necropsy study and disease diagnosis. However, dead bodies of other animal species is also received occasionally, especially during disease outbreak and in cases of veterolegal importance. Out of the total of 487 postmortems performed during the fiscal year 070/071 only 3 cases of pigs from Kailali, Dadeldhura, Kanchanpur and the rest were of poultry species. Since most of the pathological samples comprised of poultry, it is important to present the major diseases diagnosed, based on findings of postmortem lesions. The common diseases that were diagnosed through postmortem was coccidia, IBH, IBH, Ecoli, CRD, Mycotoxin, asitis, gout & Fatty kidney syndrome etc.

Post mortem report (poultry)

S.N.	Disease/ Conditions	numbers	percentage
1	IBD	206	38%
2	Gout	92	17%
3	IBH/Lechy	125	23%
4	Asitis	22	4%
5	CRD	43	8%
6	ND, other	16	2.9%
7	Coccidiosis	16	3%
8	E-coli	22	4%

Post mortem report (Pig)

S.N.	Disease	Numbers of pig	Remarks
1	Classical swine fever	2	Tikapur(Kumar Sings's) according to FMD Lab
2	Pneumonia	2	RVL Dhangadhi
3	Internal Parasite(Round Worms)	2	RVL Dhangadhi

5. Microbiology:

The samples received to microbiological examination at the Regional Veterinary Laboratory, Dhangadhi constitutes of milk, nasal swab, vaginal swab, and swab from visceral organs like liver, lungs, intestine etc. of various animal species. The media used for microbiological culture were Nutrient agar, Mc conkey agar, Blood agar, and Saboroud Dextrose agar. Bacteria and fungi were identified on the basis of colony characteristics, Gram's staining property and the structure of the organism as seen under the microscope. Due to limitation of the facility. The Result of microbiological test is presented in the table as below:

Animal	Type of sample	Number	Major bacteria identified	Remarks
Cow	Milk	98	Strep., Staph., Cory.	
Buffalo	Milk	78	Strep., Staph., Cory., Pseudomonas	
Goat	Nasal, vaginal swab	24	Strep., Staph., Bacillus	
Poultry	Liver	42	E. coli, Strep., Staph., Aspergillus, Penicillium, Candida	
	Lungs	14	E. coli, Strep., Staph., Coryne	
	Intestinal swab	6	E. coli, Strep., Staph.,	

The milk samples positive for SLST were tested for antibiotic sensitivity test following preliminary culture in order to choose right antibiotic for the treatment of mastitis. The result of antibiotic showed that Enrofloxacin was more effective.

6. Biochemical:

Altogether 328 samples were collected & analyzed in this fiscal year for estimation calcium, phosphorus, Total protein, & glucose.

7. Investigation program Setaria spp (Kumri) in Goats

A part from above activities this laboratory has performed Kumri in Goat investigation program in Kailali & Kanchanpur district. The main objective of study was to see the prevalence of Kumri in goat, especially in Hilly area & Tarai area of Kailali & Kanchanpur district. Active surveillance was done through questionnaire and serum Faecal samples were taken from infected goats. This investigation programs was conducted in sites of Kailali district.

Namely, Ramsikharjhala , Chaumala Godavari VDC, Masuriya VDC and , Phulbari VDC. In this study a total of 291 house hold were interviewed and sampled, there were 3484 goats in that area .Summary of investigation and finding are given below

Summary of goat [population

Site	Farmer number	Total goats	Kumri (sateria spp)/Paralysis	Prevalence%
Phulbari-7	28	184	48	21.85
Godavari-8	54	815	74	9.7
Ramsikhar jhala	67	1402	252	17.97
Chaumalka	35	385	96	24.93
Masuriya	21	189	39	20.63
Total	205	2975	509	17.10

This investigation programs was conducted in sites of Kanchanpur district.

Namely Krishnapur VDC. A total of 86 house hold were having 516goats were involved in this investigation programm. Summary of investigation and finding are given below

Summary of goat [population

Site	Farmer number	Total goats	Kumri (sateria spp)/Paralysis	Prevalence%
Krishnapur	86	516	96	16.66

Method of Investigation

Active Surveillance through questionnaire & Clinical examination.

Finding

1. Goats effected by Kumari (setaria spp.) were 605
2. (Kailali 509 Kanchanpur 96)
3. Affected month Bhadra toMangshir
4. prevalence Rate 17.33%

Sub clinical Mastitis Investigation Program

This program was conducted to see the prevalence of sub clinical mastitis in milking cow of Kailali Kanchanpur & Doti districts. So fresh milk samples were taken from farmers directly and tested by SLST reagent immediately.

Investigation Site

Kailali (Masuriya Phulbari,Tikapur Dhangadhi,& Darakh)

Kanchanpur (Suda VDC ,Daizzi VDC,Tribhubanbasti,& Shreepur,)

Doti (Chhatiwan VDC Budar area)

Objective - To see the prevallence of sub clinical Mastitis in cow in Kailali , Kanchanpur & Doti district .

Method – Active Surveillance & Questionnaire

- Milk sample were directly taken from farmers.
- Test Methodology : Through SLST Reagent(Coagulation)

Summary of Sub clinical mastitis Investigation program

Site	Farmer	C.Cow milk sample		L.Cow milk sample		Prevalence
		+ve/ Total	-ve/ Total	+ve/Total	-ve/ Total	
Kailali	204	29/106	77/106	31/260	229/260	16.39%
Kanchanpur	118	31/150	119/150	0/27	27/27	17.51%
Doti	36	3/19	16/19	2/29	27/29	9.60%
Total	358	63/275	212/275	33/316	283/316	16.24%

Findings –

- Total Prevalence in Cow was 16.24% .
- The low Prevalence of sub clinical mastitis in local breed .

Prevention & Control program of Khari

This program was conducted to control Khari disease in Buffalo of Baitadi district.
Program Site.

Baitadi (Shidheshwor,shidhapur,shilanga & Sakar VDC)

Total No.of Household:- 204

Total No.of Buffalo:- 207

Total No.of Affected:- 135(98 Lacting& 37Dry were affected)

Method of Control program of Khari Disease:-

- Active Surveillance, Questionar .
- Clinical Examination.
- Free Distribute Medicine
(Pentasulphate,UMB,Anthelmentic,MiniralMixture, inj Ivermectin Teeburb etc)

Farmer's Experience:

- Grazing,Feeding vally,sugar,& Honey reduce the diseases incidence.
- Usally death occurs after 3-4 years .
- Mostly seen in winter months .
- Incidence is more in stall feed animals than grazing animals.

Disease reduced affected animals use(Pentasulphate,UMB,Anthelmentic,MiniralMixture, inj Ivermectin Teeburb etc

Our Experience:

- Pregnant buffalo is not affected but Lcting & dry Buffalo were affected.

- Improvement of stall/shed of animals.
- Ventilation is necessary in shed.
- Pakka floor necessary in shed.
- Routine Deworming.
- Diseased animals daily used Pentasulphate, UMB & vally Khari disease control .

1. Laboratory Services

1.1 Parasitological Examination

Faecal samples were examined adopting both qualitative and quantitative methods. In the fiscal year 2070/71 altogether 305 faecal samples from sheep and goats were examined. Whereas 1659 intestinal and caecal scrapping of poultry during necropsy examination revealed that 302 (18.20%) cases were found positive for coccidiosis.

Twenty five skin scrapings from dogs were received for the examination and identification of mites. Among these samples, 6 samples were found positive for the mites in dogs .

1.2 Microbiological Examinations

Microbiological examinations include the isolation and identification of bacteria and fungi from the pathological samples received in the laboratory. Bacteriological culture and antibiotic sensitivity tests were performed of the samples received for microbiological investigation. During 2069/70 a total of 820 samples were examined in microbiology unit of the laboratory.

1.2.1 Clinical Mastitis:

Out of 751 samples of microbiological tests 515 samples were milk for the test of Clinical Mastitis. These entire 515 samples were subjected for the Sodium Lauryl Sulphate Test (SLST) screening test for detecting mastitis in the animals. In SLST test of milk sample 283(54.64%) samples of animals were found to be positive. After the SLST screening test these positive samples were subjected for the microbiological culture and antibiotic sensitivity test. After culture the clinical mastitis cases gave the following result:

Table : Different isolates of Clinical Mastitis

S.N.	Name of the isolates	No. of Cases	Percentage of cases
1	<i>E.coli</i>	92	44.23
2	<i>Salmonella sps</i>	1	0.004
2	<i>Staphylococcus sps.</i>	35	17.32
3	<i>Streptococcus sps.</i>	46	22.77
4	<i>Bacillus sps</i>	16	7.92
5	Proteus	3	1.48
6	No. growth	9	4.45
Total		202	100

All the organisms shown in the culture were subjected for the antibiotic sensitivity test and gave the following result

Table : Antibiotic Sensitivity pattern of mastitis milk samples

Antibiotics	HS	MS	R
Amoxicillin/Ampicillin	13	12	79
Oxytetracycline	17	17	75
Enrofloxacin	96	8	0
Gentamicin	16	36	23
Cefotaxime	10	22	7
Ceftiaxone	15	23	27
Levofloxacin	60	5	0

HS- Highly Sensitive, MS- Medium Sensitive, R-Resistant

1.3 Pathological Examinations

Pathological examinations mostly consisted of necropsy examination of carcasses presented in the laboratory where poultry dominates all. In the pathology unit, the cause of death of chickens presented was generally drawn on the basis of both the post mortem lesions observed and laboratory investigation of samples collected during necropsy examinations.

Table: Diseases of chickens diagnosed on the FY 2069/70 are summarized in the following

Diseases / conditions	Number of cases	Percentage
Colibacillosis	802	30.05
Ascites	221	8.28
CRD	452	16.94
Coccidiosis	285	10.68
Infectious Bursal Disease	409	15.32
Salmonellosis	103	3.86
Mycotoxycosis	161	6.03
Avian Nephritis	5	0.18
New Castle Disease	52	1.94
Litchy Heart Disease	25	0.93
Visceral Gout	18	0.67
Ascariasis	9	0.33
Others	121	4.53
Total cases (n) = 2668		

It can be seen during the FY 2069/70, Colibacillosis was the most prevalent poultry disease followed by chronic respiratory disease and infectious bursal disease. There was increase in the cases of Colibacillosis, which might be due to the increased number of farming and poor management condition of the farming system. Among 960 samples from poultry tested for avian influenza, 15 samples were found to be Flu A positive on rapid test. 2 of the 15 positive samples were H5 positive confirmed by Central Veterinary Laboratory wher as 13 samples were found to be H9 positive.

1.4 Serological examinations

Serological examinations mainly consisted of plate agglutination test of chicken serum to detect antibody against *Mycoplasma gallisepticum* and *Salmonella pullorum* organisms. Similarly, serum samples from cattle, buffalo, sheep, goats and dogs were tested for brucella antibodies using Rose Bengal Plate Agglutination Test (RBPT). During the fiscal year 2070/71, the serum samples tested and their results are presented as follows:

Table : Serological Test Result in RVL in FY 2070/71

Species	Number of Serum	Tested For	Test Method Applied	Results
Cattle/Buffalo	115	Brucellosis	RBPT	All Negative
Goats	535	Brucellosis	RBPT	All Negative
Dog	15	Brucellosis	RBPT	All Negative
Poultry	281	Salmonellosis	PAT	74+ve
Poultry	255	Mycoplasmosis	PAT	61+ve

Table : Following is the virological test results of samples of different species

Species	Number of Serum	Tested For	Test Method Applied	Results
Goats	270	PPR	Pen side test	15 +ve
Dog	113	Rabies	Rapid test	7+ve
Poultry	97	IBD	IBDV rapid test	65+ve
Poultry	103	Newcastle d/s	NDV rapid test	20 +ve

1.5 Haematological Examinations

Hematological unit of the laboratory is well equipped to determine a range of hematological parameters such as Total Erythrocyte Count (TEC) and Total Leukocyte Count (TLC), Differential Leucocytes Counts (DLC), Erythrocyte Sedimentation Rate (ESR), determination of hemoglobin (HB) and Packed Cell Volume (PCV) and staining of blood smears for blood protozoa and bacteria. A total of 100 blood samples from animals were examined for different hematological parameters.

1.6 Biochemical examinations

Biochemical examinations included biochemistry of serum and routine and microscopic examination of urine. Multistick strip was used for routine urine analysis. Microscopic examination of urine was done after centrifugation of the urine samples.

1.7 Sample Collection and Dispatch

During 2070/71, a total of 242 serum samples, blood, fecal samples, Swabs, Dead birds, brain and tissue samples of different animal species and poultry were collected from the disease investigation sites. A total of 442 various samples were dispatched to Central Veterinary Laboratory, Kathmandu and National FMD and TADs laboratory, Kathmandu for confirmatory disease diagnosis.