# ANNUAL REPORT 2019-2020



VETERINARY RESEARCH INSTITUTE ZARRAR SHAHEED ROAD, LAHORE CANTT

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### MESSAGE BY THE DIRECTOR GENERAL (RESEARCH) LIVESTOCK AND DAIRY DEVELOPMENT DEPARTMENT PUNJAB



of immense It is matter pleasure that Veterinary Research Institute. Lahore has been ISO 9001 :2015 certified. L&DD department, Punjab discarded old strategy of treatment of diseased animals and made paradigm shift from curative to preventive to ensure animal health and to save million of mpees spent on treatment of diseased animals. VRI is playing a pivotal role to make

sustainable preventive policy of L&DD department, Punjab. The recent advancements in biologics production have tremendously reduced the incidence of infectious diseases and ultimate loss of livestock farmers.

Let's us work hard to upgrade our institutions to get their output to the level of "good manufacturing practices" under auspices of OIE standards to prevent infectious and zoonotic diseases in livestock sector and consequently achieve disease free status, which will lead to export value added livestock and poultry products in regional and international markets.

#### Dr. Muhammad Iqbal

#### MESSAGE BY THE DIRECTOR VETERINARY RESEARCH INSTITUTE, LAHORE CANTT



Livestock is a backbone of agriculture economy of Pakistan & playing the pivotal role in the poverty alleviation & food security. The increase in livestock production is possible through disease control, better nutrition & management. It is great honor for me to lead the Veterinary Research Institute (VRI) which is the premier institute of Punjab province for the production of Veterinary Vaccines and antigens along with allied Research & Development. Veterinary Research Institute is contributing significantly to reduce and in control of many diseases. It is an honor to add, this institute has also feather in its cap for eradication of Rinderpest disease from Pakistan during the year 2003 through its sustainable hard-

work and devotion of scientists. The Organization has a long & successful history of & controlling diseases and vaccine available today in Institute represents years preventing of innovative research by the scientists. I have firm belief that under the vision of L&DD Department, the organization will achieve its through the provision goals of good quality vaccine. I admire the great efforts of the team of VRI and grateful for their constant support and hard work towards production of quality vaccines and their dedication in improvement of the quality through Research & Development. The VRI team has tackled substantive questions, challenges & built a significant reputation. Veterinary Research Institute is ISO 9001 :2015 certified & 17025 accredited. I wish a lot of success and progress for this research institute and its team.

#### Dr. Sajjad Hussain

## **EXECUTIVE SUMMARY**

Veterinary Research Institute, Lahore is the inaugural and leading research organization in the counuy, administratively controlled by the Livestock and Dairy Development Department, Government of the Punjab. The Institute was established in 1963 and is situated at Zarrar Shaheed Road, Lahore Cantt. The institute pledges research on important animal diseases prevalent throughout the province, produces vaccines and diagnostic agents for their effective control.

The core functions of VRI are research and development activities along with production of vaccines for the control of different infectious diseases. Institute imparts trainings to the veterinary staff and interns from different institutes. The total non- development budget allocation for the year 2019-20 was 311.728 Million.

The institute produced 225.448 Million doses of different vaccines along with Million doses of diagnostics for different tests. The Institute also supplied different biologics to other provinces including AIK. and Gilgit Baltistan along with technical Support.

Total area of VRI is 25 acres of land comprising of different sections and a housing colony for the staff.

VRI, Lahore is rich in highly qualified and trained human resource consisting of technical and nontechnical staff. The total strength of technical staff in BS-17 to BS-20 is 82 and strength of staff in BS-1 to BS-16 is 313.

All the labs of VRI are ISO 9001 :2015 Certified. Central Reference Lab and Quality control lab in the institute are for the testing of morbid samples and quality control of the biologics produced in the institute. Institute has successfully mechanized the production of biologics to improve the quality and efficiency of work through different development projects.

Capacity building of the staff is done through various local and foreign trainings. A day care center has been established for the children of the women working in the institute.

VRI, Lahore continued it's R&D and biologics production activities during Covid-19 Pandemic with skeleton Staff to fulfill the field demand.

# CORE TEAM Additional Principle Veterinary Officers





Dr. Rizwan Qayyum



Dr. Umber Rauf





Dr. Azam Ali Nasir



Dr. Asfa Rasool

#### Senior Veterinary Officers







Dr. Hafiz Muhammad Numan

Dr. Muhammad Asim

Dr. Bushra Zamir



Dr. Zain Ul Abidin







Dr. Sobia Amir



Dr. Iffat Huma



Dr. Nadeem Akram



Dr. Summya Sattar







Dr. Abdul Wahab





Dr. Nida Arooj



Ms. Asma Aziz



Dr. Aqsa Mushtaq

Dr. Muhammad Asif





Dr. Waseem Shahzad

Dr. Nida Luqman

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Dr. Sumbel Aslam

#### **Veterinary Officers**







Dr. Ali Abbas



Dr. Shahzad Qadir



Dr. Sadia Sarfraz



Dr. Nofil Mustafa



Dr. Tayyaba Naz



Dr. Amjad Iqbal



Dr. lqra Zafar



Dr. Hafiza Zain Ul Fatima



Dr. Asma Kausar



Dr. Shakhseema Shaukat



Dr. Ayesha Qadri







Dr. Zahid Fareed

Dr. Atta Ullah



Dr. Saqib Tanveer

Dr. Hamza Khalid

Dr. Sami Ullah



Dr. Hafiz M Waqar



Dr. M Usman Ashraf

Dr. Muhammad Azeem

Dr. Zubair Latif



Dr. M Saqib Hussain



Dr. Afeefa Shafique



Dr. Saba Waqar



Dr. Iqra Tahira



Dr. Umar Waqas

### ORGANOGRAM



### **MISSION STATEMENT**

"To improve the health and productivity of livestock and poultry through research on emerging & re- emerging diseases & development of quality vaccine & diagnostic reagents."

### Objectives

The main objectives of the institute include:

- Large scale production of quality biologics for the control of infectious diseases of livestock and poultry.
- Research studies in the related disciplines of animal health and biologics produced in VRI.
- Studies related to prevailing and newly emerging diseases of livestock and poultry
- Development and standardization of modern techniques for research, production of biologics and to improve the quality of biologics being produced at VRI.
- Training of in-service veterinarians, para Veterinary staff, post-graduate students and graduate interns from Veterinary / other universities from all over the Punjab

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### Performance of the VETERINARY RESEARCH INSTITUTE 2019-2020

### **1. BUDGET**

A. Non Development Budget

(2019-20 )					
Name of Head / Function	Demanded	Allocation	Modified Allocation	Released/ Final Allocation	Utilization
Total Employees Related	169.188	162.682	149.221	146.274	145.263
Total Operating Expense	446.849	237.011	252.617	165.454	165.368
Total:-	616.037	399.693	401.838	311.728	310.631

B. Development Budget

Year	Release (in Millions)	Expenditure (in Millions)
2019-2020	20.877	20.875

### **2. BIOLOGICS PRODUCTION**

#### a. Viral Vaccine Production (Million Doses)

Sr.	Name Of	Annual	Production
No	Vaccine	Target	
1	Peste des petitis Ruminants	21.77	11.75
2	Sheep Pox Vaccine	On Demand	1.16
3	Goat Pox Vaccine	On Demand	0.785



#### b. Bacterial Vaccines Production (Million Doses)



#### c. Avian Vaccines Production (Million Doses)

Sr. No	Name Of	Annual	Production
	Vaccine	Target	
1	Newcastle	105.00	96.39
	Disease		
	Vaccine		
2	VRI- FLU	On	2.00
	Vaccine	Demand	

![](_page_15_Figure_2.jpeg)

#### d. Diagnostic Agents Production

NAME OF PRODUCT	PRODUCTION
Anthrax Spore Vaccine	20000
Antinax Spore Vaccine	Doses
Mallein Concentrated	34625
	Doses
Rose Bengal Plate Test	93390
Antigen	Tests
Milk Ring Test Antigen	5750
	Tests
Brucella Concentrated	3000
Antigen	Tests
Mallein Ordinary	2875
	Doses

![](_page_15_Figure_5.jpeg)

### b. Auxiliary Activities

Sr. No.	Activities	Accomplishments
1	Media, Reagents & solutions produced	79592 Liters
2.	Diluent Produced	18.3 Million ml
3	No. of lab animals maintained & produced	3185
4	No. of Vaccine Doses Lyophilized	128.7726 Million Doses
5	Haemorrhagic Septicaemia Antigen	6160 ml

#### c. Quality Control Tests for VRI Products

The quality of following vaccines was checked:

Sr.	Vaccine/Antigen	2019-20
No.		Number of Batches tested
1	Hemorrhagic Septicemia Vaccine	78
2	Black Quarter vaccine	29
3	Enterotoxaemia vaccine	55
4	Anthrax Spore vaccine	02
5	Brucella antigen	07
6	Goat Pox/Sheep Pox Vaccine	05
7	Contagious caprine Pleuropneumonia Vaccine	34
8	Mallein antigen	05
9	Newcastle Disease vaccine	44
10	Peste des petites ruminants (PPR) Vaccine	23
11	Diluent	37
12	Avian Influenza+ND	03

\*The standard of IS0-9001-2015 and IS0-17025 in lab management system and lab testing was observed.

#### Total Samples Tested at Provincial Nutritional Lab VRI LAHORE

Sr.No.	Type of Sample	Number of samples	
1.	Wanda (Concentrate Cattle Feed)	296	
2.	Silage	88	
3.	Fodder	259	
4.	Poultry Feed	81	
5.	Wheat Grains/Dalia	13	
6.	Wheat Bran (Chokar)	37	
7.	Cotton Seed Cake (Khal)	183	
8.	Straws (Wheat Straw and Rice Straw)	60	
9.	Rice Broken	5	
10.	Soybean Meal	2	
11.	Grams	1	
12.	Bakery Waste	8	
13.	TMR	2	
14.	Cat Feed	1	
15.	Goat Feed	6	
16.	Rape Seed Cake	13	
17.	Beet Pulp	1	
18.	Sugar cane Mud	6	
19.	Formalin	16	
20.	Roti Tukra	45	
21.	Sulphuric Acid	2	
22.	Milk	76	

# a) Total Samples tested at Nutritional lab During 2019-20

23.	Maize Grains/Dalia	12
24.	Soybean Hulls	2
25.	Нау	2
26.	Rice Polish	3
-27.	Fish Feed	11
28.	Etanol	1
29.	Methanol	1
30.	Barley	1
31.	Canola Meal	1
32.	Antigen	9
33.	Mineral Mixture	3
34.	Water	97
	Total Number of samples Tested	1344

![](_page_19_Figure_1.jpeg)

#### b) Type of Tests Performed

Sr.No.	Type of Test
1.	Crude Protein (%)
2.	Moisture (%)
3.	Ash (%)
4.	Crude Fat (%)
5.	Crude Fiber (%)
6.	Aflatoxins (ppb)
7.	рН
8.	TDS (ppm)
9.	Cyanides presence in fodder
10.	Nitrates presence in Fodder samples (ppm)
11.	Presence of grains in silage
12.	Observance of half milk line in silage
13.	Aroma of silage
14.	Colour of silage
15.	Purity of Chemicals

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#### 1-No. of Field Samples processed for molecular confirmation.

# 2. No. of seed & vaccine samples processed for molecular confirmation in CRL/R&D VRI.

![](_page_21_Figure_3.jpeg)

#### **RESEARCH PROJECTS (2019-20) VRI**

Sr. No.	Title	Objective	Main Section Involved
	Preparation and Evaluation of	1) Standardization and production of Brucella Vaccine.	Antigen Section
1	Brucella abortus vaccine at VRI Lahore	<ol> <li>Evaluation of vaccine in Lab Animals.</li> <li>Evaluation of vaccine in field conditions.</li> </ol>	(Collaboration: CRL & QCL)
2	Determination of Post- Vaccine titers of Enterotoxemia by IHA and its relationship with toxin titration in Mice from Live Culture	<ol> <li>To standardize IHA to find out the post vaccine titers of epsilon antitoxin in the serum and its comparison with ELISA</li> <li>Co-relation of Epsilon toxin concentration in the live culture with the potency of vaccine prepared from that culture</li> </ol>	Anaerobe Section
3	Preparation and Evaluation of Cost Effective Oil adjuvants for Poultry Vaccines	<ol> <li>To prepare cost effective poultry vaccines Avian Influenza (H9) using Montanide (ISA 70 MVG), Eolane 130, Eolane 150 and Eolane 170 oils as adjuvants.</li> <li>Comparative Evaluation of oil based Avian Influenza (H9) vaccines prepared with Montanide, Eolane 130, Eolane 150 and Eolane 170 oil adjuvants by HA and HI tests in cockerels.</li> </ol>	Avian Influenza Section (Collaboration: R&D Section)
4	Development of Oil Based Hemorrhagic Septicemia Vaccine using Eolane as Adjuvant (PARB Funded)	Preparation and evaluation of cost effective oil based Hemorrhagic septicemia vaccine using cheaper oil adjuvant (Eolane, Total, France)	R&D Section (Collaboration : Biochemistry Section, Dr Ihtisham ul Haq, CVAS Jhang)

5	Comparative Evaluation of four different adjuvants by using HS BQ combo vaccines in rabbits and calves	<ol> <li>Preparation of HS BQ combo vaccine by using four different adjuvants (Montanide ISA-50 V2, Eolane 170, Aluminium Hydroxide Gel, Alum)</li> <li>Evaluation of efficacy in lab and large animals</li> </ol>	R&D and Anaerobe Section
6	Preparation and evaluation of Hemorrhagic Septicemia - Black Quarter Combo Oil adjuvanted vaccine	To prepare oil based HS BQ combo vaccine by using Montanide (ISA 50 V2) and Eolane 170 to evaluate its efficacy in buffalo calves at farm level	R&D and Anaerobe Section
7	Standardization of Lypholization cycles for Freeze drying of Peste Des Petits Ruminants vaccine and Sheep Pox virus vaccine	Evaluation of different lypholization cycles in order to identify optimal lypholization conditions that will deliver maximum retention of viral infectivity titer.	Cell Culture and Flury Section Collaboration (Freeze: Drying Section)
8	Molecular Detection and differentiation of Fowl adenovirus affecting poultry in Pakistan and determination of their homology with vaccine strains.	<ol> <li>Molecular Characterization of Fowl adeno viruses suspected cause of outbreaks of IBH/HPS in poultry in Pakistan</li> <li>Molecular characterization of strains of Fowl adeno virus serotypes associated with suspected outbreaks of HPS available in Punjab</li> <li>To determine homology of fowl adeno virus serotypes associated with suspected outbreaks of HPS in Pakistan with fowl adeno-virus serotype 4 strains present in commercial vaccines.</li> </ol>	CRL Section
9	Molecular characterization of fowl pox virus from morbid samples of local poultry and its adaptation on embryonated eggs	<ol> <li>Molecular epidemiology of fowl pox virus associated with outbreaks in rural poultry</li> <li>Adaptation of virus on embryonated eggs to develop the seed bank</li> </ol>	CRL Section
10	Molecular characterization of	1. Optimization of PCR to differentiate between Pox and Orf virus	CRL & Flury

	sheep pox virus and goat pox virus, their isolation and adaptation	2. 3. 4.	Optimization of PCR to differentiate between Sheep Pox and Goat Pox in one step Molecular characterization of Orf virus from small ruminants Molecular characterization of Sheep Pox and Goat Pox Virus.	Section
11	Molecular characterization of Orf virus from Ruminants, Its isolation and adaptation	<ol> <li>1)</li> <li>2)</li> <li>3)</li> </ol>	PCR confirmation of Orf virus in morbid samples collected from sheep, goat and camel inflicted with papules, pustules and scab like lesions from different districts all over the Punjab province and its differentiation with pox virus. Molecular characterization of Orf virus to determine genetic diversity of Orf virus strains effecting different species of ruminants in different districts of Punjab Isolation and adaptation of circulation Orf virus strains from characterized morbid samples.	CRL & Flury Section
12	Isolation and Molecular Identification of Different Lactobacillus strains from chicken and their application as probiotics in commercial Poultry Feed	1) 2) 3)	Isolation & Molecular characterization of Lactobacillus species lactobacillus rueteri, lactobacillus salivarius, L. rhamnosus, and lactobacillus bulgaricus. Standardization of Lactobacillus probiotics. Evaluation of probiotics on poultry health and its comparison with antibiotic growth promoters	QCL Section (Collaboration: Biochemistry Section)

13	Comparative Immunogenic Response of Live Vaccine of La-Sota and Mukteswar Strains of Newcastle Disease Virus by Different Routes of Inoculation in Layer chicken	<ol> <li>Determination of immunogenic effects of ND Lasota vaccine inoculated by ocular drops, injection and drinking water through serology and histopathology.</li> <li>Determination of effectiveness of different routes of inoculation of ND Mukteswer (injection, ocular, drinking water) by serology and histopathlogy.</li> <li>Compare the immunogenic response following ocular drops of ND LaSota and booster by injecting ND Mukteswer vaccine</li> <li>Compare the immunogenic response following inoculation of both ND LaSota and ND Mukteswer vaccine.</li> <li>Determination of adverse effects of both vaccines by Histopathlogy.</li> </ol>	Poultry Vaccine Section
14	Optimization, Standardization and Biomass production of <i>Pasteurella</i> <i>multocida</i> serotype B:2 on Fermentation Technology	To optimize the growth conditions to get maximum biomass by using fermentation technology	HS Section
15	Molecular epidemiology of the causative agent of contagious caprine pleuropneumonia (CCPP) infection in Punjab Pakistan	<ol> <li>To isolate and identify the actual causative agent for CCPP in Pakistan</li> <li>To determine the genetic variability among different isolates responsible for CCPP</li> <li>To prepare a vaccine containing the field isolate of CCPP</li> <li>To prepare cost effective inactivated oil based vaccine of CCPP in order to meet the International standards meant for its preparation.</li> </ol>	Mycoplasma Section

### **ISO 9001: 2015 CETIFICATION OF LABS**

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### Minister/ Secretaries Visit To VRI, Lahore

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Secretary L&DD, Mr. Nadeem Irshad Kayani Visit to VRI.

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Secretary L&DD, Capt (Retd) Saqib Zafar Visit to VRI

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## Disinfection of Premises During Covid-19 Pandemic

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### World Rabies Day (September 28, 2019)

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![](_page_30_Picture_2.jpeg)

Interactive session & awareness walk about Rabies at VRI, Lahore.

# Visit of Different Delegations/ Trainings/ Meetings at VRI, Lahore

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### Independence Day & Defense Day

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![](_page_32_Picture_2.jpeg)

Flag hoisting & Cake Cutting ceremony at VRI, Lahore.

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### Clean & Green Pakistan

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#### **Research Publications 2019-2020 (Abstracts)**

#### 1. Preparation And Evaluation Of Haemorrhagic Septicaemia Oil Adjuvanted Vaccine With A New Ready To Use Oil Adjuvant For Cattle And Buffaloes

#### Journal: Buffalo Bulletin (October-December 2019) Vol.38 -4

Authorfs: W. Shahzad, B. Zameer, S. Naz, M.S. Hussain, A. Aziz, R. Munir, S. Hussain, Z.A. Qureshi and M. Iqbal.

#### Abstract:

Haemorrhagic Septicaemia (HS) caused by Pasteurella multocida serotype B:2 is an economically important disease of cattle and buffaloes, which causes heavy economic losses due to sudden death of animals in developing countries like Pakistan. In this country, animals were being vaccinated by alum (adjuvant) precipitated vaccine twice a year. Immunity induced through this prophylactic measure lasts for 3 to 4 months only, which reflect an un-protective state of the animals between two vaccinations. A new HS oil based vaccine has been developed by adding an adjuvant Montanide ISA-50V2 with the ratio of 1:1. The bacterial dry weight has been adjusted to 2 mg/ml which results in to reduction of dose per animal from 5 ml (alum precipitated) to 2 ml. The addition of enrichments and aeration (sparging and vortexing) has resulted in dense bacterial growth

of Pasteurella multocida. The new vaccine haspassed sterility, safety and potency tests as per OIE, 2017. This new product has low viscosity and single shot is expected to confer solid immunity against HS for one year. Active Mouse Protection Test, Passive Mouse Protection test and Indirect Haem-Agglutination Tests have been used to evaluate its potency. Properties like easy to inject with no side effects such as swelling at the injection

site, have motivated the livestock owners to use this new product to protect their animals against fatal HS disease which will ultimately result in the increased productivity of livestock in Punjab, Pakistan.

#### 2. Comparative Efficacy of Different Inactivated Hydro-Pericardium Syndrome Vaccines Prepared from Infected Liver and Vero Cell Line Adapted Adeno Type 4 Virus.

#### Journal: World Journal of Vaccines, 10, 1-16 (2020).

Authors: Muhammad Danish Mehmood, Huma Anwarul-Haq, Faisal Amin, Sajjad Hussain, Ejaz Rafique, Muhammad Usman Ghani, Muhammad Ismail, Fareeha Ghaffar

#### Abstract:

Hydro-Pericardium Syndrome (HPS) is viral problem of commercial poultry caused by aviadeno virus type-4. In Pakistan the problems have been controlled by administering inactivated infected liver homogenate vaccine (ILHV). The use of liver based HPS vaccines remained potential threat for having hypersensitivity reactions in poultry. The current study was carried out to compare the

serological potency of HPS ILHV to vero cell line adopted vaccine in term of anti HPS-ELISA antibody titers. 14 HPS virus vaccines were prepared based on different concentration of antigen, type of adjuvants and source of virus substrate. Total of 160 birds were divided into 16 groups each containing 10 birds. At day of 14th age each bird of every group was injected with 0.3 ml dose of respective vaccine. It was observed that HPS infected liver based vaccine having 1 × 105.6, 1 × 105.6 and 1 × 103.6 bird lethal dose 50 induced 1092.10, 875.25 and 702.2 anti-HPS ELISA antibody titer respectively. The 20, 25 and 30 doses/gm HPS infected liver vaccine induced 110.4, 1071.9 and 1037.8 anti-HPS ELISA antibody titer respectively. Montanide based tissue culture HPS vaccine showed significantly higher 1148.45 anti-HPS ELISA antibody titer to aluminium hydroxide based vaccine (137.2) (P < 0.05). It is concluded that montanide based HPS vaccine prepared from tissue culture technique having biological titer  $\geq 1 \times 105.6$  TCID50 is serological potent against field infection. The vaccines based on such formulation could be prepared in future for effective immuno-prophylaxis against HPS virus.

# **3.** Hematological Changes and Comparative Efficacy of Allopathic and Herbal Drugs on Coccidiosis in Rabbits.

#### Journal: Baltica Journal (2020).

Authors:

Adnan Ayan1, Irfan Ahmed, Jamal Muhammad Khan, Shahzad Munir, Mubashir Hussain, Ahmad Kamran Khan, Abdullah Jalal, Muhammad Abdul Qudus, Muhammad Irfan Saleem, Ahmad Sheraz, Sajjad Hussain, Mir Zulqarnain Talpur, Kashif Rahim, Zulqarnain Baloch

Abstract:

Objectives: Rabbit coccidiosis is caused by Eimeria sp. inhabiting the liver and intestine. The present study investigated the coccidiosis in rabbits in terms of age, humidity, sex variations and temperature and chemotherapy was used in the form of allopathic and herbal drugs. Methods: A total of 112 rabbits possibly infected during coccidiosis outbreak were obtained from different locations of Lahore, Pakistan. The faecal examination was performed using direct smear, floatation to observe the oocyst of Eimeria spp. Results: It was observed that females were slightly more susceptible (47.2 %) than males (44.7 %); also the incidence of coccidiosis was more prevalent in baby kits from 1-3 months (25.3 %) as compared to adults (15.2 %). Monthwise analysis depicted that, the infection was more prevalent in the month of March, and July (58.3 %, 60.7 %, and 73.9 % respectively due to high humidity. Coccidiosis also changes in blood parameters with increased level of WBCs (20.8 %), neutrophils (17.5 %), monocytes (29.1 %) and basophils (11.3 %) while decreases RBCs (44.9 %), lymphocytes (18.3 %) and haemoglobin level (35.4 %). Chemical drugs minimize OPG (Oocyst per Gram) dramatically such as Sulphadimidine sodium and toltrazuril efficiently decreases the OPG to 48 % and 74.4 %, respectively. Herbal drugs in the form of Nigella sativa L. seeds (kalonji) minimized the OPG gradually 57 % and 53.4 % oocysts respectively in 14 days. Garlic extract reduces 57.8 % oocytes in faeces. Conclusions: The chemotherapy suggested that toltrazuril is more efficient for quick action compared to Sulphadimidine Sodium. In addition, we found that garlic and black seeds were more effective in reducing the OPG level.

# 4. Comparison of Thioglycolate broth with liver extract (BQ Vaccine Media) with traditionally used culture media for Mass Scale production of *Clostridium perfringens* Type D

Journal: J Biol Today's World 8 (3)

Authors: Azam Ali Nasir, Sohail Manzoor, Nofil Mustafa, Asma Kausar, Muhammad Usman Ashraf, Muhammad Imran

#### Abstract:

Thioglycollate broth with Liver Extract (BQ vaccine medium) was used to produce mass scale yield of Clostridium perfringens Type D. Upon growth of Clostridium perfringens in different culture media the OD value of bacterial growth in BQ vaccine medium, Reinforced Culture Medium (RCM), Cooked Meat Medium (CMM) and Nutrient Broth (NB) was 2.260, 2.184, 2.096 and 1.984 respectively. Mice Lethality result of growth in BQ vaccine medium was equal to that of RCM while growth in CMM and NB had very less lethality. ELISA percentage positivity for Epsilon toxins in the supernatant of growth in BQ medium at 450 nm wavelength at dilution of 320 and 640 was higher than that of all the culture media under comparison. It was concluded that BQ was an excellent culture medium for the development of Enterotoxaemia vaccine producing maximum concentration of epsilon toxin than any culture media used before.

# 5. Clinico Haemto- Biochemical and molecular diagnostic investigations of peste des petitis ruminants in goats

Journal: PVJ 40 (3)

Authors: Syed Abdul Khaliq, Mudassar Mohiuddin, Mudasser Habib, Riaz Hussain, Muhammad Abbas, Xiaoxia Du, Azam Ali Nasir, Ayesha Mohi ud Din, Ahrar Khan and Jiang Bayi

#### Abstract:

Peste des petits ruminants (PPR) is an acute, infectious and devastating disease of small ruminants, especially for goats. Recently, an outbreak of PPR occurred at a goat farm in Nankana District, Punjab province, Pakistan with 100% (n=105) morbidity and 24% (25/105) mortality. The goats showed characteristic signs of PPR including high temperature, oculo-nasal discharges, diarrhea and ulcerative lesions in the oral cavity. The clinical signs, pathological lesions, hematological values, and serum biochemistry were studied. On postmortem examination, severe pneumonia and enteritis were observed in infected animals. There was marked lymphopenia, decreased erythrocytes level with increased mean corpuscular hemoglobin volume (MCV). The release of albumin and pus cells in urine indicated the kidney damage. Clinical outcome, gross lesions, and histopathological findings were suggestive of Peste des petits ruminants virus (PPRV) infection, which was confirmed by the application of antigenically conserved N genebased RT-PCR. More than 70% of clinically infected animals were found positive for PPR virus (PPRV) using the RT-PCR. Further investigations carried out to understand the phylogenetic relationship revealed lineage IV PPR viruses involved in the outbreak having more than 90% similarity with isolates previously reported from Pakistan. Pakistan is still in the endemic state for PPR as various outbreaks have been reported from various regions of the country. Regular monitoring of PPR disease and viruses spread are essential for the implementation of appropriate control actions and to know the risk assessment.

## 6. The adverse effects of carbofuran are efficiently counteracted by the supplementation of star anise (*Illiciumverum*) in broiler chicks.

#### **Journal: Toxin Reviews**

#### Authors:

Ashiq Ali, Aisha Khatoon, Zain Ul Abidin, Sajjad Hussain, Muhammad Kashif Saleemi, Rao Zahid Abbas, Muhammad Tariq Javed, Shafia Tehseen Gul & Farzana Rizvi,

#### Abstract:

This study was designed to investigate carbofuran (CF)-induced pathological alterations in broiler chicks and ameliorative potential of Star anise against these anomalies. Ninety-day old broiler chicks were procured from a local hatchery, equally divided into six groups and were given different combinations of CF (6 and 10 mg/kg) and Star anise (6 g/kg). Parameters studied were feed intake, mortality, body weight gain, and relative organ weights along with some hematological and serum biochemical parameters. Results of this experiment confirmed a mitigative potential elucidated by Star anise against CF-induced toxicopathological, hematological, serum biochemical, and histopathological alterations in broiler chicks.

#### 7. A Study on Pathological Effects of Acholeplasma laidlawii Isolated from Buffaloes in Mice Model

#### Journali: Pak. j. life soc. Sci. (2019), 17(1)

Authors: Syed Khurram Fareed, Sobia Naseem Siddiqui, Mehir Un Nisa Iqbal, Faiz Muhammad, Sajjad Ali, Taseer Ahmed Khan, Nabeel Ijaz and Aqeel Ahmad

Abstract: Respiratory distress has become a hot issue that is causing severe infection in livestock industry of Pakistan. The exact and timely diagnosis is incredible to treat the disease. However, Acholeplasma (A.) laidlawii is found very significant from buffalo lungs but being a ubiquitous organism, its pathogenic description is not completely understood. The study was designed to validate the involvement of A. laidlawii in respiratory diseases in buffaloes. For this purpose, experimental trials on mice were conducted to confirm the involvement of the organism in respiratory tract infection. It was re-isolated from experimentally infected mice, showing lesions in respiratory tract (83.3%), proving Koch's postulates. Statistically, the experimental group-A (subcutaneous route) showed significant difference (P0.05) in all cases. Based on current study, it may be concluded that the organism is opportunistic, and can produce either disease or lesions on argeted organs in stressed animals, particularly buffaloes.

# 8. Abattoir based Sero-Survey of *Mycobacterium avium* subspecies Paratuberculosis in Bovines in District, Faisalabad-Pakistan.

Journal: Pakistan Journal of Zoology, 52(1).

Authors: Rais Ahmed, Muhammad Khalid Mansoor, Iftikhar Hussain, Muhammad Saqib, Muhammad Hammad Hussain, Amjad Islam Aqib, Haleema Sadia, Javed Muhammad, Asma Irshad, Kashif Prince, Muhammad Zain Saleem and Abdul Whab Manzoor

Abstract:

Bovine paratuberculosis is a chronic disease of cattle and buffaloes, causing progressive weight loss, persistent diarrhea and finally death. Due to zoonotic nature of the disease, workers in slaughterhouses are at a high risk of infection. In this study, the status of paratuberculosis was assessed in a slaughterhouse located in district Faisalabad, Pakistan. A total of 455 blood samples were collected from slaughtered cattle and buffaloes at random and then tested by a commercially available ELISA kit for the detection of antibodies against *Mycobacterium* avium subsp. paratuberculosis (MAP). Data were analyzed using chi square for ELISA and odds ratio for finding association of species, gender, body weight, age and body condition score (BCS) with prevalence of antibodies against MAP. Seropositivity against MAP was significantly (P<0.05) detected. Male and female animals were equally susceptible to the disease (OR: 0.908, 95% CI= 0.6111-1.350). Age groups were not found associated with chances of being seropositive. Seropositivity was significantly (P<0.05) higher in animals under 300kg body weight. Chances of being seropositive were found more in animals belonging to BCS 1 (OR: 13.25, 95%CI=6.29-27.88) and BCS 3 (OR: 4.78, 95%CI=2.36-9.70) than BCS 4 and above. In conclusion, gender, body weight and body condition score of the animals are positively associated with the occurrence of the disease. These data will not only help in screening of animals but will also be instrumental in future for isolation of MAP to make vaccine against paratuberculosis using local isolates.

# 9. Prevalence of *Helicobactor pylori* infection and its associated diseases in low socio-economic workers in tertiary care hospital of Lahore, Pakistan.

#### Journal: Biomedical Letters 2019; 5(1).

Authors: Noreen Sarwar, Rais Ahmed, Marya Saadullah, Kamran ullah Khan, Shagufta Kamran, Faisal Ameen Baig, Muhammad Khalid Mansoor, Amjad Islam Aqib, Kashif Prince, Muhammad Zain Saleem, Abdul Whab Manzoor, Haleema Sadia

Abstract: Helicobacter pylori is endemic worldwide and causes gastric ulcers, gastroesophageal reflux disease (GERD) and gastric carcinoma. Purpose of the present study was to determine the prevalence of H. pylori and its associated diseases among low socio-economic population attending a tertiary care hospital in Lahore, Pakistan. Total 603 patients with gastrointestinal (GI) tract problems were included from March 2015 to December 2016. Strip detection test, urea breath test (UBT), endoscopy and biopsies were performed to confirm the presence of H. pylori. Out of 603 patients, 48 patients were positive for H. pylori and the prevalence was assessed up to 7.9 %. Patients han females (31.3%). Age and gender betwaged >40 years were more affected than other young age groups. Males (68.8%) were more affected teen the two groups did not show significant association with H. pylori infection (p>0.05). But they were found to be inversely proportional to each other (p0.05) was not found to be statistically associated with the presence of H. pylori but hiatal hernia was strongly associated.

# **10. Elucidating the Genetic Diversity of Prevalent Strains of Peste des PetitsRuminants Virus in Gilgit-Baltistan Province, Pakistan**

#### Journal: Pakistan Veterinary Journal

Authors: Majeeda Rasheed, Tanveer Akhtar, Nabila Roohi1, Nida Arooj, Mashal Rasheed, Muhammad Farooq and Muhammad Yousaf

Abstract: Peste des petits ruminants caused by small ruminant morbillivirus (SRMV), is a highly contagious disease of small ruminant. It is endemic in Gligit-Baltistan (GB) territory of Pakistan where a number of clinical cases are being reported frequently. However, so far, the phylogenetic relationship of prevailing strains in this particular geography has remained elusive. We carried out a study to characterize partial N gene of PPR viruses from outbreaks during years 2017-18 from GB. Out of 30 clinical samples analyzed, 27 percent (n=8) were positive for N gene (351). However, owing to close relationship expected among study sequences, only two samples from sheep and one from goat were sequenced by phylogenomic analysis using a range of bioinformatics tools. Phylogeny analysis revealed a close relatedness with the previously reported viruses with lineage IV. Sequence composition showed high level of homology with circulating viruses suggesting that that these viruses do not undergo rapid genetic changes in N gene. However, a number of genomic and residue substitutions were noted within the prevalent viruses as compared to those reported previously. This study provides the first genetic evidence of SRMV strains sequence analysis of partial N gene involved in recent outbreaks in the GB region. Future studies are necessary to further ascertain the study outcomes and elucidate the molecular epidemiology of prevalent strains in the said geographical area for better disease control and management interventions.

#### **11.** Serological study for the detection of antibodies against leptospira in goats.

#### Journal: Pakistan Veterinary Journal

Authors: Muhammad Umair Aziz, Muhammad Ijaz, Arslan Ahmed, Awais Ghaffar, Hammad Nayyar Ghauri, Muhammad Zeeshan Zafar, Muhammad Altaf, Farah Nadia Sheikh, Imtiaz Ahmad and Waseem Shahzad

#### Abstract:

Leptospirosis is globally distributed disease of zoonotic significance, caused by pathogenic spirochetes of the genus leptospira. There is no study regarding the seroepidemiological investigation of leptospirosis in goats of Pakistan. The study was intended to assess the sero-prevalence and risk factors of leptospira along with the alterations in hematobiochemical parameters between seropositive and seronegative goats. Serum samples from 155 goats were collected aseptically from tehsil Pattoki and screened by indirect ELISA to detect antileptospiral antibodies. A total of 15 herds were selected and from each herd 10-15 goats were taken for sampling by convenient sampling technique. The records regarding hypothesized risk factors were analyzed by logistic regression model on SPSS. The overall seroreactivity to leptospira was 21.29%. The potential risk factors contributing towards disease occurrence were; age of animals, different types of rearing systems, grazing status, call of veterinary professionals and body condition scores of animals.

# 12. Pathotyping and genetic characterization of a highly virulent Newcastle disease virus strain isolated from recent outbreak in District Okara, Pakistan.

#### Journal: Pak Vet J, 39(3): 353-358-173

Authors: Asif Mehmood, Muhammad Hidayat Rasool, Aftab Ahmad Anjum, Muhammad Asif Zahoor and Muhammad Shafique

#### Abstract:

In Pakistan, growth of poultry industry is confronted by many viral diseases and Newcastle Disease (ND) is on top. The present study was conducted for pathotyping and genetic characterization of NDV isolated from an outbreak in District Okara region of Punjab, Pakistan. Post mortem examination of fresh carcasses (n=10) was performed and various organs like trachea, spleen, lung tissues, and cloacal contents were collected from birds showing typical lesions of the disease aseptically. Tissue homogenate was inoculated in 9 days old embryonated chicken eggs and allantoic fluid was harvested. The presence of virulent virus was checked by hemagglutination, hemagglutination inhibition using hyperimmune serum and ICPI assays. NDV was purified using sucrose gradient ultracentrifugation and confirmation was done by F-gene amplification, SDS-PAGE and mass spectrometry of fusion protein. The HA titer was 1:256, and hemagglutination inhibition was achieved using Anti NDV serum. A band of  $\approx 800$  bp of F gene (Acc. No. Okara/Pakistan/MH607122) was amplified which revealed 99% similarity with UVAS/Pak/2015/MF437287, UVAS/Pak/2016/KX791187 and Tehran/Iran/MG871466 strains. SDS-PAGE yielded five bands including Fusion protein with the estimated mass of 58896.0 Da. The presence of fusion protein and ICPI value was 2 which confirmed virulence of isolated strain resulting in high mortality. Thus, it is the need of the hour to characterize the prevalent indigenous strains of NDV to develop some novel vaccine strategies for effective control.

# **13.** Antibody Response of Goats to Gel Based Combined Vaccine Against Peste Des Petits Ruminants, Contagious Caprine Pleuroneumonia& Foot and Mouth Disease

#### Journal: JAPS, 29(4)

Authors: M. Khalil, K. Muhammad, J. Nazir, A. Z. Durrani, F. A. Khan, S. Sarfaraz, R. Riaz and A. Zeb

Abstract: Present study was designed to prepare and evaluate gel based combined vaccine against Peste des Petits Ruminants (PPR), Contagious Caprine Pleuropneumonia (CCPP), Foot and Mouth Disease "O serotype" (FMDO) to mitigate the cost and frequency of the vaccination. Gel based vaccines contain either single immunogen (monovalent) of PPR, CCPP or FMDO, two (bivalent) PPR+FMDO, PPR+CCPP, FMDO+CCPPP or three (trivalent) PPR+CCPP+FMDO. Each dose (0.5 ml) of either of the vaccines contained 105 tissue culture infective dose (TCID50) units of FMDO virus, 107 TCID50 units of PPR virus and 0.15 mg of Mycoplasma mycoides capri. Each of the vaccine was injected subcutaneously to each of the goats (n=4). Antibody response of goats to FMDO, PPR and CCPP was determined by complement fixation test (CFT), virus neutralization test (VNT) and enzyme linked immunosorbant assay (ELISA), respectively at 0, 60, 120 and 180 days post-priming. Antibody response of the goats to either of the immunogen was not significantly different irrespective to the form of vaccine (monovalent, bivalent or multivalent vaccine) (P>0.05). It indicated that either of the immunogen did not interfere the immunogenesis process of other immunogens in the same vaccine.

# **13.** Immunomodulatory Effect of Newcastle Disease Virus on Inactivated *Mycoplasma Gallisepticum* Vaccine Response In Chickens

#### Journal: Pakistan Journal of Zoology.

Authors: Rabia Riaz, Khushi Muhammad, Masood Rabbani , Muhammad Amjad Iqbal, Aamir Riaz Khan , Sadia Sarfaraz, Mudassar Naseer and Khalid Majeed

#### Abstract:

In Pakistan the poultry sector is an important and vibrant segment of agriculture in Pakistan with a significant contribution to the national GDP (1.3%). Mycoplasma gallisepticum (MG) causes avian mycoplasmosis and is only controlled through mass vaccination. Antibody response of birds to inactivated MG vaccine is poor. On 7th day of age (broilers: n=40) each bird of group A, B, C and D (n=8) was vaccinated (subcutaneous: 0.3ml) with G-MG, O-MG, G-MG+NDV (New castle Disease Virus) and O-MG vaccines, respectively. Serum samples were collected on 21, 28 and 35 days post vaccination for anti-MG-ELISA antibody titer. Cumulative mean anti-MG-ELISA antibody titer of birds to G-MG, O-MG, G-MG+NDV, O-MG+NDV and control was 80.30, 137.49, 91.59, 169.76 and 0.15 units respectively on 35 days post-vaccination. It is concluded that NDV has insignificant immunomodulatory effect on antibody response of birds to the MG vaccine.

### Way Forward/Research Priority Area

- Progressive control of the PPR in the Punjab leading towards its eradication under the road map of OIE and FAO 2030.

- 100% Vaccine Production for control of HS, BQ, ETV, CPPV, ND, etc.
- Development of Brucellosis Vaccine.
- Development of Rabies Vaccine.
- Development of HS+BQ Combo Vaccine.
- Preparation and Standardization of cost effective vaccine adjuvant.
- Development of Recombinant vaccine for Poultry .

## **TARGETS & THRESHOLD**

Following are the approximate demand of vaccines according to notified schedule for the year 2020-2021

Sr. No.	Name of Vaccine	Demand for the year 2020- 21 (Million doses)
1	Haemorrhagic Septicaemia Vaccine	28.60
2	Black Quarter Vaccine	14.66
3	Enterotoxaemia Vaccine	35.00
4	Caprine Pleuropneumonia Vaccine	14.55
5	New Castle Disease Vaccine (Lasota)	54.00
6	Peste Des Petits Ruminants Vaccine	20.50
7	*Other Vaccine (ASV, SPV, GPV)	On demand
8	Diluent	20.50
9	Diagnostic Agents (in doses)	On demand