



ANNUAL REPORT

2018-2019



VETERINARY RESEARCH INSTITUTE
ZARAR SHAHEED ROAD,
LAHORE CANTT

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



It is matter of immense 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 rupees 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 control the prevalence 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 preventing & controlling diseases. The vaccine available today in Institute represents years of innovative research by the scientists. I have firm belief that under the vision of L&DD Department,

the organization will achieve its goals through the provision of good quality vaccine. I admire the great efforts of the team of VRI and I am 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 of this research institute and its dedicated team.

Dr. Sajjad Hussain



EXECUTIVE SUMMARY

Veterinary Research Institute, Lahore is the inaugural and leading research organization in the country, 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 2018-19 was 417.146 Million.

The institute produced 206.2257 Million doses of different vaccines along with 0.2 Million doses of diagnostics for different tests. Other than Punjab province, institute supplies biologics to other provinces including AJK 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. Total strength of technical staff in BS-17 to BS-20 is 50 and strength of staff in BS-1 to BS-16 is 256. Total number females working in VRI, Lahore is 40.

All the labs of VRI are ISO 9001:2015 Certified. There is a Central Reference Lab and Quality control lab in the institute 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.



CORE TEAM

Additional Principle Veterinary Officers



Dr. Sarwat Naz



Dr. Rizwan Qayyum



Dr. Umber Rauf



Dr. Azam Ali Nasir



Dr. Asfa Rasool

Senior Veterinary Officers



Dr. Muhammad Asim



Dr. Bushra Zamir



Dr. Zain Ul Abidin



Dr. Sajjad Ali



Dr. Sobia Amir



Dr. Iffat Huma



Dr. Nadeem Akram



Dr. Hina Afrooz



Dr. Summya Sattar



Dr. Aqsa Mushtaq



Ms. Asma Aziz



Dr. Waseem Shahzad



Dr. Muhammad Asif

Veterinary Officers



Dr. Sheraz Shahid



Dr. Ali Abbas



Dr. Shahzad Qadir



Dr. Sadia Sarfraz



Dr. Nofil Mustafa



Dr. Tayyaba Naz



Dr. Amjad Iqbal



Dr. Hafiza Zain Ul Fatima



Dr. Shakhseema Shaukat



Dr. Iqra Zafar



Dr. Hamza Khalid



Dr. Hafiz Waqar



Dr. Afeefa Shafiq



Dr. Ayesha Qadri



Dr. Sami Ullah



Dr. Zahid Fareed



Dr. Zubair Lateef



Dr. Muhammad Usman



Dr. Atta Ullah



Dr. Muhammad Azeem



Dr. Saqib Hussain



Dr. Saqib Tanveer



Dr. Saba Waqar



Dr. Iqra Tahira



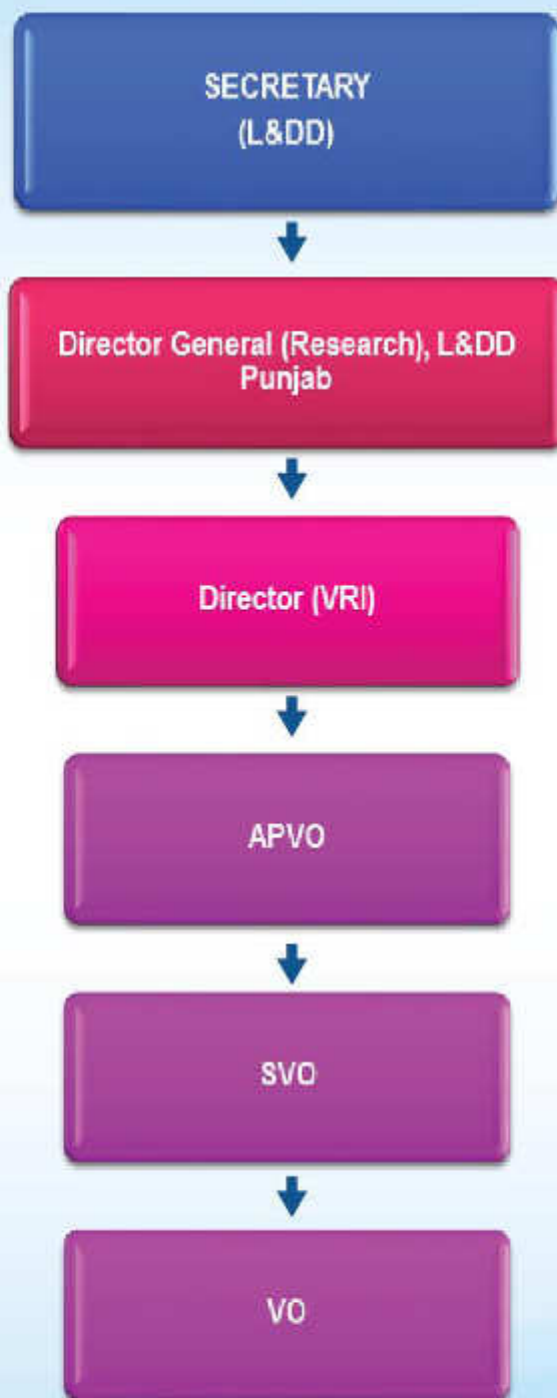
Dr. Asma Kausar



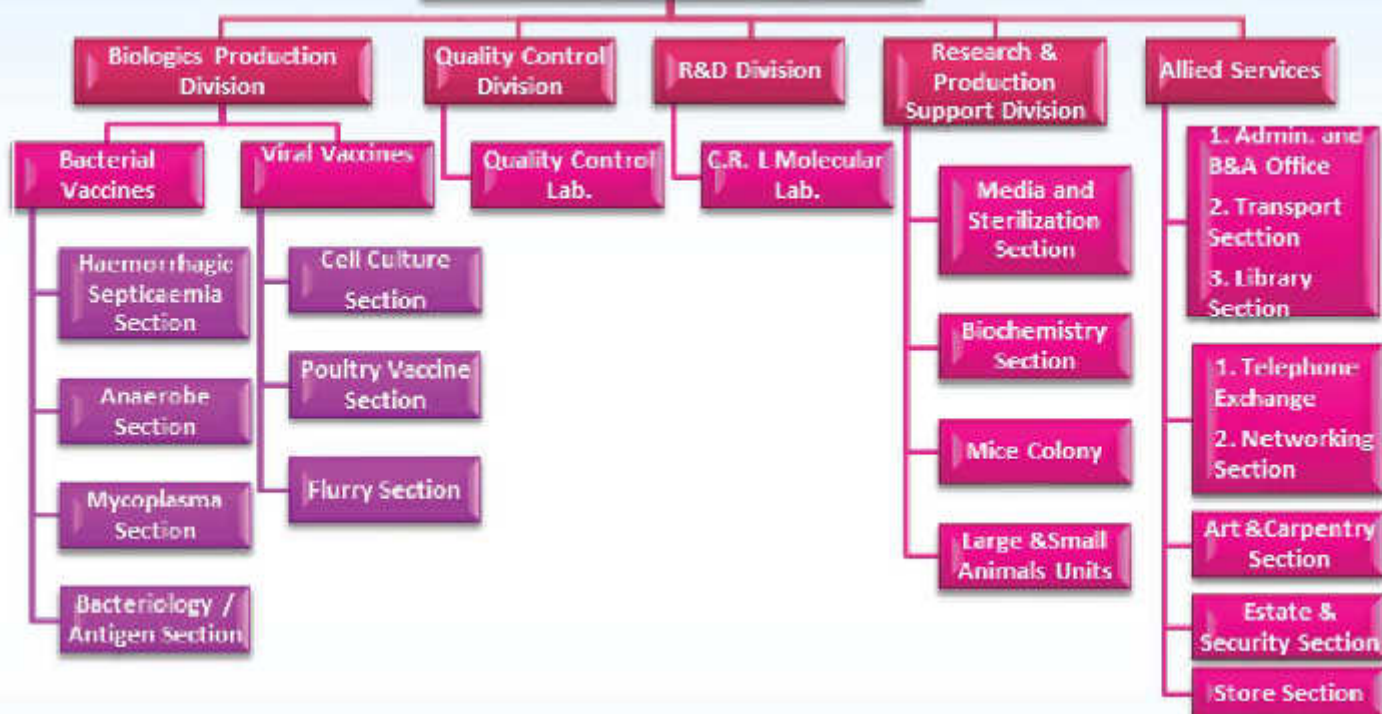
Dr. Umar Waqas



ORGANOGRAM

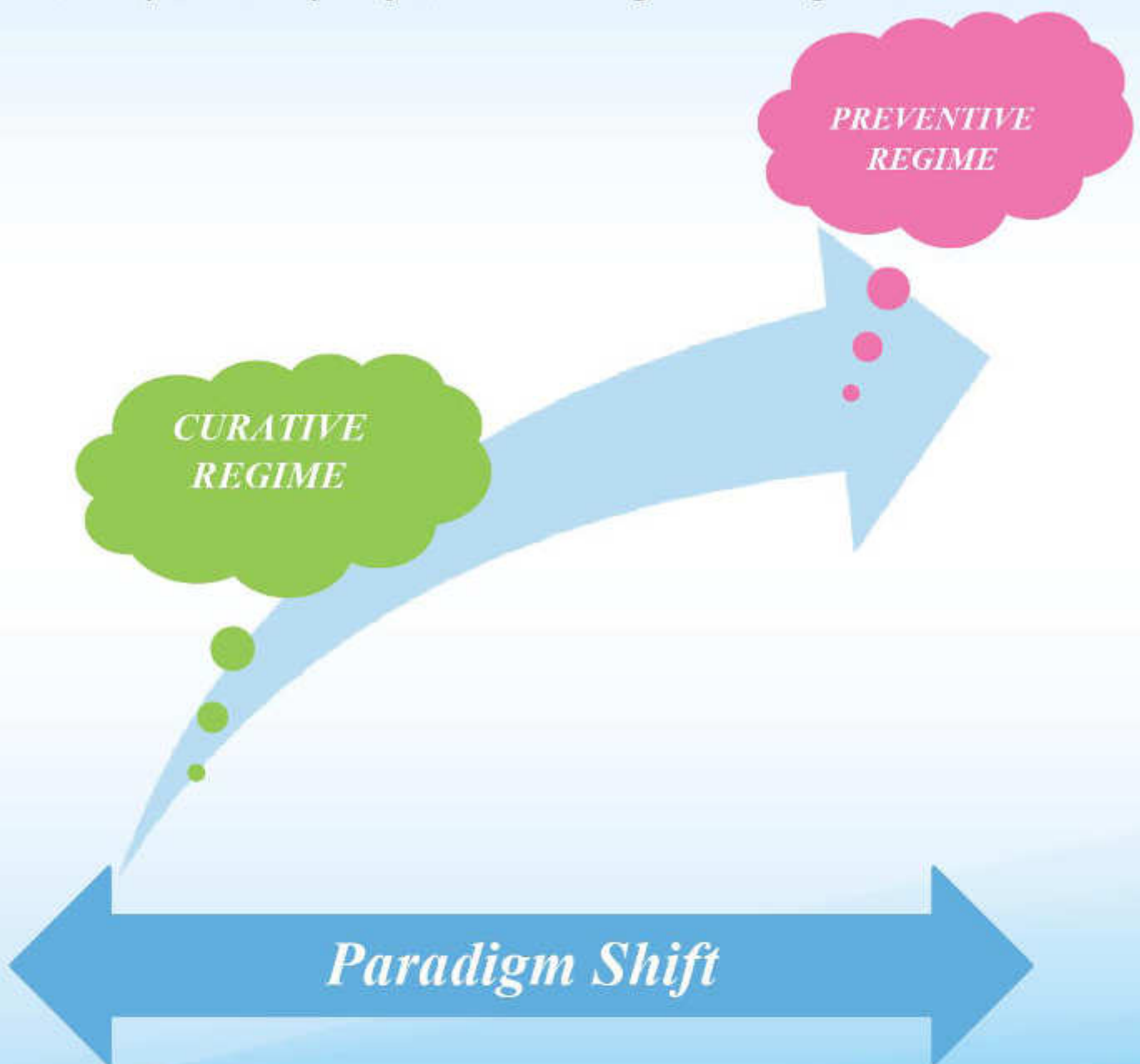


DIRECTORATE OF VRI



MISSION STATEMENT

“To improve the health and productivity of livestock and poultry through research on emerging & reemerging diseases & development of quality vaccine & diagnostic reagents.”



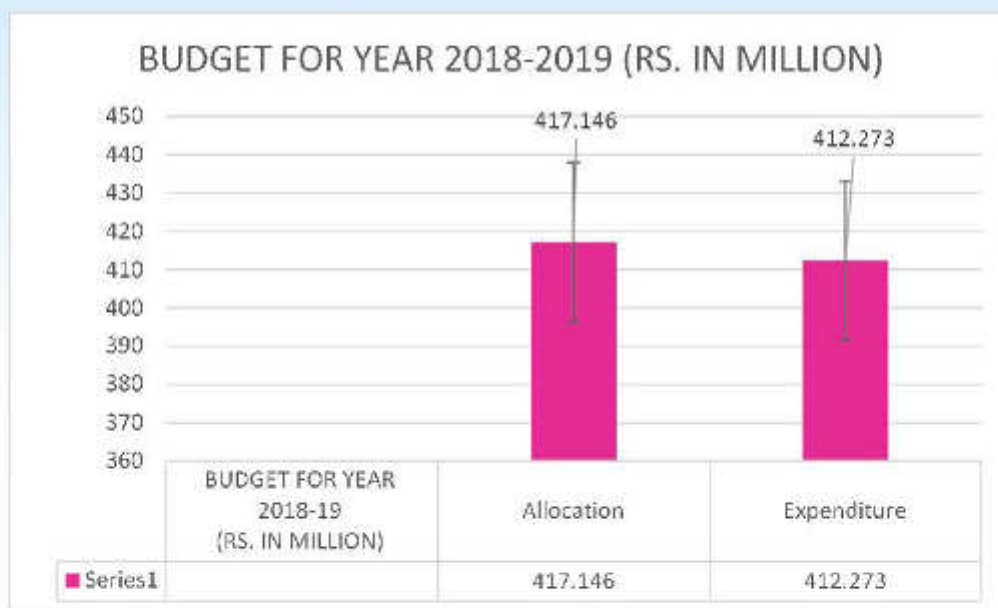
Objectives

- 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



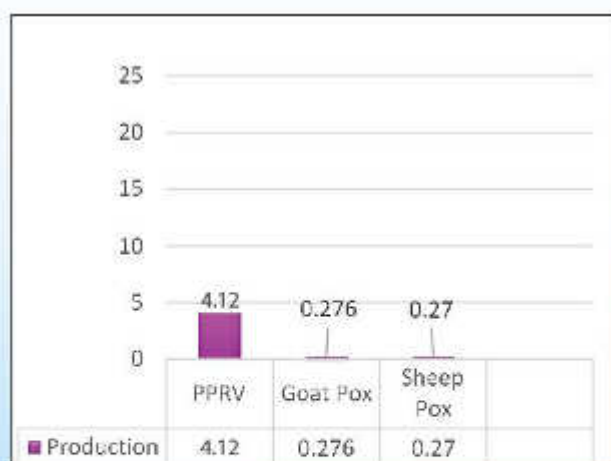
Performance of the Veterinary Research Institute 2018-2019

BUDGET



1. BIOLOGICS PRODUCTION

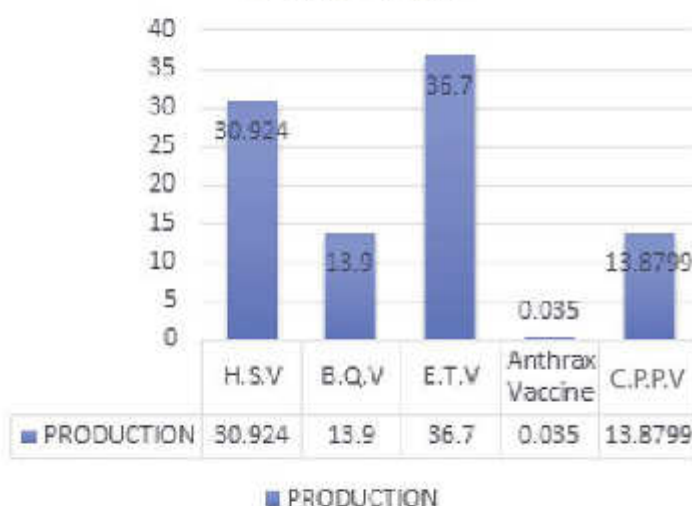
a. Viral Vaccine Production 2018-2019



Sr. No	Name Of Vaccine	Annual Target (In Millions)	Production (In Million)
1.	Peste des petitis ruminants (PPRV)	21	4.1214
2.	Sheep Pox vaccine	On Demand	0.27
3.	Goat Pox Vaccine	On Demand	0.276

b. Bacterial Vaccines Production 2018-2019

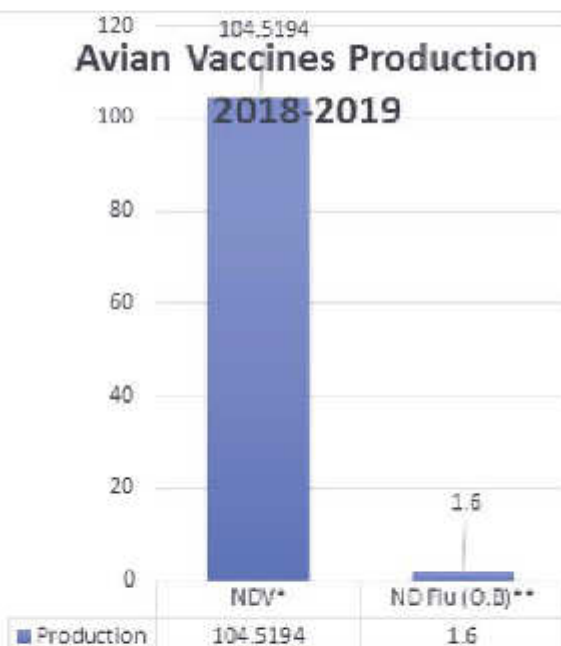
Bacterial Vaccines Production 2018-2019



Name of Vaccines	Annual Target (in million)	Production (in million)
Haemorrhagic Septicaemia Vaccine	28.37	30.924
Black Quarter Vaccine(BQV)	12	13.90
Enterotoxaemia Vaccine (ETV)	39.84	36.70
Anthrax Vaccine	On Demand	.035
Caprine Pleuropneumonia Vaccine (CPPV)	14.39	13.8799
Total	94.6	95.4389

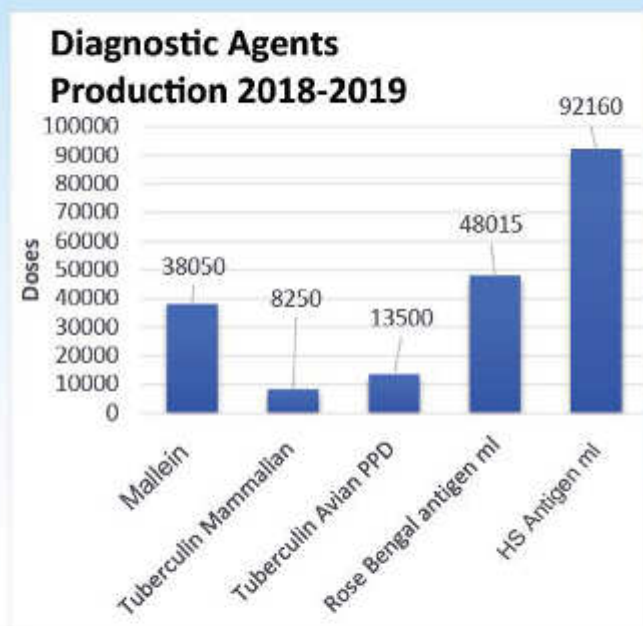
c. Avian Vaccines Production 2018-2019

Avian Vaccines Production 2018-2019



Sr No	Name of Vaccine	Annual Target (In Million)	Production (In Million)
1.	Newcastle Disease Vaccine	92.52	104.5194
2	VRI ND FLU Vaccine	As per demand	1.6
	Total		106.1194

d. Diagnostic Agents Production 2018-2019



Sr. No	Diagnostic Antigen	Annual Target	Production (test Doses)
1.	Mallein Doses	On Demand	38050
2.	Tuberculin (Mammalian)Doses	On Demand	8250
3.	Tuberculin (Avian PPD)Doses	On Demand	13500
4.	Rose Bengal (33 test/- ml)	On Demand	48015
5.	HS Antigen ml (20ml/-320 test)	On Demand	92160
	Total		199,975

b. Auxiliary Activities

Sr. No.	Activities	Achievement
1	Media, Reagents & solutions produced	137049 liters
2.	Diluent Produced	75000 vials
3	No. of lab animals maintained & produced	2587
4	No. of poultry birds maintained	1269
5	No. of Vaccine Doses Lyophilized	125.94 Million Doses
6	No. of Seed Vials Lyophilized	1560
7.	Haemorrhagic Septicaemia Antigen	5760 ml

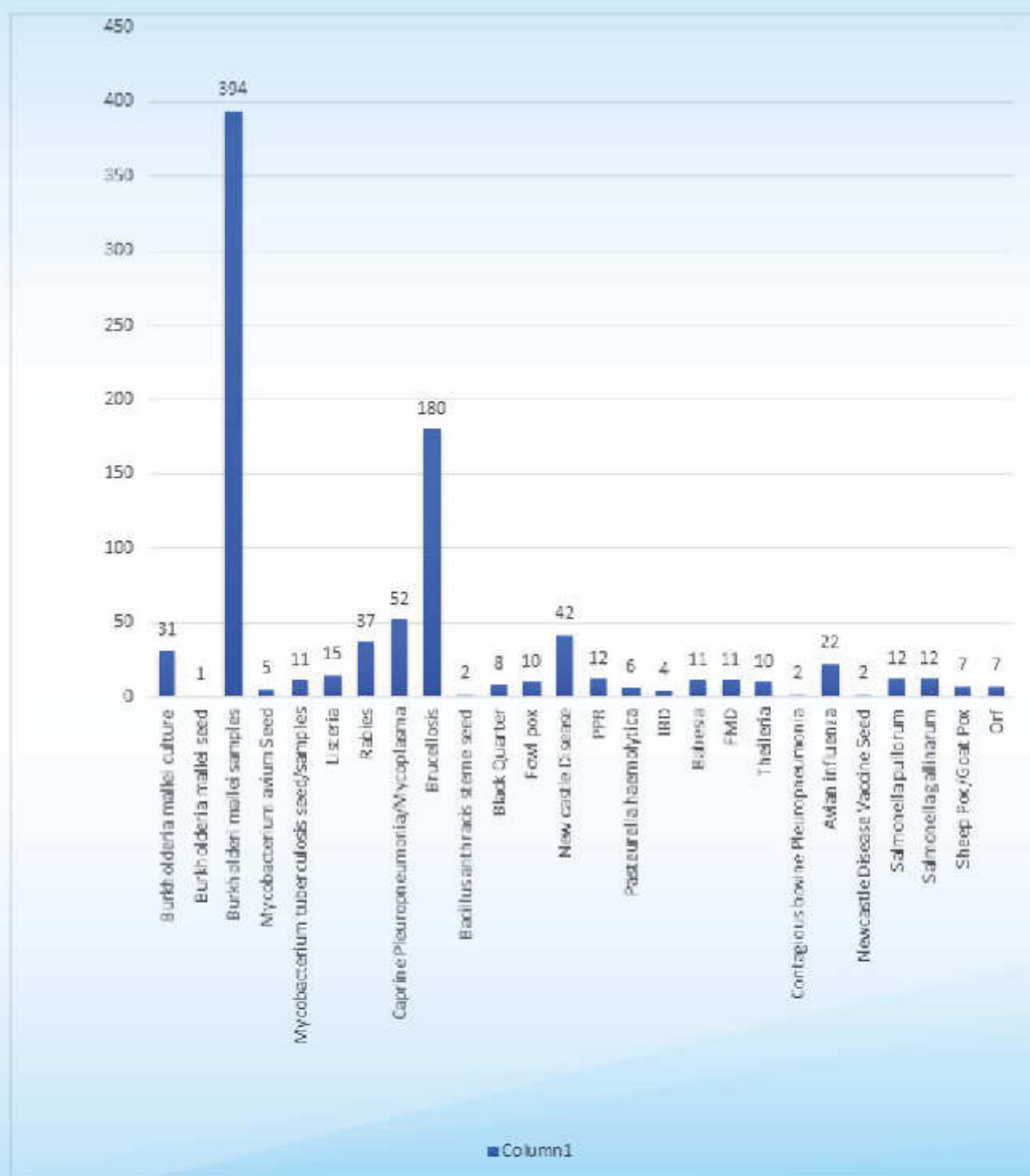
c. Quality Control Tests for VRI Products

Sr. No.	Vaccine	No. of Batch tested	Test performed
1.	H.S Vaccine	67	Sterility test, safety test
2.	B.Q.V	27	Sterility test, safety test
3.	E.T.V	55	Sterility test, safety test
4.	Anthrax Vaccine	03	Spore count, Sterility test, safety test
5.	C.P.P Vaccine	39	Sterility test, safety test
6.	Sheep pox	02	Sterility test, safety test
7.	Goat pox	02	Sterility test, safety test
8.	P.P.R Vaccine	48	Sterility test, safety test
9.	Avian Influenza+ND	04	Sterility test, safety test
10.	N.D.V	57	Sterility test, safety test, EID ₅₀ /ELD ₅₀
11.	FMD Serum	03	Sterility test, safety test
12.	Mallein	08	Sterility test, safety test
13.	Tuberculin	02	Sterility test, safety test
14.	Brucella Antigens	03	Spot Test
16.	Diluent	15	Sterility test

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

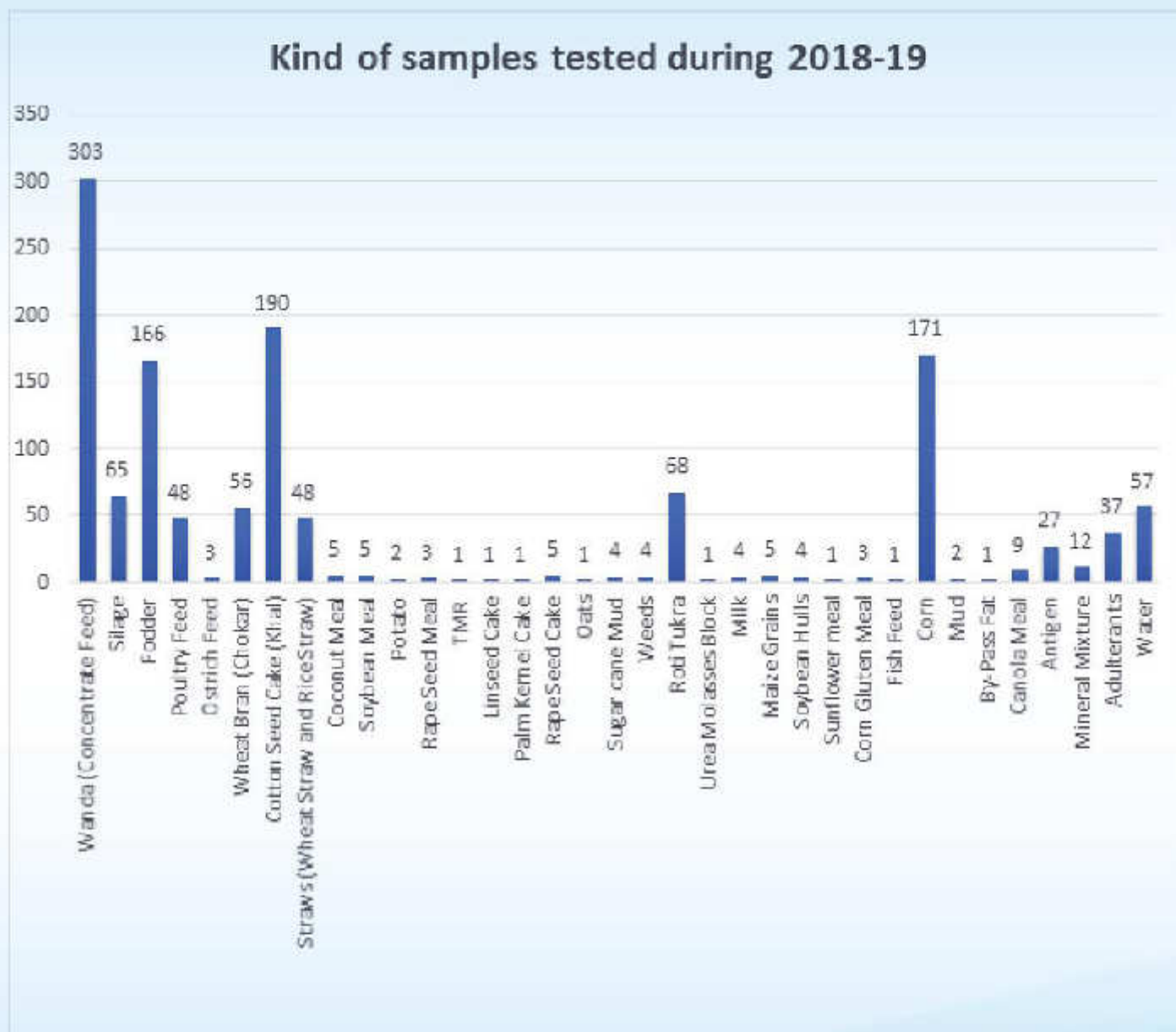
2. RESEARCH & DEVELOPMENT

Detail of Total 906 Vaccine/Morbid Samples Processed for Identification and Confirmation



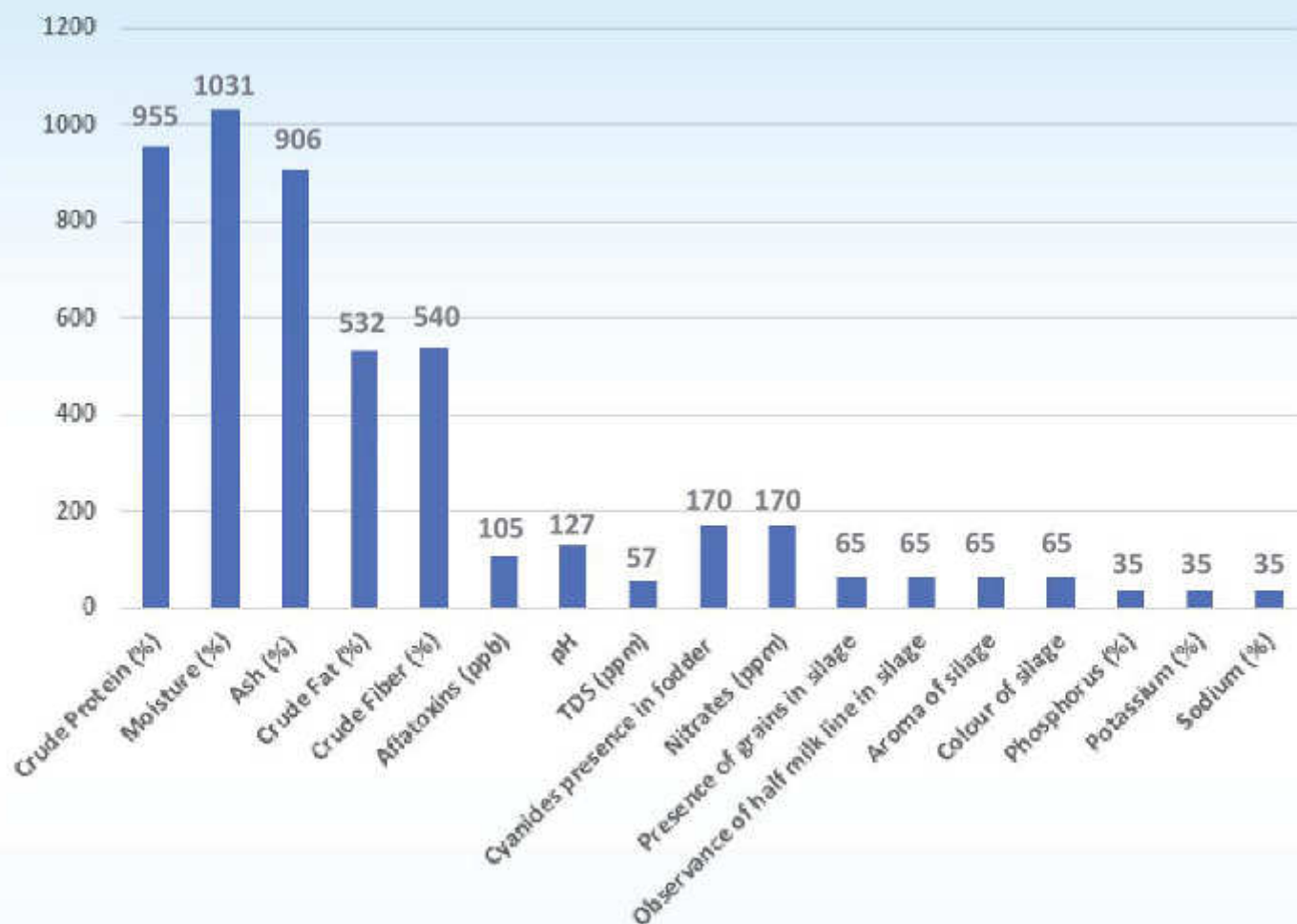
Total Samples Tested at Provincial Nutritional Lab VRI LAHORE

a) Total Samples tested at Nutritional Lab During 2018-19



b) Type of Tests Performed

Number of tests Performed during 2018-19





RESEARCH PUBLICATIONS 2018-2019

Name of Officer	Research Title	Name of Journal	Publish Year
Dr. Sajjad Hussain	Molecular Detection and Sequencing for S1 Glycoprotein Gene of Bronchitis Virus of 2016 Epidemic from Sindh and Punjab	Advances in Bioscience and Biotechnology.	2018
Dr. Sajjad Hussain	In Vivo Antiviral Effect Of Melaleuca Alternifolia (Tea Tree Oil) And Olea Europaea (Olive Leaf Extract) On Vero Cell Adapted Avian Influenza Virus.	International journal of pharmacy and pharmaceutical research	2018
	Antiviral Activity And Its Prospective Mechanism Of Action On Newcastle Disease Virus Using Crude Extract Of Four Medicinal Plants.	World Journal of Pharmaceutical Research,	2019
	Development and optimization of virus neutralization test in chicken embryonated eggs for indirect identification of avian influenza and Newcastle disease virus	Journal of Drug Delivery and Therapeutics.	2018
	Detection of influenza A virus contamination in Newcastle disease live virus vaccines and their pathological effects on visceral organs	Biointerface Research In Applied Chemistry	2019
Dr. Zain-ul-Abidin	An extensive review of experimental ochratoxigenesis in poultry: II. Hemato-biochemical and immunological alterations along with other health issues.	Toxin Reviews	2019
	Confirmation of rabies infection by mouse inoculation test and reverse transcriptase polymerase chain reaction in suspected samples of cow and mule	Pakistan Journal of Zoology	2018
	An extensive review of experimental ochratoxigenesis in poultry: I. Growth and production parameters along with histopathological alterations.	World Poultry Science Journal	2018
	Mycotoxicosis: Diagnosis, prevention and control: Past practices and future perspectives	Toxin Reviews	2018
	Dietary L-carnitine and vitamin E; a strategy to combat ochratoxin-A induced immunosuppression,	Toxicon	2018
	Effects of feeding bentonite clay upon ochratoxin A induced immunosuppression in broiler chicks.	Food Additives and Contaminants	2018

	Dietary L-carnitine and vitamin E; a strategy to combat ochratoxin-A induced immunosuppression,	Toxicon	2018
	Effects of feeding bentonite clay upon ochratoxin A induced immunosuppression in broiler chicks.	Food Additives and Contaminants	2018
Dr. Asma Kausar	An insight into the ecobiology, vector significance and control of Hyalomma ticks (Acari: Ixodidae): A Review	Acta Tropica	2018
Dr. Azam Ali Nasir	Bacterial count and predisposing factors of Clostridium perfringens (CPA gene) infection alongwith antimicrobial sensitivity in diarrheic sheep in Pakistan	Tropical bio medicine	2018
	Investigation of different serotypes of FMD in vaccinated buffaloes in southern areas of Punjab Province.	Pakistan Veterinary Journal	2019
Sumiyya Sattar & Hina Afroz & Asfa Rasool	A novel approach for development, standardization, and safety testing of enriched alum-precipitated vaccine against hemorrhagic septicemia in different breeds of cattle.	Tropical Animal Health and Production	2018

ABSTRACTS

1. Molecular Detection And Sequencing For S1 Glycoprotein Gene Of Bronchitis Virus Of 2016 Epidemic From Sindh And Punjab.

Ahmad Umer Sultan, Muhammad Danish Mehmood, Rameez Hassan, Huma Anwar, Sana Noreen, Faisal Amin, Sajjad Hussain
Abstract.

Infectious Bronchitis (IB) is highly contagious disease of commercial poultry causing substantial economic losses by producing poor quality meat in broilers and effecting production in breeder birds. The causative agent has been reported as most hazardous pathogen among other infectious agent even after being immunized with multi-variant strain vaccine. Currently, different strain such as H-120, 4/91 and D274 have been used extensively for immunoprophylaxis against velogenic strain across Pakistan with minimal protection reported. In current study PCR analysis was used to investigate the molecular nature of IB isolates from Punjab and Sind province of Pakistan in 2016 epidemics. Total of 100 tracheal samples were considered for virus inoculation in 10 days old chicken embryonated eggs. The IBV infected amniotic fluid was neutralized with monoclonal antisera of H-120, 4/91 and D274 strains. The IBV screened samples were subjected



for RNA extraction and subsequent to PCR using type specific primer of each strain. The amplified product of 840 bp was sequenced through Sanger sequencing. On the basis of PCR results, four similar amplified products from both regions were obtained showing similarities in agarose gel electrophoresis, but they differ from each other on the basis of nucleotides sequence. Phylogenetic analysis revealed that nucleotide sequences of isolates from Karachi were similar to the IBV H-120, Mass-41 and Connecticut 46 reference strains. Whereas, isolates from the Punjab province are analogous to the Mans-2, Mans-3, 9/41(UK) but did not show significant similarity with other reference strain. Therefore, it is recommended that use of M-41 and H-120 in vaccine production could be effective measure against velogenic infectious agent in Sindh particularly in Karachi, whereas, it would be better to incorporate either of the variant GQ281656.1, AY279533.1 in vaccine because of their highest level of resemblance with genetically sequenced isolates from Lahore and its surroundings.

2. In Vivo Anti-Viral Effect Of *Melaleuca Alternifolia* (Tea Tree Oil) And *Olea Europaea* (Olive Leaf Extract) On Vero Cell Adaptedavian Influenza Virus

Muhammad Danish Mehmood, Huma Anwar, Sana Noreen, Faisal Ameen, Saud-ul-Hassan, Sajjad Hussain

Abstract:

Viral problems have been in focused of the scientists due to their high metabolic rate, drug resistance and unique nature of pathological mechanism. The current study was under taken to evaluate the in vivo antiviral potential of purified extracts of medicinal plants by means of AIV Haemagglutination (HA) titer in-vivo vero cell line culture. Different doses were interacted with H7N3 (TCID₅₀ = $1 \times 10^{5.6}$, HA= 264 HA unit/ml) and H9N2 (TCID₅₀ = $1 \times 10^{6.2}$, HA= 264 HA unit/ml) strain of the AIV in 90% saturated vero cell line It is evaluated that 10ul of *Melaleuca alternifolia* (TTO) and *Olea europaea* (OLE) pre-treated vero cell line showed significant anti influenza H7N3 (0.5+43) and H9N2 (1.0+45) mean HA titer as compare to non-medicated control group (320.0±128.0) inoculated with the same dose of AIV provided with same conditions. Thus, maximum dose (10µl) of *Melaleuca alternifolia* showed significantly similar antiviral mean HA titer (0.5±1.0) ($p < 0.05$) response on vero cell infected H7N3-AIV when treated 24 hour, but significantly higher than *Olea europaea* (3.0±1.1) in terms of mean HA titer. However, either of the selected plants did not show any cell toxicity by means of cytopathic effect (CPE) and Haemagglutination activity alone or synergistically even at 100µg/ml. Results based on the current study would suggest the use of *Melaleuca alternifolia* and *Olea europaea* in poultry as prevention may help to control AIV outbreaks.

3. Antiviral activity and its prospective mechanism of action on newcastle disease virus using crude extract of four medicinal plants.

Muhammad danish mehmoor, ejaz rafique, huma anwar, sana noreen, mehreen gul and sajad hussain

Abstract:

Viral problems have been in focused of the scientists due to their high metabolic rate, drug resistance and unique nature of pathological mechanism. The failure of novel synthetic allopathic antiviral drugs impels the scientists to investigate other sources of alternative antiviral agents. Herbal extracts has various inhibitory effects against avian viruses particularly Newcastle disease virus (NDV). The current study was under taken to evaluate in-vitro NDV potential of crude extracts of different medicinal plants by means of Haemagglutination (HA) titer in vero cell line culture. To study the potential NDV activity, Vero cell line were treated with different doses (25µl, 50µl, 75µl and 100µl) of solvent free crude extracts of *G. glabra*, *P. emblica*, *A. sativum* and *A. indica* and interacted with 1000 TCID₅₀ of the lasota virus during infection at different time periods. Minimum dose (25µg/ml) of crude ethanolic extract of *A. indica* when inoculated on to vero cell line before NDV exposure showed significantly higher (48 HA titer) anti-NDV response as compare to *A. sativum* (64 HA), but significantly lower than *P. emblica* (20 HA titer). Similarly, minimum dose (25µg/ml) of crude ethanolic extract of *P. emblica* after one hour post NDV exposure showed significantly higher (Mean NDV HA Titer 24) anti-NDV response as compare to *A. indica* (48 HA Titer) and glycyrrhiza, but similar to *A. sativum* (48 HA). Whereas, maximum dose (100µg/ml) of *P. emblica* showed significantly similar antiviral (3HA) response on vero cell infected NDV when treated one hour before with *A. indica*, *A. sativum* (3 HA), but significantly higher than *G. glabra* (128 HA) in terms of mean HA titer. Maximum tested dose (100µg/ml) of *P. emblica* post one hour NDV exposure showed significantly similar antiviral (2.5 HA) response on vero cell to *A. indica* (6HA) and *A. sativum* (4 HA), but significantly higher than *G. glabra* (192). However, either of the selected plants did not show any cell toxicity by means of cytopathic effect (CPE) and Haemagglutination activity alone or synergistically even at 100µg/ml. It is evaluated that simultaneous treatment (25µg/ml) with crude extract of *A. sativum* to expose NDV (EID₅₀=10⁵) in 90% saturated vero cell line showed substianal antiNDV activity.

4. Development and Optimization of Virus Neutralization Test in Chicken Embryonated Eggs for Indirect Identification of Avian Influenza And Newcastle Disease Virus.

Huma anwar, muhammad danish mahmoor, sana noreen, muhammad ismail, sajjad hussain

Abstract:

Avian viral problems have been consistently reported in commercial poultry of Pakistan causing heavy economic losses to the poultry farmers. Authentic idenfication and confirmation of the causative agent is always been question mark for the selection of vaccinal strain in this regard. Current study was therefore undertaken to optimize the virus neutralization test for the serological survey of vaccinated poultry particularly for avian influenza virus's subtypes and



Newcastle disease virus. Various physiochemical factors such as concentration of antigen and antibody, Incubation temperature and incubation period for in vitro and in-vivo reaction of antigen and antibody were optimized in chicken embryonated eggs. Serum samples were obtained from vaccinated breeder birds of five commercial poultry breeder companies and subjected for VNT using different concentration of three antigen and their respective homologous antibodies under optimized conditions. AIV H9 (EID₅₀-1×10^{9.0}/ml) and NDV (EID₅₀-1×10^{8.2}/ml) having biological titer of 10⁻⁷ /50ul HA units were neutralize with 10⁻²/50ul HIU of antibody and incubated at 37°C for 30 minutes was injected subsequently into 10 day old chicken embryo followed by incubation at 37°C for 38 hours showed ≥90% neutralizing specificity. Furthermore, sera obtained from five AIV-H9, AIV-H5 and NDV exposed commercial poultry farms revealed that Big bird broiler, Big bird breeders and A&S chicks are 100% sensitive and specific whereas, Gateway chicks and Waqas poultry breeders showed 100% homology for AIV-H5 virus but do not confers similarity with prevailing AIV-H9 and NDV field strains. Therefore, high sensitivity, reproducibility and specificity VNT, it could be a tool for indirect detection of homology between vaccinal strain and wild virus antigen using known antisera. Particularly, for those organisms possess natural ability to mutate in the adverse climatic conditions.

5. Detection of Influenza a Virus Contamination in Newcastle Disease Live Virus Vaccines and their Pathological Effects on Visceral Organs.

Munir Hussain, Muhammad Saeed Imran, Asim Aslam , Nafeesa Yasmeen, Sajjad Hussain , Shahzad Munir , Mubashir Hussain, Ahmad Kamran Khan, Muhammad Usman, Abdullah Jalal, Muhammad Ameen Jamal, Mir Zulqarnain Talpur , Irfan Ahmed, Zulqarnain Baloch,

Abstract:

To investigate this perception that the NDV live virus vaccine could be the source of Avian Influenza A virus (H9) contamination. Sixteen samples of ND live virus vaccines were purchased from the local market. Prior to use in birds, the samples were tested for Avian Influenza A virus contamination through RT-PCR and used in live birds for any gross pathology and histopathology changes. All the samples were negative against Avian Influenza A virus. Furthermore, these vaccines were also used in broiler and desi chicks at day 5 and day 21 through eye drop and drinking water route respectively. Then, these birds were slaughtered at day 10, 20, 30 and 40 for any gross pathological and histo-pathological changes against Avian Influenza (H9). There were no macroscopic and microscopic lesions observed in visceral organs like trachea, lungs, liver and spleen for Avian Influenza. The results of the study using RT-PCR indicated that the ND live virus vaccine both (local and imported) was free of Avian Influenza A virus (H9). There was a perception among some technical persons that some Avian Influenza outbreaks in the field might be through the source of Newcastle Disease live virus vaccine. This theory regarding contamination of Avian Influenza A virus in Newcastle Disease



live virus vaccines found to be wrong on the basis of this study and these commercial vaccines placed in the market are safe to use against Newcastle Disease and are not source of Avian Influenza outbreaks in the field.

6. An extensive review of experimental ochratoxicosis in poultry: ii. Hemato-biochemical and immunological alterations along with other health issues.

A khatoon, z ul abidin - toxin reviews, 2019

Abstract:

Ochratoxins, primarily produced by certain species of *Aspergillus* and *Penicillium* are considered to be important threats for the health of poultry chicks. These fungal species are present as storage fungi within the stored feed ingredients and when they get proper growth requirements like aerobic condition coupled with high moisture contents lead to the production of ochratoxins as their secondary metabolites. In this second review of the series “An extensive review of experimental ochratoxicosis in poultry” OTA associated alterations in hematological, serum biochemical and immunological parameters have been addressed along with certain other important health issues associated with it in poultry.

7. Confirmation of rabies infection by mouse inoculation test and reverse transcriptase polymerase chain reaction in suspected samples of cow and mule.

Zain ul Abidin, Aisha Khatoon, Abdul Whab Manzoor, Nida Arooj, Sajjad Ali and Muhammad Numan

Abstract:

Rabies, a Lyssavirus infection of Rhabdoviridae family, is a potential neurotropic disorder affecting all mammals and humans. This infection spreads through biting of infected and/or carrier animals to healthy ones including humans. Incubation period of this infection is quite variable ranging from a few days which can last up to one year in few cases. This case report presents the diagnosis and screening of suspected rabies samples of cow and mule. Clear behavioral changes along with paralysis of tail and hind legs were noticed in the mice of both groups between 11-15 days post inoculation while all the mice were found dead between 15-18 days post inoculation in mouse inoculation test (MIT). Reverse transcriptase-polymerase chain reaction (RT-PCR) performed for both samples gave a product of 443-bp amplifying the highly conserved “N”-region gene of virus confirming the rabies infection in both cases.

8. An extensive review of experimental ochratoxicosis in poultry: i. Growth and production parameters along with histopathological alterations.

A.KHATOON & Z.ABIDIN

Abstract



The presence of certain mycotoxins within poultry feed has a negative impact upon the growth and the quality of the final product in the form of meat and eggs. More than 300 different chemically diverse mycotoxins have been identified, but ochratoxins and aflatoxins are considered to be most harmful to the poultry industry. Ochratoxin, more importantly ochratoxin A (OTA) is produced by different species of *Aspergillus* and *Penicillium* spp. which are present as storage fungi within the stored grains and feed ingredients. Body weight gain has been found to decrease in a dose dependent manner when infected at rates of 0.5-29.4 mg/kg for 7-60 days in different experimental studies. Decreased feed intake has been observed at levels of 0.5-4 mg/kg OTA fed for 21-60 days, while egg production, hatchability, eggshell thickness and egg mass production is severely affected when 0.5-4 mg/kg OTA in feed was fed for 28-84 days. However, 0.5-20 mg/kg OTA given for between two and 10 weeks of age was sufficient to produce histopathological alterations in the liver, kidney, thymus, bursa of Fabricius, spleen, lungs and heart. The research shows that OTA adversely affects every organ in birds and, in the following review, OTA associated alterations in growth parameters, production performance and histopathological disturbances of different body organs are discussed.

9. Mycotoxicosis: Diagnosis, Prevention And Control: Past Practices And Future Perspectives

A Khatoon, Z Abidin - Toxin Reviews, 2018

Abstract

Mycotoxins pose severe health hazards in animals, humans, and poultry birds. More than 400 chemically different mycotoxins have been identified to date. Twenty-five percent of world's crops are potentially contaminated with mycotoxins. Luckily, nature has provided the ruminants a unique property of inactivating and detoxifying most of the mycotoxins with the help of microflora and microfauna present within their ruminal fluid; however, unfortunately avian species lack such ability putting them at high risk to the deleterious effects of mycotoxins. This review elaborates different strategies for diagnosis, prevention and control of mycotoxins.

10. Dietary L-Carnitine And Vitamin E; A Strategy To Combat Ochratoxin-A Induced Immunosuppression,

Abstract

This study aimed to evaluate the effect of dietary ochratoxin A (OA), in the presence and absence of L-carnitine (LC) and vitamin E (VE), on the humoral immune responses of White Leghorn cockerels (WLC). One-day old white male Leghorn chicks were divided into 12 groups, having 20 birds each and were offered ration contaminated with OA (1.0 or 2.0 mg/kg feed) alone and concurrently with LC (1.0 g/kg) and/or VE (0.2 g/kg), for 42 days. The humoral immune responses were accessed by lymphoproliferative response to avian tuberculin, in-vivo phagosomes activity to carbon particles and antibody response to the sheep red blood cells (SRBCs). The dietary addition of OA alone suppressed the humoral immune responses, however, the exposure of birds to 1.0 mg/kg OA in the presence of LC

and/or VE showed a significant reduction in OA induced immunotoxicity. This protective response was absent in the birds fed 2.0mg/kg OA in the presence and absence of LC and/or VE. Histopathological and morphometric examination of the bursa of Fabricius exhibited a decrease in the severity and frequency of OA induced lesions in the presence of dietary LC and/or VE. The use of LC and VE as dietary supplement, can effectively overcome OA (≤ 1.0 mg/kg) induced immunosuppression.

11. Effects Of Feeding Bentonite Clay Upon Ochratoxin A Induced Immunosuppression In Broiler Chicks.

A Khatoon, MZ Khan, Z Abidin... - Food Additives & ..., 2018

Abstract

A presence of mycotoxins in feed is one of the most alarming issues in the poultry feed industry. Ochratoxins, produced by several *Aspergillus* and *Penicillium* species, are important mycotoxin regarding the health status of poultry birds. Ochratoxins are further classified into to several subtypes (A, B, C, etc) depending on their chemical structures, but ochratoxin A (OTA) is considered the most important and toxic. Bentonite clay, belonging to phyllosilicates and formed from weathering of volcanic ashes, has adsorbent ability for several mycotoxins. The present study was designed to study the effects of bentonite clay upon OTA-induced immunosuppression in broiler chicks. For this, 480 day-old broiler chicks were procured from a local hatchery and then different combinations of OTA (0.15, 0.3, or 1.0 mg/kg) and bentonite clay (5, 10, and 20 g/kg) were incorporated into their feed. At 13, 30, and 42 days of age, parameters such as antibody responses to sheep red blood cells, in situ lymphoproliferative responses to mitogen (PHA-P), and in situ phagocytic activity (i.e., via carbon clearance) were determined respectively. The results indicated there was a significant reduction of total antibody and immunoglobulin titres, lymphoproliferative responses, and phagocytic potential in OTA-treated birds, suggesting clear immunosuppression by OTA in birds in a dose-dependent manner. These results were also significantly lower in all combination groups (OTA with bentonite clay), suggesting few to no effects of feeding bentonite clay upon OTA- induced alterations in different immune parameters.

12. An Insight Into The Ecobiology, Vector Significance And Control Of Hyalomma Ticks (Acari: Ixodidae): A Review

Sajid, M. S., A. Kausar, A. Iqbal, H. Abbas, Z. Iqbal, and M. K. Jones

Abstract

Ticks (Acari:Ixodoidea) are important ectoparasites infesting livestock and human populations around the globe. Ticks can cause damage directly by affecting the site of infestation, or indirectly as vectors of a wide range of protozoa, bacteria and viruses which ultimately lead to lowered productivity of livestock populations. *Hyalomma* is a genus of hard ticks, having more than 30 species well-adapted to hot, humid and cold climates. Habitat diversity, vector ability, and emerging problem of acaricidal resistance in enzootic regions typify this genus in various countries around the world. This paper reviews the epidemiology,

associated risk factors (temperature, climate, age, sex, breed etc.), vector role, vector-pathogen association, and reported control strategies of genus *Hyalomma*. The various proteins in saliva of *Hyalomma* secreted into the blood stream of host and the prolonged attachment are responsible for the successful engorgement of female ticks in spite of host immune defense system. The various immunological approaches that have been tried by researchers in order to cause tick rejection are also discussed. In addition, the novel biological control approaches involving the use of entomopathogenic nematodes and *Bacillus thuringiensis* (*B. thuringiensis*) serovar *thuringiensis* H14; an endotoxin, for their acaricidal effect on different species and life cycle stages of *Hyalomma* are also presented.

13. Bacterial Count And Predisposing Factors Of Clostridium Perfringens (Cpa Gene) Infection Alongwith Antimicrobial Sensitivity In Diarrheic Sheep In Pakistan

Hussain, K., M. Ijaz, A. Z. Durrani, A. A. Anjum, A. A. Nasir, S. H. Farooqi, A. I. Aqib, and A. S. Ahmad

Abstract

Clostridium perfringens (*C. perfringens*) is a normal inhabitant in the gut of animals. It may proliferate rapidly in favorable conditions and produces lethal toxins. These toxins may cause lethal effects in the intestines and systemically it may cause enterotoxaemia. In disease conditions, the presence of *C. perfringens* CFU/g in fecal sample can be of diagnostic value. This study aims to determine the bacterial counts and predisposing factors of *C. perfringens* (targeting CPA gene) infection in addition to an in-vitro antimicrobial trial in entero-toxic sheep in Pakistan. A total of 192 diarrheic sheep irrespective of age, gender and breed were selected and the CFU/g was determined from the fecal samples. The study showed that 34.9% of the samples had elevated level of bacterial count compared to the normal (10⁴-10⁷ CFU/g). Out of the total, 7.8% of the samples had subnormal bacterial count (CFU/g), while, 57.3% of the samples showed bacterial counts in the normal ranges. The confirmation of selectively isolated *C. perfringens* was done by amplification of 324bp CPA gene fragment using polymerase chain reaction (PCR). The in-vitro antimicrobial sensitivity trials showed that penicillin, ciprofloxacin and ceftriaxone are 100% efficacious against *C. perfringens*, while, bacitracin, ampicillin and amoxicillin were found to be least effective. The key determinants in this study which support the in-vivo growths of *C. perfringens* were; carbohydrate rich diet and overcrowding with the odds ratios (OR) of 5.44 and 2.26, respectively. This study concludes that *C. perfringens* is highly prevalent in sheep population of Pakistan. The incidence of enterotoxaemia can be minimized by controlling the factors which enhance its in-vivo growth. The diseased animal associated with elevated *C. perfringens* levels can be effectively cured using any one of the penicillin, ciprofloxacin and ceftriaxone.



14. Investigation Of Different Serotypes Of Fmd In Vaccinated Buffaloes In Southern Areas Of Punjab Province

Riaz Hussain, Fazal Mahmood, Bilal Aslam, Abu Baker Siddique, Azhar Rafique, Syed Abdul Khaliq, Iahtasham Khan, Sadaf Imran, Mansoor Mubeen, Jahanzaib and Azam Ali Nasir

Abstract :

Foot and mouth disease (FMD) is highly endemic in Pakistan, which induces heavy economic loss to livestock holders in term of decrease milk production, high morbidity and mortality in large and small ruminants. In present study, we observed the clinico-pathological lesions during an outbreak of FMD in vaccinated buffaloes. Overall 31.56% morbidity (119/377), 4.77% mortality (18/377) and 15.12% case fatality (18/119) was recorded during the current outbreak. The morbid animals were lethargic, depressed and exhibited marked lameness, profuse salivation, myositis of tongue, vesicular fluid and epithelial sloughing. Vesicular fluid and epithelial sloughing collected from clinically sick animals were positive for FMDV predominantly O strain and Asia I through ELISA technique. Grossly, mandibular lymph nodes were swollen and hemorrhagic in infected animals. The mucosa of abomasum was severely congested, ulcerated and showed hyperemic edges with centrally yellow necrotic areas. Severe purulent inflammation of jejunum and petechial hemorrhages over base of heart were also observed. Histologically degenerative changes in keratinocytes in stratum spinosum, marked acanthosis and intracellular accumulation of eosinophilic, a cellular, transudate forming characteristic vesicles and bullae were observed in epidermis. The mandibular lymph nodes exhibited prominent capillaries engorged with erythrocytes, necrosis and dendritic cells with engulfed necrotic cells. Hemosiderin accumulation was also observed. Fusion and stunting of intestinal villi along with infiltration of inflammatory cells were characteristic lesions in infected animals. From the findings of our study it can be suggested that clinical signs, necropsy lesions and histopathological changes are valuable and useful tools for the diagnosis of foot and mouth disease in *Bubalus bubalis*. Moreover, strict surveillance, use of good quality vaccines, regular monitoring and geographical distribution of various serotypes of FMDV are valuable tool for establishment of effective control procedures.

15. A novel approach for development, standardization, and safety testing of enriched alum-precipitated vaccine against hemorrhagic septicemia in different breeds of cattle.

Farooq MZ, Sattar S, Afroz H, Rasool A

Abstract:

Hemorrhagic septicemia is a fatal disease of cattle and buffalo all over the world including Pakistan and it causes heavy economic losses every year. The poor farmers cannot bear this loss in the form of less milk production and heavy expenditures on the animal treatment. An enriched alum-precipitated vaccine with reduced dose was prepared and



standardized and safety testing of enriched vaccine was performed in Swiss albino mice as well as in natural host. In this experiment, a total of 36 cattle both male and female of different age groups ranging from 4 months to 4 years were used. All these animals belong to all major cattle breeds of Pakistan including Sahiwal, Red Sindhi (crossbred and purebred), Dhani (crossbred and purebred), Lohani (crossbred and purebred), and Cholistani, and exotic breeds including

Holstein Friesian and Jersey. These animals were examined for current immune titer prior to vaccination. Animals were vaccinated subcutaneously with 2 mL and 4 mL dose of new vaccine and were observed for any untoward reaction for 48 h. All the animals were kept under close observation for the next 30 days and all were found safe. The experiment was designed to reduce the dose of the vaccine to 2 mL by using BHI as a growth medium, as well as to increase the number of doses prepared in the same infrastructure, hence reducing the cost of vaccine production. The study proved that vaccine with increased biomass in reduced dose is safe in local as well as in exotic breeds of cattle.



Internship/ Educational Trainings (Number Of Students)

- *GCU, Faisalabad* *B.S(Micro) 04*
- *UVAS, Lahore* *B.S(Micro) 05*
- *UVAS, Lahore* *DVM 09*
- *Islamia University* *DVM 04*
of Bahawalpur
- *Bahauddin Zakariya* *DVM 02*
University, Multan
- *University of Agriculture* *DVM 03*
Faisalabad
- *PMAS, ARID, Agriculture* *DVM 08*
University, Rawalpindi
- *Lahore Garrison* *MSc 07*
Zoology

Total =42

Capacity Building of Officer

Sr. No.	Name of Officers	Designation	Title of Training/ workshop/ Short Course	Venue of Training
1.	Dr. Sajjad Hussain	APVO	Project Management	PPMI, Islamabad
			Promotion link training (Mandatory training for Promotion BS-19 to BS-20)	UVAS Lahore
			Workshop on (How to include standard quality and specification in public procurement)	PC Hotel, Lahore
2.	Dr. Muhammad Anees	APVO	Human Resource Management	MPDD Lahore
3.	Dr. Sarwat Naz	APVO	Effective Office Management	MPDD Lahore
			Decision Making	MPDD Lahore
4.	Dr. Azam Ali Nasir	APVO	PEEDA Act 2006	MPDD Lahore
			Leadership skills	MPDD Lahore
			Crises Management	MPDD Lahore
5.	Dr. Asfa Rasool Bhatti	APVO	Communication skills	MPDD Lahore
6.	Dr. Muhammad Asim	SVO	Human Resource Management in Public Sector	PPMI Islamabad
			Managing Legal Affairs/issues in Government Department	PPMI Islamabad
			Human Resource Management	MPDD Lahore
			8 weeks mandatory training	UVAS, Lahore
			3 rd PPR vaccine producer workshop	Amman, Jordan
7.	Dr. Zain ul Abidin	SVO	Project Management	MPDD Lahore
			PEEDA Act	MPDD Lahore



8.	Dr. Hina Afroz	SVO	8 weeks mandatory training	UVAS, Lahore
9.	Dr. Sajjad Ali	SVO	Financial Management Skills	MPDD Lahore
			Brucella Vaccine Production hands on training	Istanbul Turkey
10.	Dr. Sobia Aamir Chughtai	SVO	Preparation of PC-I	PPMI Islamabad
11.	Dr. Aqsa Mushtaq	SVO	Preparation of PC-I	PPMI Islamabad
			8 weeks mandatory training	UVAS, Lahore
12.	Dr. Waseem Shahzad	SVO	Result Based Management	MPDD Lahore
13.	Dr. M. Najji Ullah Khan	SVO	Procurement Rules	MPDD Lahore
			Communication skill	MPDD Lahore
			Financial and Audit Management in Public Sector	PPMI Islamabad
14	Dr. Muhammad Usman Ashraf	VO	Capacity building of laboratory staff	NVL Islamabad
			Preparation of PC-I	PPMI Islamabad
			MS Project	PPMI Islamabad
15	Hafiz Muhammad Waqar Ahmad	VO	Advance IT skills	MPDD Lahore
16	Dr. Saqib Tanveer	VO	Public Procurement Policies and PPRA Rules	PPMI Islamabad
17	Dr. M Shahzad Qadir	VO	Brucella Vaccine Production hands on training	Istanbul Turkey
18	Dr. Nofil Mustafa Javaid	VO	Information Technology	MPDD Lahore
19	Dr. Iqra Zafar	VO	8 th Chinese Language Course	MPDD Lahore
20	Dr. Muhammad Ali Qureshi	VO	Environmental Impact Assessment for development projects	NCRD Islamabad
21	Dr. Muhammad Amjad Iqbal	VO	PC-I	MPDD Lahore



Pakistan National Accreditation Council

Ministry of Science & Technology
Government of Pakistan
Islamabad



Certificate of Accreditation

is awarded to

**Veterinary Research Institute Quality Control Laboratory
Zarar Shaheed Road, Lahore, Pakistan**

in accordance with the requirements of ISO/IEC 17025:2005
The accreditation is subject to regular surveillance and compliance
to the requirements of PNAC.
For scope of accreditation, see appendix.

Accreditation Certificate Number: LAB 117

Date of Issue:
23-12-2016


Director General

Valid until
22-12-2019



ISO 9001: 2015 CERTIFICATION OF LABS



QSP-09/F-03

Certificate of Conformity

This is to certify that the Management System of

M/s Veterinary Research Institute

Located at:

Veterinary Research Institute, Zarrar Shaheed Road, Lahore Cantt

has been assessed and certified against the requirements of

Quality Management System ISO 9001:2015

Certified Scope

Production of Veterinary Biologics (Veterinary Vaccines & Antigens)

03-04-2018
Certified Since

03-04-2019
Issued on

02-04-2020
Valid till

Q – 03041950
Certificate Number

Authorized by

Shafqat Iqbal
Chief Executive Officer



**Certification Services
Pakistan (Pvt) Ltd.**

Certifications, Trainings, Inspections



1st May 2018

Company Address: NIE Complex, NECOP Building 1st Floor, Plot No. 17, Street No. 06, H-9/I, Islamabad, Pakistan
Telephone #: 92-51-8438844-5 Fax #: 92-51-4865360, 92-51-8357207, Email: info@cesp.com.pk, Web: www.cesp.com.pk



Annual Development Project during 2018-19

In process

Name of Project:-

Establishment of State of the art Labs at Veterinary Research Institute and Foot & Mouth Disease Research Center to meet the International Standards of Biologics Production and Research & Development

Gestation Period:- 36 Months (2017-2020)

Budget Of project:- (2018-2019)

Year	Release (in Millions)	Expenditure (in Millions)
2018-2019	123.375	121.284

Objectives of project: (2018-19)

1. Automation in Filling and labeling of live vaccines.
2. Foreign training of two officers on Brucella Vaccine production from Turkey.
3. Complete replacement of water and sewerage lines.
4. Renovation of Mycoplasma, Poultry Vaccines Section and Media Sections buildings.
5. Renovation of the Main Roads of VRI.
6. Installation of HVAC System in Cell Culture Section.

REVAMPING OF BUILDINGS

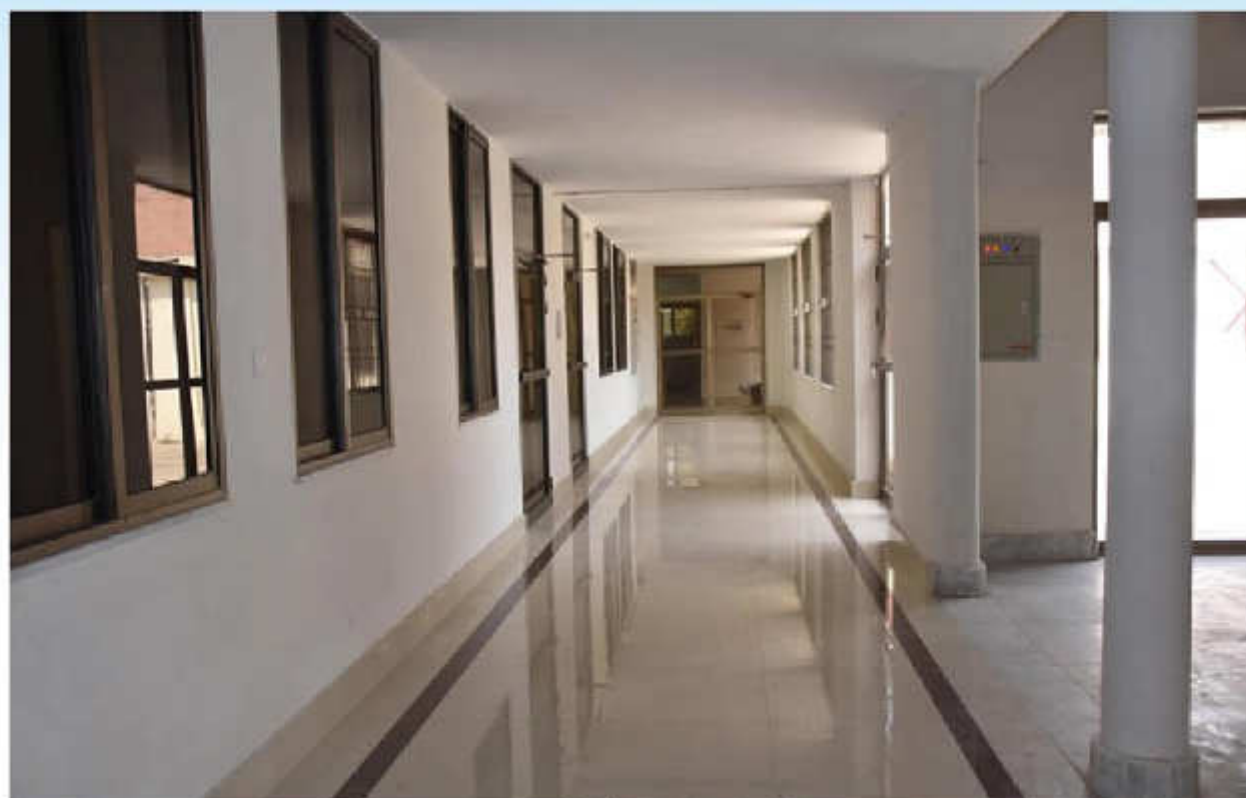


Renovation of Cell Culture Section Building

RECONSTRUCTION & MAINTENANCE OF ROADS



RECONSTRUCTION OF MEDIA, NDV CORRIDOR



REFURBISHMENT OF LABS



Renovation of Media Preparation Room



Renovation of Mycoplasma Section



Renovation of Cell Culture Section Lab



Renovation of NDV Section Lab

DAY CARE CENTRE AT VRI, LAHORE

Took initiative to empower the women



Women in Pakistan are playing important role for prosperity in the Country due to enhance opportunity and enabling environment. The share of the Women in labour market is increasing day by day. Keeping in view enhance participation Government OF Punjab decided to facilitate working Women by establishing Day Care Centers in the respective Organizations. Women in Veterinary Research Institute Lahore are serving since 1963. It was very unfortunate for this institute that there was no day care center facility for the women working in the institute. The initiative taken by the government enlightened the idea of development and operationalization of a Day care centre at VRI Lahore as 40 no. of women serving the institute. The administration of VRI Lahore decided to establish a day care Centre at the premises of Veterinary Research Institute, Lahore with the collaboration of Government of The Punjab.

In April 2019 with the efforts of administration of L&DD department VRI, Lahore got the success of establishing a fully funded Day Care Centre for the betterment and efficient utilization of women workers serving the institute.

PRIME MINISTER'S CLEAN AND GREEN PAKISTAN

Under Prime Minister Clean and Green Pakistan Initiative, 836 new trees and 523 plants were planted at VRI, Lahore premises.



Tree Plantation by advisor to CM Mr. Faisal Hayyat and Secretary L&DD Punjab

DELEGATION VISITS



Visit of Mr Opal to Veterinary Research Institute Lahore



Sectional visit of Minister Livestock and Adviser To Chief Minister





VRI stall in Mela Movashiyaan 2018 at BRI, Pattoki.

Way Forward/Research Priority Areas

- 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 2019-20:

Sr. No.	Name of Vaccine	Proposed Target 2019-20 (millions)
1.	HSV	35.00
2.	BQV	15.00
3.	ETV	36.50
4.	CPPV	14.00
5.	PPR	21.77
6.	NDV Lasota	105.00
7.	Anthrax Spore Vaccine	As per demand
8.	SPV	As per demand
9.	GPV	As per demand
10.	ND+H9 (OIL Based)	As per demand
11.	Fowl Pox	As per demand