



Joint FAO/IAEA Programme  
Nuclear Techniques in Food and Agriculture

# Animal Production & Health Newsletter



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## International Symposium on Sustainable Animal Production and Health Current Status and Way Forward

22-26 June 2020, Vienna, Austria

[www.iaea.org/events/aphs2020](http://www.iaea.org/events/aphs2020)





## To Our Readers

Dear colleagues,

The first six months of this year have been a busy time for all personnel in the subprogramme. Apart from our regular Coordinated Research Projects (CRP) activities and our technical support given to on-going national and regional Technical Cooperation (TC) projects, we were also involved in the initiation (together with TC Country Officers) of the 2020–2021 biennial TC project cycle. In addition, when carrying out our 2018–2019 mid-year programmatic performance evaluations, we could identify the areas where good performance was achieved as well as areas where further improvements are needed. We hoped our inputs will serve the best interests of our Member States for the present programme cycle 2018–2019.

In response to many requests from our readers, I have decided to give a brief overview of our subprogramme as background. The focus of the Animal Production and Health Subprogramme activities is on enhancing food security by supporting sustainable livestock production systems in developing countries. This is to be achieved by strategic and applied research, technology transfer and capacity building. The three principal components of the subprogramme are animal nutrition, animal reproduction and breeding and animal health. Problems are identified and solutions developed through the use of strategically applied nuclear-based tools, in conjunction with conventional technologies to:

- characterize and optimally utilize locally available feed and feed resources to enhance maximum energy conversion, whilst protecting the environment and minimizing methane and CO<sup>2</sup> emissions;

- enhance animal reproduction and breeding through the introduction of artificial insemination, embryo transfer and productive breed selection, and the characterization of livestock genetic make-up to drive the integration of locally adapted animal breeds with trait selected exotic breeds to satisfy the increasing demand for more and better-quality animals and animal products;
- assess and reduce the risk of transboundary animal and zoonotic diseases to livestock through the implementation of early and rapid diagnosis and control technologies and their use in national and international control and eradication programmes.

The above activities are complemented by tools developed for computerized data management in disease diagnosis and animal production; use of geographic information systems in management of farm resources and diseases; and distance learning through information communication technologies in the related areas. The FAO/IAEA Veterinary Diagnostic Laboratory (VETLAB) Network is instrumental to the development, validation and dissemination of technologies, know-how and expertise worldwide. Please note the ‘Sustainable Animal Production and Health’ symposium that will take place from 20 to 26 June 2020 at the IAEA’s Headquarters in Vienna, Austria.



Gerrit Viljoen  
Head, Animal Production and Health Section



APH team at the APH Laboratory in Seibersdorf, Austria

# International Symposium on Sustainable Animal Production and Health: Current Status and Way Forward

## 22 to 26 June 2020

### Background

Systems of livestock production in developing countries are becoming progressively more intensified as producers and traders respond to increasing demands from consumers in urbanized societies for milk, meat, other livestock products and animals. This includes the challenges of increasing productivity without degrading feed and genetic resources, and of ensuring that diseases of a transboundary or zoonotic nature are early recognized and brought under control. Increasing demand can only be met through the selection of animals that produce more meat and milk and show disease resistance and heat tolerance; the optimal utilization of local resources that simultaneously protects animal biodiversity and the environment; and the protection of animals and their caretakers from diseases.

It is necessary to assess and manage the risks and the opportunities arising from intensification and to control emerging and re-emerging animal and zoonotic diseases to minimize adverse effects on farmers' livelihoods. In turn, this requires developing capacities to adapt and foster the application of the appropriate production and protection enhancing technologies, as well as sound and mutually supportive policies.

The symposium will draw on lessons learned and current best practices to provide a roadmap for the sustainable improvement of animal production whilst protecting the environment. The focus of the symposium will be on the contributions and impact of nuclear technologies and applications.

### Objectives

The objectives of the symposium are to provide information and share knowledge on modern and novel technologies in animal production and health, and their application to support sustainable livestock production systems; identify capacity and research needs and address gaps and new opportunities for the effective transfer of nuclear and nuclear derived/related technologies; and build capacities for ameliorating or solving factors that are limiting livestock productivity and causing animal diseases.

### Topics

- The use and application of atomic, nuclear, and nuclear derived/related technologies in animal nutrition, reproduction, breeding and health
- Feed resources and feeding strategies under grazing and cut-and-carry production systems in developing countries
- Assisted reproductive technologies for enhanced livestock productivity
- Radiation hybrid mapping and gene-marker selection in animal characterization and breeding programmes
- Early and rapid animal disease diagnostic tests in national and decentralized laboratories
- Threats to food security, public health and international trade due to emerging and re-emerging transboundary and zoonotic animal diseases
- The role of the FAO/IAEA Veterinary Diagnostic Laboratory (VETLAB) Network in capacity building, transfer of technology, and sharing of knowledge
- Irradiated vaccines
- The use of stable isotopes to trace and monitor migratory animals and their role in the introduction of transboundary animal and zoonotic diseases

### Programme Structure

The symposium will be held in English. The programme will comprise keynote addresses, plenary presentations by invited speakers, satellite meetings on selected topics, and group discussions.

### Target Audience

The symposium welcomes scientists, academics, research managers, laboratory personnel, policy-makers from governmental and non-governmental organizations, private sector actors and representatives of international and national donor agencies, foundations and trusts.

**More information:** [www.iaea.org/events/aphs2020](http://www.iaea.org/events/aphs2020)

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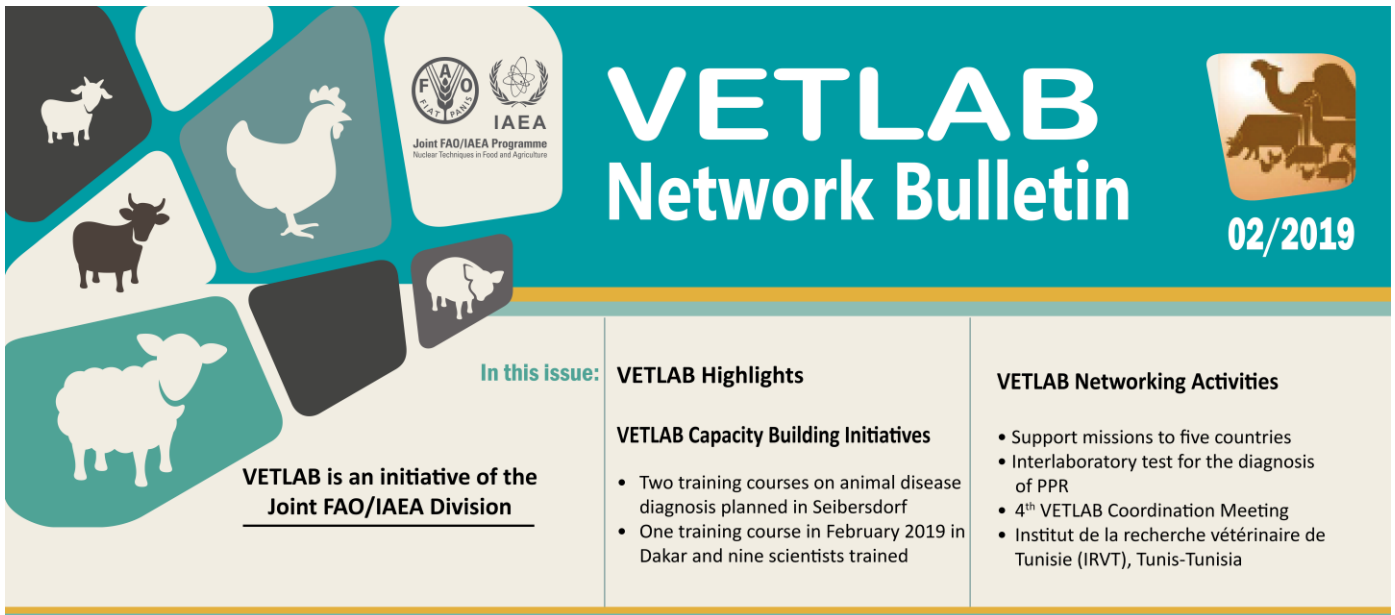


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# VETLAB Network Bulletin

02/2019

**VETLAB is an initiative of the Joint FAO/IAEA Division**

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## To the readers

In the past six months the VETLAB network has supported partner laboratories facing various challenges. African Swine Fever (ASF) spread in East Asia causing severe losses. The efforts made by the VETLAB Network to create preparedness and strengthen diagnostic capacities among countries at-risk has significantly contributed to the rapid detection and response of countries recently affected. For example, before the ASF spread in Asia, two training courses targeting VETLAB partner laboratories addressed ASF detection, genotyping the virus and determining its variants. The participants received validated ASF laboratory procedures and reagents to implement the tests in their laboratories. Furthermore, directors of VETLAB partner laboratories received updated information on the latest developments on ASF diagnosis and characterization enabling them to implement and improve diagnostic strategies in the respective laboratories.

In West and Central Africa, equine infectious diseases such as equine influenza and African Horse Sickness are spreading causing death and losses in horse and donkey populations. Beyond the support in terms of laboratory protocols and reagents, the network platform was used by the laboratories to exchange data and information on the disease situation, the diagnostic approach and tests used and the main challenges.

Highlighted in this bulletin there are also examples of the application of the multiplex PCR-based tests on clinical specimens. These tests can effectively contribute to syndromic surveillance and differential diagnoses of important diseases currently affecting or threatening several member states in Asia and Africa, such as Peste des Petits Ruminants, Contagious Caprine Pleuropneumonia and African Swine Fever.

Looking forward for contributions of your laboratories to the next VETLAB activities!

## VETLAB Highlights

### VETLAB Networking in Action I: Supporting ASF diagnosis in Eastern Asia

Since its emergence in China in August 2018, ASF virus has hit several countries in Asia, causing the destruction of more than 2,000,000 pigs. The VETLAB Network reacted through the emergency delivery of SOPs, reagents, consumables and small equipment to the affected countries and those at higher risk such as Myanmar and Thailand for the rapid implementation of specific laboratory diagnostic tests. Experts field missions are ongoing.

### VETLAB Networking in Action II: Enhanced ASF laboratory preparedness

Through the organization of training, distribution of laboratory procedures and standards, laboratory preparedness to ASF emergence has been enhanced in VETLAB partner laboratories, particularly in countries at-risk and with no previous experience on ASF infection. These efforts enabled countries like Mongolia to rapidly detect, confirm and contain the first introduction of ASF early this year.

### VETLAB Networking in Action III: Facing equine epidemics and severe mortalities in West and Central Africa

Several countries in the region have been confronted with severe cases of respiratory disease and mortality in donkeys and horses. Depending on the countries, cases are associated with epidemics of equine influenza, African Horse Sickness and Strangles. Through the VETLAB network, partners from affected countries are exchanging information and protocols. The network is supporting laboratories in the provision of reagents and controls to rapidly diagnose and confirm cases.

### Pseudocowpox disease in Africa

The application of an HRM assay for the simultaneous detection and differentiation of poxviruses, developed at APHL and transferred to several VETLAB laboratories, revealed the occurrence of distinct infections causing skin lesions and disease in African cattle. BNVL in Botswana, LNRV in Senegal and CVRI in Zambia detected pseudocowpox virus, while bovine papular stomatitis virus was diagnosed by CVL in Mozambique.

### Highly pathogenic avian influenza (H5N8) virus caused high mortality in African penguins (*Spheniscus demersus*) in Namibia

In January 2019, an unusually high mortality of African penguins was reported in Halifax Island, 100 m from the mainland. Samples were processed by the Central Veterinary Laboratory, Windhoek by RT-qPCR and indicated the presence of Highly Pathogenic Avian influenza (HPAI) subtype H5N8. Sequencing services supported by VETLAB enabled the confirmation of the H5N8 infection for the first time in Namibia.



## VETLAB Capacity Building Initiatives

and Quality Control for Molecular Assays (9 - 20 September 2019). The purpose of the event is to strengthen the Member States' veterinary diagnostic and research laboratory capacities in introducing, validating and monitoring assays for routine use. This will facilitate the implementation and maintenance of quality systems in the VETLAB Network partners' laboratories.

Surveillance of Transboundary Animal Diseases (4 - 15 November 2019). The training will focus on the application of rapid multiplex serological and

molecular assays for the differential diagnosis and syndromic surveillance of transboundary animal and zoonotic diseases.

### Training in VETLAB Countries

Training on nuclear derived technologies for animal pathogen detection, instrument calibrations and troubleshooting was held in LNERV, Dakar from 11 to 20 February 2019. Nine participants from LNERV, Dakar and ONARDEL, Mauritania attended the training conducted by an APHL expert.

### Training in Seibersdorf

Training Course on Transboundary Animal Disease Diagnoses: Validation, Implementation, Monitoring

## VETLAB Networking Activities

### Interlaboratory test for the diagnosis of PPR

The APH is organizing the yearly PPR interlaboratory comparison focusing on its serological and PCR-based detection. Participating laboratories in Africa, Asia and Europe should expect to receive the panel in October 2019.

### Support missions

Five VETLAB laboratories (Malaysia, Morocco, Senegal, Tunisia, Vietnam) were visited by APHL staff during the last semester 2019. The visits aimed to strengthen the VETLAB activities and network among the different laboratories as well as transferring novel protocols and technologies.

### 4<sup>th</sup> Coordination Meeting with Directors of Veterinary Laboratories of Africa and Asia

It will take place from 19 to 23 August 2019 in Vienna, Austria. The purpose of this meeting will be to review the achievements for 2018-2019 and formulate new work plans for the participants' respective laboratories. This meeting is a forum for participants to share experience and knowledge, and to identify activities of common interest for enhancing capacity of these veterinary laboratories.

### The VETLAB Network Laboratories:

#### Institut de la recherche vétérinaire de Tunisie (IRVT), Tunisia

IRVT operates under the Ministry of Agriculture, Hydraulic Resources and Fisheries and comprises a Central Laboratory in Tunis and four regional laboratories (Bousalem, Sousse, Sfax and Gabes). The main missions of IRVT are to support the epidemiological surveillance, and the official control of legally notifiable infectious diseases. IRVT is also involved in the official control of foodstuffs and testing for parentage verification and genetic identification of horses, cattle and carnivores.

The regional laboratory in Bousalem mainly analyses milk and feed concentrates. The laboratory in Sousse focuses on the control of foodstuffs and the one in Sfax oversees the quality and safety control of fishery products for export and in the search for marine biotoxins in bivalve molluscs.

As part of the accreditation of some of our laboratories, the institute is deploying efforts to bring the quality management system of the food microbiology laboratory in Tunis and the biotoxins laboratory in Sfax into compliance, for accreditation under ISO 17025.

The virology laboratory in Tunis, which aspires to become an OIE reference laboratory, has an OIE twinning project for the diagnosis of bluetongue and regularly takes part in interlaboratory tests for various viral diseases. In addition,



IRVT, Tunisia hosted the OIE meeting for the laboratory focal point in September 2018. Several VETLAB partners attended the meeting



The staff of the Veterinary Research Institute in Ipoh, Malaysia at the time of APHL expert visit (May 2019)

the laboratory has hosted several training courses on viral diseases of fish and livestock with the support of the Arab Organization for Agricultural Development (AOAD) and received trainees from Arab countries and the Comoros Islands.

As part of the upgrade of its laboratories, the IRVT has set up a Laboratory Information Management System (LIMS) software complying with ISO 17025. This will contribute to the traceability and data quality of IRVT.

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## Forthcoming Events

### **Regional Training Course on Collection, Sorting, Storage and Identification of the Most Important Vectors for Vector Borne Diseases (RER5023)**

Ivancho Naletoski

A regional training course on the collection, sorting, storage and identification of the most important vectors for vector borne diseases will be held in Ankara, Turkey from 1 to 12 July 2019. Thirty-four participants from the Member States of the IAEA are expected to attend the training course.

The course will be led by four international experts, as well as four local experts from the host institution (Hacettepe University, in Ankara, Turkey). The course will be comprised of field exercises in which participants will be divided in groups, according to the group of priority arthropod vectors. During the second week of the training course, a production of a teaching video has been planned.

The video should cover all critical points of the already produced SOPs (Standardized Operating Procedures) for capture and identification of priority vectors. It will be available as an open-source product on the IAEA webpage and should complement the use of the appropriate procedures in the Member States laboratories.

### **Regional Training Course on Genetics of Parasite Resistance in Sheep and Goats: Bioinformatics Analysis of Genomic Data to Assess Population Structure, Genotype-Phenotype Association and Genomic Prediction (RLA5071)**

Mario Garcia Podesta

The training course will take place from 1 to 12 July at the Seibersdorf Laboratory, Austria.

The purpose of the event is to improve the skills on bioinformatics analysis of large sets of genomic data to assess population structure and genetic admixture in livestock; to enhance technical skills in analyzing and interpreting genomic data for association with phenotypes

and genomic prediction related to breeding sheep and goat for parasite resistance; and to build the technical capacity in establishing gene bank of performance recorded animals and improve the application of genomics for breeding local sheep and goat with enhanced host resistance against parasites.

### **Second Research Coordination Meeting on Quantification of Intake and Diet Selection of Ruminants Grazing Heterogeneous Pastures Using Compound Specific Stable Isotopes (D31029)**

Mario Garcia Podesta

The research coordination meeting (RCM) will take place in Piracicaba, Brazil, from 12 to 16 August 2019.

The purpose of the event is to conduct a midterm review of the project, record current achievements and coordinate and finalize the plan of future activities.

### **Fourth Research Coordination Meeting on Early Detection of Transboundary Animal Diseases (TADs) to Facilitate Prevention and Control through a Veterinary Diagnostic Laboratory (VETLAB) Network (D32032)**

Ivancho Naletoski

The research coordination meeting (RCM) will take place from 19 to 23 August 2019 in Vienna, Austria.

Partners of the project will gather to review the results and achievements generated so far through the RCM, namely the produced reference materials, the dissemination of the sequencing services and the use of the iVetNet among counterparts, as well as the results of the testing of the multi pathogen molecular assays disseminated to selected counterparts of the VETLAB Network.

During the same week, the directors of the VETLAB Network will meet in Vienna, to review the activities of the VETLAB Network over the previous year and determine priorities for next year. As usual, the two groups will merge together during the last day of the meetings and exchange information of interest for both groups.



## **Coordination Meeting with Directors of Veterinary Laboratories in Africa and Asia that Are Supported by the African Renaissance Fund (ARF) and the Peaceful Uses Initiative (PUI)**

Charles Lamien and Giovanni Cattoli

The coordination meeting will take place from 19 to 23 August 2019 in Vienna, Austria.

This will be the fourth joint technical meeting of the VETLAB Network with directors of veterinary laboratories in Africa and Asia that are supported by the African Renaissance Fund and the Peaceful Uses Initiative to strengthen animal disease diagnostic capacities.

The purpose of this meeting will be to review the achievements for 2018–2019 and formulate new work plans for the participants' respective laboratories for 2019–2020.

In addition, this meeting will be a forum for participants to share experience and knowledge, and to identify activities of common interest for enhancing capacity of veterinary laboratories in Africa and Asia.

As usual, the meeting will be held in parallel with the forth RCM of the VETLAB CRP D32032 to allow interaction between the laboratory directors and the CRP experts and their critical assessment of the CRP progress.

## **Training Course on Transboundary Animal Disease Diagnoses: Validation, Implementation, Monitoring and Quality Control for Molecular Assays**

Charles Lamien and Giovanni Cattoli

The training course will be held from 9 to 20 September 2019 at the Animal Production and Health Laboratory in Seibersdorf, Austria.

The purpose of the event is to strengthen the Member States' veterinary diagnostic and research laboratory capacities in introducing, validating and monitoring assays for routine use.

This will facilitate the implementation and maintenance of quality systems in the Veterinary Diagnostic Laboratory (VETLAB) Network partners' laboratories.

## **Training Course on the Detection of Multiple Pathogens for the Differential Diagnosis and Syndromic Surveillance of Transboundary Animal Diseases**

Charles Lamien and Giovanni Cattoli

The training course will be held from 4 to 15 November 2019 at the Animal Production and Health Laboratory in Seibersdorf, Austria.

The purpose of the event is to strengthen the capacity of Member States' veterinary laboratories partnering the Veterinary Diagnostic Laboratory Network (VETLAB Network) in applying rapid multiplex serological and molecular assays for the differential diagnosis and syndromic surveillance of transboundary animal and zoonotic diseases.

## **Past Events**

### **Technical Assistance on Animal Nutrition to Project 'Improving the Livelihood of Stallholder Livestock Farmers by Developing Animal Feeding Strategies for Enhanced Food Security' (MYA5026)**

Daniela Battaglia

In Myanmar, feed is the main driver of livestock production. It accounts for up to 70 percent of the total cost of operations. Poor or unbalanced feeding adversely affects animal productivity, health, behaviours and welfare. In addition, it has a higher environmental impact. Efficient use of locally available feed resources is key to efficient animal production and food security. But you can only manage when you can measure. Therefore, the generation of feed-related data such as feed inventory, feed balance, feed nutritional quality and safety, is of primary importance.

For these reasons, from 11 to 13 February 2019, a technical assistance mission was carried out at the University of Veterinary Science of Yezin in Myanmar.

Its purpose was to assess the state of the feed analysis laboratory infrastructure and equipment provided by the project and their use, as well as the quantity and quality of

analytical results within the framework of to advise on the best possible use of locally available feed resources for feed formulation and to improve animal nutrition and productivity.

The project activities and outputs so far are: a survey on available feed resources in Pyawbwe; the collection and analysis of ruminant feed samples; field trials for the comparison of the nutritive values of several forages and pulses as well as the conduction of trials to measure their methane production; training of trainers (planned to be extended also to community animal health workers and farmers); research on the formulation of balanced diets with locally available feed resources (e.g. rice straw).

The need to develop a database with feed analytical results and to strengthen contact and information and knowledge sharing with all stakeholders (e.g. farmers, extension workers) was highlighted. The technical mission concluded with a lecture that provided an overview on feed inventories; balanced feeding to improve livestock productivity; feeding trials; feed analysis and quality control; feed safety and related international standards and requirements; feed ratio formulation; livestock feeding strategies; and animal research methodologies.

## Regional Training Course on the Use of Nuclear Derived Techniques in Diagnosis of the Peste des Petits Ruminants in Sheep and Goats (RER5023)

Ivancho Naletoski

The course was held from 11 to 15 February 2019 at the Animal Production and Health Laboratory in Seibersdorf, Austria. A total of twenty-four participants from Albania, Armenia, Azerbaijan, Belarus, Bosnia and Herzegovina, Bulgaria, Croatia, Cyprus, Georgia, Greece, Hungary, Latvia, Lithuania, Montenegro, North Macedonia, Poland, Portugal, Moldova, Romania, the Russian Federation, Serbia, Slovakia, Slovenia and Tajikistan attended the course.



Participants of the training course in Seibersdorf, Austria

The topics covered serological and molecular techniques for detection of peste des petits ruminants (PPR) in sheep and goat, as well as techniques for advanced differentiation of the PPR strains, such as genetic sequencing, bioinformatics and phylogeny.

The training course was led by a team of international experts from the French Agricultural Research Centre for International Development (CIRAD) in Montpellier, France; the Royal Veterinary College in London, the UK; the National Laboratory for Livestock and Veterinary Research (ISRA/LNERV) in Dakar, Senegal, and the FAO Headquarters in Rome, Italy.

## Strengthening Africa's Regional Capacity for Diagnosis of Emerging or Re-emerging Zoonotic Diseases, including Ebola Virus Disease (EVD), and Establishing Early Warning Systems (RAF5073)

Bharani Settypalli

From 11 to 20 February 2019, Mr Bharani Settypalli, from the Animal Production and Health Laboratory in Seibersdorf, conducted an expert mission to Dakar, Senegal.

The purpose of the mission was to train the staff members of the National Laboratory for Livestock and Veterinary Research (LNERV/ISRA) on animal pathogen detection and support the laboratory in instrument calibrations and troubleshooting.



Hands-on training in Dakar, Senegal

Nine participants representing LNERV/ISRA, the Ministry of Livestock and Animal Production of Senegal, and the National Office for Livestock and Veterinary Research (ONARDEL) of Mauritania received training on real-time PCR based multiplex assays for differential diagnosis and multiple pathogen detection in small ruminants and swine.



The participants were also trained on detection of zoonotic pathogens from vectors including the detection of Rift valley fever virus. The sequencing protocols for characterization of capripox, peste des petits ruminants and African swine fever viruses, and the work flow from amplicon preparation, sequencing data analysis to phylogenetic analysis were demonstrated. The gaps in the molecular diagnostics workflow at LNERV were assessed and discussed. Staff members were trained on calibration and maintenance of real-time PCR detection and nucleic acid quantification systems.

## Regional Training Course on Standard Operational Procedures (SOPs), Implementation and Maintenance of ISO 17025 Standard (RER5073)

Ivancho Naletoski

The training course took place from 25 February to 1 March 2019 in Windhoek, Namibia. Thirty-five participants from Algeria, Angola, Benin, Botswana, Burkina Faso, Burundi, Cameroon, Congo, Congo DRC, Ivory Coast, Egypt, Eswatini, Ethiopia, Ghana, Madagascar, Malawi, Mali, Mauritania, Morocco, Mozambique, Namibia, Niger, Nigeria, Senegal, Sierra Leone, South Africa, Uganda, Zambia and Zimbabwe attended the course.

The iVetNet Information Platform was developed by the Animal Production and Health Section to store personal, institutional, environmental and equipment records, relevant for the ISO 17025 standard, control records for the environment and the equipment, as well as exchange of validated SOPs for the diagnosis of animal and zoonotic diseases, with build in quality assurance and quality control records.

The participants practiced step-by-step procedures on the use of iVetNet and discussed its potential improvements and upgrades.

The course was led by an international expert and the technical officer of the course.



Participants at the training course on the use of iVetNet Information Platform in Windhoek, Namibia

## Regional Workshop on the Collection and Identification of the Most Important Vectors for Transmission of Priority Vector Borne Diseases (RER5023)

Ivancho Naletoski

The workshop took place from 25 to 29 March 2019 at the Institute for Diagnosis and Animal Health, in Bucharest, Romania. Thirty-eight participants from Albania, Armenia, Azerbaijan, Belarus, Bulgaria, Croatia, Cyprus, Estonia, Georgia, Greece, Hungary, Kazakhstan, Kirgizstan, Latvia, Lithuania, Montenegro, North Macedonia, Poland, Portugal, Moldova, Romania, the Russian Federation, Serbia, Slovakia, Slovenia, Turkey, Turkmenistan, Ukraine and Uzbekistan attended the workshop.

The objective of the workshop was to present, discuss and exchange up-to-date techniques for collection and identification of the most important vectors of the priority diseases for the project (culicoides, mosquitoes, sand flies and ticks), as well as to present the current principles of vector control in the context of vector borne diseases.

The event was led by a team of experts, one for each vector group, as well as an expert in vector control.



Participants of the workshop in Bucharest, Romania

## Research Institute of the Tajik Academy of Agricultural Sciences in Dushanbe, Tajikistan – Project Design TAD2018003

Ivancho Naletoski

The Veterinary Research Institute (VRI) of the Tajik Academy of Agricultural Sciences in Dushanbe, Tajikistan, has applied for a new Technical Cooperation project for the cycle 2020–2021. After initial review and evaluation by the management of the IAEA, a fact-finding mission was organized from 1 to 5 April 2019 to review the existing capacities of VRI and fine tune the project workplan to optimally upgrade the existing capacities.

The counterparts agreed to upgrade of the capacities for diagnostic tests (both serological and molecular); strengthen the biobanking capacities of the institute (storage of live pathogens, nucleic acids and sera from field samples); and facilitate the implementation of the ISO 17025 standards in the local laboratories.

The tasks of the project will be implemented through a set of fellowships and scientific visits, delivery of equipment and consumables for the priority diseases in the country and expert missions to support the implementation of up-to-date technologies in the local laboratories.



*Participants at the Tajik Academy of Agricultural Sciences in Dushanbe, Tajikistan*

## **Regional Training Course on Statistical Analysis of Data Related to Gastro-Intestinal Parasite Management in Sheep and Goat (RLA5071)**

Mario Garcia Podesta

Gastro-intestinal (GI) parasites pose a major threat to livestock productivity, particularly in tropical and subtropical regions. Parasite infection is one of the major constraints for sheep and goat rearing in Latin America. Apart from the loss due to mortality and slow growth of meat, the growing level of anthelmintic resistance in parasites is a serious challenge.

The Regional ARCAL Technical Cooperation Project RLA5071 on “Decreasing the Parasite Infestation Rate of Sheep” started in 2016 to implement breeding programmes to improve the genetic potential of local sheep for enhanced parasite resistance characteristics. As part of the project, this training course was organized from 6 to 12 May 2019 in Heredia, Costa Rica. Mr Victor Montenegro was the Course Director.

A total of 19 participants from 10 countries (Argentina, Bolivia, Costa Rica, Cuba, Dominican Republic, Mexico, Paraguay, Peru, Uruguay and Venezuela) were provided with hands-on training on descriptive statistics, data quality control, statistical models, graphical representation of data,

hypothesis testing, main parametric and nonparametric tests, correlations and regressions, and software management for statistical analysis; analysis of biological data related to management of gastrointestinal parasites in livestock; and on the designing of experiments on veterinary parasitology, reproduction, nutrition and genetics.

The IAEA external lecturer, Mr Jorge Piaggio, from Uruguay, efficiently covered all the theoretical and practical sessions. He distributed several datasets and protocols to be used as examples and guides during the practical sessions.

The Stata software was selected for use in the training course. Perpetual single user licenses were distributed to course participants.



*Meeting participants in Heredia, Costa Rica*

## **National Training Course on the Use of Routine Serological Techniques for Diagnosis of Animal Diseases, including those with Zoonotic Potential (NEP5005)**

Ivancho Naletoski

The national training course was held from 6 to 17 May 2019 at the facilities of the Central Veterinary Laboratory (CVL) in Kathmandu, Nepal. Twelve participants from the CVL (4) and the regional veterinary laboratories from Bharatpur (1), Biratnagar (1), Chitwan (1), Itram (1), Kailali (1), Kaski(1), Pokhara (2) and Surkhet (1) attended the course.

The training course covered a large scope of serology techniques, such as agglutination tests (rose Bengal and serum agglutination), complement fixation test, agar gel immunodiffusions, indirect and competitive ELISA, topics on validation and verification of diagnostic tests, as well as quality assurance and quality control.

The event was led by an international expert, Ms Ana Maria Nicola from the OIE reference laboratory for brucellosis in Buenos Aires, Argentina.



## Regional Training Course on the Use of Real Time Geo-visualization in Monitoring Vectors and Vector Borne Diseases (RER5023)

Ivancho Naletoski

The regional training course was held from 13 to 17 May 2019 in Jelsa, Hvar Island, Croatia. Thirty-three participants from Azerbaijan, Bosnia and Herzegovina, Bulgaria, Croatia, Cyprus, Czech Republic, Georgia, Greece, Kazakhstan, Kirgizstan, Latvia, Montenegro, North Macedonia, Portugal, Moldova, Romania, the Russian Federation, Serbia, Slovakia, Turkey and Turkmenistan attended the course.



*Participants practicing use of GIS technologies for monitoring vector borne diseases*

The course delivered theoretical and practical exercises on the basics of Geographic Information Systems (GIGs), data management and data presentation using GISs, event (disease and vector) monitoring using GISs, use of GISs in disease zoning (infected and surveillance zones), basic statistics integrated in GISs, tools for animating GISs data, as well as selected topics from the use of satellite remote sensing imagery for monitoring / modelling of vector borne diseases.

The course was led by three international experts from the topics mentioned above.

## Expert Meeting to Establish Online Open-Source Software for Species Identification of Priority Arthropod Vectors (RER5023)

Ivancho Naletoski

The expert meeting on the development of an online open-source software for species identification of priority arthropod vectors was held at the IAEA Headquarters on 23 May 2019. Four international experts, each for one group of arthropod vectors (culicoides, mosquitoes, sand flies and ticks) attended the meeting.

The meeting was dedicated to the development of internet keys for identification the insect vectors. The internet-based keys, developed by the experts, should lead the responsible researchers at the veterinary laboratories towards the rapid and accurate morphological identification of the vectors.

The vectors for which morphological keys will not be able to accurately determine the species, DNA barcoding procedures will be developed, and the results shared onto the online barcoding databases for species identification (barcodes of life: <http://www.boldsystems.org/>).

These procedures complement the currently developed, harmonized Standardized Operating Procedures (SOPs) for vector capture, subject of the regional workshop in Bucharest (read more on page 11 of this edition). Practical exercises on both sets of SOPs – vector capture and identification – are being planned for the upcoming training course in Ankara, Turkey, scheduled from 1 to 12 July 2019.

## Second Research Coordination Meeting on Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers (D32033)

Hermann Unger

The research coordination meeting (RCM) took place from 20 to 24 May 2019 in Vienna, Austria.

All research contract holders presented the work performed in the last two years, showing that a good progress was achieved.



*Meeting participants at the APH laboratory in Seibersdorf, Austria*

For Mannheimia, the irradiation dose was established (20kGy) and the vaccinations worked without side effects. The irradiation of influenza is progressing well and a trial on chicken is currently being carried out to investigate the immune response and protection.

For fowl pox, the best irradiation dose was 2,4kGy and, in a vaccination challenge, trial birds had similar symptoms and survival rates regardless of having received commercial or irradiated vaccines.

The already very successful tests with the irradiated *hemonchus contortus* vaccine will soon be repeated with a much larger group of animals and the immunology expanded to find correlates for protection. The scientific meetings were supported by B. Kaspers, bird immunologist from Munich LMU and by S. Ulbert, Fraunhofer Inst, Leipzig, expert in novel irradiation technologies.

A three-day immunology workshop ensued at the APH labs in Seibersdorf. Immunological approaches applying immunostaining, cell-sorting, cytokine assays, qPCR for cytokines and ELLISPOT were discussed and techniques were transferred in hands-on exercises. The curriculum prepared for this workshop was amended during the course and now serves as the immunology manual for each lab. Workplans were prepared and the immunological tests were handed out to all participants.

## Training Workshop on Measuring Cell Mediated Immunity in Livestock Species

Giovanni Cattoli

There is an urgent need to improve methods to evaluate vaccine responses in livestock animals. This is because currently available methods are mainly focused on measuring antibody responses instead of cell mediated immunity (CMI). In human immunology, this is well developed, which allows to better evaluate vaccines that target humans.

The immunology and vaccine development program at the Animal Production and Health Laboratory (APHL) recognized this gap of knowledge and has been developing several assays and tools to measure CMI. The next step of this program was to provide scientists Member States with training on how to use these tools and technologies.

A three-day training workshop on measuring CMI in livestock species was held from 22 to 24 May 2019 in the IAEA Laboratories in Seibersdorf, Austria. Seven scientists from Ethiopia, Sudan, Iran, Egypt, Sri Lanka, Bangladesh and Italy participated in this initial training course.

In parallel to the training workshop, APHL developed a comprehensive booklet that includes theoretical knowledge, SOPs and videos explaining salient points of assays which was given to participants.

The assays in the workshop targeted all the major livestock species including poultry. The participants had the opportunity to practice blood and tissue processing and aseptic cell culture techniques which forms the basis for the CMI assays. Multiple technologies including real-time

PCR, ELISPOT and flow cytometry was demonstrated and practiced during the workshop. In addition, participants received a package of reagents to initiate these cutting-edge technologies in their prospective laboratories.

All the participants were counterpart scientists in the coordinated research project (CRP) D32033 – Irradiation of Transboundary Animal Disease Pathogens as Vaccines and Immune Inducers. Therefore, access to the training program will enhance the quality of experiments done in developing these vaccines.



*Hands-on training for contract holders of CRP D32033 in Seibersdorf, Austria*

## Stories

### The Semen Processing Laboratory in Cambodia Facilitates the Expansion of the Artificial Insemination Service to Small Cattle Holdings

A 1-hour video prepared by Dr Vonika Vu, Director of Animal Health and Veterinary Public Health (DAHVP), General Directorate of Animal Health and Production, Phnom Penh, Cambodia, showing the activities carried out for improving cattle productivity in the country with the support of IAEA TC KAM5003. Dozens of livestock extension workers have been trained on the artificial insemination technique and farmers have been sensitized on the advantages of this biotechnology tool for improving the genetic quality and productivity of their animals.



[Click here](#) to watch



## Establishing Genetic Characterization and Breeding Capability for Livestock in Burkina Faso

The government of Burkina Faso support and INERA's commitment for the establishment of a molecular genetic laboratory with the technical support of the IAEA has resulted in a well-equipped laboratory. From a single room where basic research work was done on animal health, reproduction, nutrition and breeding, INERA has now built a separate unit for molecular genetic work with a complete workflow for animal breed genotyping operated by a group of qualified staff. The facilities of the molecular genetic laboratory and the expertise of the staff has attracted the attention of neighbouring West African countries which are sending their samples for analysis and requesting training on DNA technology.



[Click here](#) to read more

## Fast Response to Highly Pathogenic Avian Influenza (HPAI) Outbreak Avoids Economic Losses in the Democratic Republic of the Congo (DRC)

At the end of May 2017, the DRC was faced with what appeared to be its first-ever outbreak of highly pathogenic avian influenza (HPAI). Rapid confirmation was critical. Animal and public health officials needed to know if it really was the avian influenza virus, and, if so, which strain of the virus was present. The Central Veterinary Laboratory in Kinshasa was prepared with sampling and testing protocols to deal with the emergency.

The Joint FAO/IAEA Division had previously provided the lab with diagnostic equipment and training for staff both in the lab and in the field. Thus, when samples from the outbreak area were brought for analysis, the lab was prepared, and it quickly determined that, yes, it was avian flu. While previous outbreaks in Africa had meant the loss of millions of birds, the fast reaction in the Congo meant the outbreak was contained with minimal poultry losses and no human casualties.



[Click here](#) to read more

## Laboratory in Ghana Ready to Handle Diagnosis of Major Animal Disease Outbreaks in West Africa

Hundreds of animal diseases can now be rapidly and accurately diagnosed by specialists at the Accra Veterinary Laboratory (AVL) in Ghana, helping to better protect farmers' livestock and food security. The AVL enhanced facilities and team's new skills, developed through support from the IAEA in cooperation with the Food and Agriculture Organization of the United Nations (FAO), are also an important new resource for many other countries in West Africa.



[Click here](#) to read more

## Hunting for Viruses in Sierra Leone with the Help of Nuclear Technology

It sleeps upside down, comes to life at night and can carry Ebola. What is it? A bat. After suffering from a devastating Ebola outbreak in 2014, veterinary scientists in Sierra Leone are training their peers from around Africa to catch, sample and diagnose, using nuclear-derived techniques, potentially virus-transmitting bats.



[Click here](#) to read more

## IAEA Helps Small-holder Farmers to Improve Twin Births and Mutton Production in Local Sheep Flocks of South India

Scientists have long known that certain sheep breeds (Garole sheep in India, Boorola Merino in Australia, Han sheep in China, and others) have high genetic potential to produce twins and triplets due to the presence of a naturally occurring mutation called FecB. Thanks to advances in nuclear-related molecular technologies, it is now possible to select and breed such animals accurately and improve their prolificacy (number of lambs born per birth), and hence mutton production. The Animal Production and Health Laboratory of the Joint FAO/IAEA Division recently developed a cost-effective, real time, polymerase chain reaction-based genotyping technology to accurately identify sheep carrying the twinning variant of FecB gene.



[Click here](#) to read more

## Using Nuclear Science to Improve Animal Breeding

Animals, so important for food security, provide us with milk, with eggs, with meat. But as global population growth has accelerated, so has the demand for animal produce, putting a strain on both farmers and the environment. Simply raising more animals is not an option; it would put even greater pressure on an already strained environment. The solution lies in increasing the productivity of those animals. This is usually done by selecting and breeding superior animals; but this is a slow process, it can take several years to determine their breeding potential. Now, with advances in genomics, it has become possible to estimate the breeding potential of an animal on the day of its birth, simply by looking at its DNA - or genome map. To develop these genomic maps, a nuclear technique called hybrid radiation mapping is used. The video shows how this can be possible.



[Click here](#) to watch

## Bulgaria Stops Spread of Animal Disease with Help of IAEA and FAO

Bulgaria has halted the spread of the Peste des Petits Ruminants (PPR) — a virus disease that can devastate livestock — thanks, in part, to the support of the IAEA and the Food and Agriculture Organization of the United Nations (FAO). This was the first time PPR was recorded in the European Union, so halting its spread early is of regional importance.



[Click here](#) to read more



## Improving Animal Disease Diagnosis and Cattle Fertility in Lesotho

### The challenge

Lesotho is a country which depends mostly on animals for living. Apart from gaining meat as protein source they also use animals for drought power especially cattle and horses. Horses are considered a pride of Basotho linked traditionally as the main source of transport especially in the mountainous areas. Lesotho is also the main exporter of wool and mohair that bring income to the country and contribute hugely to the country's GDP 62% of Agriculture sector.

The Ministry of Agriculture and Food Security, through the Department of Livestock Services (DLS), is responsible for the promotion of livestock production and health through its Food Security Policy (2005) and Lesotho Zero Hunger Strategy review (2018), which is linked to Sustainable Development Goal 2. The Central Veterinary Laboratory (CVL) is the section under The Animal Health Division of DLS. The laboratory in collaboration with other sister-sections and other stakeholders is responsible for disease diagnosis, research, surveillance regulation of movement of animal and animal products to ensure food safety (Public Health Order, 1970). In order to fulfil the mandate, DLS, under the support of the IAEA, initiated a Technical Cooperation (TC) project to build capacity for diagnosis of animal diseases and enhance artificial insemination in livestock.

The project was aimed at improving animal disease management and promoting production and productivity of sheep and goats through the use of nuclear and molecular technology. Unlike conventional laboratory tests, the technology offers timely and accurate diagnosis of animal diseases of transboundary, economic, public health and animal welfare importance. In addition, the technology involved with animal synchronization, harvesting, processing, analysis and storage of semen and measurement of reproduction-indicator hormones to determine pregnancy and breeding soundness to support ongoing initiative to enhance production of fiber, under the project "Wool and Mohair promotion project". The IAEA provided support for training of staff members and procurement of reagents and equipment.

### The impact

CVL has the capacity and required competencies for the diagnosis of animal diseases which include brucellosis, anthrax, Newcastle through the use of molecular and ELISA technology. This capacity has led to improved turn-around time, reduced reliance on other countries for disease confirmation as a requirement during enforcement of regulations such as the culling of brucella-positive animal, enhanced disease surveillance toward improved disease management and facilitated access to international market according to OIE requirements. In addition, total number of samples that are routinely received, processed and tested monthly has also increased by over 25 % due to adoption of the technology.

### Reproduction

Adoption of technology for semen evaluation has led to better management of semen for livestock farming communities. The technology has led to the discovery and addressing of the practices which previously resulted in poor conception after artificial insemination. According to the findings, the greater part of the semen in the mobile tanks was dead. This be may attributed to failure to maintain required nitrogen levels and repeated or prolonged exposure of semen straw to environmental temperature in the mobile cans.

The capacity will enable better selection and preservation of breeds towards improved production, adaptability and adoption of climate-friendly farming practices.



*Technician performing ELISA test on Brucellosis disease*

## Genetic Sequencing Services Available for all Counterparts in Member States

The Animal Production and Health (APH) Subprogramme of the Joint FAO/IAEA Division (NAFA) is engaged in the technical cooperation and coordinated research programme related to the control and eradication of animal and zoonotic disease. Both programmes include various institutions (primarily laboratories) in many IAEA Member States (MS). Additionally, APH has been involved in the official programmes of international organizations, targeting control and/or eradication of important animal diseases, such as the avian influenza, brucellosis, foot-and-mouth-disease (FMD), lumpy skin disease, Newcastle disease virus, peste des petits ruminants (PPR), rabies and others.

The animal health projects of APH are dedicated to the transfer of nuclear and nuclear related technologies for early detection of animal diseases, which mainly cover detection and differentiation of animal and zoonotic pathogens.

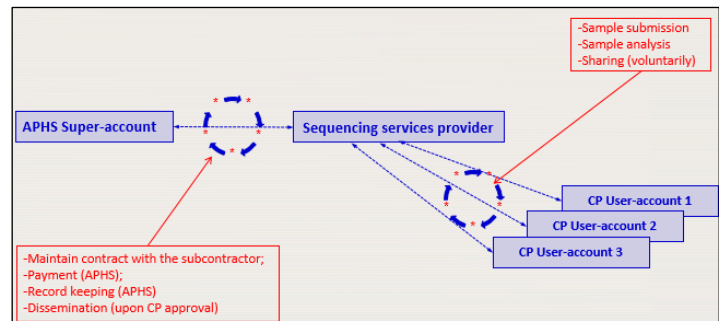
The development of technology in recent years has resulted in the establishment of much more sophisticated techniques (genetic sequencing), which enable the detection of detailed layouts of pathogen genomes. By comparison, it is possible to build detailed phylogeny trees of the circulating pathogens and understand the pathogen evolution and epidemiology of the diseases.

Simultaneously, the global scientific community has developed transparent online genetic databases which allow for comparison of millions of sets of genetic information (NCBI Genbank), thus enabling precise clustering and classification of the pathogens.

The genetic sequencing protocols require expensive hardware and continuous training. Considering the relatively low workload (only few samples per outbreak are sequenced), installation of expensive hardware into multiple laboratories is therefore not feasible.

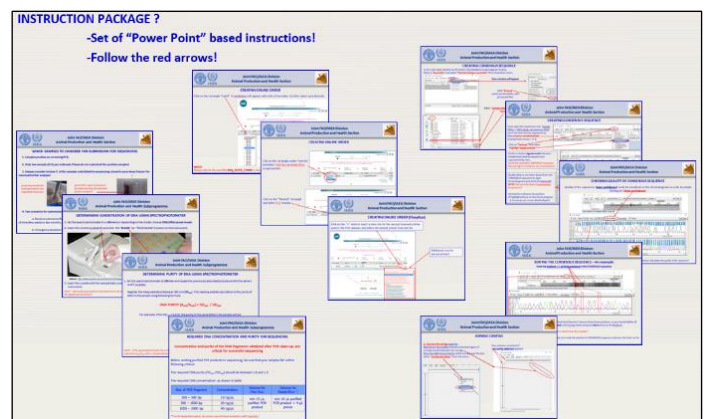
As a solution, APHS has developed a standardized multi-step procedure for sequencing services through an external service provider, comprised of instructions for sample preparation and evaluation (Polymerase Chain Reaction – PCR amplicons), sample shipment, sequence assembly, sequence alignment, and development and interpretation of phylogenetic trees of pathogens.

The sequencing service is enabled for registered users, counterparts in the APH subprogramme.



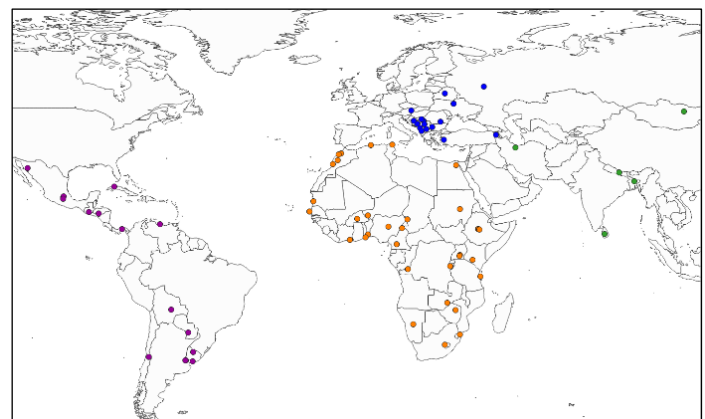
*Schematic workflow of the procedure for sequencing services*

The multi-step procedures are prepared in a format of Power Point presentations with detailed instructions for each step of the workflow. A simplified overview of the instructions is shown below.



*Simplified overview of the instruction packages*

Briefly, the users are initially registered with the external sequencing service provider as members of the APH - VETLAB Network which enables them for rapid and free of charge sequencing service, including the shipment costs.



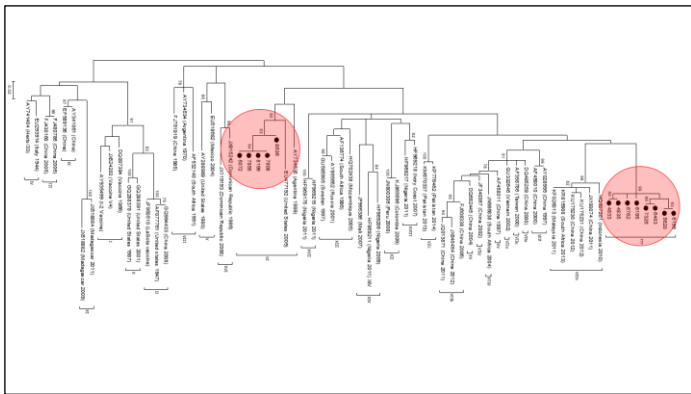
*Registered users of the sequencing service (Green-Asia and Pacific, blue-Europe, orange-Africa, purple-Latin America)*

Samples are initially tested in the local laboratories using validated polymerase chain reaction (PCR) protocols, in order to amplify the specific genomic segments. Positive signals are verified for compliance with the quality requirements of the sequencing service provider and shipped through an express courier (DHL or Fedex) to the sequencing service provider (transport takes 1-2 days in most cases).



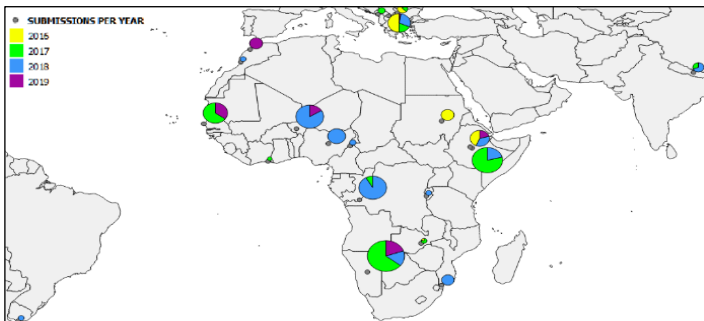
It is important to mention that the whole procedure does not include work with live pathogens, which significantly simplifies the sample manipulation and international transport. Results of the sequence analysis are usually obtained within 24 hours and the raw data sent back to the submitter immediately via email.

Upon receipt, the users perform sequence assemblies using an open source software (Staden), sequence alignment using the online available genetic databases (NCBI Genbank) or using a standard collection of reference genetic sequences. Phylogeny of the pathogens is also performed using an open source software (Mega 7). If the users are willing to share the obtained genetic sequences on NCBI Genbank, the workflow includes instructions for Sequin, an official open-source software of NCBI Genbank for submission of sequencing data and generation of unique Genbank accession numbers available online (requirement for publication in peer reviewed journals).



Phylogenetic analysis of the same pathogen detected from two different outbreaks from different geographical regions in the country, showing that the strains belong to two different clusters (red circles)

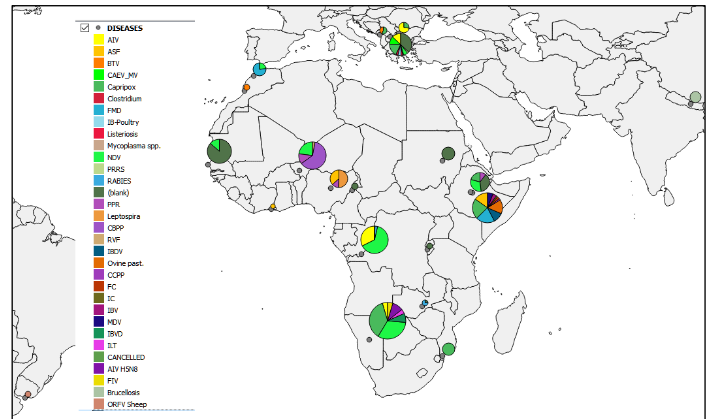
The sequencing services were launched in 2016. Until 31 May 2019, a total of 1704 submissions were recorded, starting with 154 in 2016, followed by 593, 698 and 220 in 2017, 2018 and 2019 respectively



Submissions of samples per year. Colours indicate the year of submission (yellow= 2016-total 164 submissions; green=2017; blue=2018; purple=2019). \ Sizes of the pies relative to the total numbers of submissions

In three years of operation, users have submitted samples for 32 diseases. The top five diseases of interest were Newcastle disease of poultry (351 or 20.6%); capripox – sheep & goat pox and lumpy skin disease (280 or 16.4%);

contagious bovine pleuropneumonia (124 or 7.3%); avian influenza (118 or 6.9%); and foot and mouth disease (82 or 4.85%).



Approximation of the variety of diseases submitted by the users. Sizes of the pies relative to the total numbers of submissions

Aside from the instruction package, APH has established a set of diagnostic consumables comprised of extraction, amplification and purification kits for PCR, as well as a set of primers which is distributed to the member states laboratories through the regular subprogramme activities (VETLAB Network support) or the ongoing technical cooperation (national or regional) projects.

The primer list is periodically updated, based on the priority diseases in MS. The current primer list includes 32 pathogens of the priority animal and zoonotic diseases with 69 genetic targets (for evaluation of some pathogens, multiple targets are required). The vast majority of the targets are the commonly accepted genetic regions, such as the VP 1 region of the FMDV, the cleavage site of the AIV, p72 or p54 of the African swine fever etc, which enables for easy comparison of the strains with different origin and at different laboratories.

The result of the sequencing services has been used to complement scientific publications. So far, its users have published 9 papers in international peer-reviewed journals.

Access to genetic sequencing is of major interest for the laboratories. However, it is not applicable for routine differentiation of all pathogens. Many platforms, especially for bacterial pathogens, have been developed over the last few decades, such as the Variable Number Tandem Repeats (VNTR), the Multi-locus VNTR analysis, Multi-Locus Sequence Analysis (MLSA), as well as in the recent years, mobile Whole Genome Sequencing (WGS) Platforms.

APH is working on the chance of establishing workflows for the VNTR, MLVA and WGS, under the similar principles, as used for the genetic sequencing service. Moreover, for selected, more advanced laboratories, APH is planning dissemination of the available mobile WGS platforms for initial testing and integration.

## Papers published by the users of the sequencing service of APH

Ord	Country	Publication	Link
1	Bulgaria	Stoimenov, G. et al. (2018): Outbreak of Influenza A Virus (H5N8) in Dalmation Pelicans Srebarina Reserve, Bulgaria, 2015. <i>G. Tradition and modernity in Veterinary Medicine</i> . May 16. 2018	<a href="#">Link</a>
2	Ethiopia	Demeke B. et al. (2017): Investigation of Marek's disease virus from chickens in central Ethiopia. <i>Trop Anim Health Prod.</i> 2017 Feb;49(2):403-408. doi: 10.1007/s11250-016-1208-1. Epub 2016 Dec 14.	<a href="#">Link</a>
3	Ethiopia	Mekuriaw A, et al (2017): Infectious bursal disease: outbreak investigation, molecular characterization, and vaccine immunogenicity trial in Ethiopia. <i>Trop Anim Health Prod.</i> 2017 Aug;49(6):1295-1302. doi: 10.1007/s11250-017-1328-2. Epub 2017 Jun 15.	<a href="#">Link</a>
4	Namibia	Molini U. et al (2017): First genetic characterization of newcastle disease viruses from Namibia: identification of a novel VIIk subgenotype. <i>Arch Virol.</i> 2017 Aug;162(8):2427-2431. doi: 10.1007/s00705-017-3389-y. Epub 2017 May 3.	<a href="#">Link</a>
5	Namibia	Molini U. et al. (2018): Molecular characterization of Lumpy Skin Disease virus in Namibia, 2017. <i>Archives of Virology.</i> Jun 4. doi: 10.1007/s00705-018-3891-x.	<a href="#">Link</a>
6	Namibia	Molini U. et al. (2018): Phylogenetic analysis of pigeon paramyxoviruses type-1 identified in mourning collared doves ( <i>Streptopelia decipiens</i> ) in Namibia, Africa. <i>Journal of Wildlife Diseases.</i> Mar 29. Doi: 10.7589/2007-10-246	<a href="#">Link</a>
7	Namibia	Molini U. et al. (2019): Investigation of Infectious Laryngotracheitis outbreaks in Namibia in 2018. <i>Trop Anim Health Prod.</i> 2019 May 18. doi: 10.1007/s11250-019-01918-x. [Epub ahead of print]	<a href="#">Link</a>
8	Namibia	Molini U. et al. (2019): Molecular characterization of Infectious Bursal Disease virus (IBDV) in Namibia, 2017. <i>Onderstepoort Journal of Veterinary Research.</i> Accepted	<a href="#">Link</a>
9	Uruguay	Castro ER et al. (2019): Detection and Phylogenetic Analysis of the ORF Virus from Sheep in Uruguay. <i>Annals of Clinical Virology</i> ; 2019   Volume 1   Issue 1   Article 10021	<a href="#">Link</a>

# Research Activities of the Animal Production and Health Laboratory

## Animal Genetics

### Application of Nuclear and Genomic Tools to Enable for the Selection of Animals with Enhanced Productivity Traits (CRP D31028)

#### *Development of genomic tools to construct whole genome radiation hybrid map and reference genome assembly for dromedary camels*

Genomic resources are scarcely available for genetic improvement of dromedary camels, an important livestock for nomadic pastoralists in Africa and Asia. During 2017–2018, APHL initiated the development and characterization of radiation hybrid panels to facilitate camel genome mapping. Initially, whole genome survey sequencing of camel radiation hybrid panels was attempted to perform reference genome assembly. However, with declining costs of custom genotyping technologies, APHL in collaboration with the Institute of Wildlife, Vetmed University of Vienna is now engaged in the development of a DNA microarray

tool for camelids. Whole genome camel sequence resources available with Vetmed University will be utilized for the design and development of a custom single nucleotide polymorphic (SNP) genotyping array. APHL will subsequently test and validate the DNA array for use on camels. About 5K quality control markers and ~100K SNP markers for dromedary and Bactrian camels have been identified. The process of design and fabrication of array is currently in progress. The new approach will not only help to construct the whole genome radiation hybrid map and reference genome assembly for dromedaries, but will also lead to the development of advanced, state of the art tools for camel breeding and improvement.

#### *Setting up low-cost, high throughput genome-wide SNP genotyping facility to support livestock development in member states*

Improvement of dairy cattle for increased milk production occurs mainly through two conventional approaches: (i) selection and breeding of superior genetic merit animals and/or (ii) cross breeding of local cattle with highly selected commercial taurine cattle. With recent advances in molecular technologies, efficiency of both these approaches can greatly be increased by testing and utilizing genome-wide DNA marker variations. However, application of these technologies in developing member states has been constrained by several factors that includes cost of genotyping and lack of performance records amongst others. To address the challenge, APHL initiated the setting up of semi-automated, low-cost genome-wide SNP typing



facility to support livestock development in member states. High throughput genotyping protocols were optimized by integrating automated liquid handling and Affymetrix GeneTitan systems. As a first step, cattle genotyping requirements of member state counterparts working under the IAEA coordinated research and technical cooperation projects are being supported. More than 1200 indigenous cattle from seven countries across Asia and Africa have been genotyped using 54K SNP microarray. Large sets of genotype data generated are being used by these countries to establish baseline genetic information database and analysis of basic diversity, population structure, and genetic admixture levels in their local cattle. The semi-automated genotyping methodology is also expected to help member states in accurately estimating the level of taurine inheritance in crossbred cattle and identifying optimal genotypes that can suit the existing production system.

### **Implementing Global Plan of Action for Animal Genetic Resources (AnGR)**

In continuation of Joint FAO/IAEA efforts towards implementing Global Plan of Action on animal genetic resources (AnGR), APHL supported member states in at least three major strategic priority areas: characterization, sustainable use and development, and capacity building.

#### **Development of Baseline Genetic Information in Sri Lankan Cattle**

Technical support is being provided to Sri Lanka to establish a full-fledged baseline genetic information database on its local cattle breeds. Three indigenous cattle breeds of Sri Lanka (Sri Lankan White cattle, Local Batu Haraka and Thawalam cattle) and two taurine taurine (Holstein-Friesian and Ayresshire) cattle breeds were evaluated using short tandem repeat markers, genome-wide single nucleotide polymorphic markers and mitochondrial control region variations. More than 15 million genomic data points were generated to establish basic diversity, population structure, genetic admixture levels and phylogeography of these populations. The bioinformatics analysis of genomic data on Sri Lankan cattle is currently underway. The baseline genetic information database and genomic evaluation report will play an important role in establishing reference population for phenotype-genotype association studies and future implementation of genomic selection programs for increased productivity in Sri Lankan cattle.

#### **Sustainable Use and Development**

Artificial insemination using frozen semen technology is an important means of multiplying superior germplasm for sustainable improvement of livestock productivity. Through the IAEA Technical Cooperation program, APHL

provided technical support to national artificial insemination (AI) programs in Papua New Guinea and Eritrea in terms of equipment and supplies required for frozen semen laboratories, implementation of AI under field conditions and improvement of reproductive efficiency in cows. Further, APHL supported Burkina Faso in addressing the challenges of backyard poultry production through modern genetic, breeding, nutrition and preventive healthcare interventions. Particularly, technical and scientific assistance is being provided to identify optimal indigenous chicken ecotype that suits the local production system (through characterization of local chicken ecotypes) and installation of small-scale pilot hatchery units (to address the challenge of poor hatchability of chicks).

### **Capacity Building**

APHL also continued its efforts to improve the laboratory capacity of member states to efficiently manage their locally available animal genetic resources. Institutional and technical support were provided to two countries (Papua New Guinea and Eritrea) for establishing/strengthening molecular genetic laboratories through provision of necessary equipment and laboratory supplies under the framework of national technical cooperation projects.

## **Animal Health**

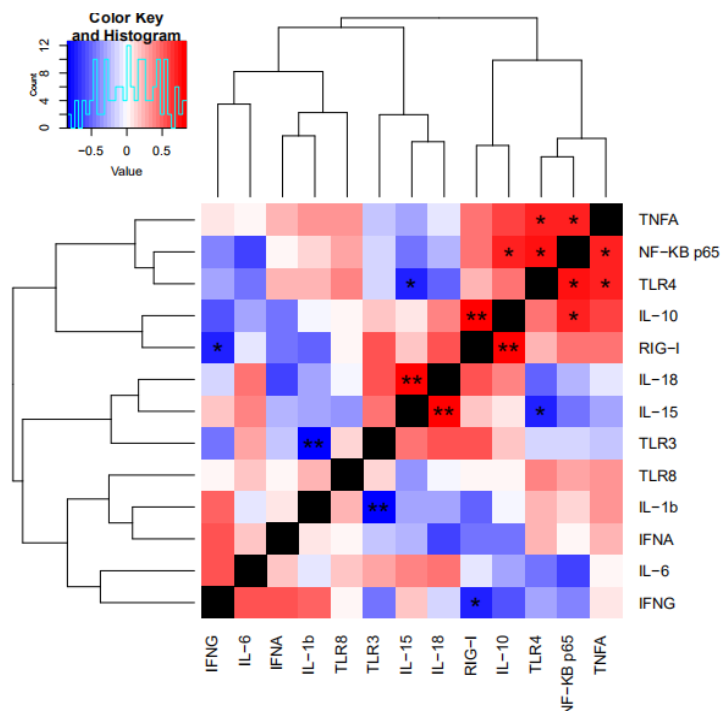
### **Studies on Innate Immune Mechanisms Against Sheeppox Virus Brings New Insights into Vaccine Development**

Sheeppox is a highly contagious disease of small ruminants that predominantly occur in Asia and Africa causing massive economic losses. It is caused by Sheeppox virus (SPPV) that belongs to genus Capripoxvirus of the family Poxviridae. Cross protection is present between capripoxviruses, therefore, live attenuated SPPV vaccines have been commonly used for vaccination against sheeppox and goatpox. Although adaptive immunity is responsible for the long-term memory, innate immune responses induced by live attenuated vaccine (LAV) is a mechanism that leads to a better protection by SPPV vaccines. Little is known on the mechanisms related to the innate immune response elicited by these vaccines.

At APHL, we delineated these mechanisms by assaying the expression level of 13 targeted genes that included pattern recognition receptors (PRRs), Nuclear factor- $\kappa$ B p65 (NF- $\kappa$ B), and cytokines in the sheep peripheral blood mononuclear cells (PBMC) exposed to SPPV and compared between wild type virus (WT) or LAV *in vitro*. We discovered that, there was significantly higher expression of PRR, RIG-1 in both WT (100-fold) and LAV (60-fold) in infected PBMC cultures compared to the PBMC that were not exposed to the virus.

The mRNA expression levels of TNF- $\alpha$ , IL-15, and IL-10 were significantly upregulated in PBMC incubated with WT or LAV, compared with matching controls. The data revealed no significant difference in expression of mRNA levels of targeted markers between the PBMC that were cultured either with WT or LAV infection, except RIG-1. Since many immune markers followed a similar pattern of expression between the two (WT and LAV) treatments, we next investigated which markers were correlated in their expression.

Indeed, heatmap analysis revealed relationships between receptors, transcription factors and cytokines. Our results showed an expression level of RIG-1, which recognizes dsRNA, highly positively correlated with IL-10 and, moreover, we observed a negative correlation between RIG-1 and IFN- $\gamma$  in PBMC. Both positive and negative correlations were statistically significant. TLR3, the other PRR that could recognize dsRNA, was moderately correlated with IL-6, IL-15 and IL-18. We also observed a highly significant negative correlation of expression between TLR3 and IL-1 $\beta$  level in PBMC. Similarly, the expression level of TLR4 signaling pathway correlated significantly and positively with TNFA and NF-kB, and significantly and negatively correlated with IL-15 in PBMC



Heat map for correlations of target genes expression using qPCR. The correlation heat map that describes the combined Pearson correlation coefficient based on distance between the two gene expression value against all target groups. \*\*  $p < 0.01$  and \*  $p < 0.05$

These results suggest the key modulating effects of RIG-1 in the induction of innate immune mechanisms against SPPV. In conclusion, we have identified some of the key innate mechanisms that follows SPPV exposure to host PBMC. These data will aid in developing killed vaccine for SPPV and other pox viral diseases in small and large animal ruminants by incorporating adjuvants that will yield a desired innate immune activation pathway.

## Laboratory Methods for Measuring Cell Mediated Immunity in Chickens

In recent years, chicken farming has become highly intensive not only in the developed world but also in the developing world. Along with that, the number of diseases that afflict the poultry industry has also increased and the best measure to control is vaccination. It is very important to measure vaccine induced immunity to understand the efficacy of both established and experimental vaccines. While measuring antibodies remains the gold standard of evaluating vaccine immunity, it is equally important to measure cell mediated immunity (CMI) especially to viral diseases such as avian influenza. The methods used to measure CMI in chickens are not well developed and standard operating procedures for such assays are not fully optimized.

To support Member States in need of such assays, APHL is currently working on improving methods to measure CMI in chicken. Chicken do not possess lymph nodes and the amount of blood that can be drawn is limited. Therefore, the best source to obtain immune cells to measure CMI in chicken is the spleen as it is the main peripheral lymphoid organ.

In collaboration with the Veterinary Medical University in Vienna, APHL has developed an optimized protocol to isolate immune cells (splenocytes) from chicken. With this protocol, it is possible to isolate a larger number of splenocytes with higher viability, which is important in immune assays. Moreover, this improved protocol allows to process spleens that have been kept overnight at refrigerated conditions.

Protocols have also been developed to use chicken splenocytes to measure vaccine induced immunity by measuring cytokine production through ELISA technique. These experimental protocols are used in measuring CMI against an experimental irradiated influenza vaccine conducted in Iran. Current, experiments at APHL are directed to develop methods measuring cytokine expression through real-time PCR and cytokine production through ELISPOT technique.



Spleen cells from chicken are isolated (left) and resuspended in proper media, counted (right) and used in different assays including ELISA, ELISPOT and quantitative PCR to measure CMI markers for immunity

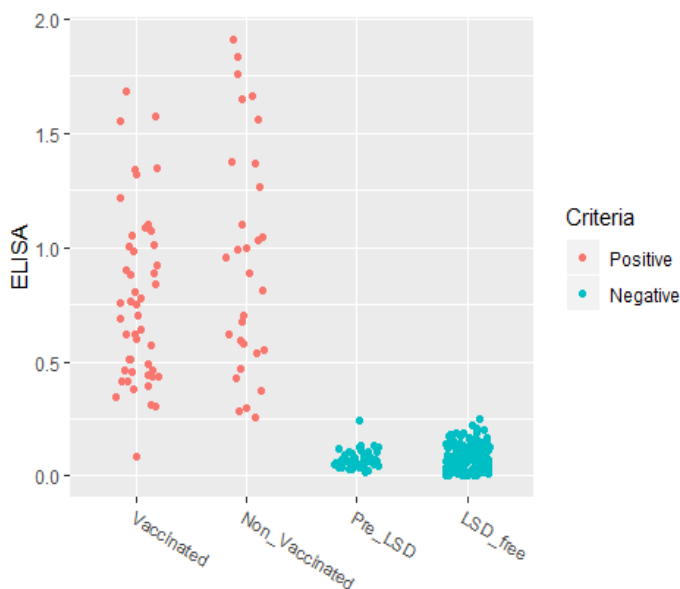


## Validation of Lumpy Skin Disease Indirect ELISA Using Field and Experimental Cattle Sera

Lumpy Skin Disease (LSD) is a contagious disease of cattle with a significant impact on the livestock trade and industry. LSD is caused by the LSD virus which together with Sheeppox virus (SPPV) and Goatpox virus (GTPV), make-up the genus *Capripoxvirus* of the family Poxviridae.

LSD was confined to Africa, with some incursions in the Middle East and Asia until 2015, when the disease was first reported in the European Union. By then, it was evident the need for a serological method to detect the disease and help with surveillance and post-vaccination monitoring. The Animal Production and Health Laboratory (APHL) worked on the development of an indirect ELISA (iELISA) for the detection of LSD antibodies which has been now validated.

The LSD iELISA developed by APHL has been tested on more than 250 negative and 75 positive bovine serum samples. The assay was able to well discriminate between LSD antibody positive and negative sera (image below), showing a sensitivity of 98.72% and a specificity of 100%. The LSD iELISA provides Member States with a rapid and effective disease diagnosis tool to control this transboundary animal disease. After a publication describing the method used, further validation on multiple origin samples will be analyzed.



Distribution of positive and negative bovine sera tested by the newly APHL developed iELISA for the detection of LSD antibodies.

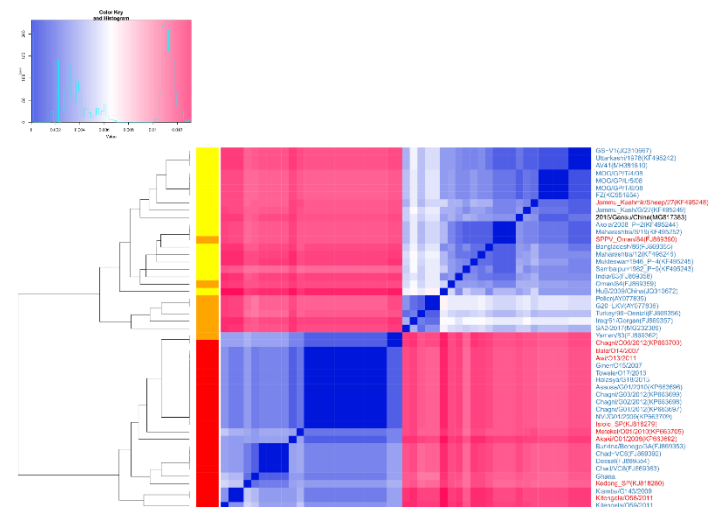
## An Approach to Discriminated Goatpox Viruses Based on their Geographical Origins

The recent incursions of Lumpy skin disease virus (LSDV) and sheeppox virus (SPPV) in Europe showed that capripoxviruses could spread from their endemic geographic locations to disease-free areas. Therefore, it is of high importance to develop molecular tools to complement traditional epidemiological methods to trace the origin of viruses when outbreaks occur in disease-free areas and detect the emergence of new strains in areas where the disease is endemic.

Scientists in APHL have investigated the use of alternative bioinformatic approaches (alignment-free) for the accurate classification of goatpox viruses – member of the capripoxvirus genus - according to their geographical origins.

An alignment-free method based on k-mer frequencies was used to compare a comprehensive set of GPCR gene sequences of GTPVs from various geographic locations. The result showed that GTPVs could segregate based on the geographic region of origin: the African GTPVs and Asian GTPVs which further split into Western and Central Asian (WCA) GTPVs, and Southern and Eastern Asian (SEA) GTPVs.

This approach will help to determine the origin of introduction in case of GTPV appearance in disease-free regions and detect the importation of new strains in GTPV endemic areas.



The heatmap for the nucleotides k-mers frequencies variations of 49 GTPVs. The Complete-linkage clustering method was used to re-order the sequences. The vertical side colors indicated where the corresponding samples came from (red for Africa, orange for West and Central Asia, yellow for East and South Asia).

## Parapoxviruses of Cattle in Africa

In a previous newsletter, APHL reported finding pseudocowpox virus (PCPV) in cattle samples submitted to the Central Veterinary Research Institute (CVRI), Zambia, on suspicion of lumpy skin disease virus infections. An HRM assay for the simultaneous detection and differentiation poxviruses of medical and veterinary importance, developed at APHL and transferred to CVRI, facilitated this detection. BNVL, Botswana, LNRV, Senegal and CVL, Mozambique, employed this HRM assay to analyse skin lesion collected from diseased cattle.

The results revealed PCPV infections in Senegal and Botswana and bovine papular stomatitis virus (BPSV) infection in Mozambique. Sequencing of the full B2L gene and phylogenetic analysis confirmed these results.

These results and our previous findings of PCPV in Zambia prove that parapoxvirus infections are common infections of cattle in Africa, although under-documented.

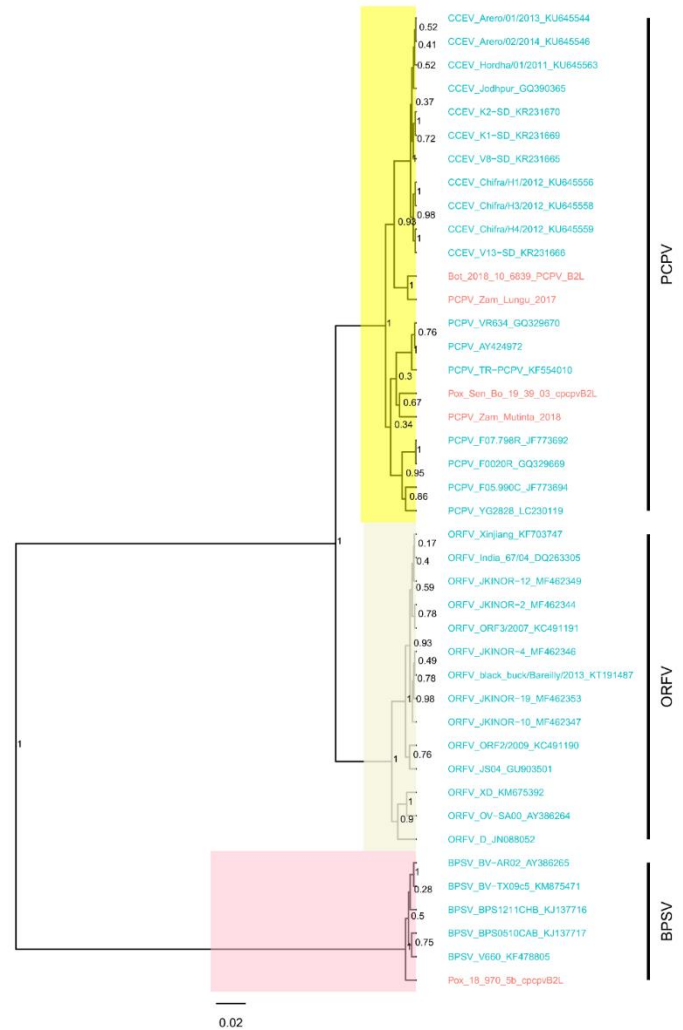
PCPV and BPSV produce pox diseases in cattle, causing losses in productivity, and these viruses can infect humans working in close contact with infected animals.

It would be of interest to further investigate the impact of these diseases on cattle productivity in African countries.

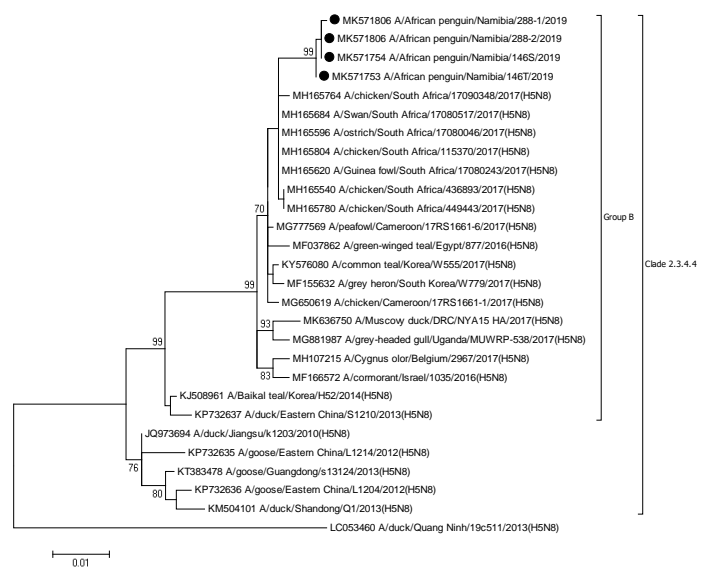
## Highly pathogenic Avian influenza (H5N8) in African Penguins – Namibia

The African penguin (*Spheniscus demersus*) is an endangered species that is found only in southern African waters with a breeding range that extends from the Eastern Cape Province, South Africa to southern Namibia. Early in January 2019, an unusually high mortality of penguins was noted during routine coastal monitoring near Halifax Island which is a protected penguin breeding site about 100m from the mainland in Namibia. By the second half of February 2019 more than 350 penguin carcasses had been found on Halifax Island confirming that this event constituted the most severe mortality event on record for this species in Namibia. An official report was made to the OIE on the 12-02-2019 by Namibian authorities (OIE 2019).

Samples were processed by the Central Veterinary Laboratory, Windhoek by RT-qPCR and indicated the presence of Highly Pathogenic Avian influenza (HPAI) subtype H5N8. Sequence analysis of the haemagglutinin and neuraminidase gene confirmed the presence of the virus in the birds and its high similarity to HPAI subtype H5N8 identified in South Africa in 2017.



Maximum clade credibility (MCC) tree based on of the partial B2L gene sequences of parapoxviruses. The posterior probabilities are plotted as respective nodes labels. The cattle parapoxvirus sequences from Zambia, Mozambique, Senegal and Botswana are highlighted based their geographical origins.



ML phylogenetic tree of partial H5 gene sequence (1061 bp) from the samples collected in Namibia combined with similar sequences available in GenBank. The sequences from this study are shown by filled black circles. Clades and groupings are shown. Only bootstrap values greater than 70% are shown



## Molecular Characterization of Peste des Petits Ruminants Viruses from Nepal

In South Asia, peste des petits ruminants (PPR) outbreaks are reported regularly in India and Bangladesh, with strong evidence existing for cross border movement of the disease between India and Bangladesh.

The goat and sheep population in Nepal is estimated to be 11 million and 800,000, respectively. Animals are imported from both India and China and movement of goats and sheep within the countries is common particularly during festive seasons. Nepal reported PPR for the first time in 1994. A National PPR Control program was launched in 2001. Although strategic mass and ring vaccination with Nigeria 75/1 PPRV vaccine (produced locally) were carried out under this program, outbreaks continue to occur on an annual basis. Despite these outbreaks, there has been limited molecular characterization of circulating virus in the country. In this study a 351 bp segment of the nucleoprotein (N) gene of the PPR virus (PPRV) was amplified and sequenced from ten samples collected between 2005 and 2016. Phylogenetic trees were estimated from these sequences using the Maximum likelihood method confirming that all of the PPRVs from the samples analysed belong to sub-clade IV of clade I of lineage IV and that they share a common origin with other PPRVs in the region.

## Fellows, Interns and Consultants

**Ms Yang Liu**, from Tsinghua University, School of Life Sciences, in China, was trained on bioinformatic analysis and phylogeny of capripox viruses and ELISA test validation for 3 months (1 March to 7 June 2019).

**Ms Mihad Fath El Rahman Mahmoud Alawad**, from the Central Veterinary Research Laboratory, in Khartoum, Sudan, was trained on applications of flow cytometry to evaluate animal immune response for three weeks (20 May to 10 June 2019).

**Mr L. G. Lokugalappatti**, from University of Peradeniya, Sri Lanka, joined APHL on 4 March 2019 as a fellow for a period of six months. Mr. Lokugalappatti will be working on advanced genomic analysis of locally available Sri Lankan cattle breeds using microarray and sequencing technologies.

**Mr Arnaud Stephane Rayangnewende** from University of Ouagadougou, Burkina Faso, joined APHL on 26 March 2019 as a PhD consultant for a period of 9 months. Mr Rayangnewende will be working on genomic evaluation of dual-purpose taurine cattle in Eastern Europe. He will also establish baseline genomic information on indigenous African zebu cattle and compare genetic admixture levels with European taurine cattle to facilitate crossbreeding programs in Africa.

**Mr Kamran Abbas**, from Virtual University, in Lahore, Pakistan, joined APHL on 26 April 2019 as a PhD consultant for a period of 8 months. Mr Abbas will be working on genomic evaluation of dual-purpose taurine cattle in Eastern Europe. He will also establish baseline genomic information on indigenous Asian zebu cattle and compare genetic admixture levels with European taurine cattle to facilitate crossbreeding programs in Asia.

## Coordinated Research Projects (CRPs)

Project Number	Ongoing CRPs	Project Officers
D31028	Application of Nuclear and Genomic Tools to Enable the Selection of Animals with Enhanced Productivity Traits	V. Tsuma M. Garcia Podesta
D31029	Quantification of Intake and Diet Selection of Ruminants Grazing Heterogeneous Pasture Using Compound Specific Stable Isotopes	V. Tsuma M. Garcia Podesta
D32031	Early and Rapid Diagnosis and Control of Transboundary Animal Diseases (TADs) - Phase II: African Swine Fever	H. Unger G. J. Viljoen
D32032	Early Detection of Transboundary Animal Diseases (TADs) to Facilitate Prevention and Control through a Veterinary Diagnostic Laboratory Network (VETLAB Network)	I. Naletoski C. E. Lamien
D32033	Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers	H. Unger G. J. Viljoen
D32034	Use of Stable Isotopes to Trace Bird Migrations and Molecular Nuclear Techniques to Investigate the Epidemiology and Ecology of the Highly Pathogenic Avian Influenza - Phase II	I. Naletoski G. J. Viljoen

### Application of Nuclear and Genomic Tools to Enable the Selection of Animals with Enhanced Productivity Traits (D31028)

Victor Tsuma and Mario Garcia Podesta

The CRP aims at enabling Member States in the application of genetic evaluation and selection involving genomic tools in artificial insemination programmes for rapid, but sustainable, improvement of livestock productivity. Ten research contracts, two technical contracts and three research agreements have already been awarded.

Two major lines of research work are planned, one for those who target crossbreeding and the other for those who keep purebred taurine populations. The crossbreeding group employs admixture analysis to assess the distribution of genetic groups of crossbreds, evaluate their performance and identify suitable genotypes for the prevailing production systems. The group with purebred taurine populations will estimate predicted transmitting ability (PTAs) of sires under local conditions, which will be correlated with genomic PTAs of sires at their origin.

Most research contract holders completed the works planned for the first two years, i.e., collection, recording and analysis of phenotypic, performance and pedigree data from a minimum of 1000 cows/heifers and sires whose semen was used to breed those animals. The technical contract on early pregnancy diagnosis has completed the laboratory works

and identified candidate conceptus-derived proteins. Regarding the technical contracts on sequencing dromedary whole genome using radiation hybrid (RH) technology, DNA has been extracted from 95 selected hamster-dromedary RH clones and is being sequenced using next generation sequencing.

The third research coordination meeting is scheduled to take place in Vienna, Austria, in June 2020.

### Quantification of Intake and Diet Selection of Ruminants Grazing Heterogeneous Pasture Using Compound Specific Stable Isotopes (D31029)

Victor Tsuma and Mario Garcia Podesta

The CRP aims at developing a practical method to predict pasture intake of ruminants grazing heterogeneous pastures and rangeland using stable isotopes to provide tools for better grassland management that enhance animal productivity and reduce impact on environment due to overgrazing, and to allow the design of effective feed supplementation strategies at farm level to optimize animal production. Eight research contracts, two technical contracts and two research agreements constitute the team.

Most research contract holders completed their animal trials and collected samples for the estimation of dry matter intake and diet composition of cattle/yak grazing on pasture/natural



grass lands using n-alkanes and their compound specific stable carbon-13 isotope in feeds and faeces. Two technical contract holders developed protocols and guidelines, which were distributed to research contract holders. A 'ring test' is being conducted with support from agreement holders from the USA and Sweden to review the proficiency of research contract holders' laboratories.

The results will be presented for review and planning future R&D in the second research coordination meeting will take place from 12 to 16 August 2019 in Brazil.

## **Early and Rapid Diagnosis and Control of Transboundary Animal Diseases (TADs) – Phase II: African Swine Fever (D32031)**

Herman Unger and Gerrit Viljoen

This CRP ended earlier this year and came up with a number of interesting results by evaluating technologies which could help control African swine fever (ASF) worldwide.

The evaluation of three different antibody ELISAs showed that all have high sensitivity and specificity when applied roughly three weeks after an infection has passed.

During an outbreak of ASF, the sensitivity drops, most likely, because circulating antibodies are captured by the abundance of viruses in blood and thus do not react with the coating antigen anymore. During an infection, PCR is the diagnostic method of choice. The evaluation of a direct PCR, i.e. without extraction of DNA, proved to be a valuable and quick method to diagnose ASF in outbreaks.

Interestingly, the most consistent results were seen when blood was diluted 1/400 but specific DNA was detected up to a log 8 dilution. This method should now be tested in outbreak situations.

In support of vaccine researches on ASF, an experimental infection setup was developed and tested in Cameroon. This would have lead to 100% mortality within a week and was evaluated with anti-viral drugs. As genotyping of ASF has only epidemiological value and does not correlate with the pathology, serotyping was planned but, due to lack of capacity to isolate viruses, this had to be aborted. Finally, a ring trial for diagnostic capacity on ASF was carried out. The results were, in most cases, perfect.

Regarding the control of ASF, a number of publications on husbandry and hygiene issues were published. Interestingly all participating laboratories reported a drop in ASF outbreaks, but this could be due to the increasingly lower mortalities observed in recent years.

All data gathered during the CRP will be published in the course of 2019.

## **Early Detection of Transboundary Animal Diseases (TADs) to Facilitate Prevention and Control through a Veterinary Diagnostic Laboratory Network (VETLAB Network) (D32032)**

Ivancho Naletoski and Charles Lamien

The Veterinary Diagnosis Laboratory (VETLAB) Network currently integrates 45 African and 19 Asian Member States which are dedicated to the sharing of knowledge and experience and to supporting each other during the implementation of international standards, routine diagnostic procedures, diagnostic approaches for specific disease outbreaks, thus facilitating emergency preparedness and response to animal health emergencies.

The concept of networking proved very successful during the rinderpest eradication campaign. Nowadays, this concept has resulted in great successes in some of the Member States where diagnostic laboratories have received the ISO 17025 accreditation. Additionally, several other laboratories in this network are in advanced phases of implementation of the ISO 17025 standard and expect accreditation soon.

The CRP targets the establishment of such standards for use in serological and molecular diagnostic techniques and produce the following outputs:

- i) A set of internationally acceptable standards for the serological diagnostic techniques for priority diseases among the partners of the VETLAB Network;
- ii) A set of internationally acceptable standards for the molecular diagnostic techniques for priority diseases among the partners of the VETLAB Network;
- iii) Procedures for simultaneous detection of multiple pathogens (multi-pathogen detection panels);
- iv) Procedure for easy access, free-of-charge genetic sequencing services for pathogens of the priority diseases among the partners of the VETLAB Network;
- v) Establish an information platform for integrated information collection, geo-visualization, analysis and decision making.

The project team is comprised of eight research partners (Argentina, Cameroon, Croatia, Ethiopia, Ivory Coast, the FYR of Macedonia, Morocco and Sudan), two technical partners (France and United Kingdom) and three agreement holders (two from France and one from Australia).

The forth research coordination meeting will take place from 19 to 23 August 2019 in Vienna, Austria.

## Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers (D32033)

Hermann Unger and Gerrit Viljoen

This CRP kicked off in early 2016 to continue exploring the possibilities of using irradiation in the development of vaccines. A major stimulus for this was the noteworthy results obtained from the previous CRP on this subject, especially yielding strong outcomes on irradiated intestinal and haemo-parasites as vaccine candidates. However, a major short coming of the initial CRP was the lack of proper immunological tools to define the immune response elicited. This issue was addressed by establishing immunology R&D at the Animal Production and Health Laboratory (APHL) in Seibersdorf, Austria, in 2015. Since then, efforts have been made to develop assays and reagents to monitor the immune responses induced by irradiated vaccines, especially on cellular immunology an area that has been neglected in livestock immunology but of immense importance.

The CRP counterparts are using the protocols, assays and reagents developed by the APHL immunology program and resulted in a big thrust in irradiated vaccine research. An additional task of this CRP is the evaluation of irradiated pathogen preparations as immune enhancers for conventional vaccines. These immune enhancers, or 'adjuvants', are sought in the vaccine market as the traditional solutions lead to severe inflammations and are to be abolished due to the side effects.

The vaccines currently being experimented cover major livestock diseases that need immediate solutions: brucellosis, haemonchus contortus, manheimia, influenza and *Salmonella gallinarum*.

The second research coordination meeting took place in Vienna, Austria, from 20 to 24 May 2019. Read more about it on page 13 of this edition.

## Use of Stable Isotopes to Trace Bird Migrations and Molecular Nuclear Techniques to Investigate the Epidemiology and Ecology of The Highly Pathogenic Avian Influenza Phase II (D32034)

Ivancho Naletoski and Gerrit Viljoen

The objective of this CRP is to evaluate the origin of wild birds that carry avian influenza (AI) and other potentially dangerous pathogens at their stopover places and match the

obtained results with the knowledge obtained through conventional migration monitoring approaches. Stable isotopes (SI) are promising huge potential when the origin (migration) of individual wild birds is required, because the probability of capturing a labelled bird with specific characteristics (disease carrier) using conventional methods is negligible. Knowledge and experience obtained through the previous project (D32030) will be of great value for the success of this project.

The use of SI in migration studies of wild animals, including wild birds, primarily in environmental protection studies and conservation activities etc. has attracted the attention of the scientific community; however, this can be used in epidemiological studies which target long-range transmissions of animal pathogens.

The development and maintenance of the IAEA Global Network of Isotopes in Precipitation (GNIP) became a significant facilitator of these studies, as it offered geo-spatial reference values for correlation of the SI ratios in the animal tissues (especially metabolically inert tissues like beaks, claws and feathers) and the isotope ratios in the environment (especially open waters).

During the first phase of the IAEA CRP D32030 (Use of Stable Isotopes to Trace Bird Migrations and Molecular Nuclear Techniques to Investigate the Epidemiology and Ecology of the Highly Pathogenic Avian Influenza), several important fragments in the linking of SI ratios of feather samples (bird migrations) with the epidemiology of AI were established.

Achievements of the CRP D32030 have shown not only that the isotope assignment works but have delivered a full package of techniques that will compact and supplement (SI component) the official wild bird monitoring programmes of Member States.

In the current project, the partners will focus on two critical issues: detecting birds that carry avian influenza viruses and eventually other dangerous pathogens and evaluating stable isotope ratios in feathers of these birds (only the pathogen carriers) to understand their origins and migration pathways.

The second research coordination meeting is scheduled to take place in Vienna, Austria, in June 2020.

## Submission of Proposals

Research contract proposal forms can be obtained from IAEA, the National Atomic Energy Commissions, UNDP offices or by contacting a Project Officer. The form can also be downloaded from the URL:

<http://cra.iaea.org/cra/index.html>



## Technical Cooperation Projects

Country TC Project	Description	Technical Officer(s)
Angola ANG5013	Applying Nuclear and Molecular Techniques for Diagnosis and Control of Transboundary Animal Diseases	G. Viljoen I. Naletoski
Burundi BDI5002	Improving Animal Production through Enhanced Application of Nuclear and Related Techniques	I. Naletoski V. Tsuma
Bangladesh BGD5030	Building Capacity to Improve Dairy Cows Using Molecular and Nuclear Techniques	V. Tsuma
Burkina Faso BKF5017	Using Modern Animal Breeding Methods, Nuclear and Genomic Tools to Improve Dairy Production in Smallholder Production Systems	K. Periasamy
Burkina Faso BKF5021	Improving Local Poultry Production through Incorporation of Nutraceuticals in Feeds and Genetic Characterization	G. Viljoen
Bosnia and Herzegovina BOH5002	Strengthening State Infrastructure for Food and Animal Food Control and Protecting Animal Health	I. Naletoski
Botswana BOT5016	Developing the Application of Immunological and Molecular nuclear and Nuclear Derived Early and Rapid Diagnosis and Control of Transboundary Animal and Zoonotic Diseases	G. Viljoen
Belize BZE5009	Establishing Early and Rapid Diagnoses and Control of Transboundary Animal and Zoonotic Diseases	G. Viljoen
Central African R CAF5009	Controlling Contagious Bovine Pleuropneumonia and Peste des Petits Ruminants	H. Unger
Central African R CAF5010	Building National Capacities for the Diagnosis and Control of Animal Diseases and for Increasing Animal Production	H. Unger
Chad CHD5005	Studying the Causes of Pulmonary Diseases in Small Ruminants	H. Unger C. Lamien
Cameroon CMR5019	Using Nuclear Techniques to Improve Milk Production	H. Unger K. Periasamy M. Garcia Podesta
Cameroon CMR5022	Controlling Transboundary Animal Diseases with Special Emphasis on Peste des Petits Ruminants	H. Unger
El Salvador ELS5012	Optimizing Livestock Production Systems through Cultivation and Efficient Use of Local Feed Resources, Monitoring of Performance and Reduction of Environmental Pollution through Solid Waste and Biogas Utilization	I. Naletoski V. Tsuma

Country TC Project	Description	Technical Officer(s)
Eritrea ERI5010	Increasing Small Scale Dairy Production through Improved Feeding, Cattle Management and Higher Conception Rates, Thereby Improving Rural Livelihood and Contributing to Food Security	K. Periasamy V. Tsuma
Ethiopia ETH5020	Enhancing the Livelihood of Rural Communities through Addressing Major Zoonotic and Economically Important Small Ruminant Diseases	H. Unger C. Lamien
Indonesia INS5042	Improving Cattle Productivity through Improved Feeding and Enhanced Reproduction	K. Periasamy V. Tsuma
INT5155	Sharing Knowledge on the Sterile Insect and Related Techniques for the Integrated Area-Wide Management of Insect Pests and Human Disease Vectors	I. Naletoski
Côte d'Ivoire IVC5038	Studying Small Ruminant Respiratory Diseases	C. Lamien H. Unger
Cambodia KAM5003	Supporting Sustainable Livestock Production	M. Garcia Podesta
Kenya KEN5038	Using Nuclear Techniques to Evaluate and Improve the Impact of Mutated Forages on the Performance of Smallholder Dairy Cows	M. Garcia Podesta V. Tsuma
Lao P.D.R. LAO5003	Using Nuclear and Molecular Techniques for Early and Rapid Diagnosis and Control of Transboundary Animal Diseases in Livestock	G. Viljoen
Lao P.D.R. LAO5004	Enhancing National Capability for Crop Production and Controlling Trans-Boundary Animal Diseases	G. Viljoen
Lesotho LES5006	Enhancing Animal Production and the Health of Sheep and Goats in Lesotho	G. Viljoen
Lesotho LES5007	Enhancing Livestock Production and Health	G. Viljoen
Madagascar MAG5024	Applying Nuclear and DNA-Based Techniques to Improve Productivity of Local Livestock Germplasm through an Efficient Artificial Insemination Programme	K. Periasamy
Malaysia MAL5031	Establishing an Environmentally Sustainable Food and Fodder Crop Production System	G. Viljoen
Mauritius MAR5025	Improving the Productivity of Dairy Cattle through On-Farm Application of Achieved Research Information on Feeding Practices	G. Viljoen
Mauritania MAU5007	Supporting Genetic Improvement of Local Cattle Breeds and Strengthening the Control of Cross-Border Diseases – Phase II	M. Garcia Podesta
Mali MLI5026	Improving the Diagnosis of Livestock Diseases	I. Naletoski C. Lamien



Country TC Project	Description	Technical Officer(s)
Mali MLI5027	Using Nuclear and Molecular Techniques for Early and Rapid Diagnosis, Epidemiological Surveillance and Control of Transboundary Animal Diseases	I. Naletoski C. Lamien
Mali MLI5029	Upgrading Capacities to Differentiate Priority Animal and Zoonotic Diseases Using Nuclear Related Molecular Techniques	I. Naletoski
Malawi MLW5002	Strengthening Capacity for the Diagnosis, Prevention and Control of Animal Diseases of Public Health Importance	H. Unger
Montenegro MNE5003	Improving Diagnosis of Animal Diseases and Food Pathogens	I. Naletoski
Mongolia MON5022	Implementing Early Diagnosis and Rapid Control of Transboundary Animal Diseases, Including Foot-and-Mouth disease (FMD) and Peste des Petits Ruminants (PPR)	H. Unger G. Viljoen
Mongolia MON5023	Enhancing Livestock Production through the Improved Diagnosis and Prevention of Transboundary Animal Diseases (TADs)	H. Unger G. Viljoen
Morocco MOR5037	Enhancing Control of Chemical Food and Feed Contaminants, Animal Disease Diagnosis and Trade in Fresh Fruits	I. Naletoski
Mozambique MOZ5007	Enhancing Mutation Breeding of Sorghum and Pearl Millet to Develop High Yield, Disease Resistance and Drought Tolerance	G. Viljoen
Mozambique MOZ5008	Strengthening National Capacity for the Application of Nuclear and Related Techniques to Improve Animal Health and Production	G. Viljoen
Myanmar MYA5024	Supporting the National Foot-and-Mouth Disease Control Programme	G. Viljoen
Myanmar MYA5026	Improving the Livelihoods of Smallholder Livestock Farmers by Developing Animal Feeding Strategies for Enhanced Food Security	G. Viljoen
Nepal NEP5004	Improving Animal Productivity and Control of Transboundary Animal Diseases using Nuclear and Molecular Techniques – Phase II	I. Naletoski
Nepal NEP5005	Strengthening Capacity in Veterinary Diagnosis	I. Naletoski
Nigeria NIR5040	Controlling Parasitic and Transboundary Animal Diseases to Improve Animal Productivity in Smallholder Farms Using Nuclear and Molecular Techniques	I. Naletoski
Pakistan PAK5050	Developing a Facility for the Diagnosis of Transboundary Animal Diseases and Vaccine Production	H. Unger V. Wijewardana
Palestine PAL5007	Upgrading Animal Feeding Laboratory in Terms of Human Capacity Building and Infrastructure	I. Naletoski

Country TC Project	Description	Technical Officer(s)
Papua New Guinea PAP5002	Genetically Characterizing and Improving Productivity of Cattle by Enhanced Reproduction and Better Feeding	K. Periasamy
Papua New Guinea PAP5003	Enhancing Genetic Characterization and Improving Productivity of Cattle by Enhanced Reproduction and Better Feeding – Phase II	K. Periasamy
Peru PER5032	Conducting Genetic Characterization of Alpacas for Resistance to Diseases	K. Periasamy
Congo, Rep. PRC5001	Monitoring Livestock Diseases and Certifying Animal Health	H. Unger
RAF0042	Promoting the Sustainability and Networking of National Nuclear Institutions for Development	I. Naletoski H. Unger
RAF0051	Supporting Specific Needs in the African Region Due to Emergencies	I. Naletoski H. Unger
RAF5068	Improving Livestock Productivity through Strengthened Transboundary Animal Disease Control using Nuclear Technologies to Promote Food Security (AFRA)	H. Unger C. Lamien
RAF5073	Strengthening Africa's Regional Capacity for Diagnosis of Emerging or Re-emerging Zoonotic Diseases, including Ebola Virus Disease (EVD), and Establishing Early Warning Systems	H. Unger I. Naletoski
RAS5078	Enhancing Food Safety Laboratory Capabilities and Establishing a Network in Asia to Control Veterinary Drug Residues and Related Chemical Contaminants	G. Viljoen
RER5023	Enhancing National Capabilities for Early and Rapid Detection of Priority Vector Borne Diseases of Animals (including Zoonoses) by Means of Molecular Diagnostic Tools	I. Naletoski
RER9137	Enhancing National Capabilities for Response to Nuclear and Radiological Emergencies	I. Naletoski
RLA5071	Decreasing the Parasite Infestation Rate of Sheep (ARCAL CXLIV)	K. Periasamy
Senegal SEN5036	Controlling <i>Mycoplasma Mycoides</i> Infection – Contagious Bovine Pleuropneumonia (CBPP) and Contagious Caprine Pleuropneumonia (CCPP)	H. Unger
Seychelles SEY5008	Building Capacity for Diagnosis of Animal Diseases using Nuclear and related Techniques	H. Unger G. Viljoen
Sierra Leone SIL5018	Strengthening Artificial Insemination and Disease Diagnosis Services Coupled with Improved Feeding to Enhance the Productivity of Cattle	H. Unger
Sierra Leone SIL5019	Strengthening Capacities for the Diagnosis and Control of Zoonoses to Improve Public Health Services and Livestock Production	H. Unger
Sri Lanka SRL5045	Establishing a National Centre for Nuclear Agriculture	H. Unger C. Lamien

Country TC Project	Description	Technical Officer(s)
Sri Lanka SRL5046	Improving Livelihoods through Dairy Cattle Production: Women Farmers' Empowerment	M. Garcia Podesta
Sudan SUD5036	Improving Livestock Production for Enhanced Food Security through Genetic Improvement of Indigenous Animal Breeds Using Artificial Insemination, Improved Nutrition and Adequate Animal Disease Control Measures	I. Naletoski M. Garcia Podesta
Syrian Arab Republic SYR5025	Enhancing the Nutritive and Reproductive Characteristics of Small Ruminants by Means of Nuclear and other Related Techniques Using Locally Available Unconventional Feed Resources	G. Viljoen
Togo TOG5001	Improving and Promoting Bovine Milk Production through Artificial Insemination	V. Tsuma
Tunisia TUN5028	Supporting Watering Strategies to Help Livestock Raised in Semiarid and Arid Regions Coping with Climate Change	M. Garcia Podesta I. Naletoski
Uganda UGA5038	Supporting National Animal Production and Productivity through the Establishment of Regional Animal Health Centres and Improving Disease Control at the National Animal Disease Diagnostics and Epidemiology Centre	H. Unger
U.R. of Tanzania URT5031	Improving Indigenous Cattle Breeds through Enhanced Artificial Insemination Service Delivery in Coastal Areas	M. Garcia Podesta V. Tsuma
Vietnam VIE5019	Applying Nuclear Related Techniques for Transboundary Animal Diseases (TADs) Diagnosis	G. Viljoen V. Wijewardana
Yemen YEM5012	Improving Diagnostic and Analytical Capabilities of the Central Veterinary Laboratory Including Residue Testing of Animal Products	H. Unger
Yemen YEM5014	Improving Management of Small Ruminants	H. Unger V. Tsuma
D.R. Congo ZAI5024	Upgrading Vaccine Production to Protect Livestock from Transboundary Animal Disease	H. Unger V. Wijewardana
D.R. Congo ZAI5027	Developing Early and Rapid Diagnosis and Control of Transboundary and Zoonotic Diseases	H. Unger
Zimbabwe ZIM5022	Establishing Molecular Epidemiology Methods, Tissue Culture and Production of Biological Reagents for the Surveillance of Livestock Diseases	I. Naletoski V. Wijewardana
Zimbabwe ZIM5024	Establishing an Artificial Insemination Center to Enhance the Rebuilding of the National Herd	V. Tsuma



# Publications

## Publications in Scientific Journals

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Alam MBB, Omar AI, Faruque MO, Notter D, **Periasamy K**, Mondal M, Sarder M, Shamsuddin M, Jianhua Cao, Xiaoyong Du, Zhenyang Wu, Shuhong Zhao. 2019. Single nucleotide polymorphisms in candidate genes are significantly associated with resistance to *Haemonchus contortus* infection in goats. *J. Anim Sci Biotechnol* 10(30). Doi: 10.1186/s40104-019-0327-8

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Zecchin B, De Nardi M, Nouvellet P, Vernesi C, Babbucci M, Crestanello B, Bagó Z, Bedeković T, Hostnik P, Milani A, Donnelly CA, Bargelloni L, Lorenzetto M, Citterio C, Obber F, De Benedictis P, **Cattoli G**. 2019. Genetic and spatial characterization of the red fox (*Vulpes vulpes*) population in the area stretching between the Eastern and Dinaric Alps and its relationship with rabies and canine distemper dynamics. *PLoS One* 14(3): e0213515. doi: 10.1371/journal.pone.0213515

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## VETLAB Network

The Veterinary Diagnostic Laboratory (VETLAB) Network, coordinated by the Animal Production and Health Section (APH) and supported through IAEA and FAO programmatic activities as well as by South Africa through the African Renaissance Fund (ARF) and by the USA and Japan Peaceful Uses Initiative (PUI), consists of national veterinary diagnostic laboratories located in 45 African and 19 Asia and Pacific Member States.

In the first semester 2019, the VETLAB Network played a key role supporting partner laboratories of Asia and Africa in strengthen and update their diagnostic platforms, preparedness and rapid response actions to important infectious diseases currently threatening livestock economies such as African swine fever, avian and equine influenza, African horse sickness and capripox infections.

For the second half of 2019, two advanced training courses on animal disease detection using multiplex assay

platforms and laboratory test validation will be organized and directly supported by the VETLAB network. In addition, an interlaboratory comparison (ring test) for PPR molecular and serological tests will be conducted.

The fourth VETLAB Directors meeting will take place in Vienna, Austria from 19 to 23 August. Directors from Asian and African countries will participate to this meeting together with experts from international reference laboratories and international organizations. APH is issuing on a regular basis the VETLAB Network Bulletin in the hope of providing a forum for participating laboratories and other stakeholders to communicate and exchange knowledge/information, to showcase achievements and to share expertise within the VETLAB Network.

The latest highlights of the VETLAB Network bulletin can be found on pages 6 and 7 of this edition.

## Impressum

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