Joint FAO/IAEA Programme
Nuclear Techniques in Food and Agriculture

Animal Production & Health Newsletter

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To Our Readers

Members of the symposium team of the Joint FAO/ IAEA Centre of Nuclear Techniques in Food and Agriculture and the IAEA Conference Service Section

Dear colleagues,

We have just concluded our "International Symposium on Sustainable Animal Production and Health – Current Status and Way Forward" with great success (see page 3). It is clear what we need to do in the future to optimize animal production and to mitigate animal diseases. The Symposium was composed of 10 working sessions distributed over five days, discussing and debating technologies, techniques, methodologies, viewpoints and what can be done in the near and medium term to improve livestock productivity and to control zoonotic and transboundary animal diseases. The symposium was attended by nearly 3000 scientists from 169 countries, who enriched the event with their questions and comments. As usual, time constraints during the symposium sessions did

not allow us to directly reply to all questions posted during a session, but each one of them was answered after the sessions.

As all of you know, the symposium was initially planned to take place already in 2020 but the COVID-19 situation forced us to postpone it until 2021 with the hope that travel restrictions would allow an 'in-person' symposium. Unfortunately, the global situation did not improve and therefore we had to implement it in virtual format with all its limitations and restrictions. The symposium ran every day from 10:00 to 15:00 Vienna time to facilitate live participation of guests from all time zones, certainly not an easy task. To ensure that all had access to the symposium material, the recorded individual presentations and full-time sessions were made available to all participants directly after they were presented to facilitate viewers that were not able connect on time. I want to thank one and all for going out of their way to ensure that we had an informative and topical symposium assessing what has been done and providing solutions and a clear way forward as to what needs to change and accomplished.

The Zoonotic Disease Integrated Action (ZODIAC) initiative – the largest technical project of the IAEA since its inception – was launched in June 2020 and will operate under a holistic approach that ranges from mining, identifying, monitoring, tracing, detection and characterization of zoonotic disease pathogens at the environment-animal-human interface, to participation in global intervention and response to a potential outbreak. ZODIAC includes strong technical support, technology packages, guidance, training and provision of equipment to Member States (MSs) and it includes building MS capacities, transfer of technologies, R&D, communication and reporting through its five pillars. Regional meetings with ZODIAC National Coordinators and relevant MS health authorities took place in February–March 2021 to provide project details and mechanisms of MS participation. Moreover, a Consultants Meeting involving 26 senior animal health experts was held in February to obtain scientific and research advice and guidance for strengthening applied research to develop and validate the needed laboratory tools within the animal sector at the animal-human interface. Based on this meeting, four ZODIAC Coordinated Research Projects were developed, one for each region based on regional priorities and regional challenges.

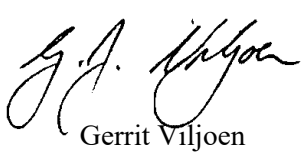
On top of that, the Animal Production and Health Subprogramme continues its focus on enhancing food security by supporting sustainable livestock production systems in developing countries. This is to be achieved by strategic and applied research, technology transfer and capacity building. The three principal components of the subprogramme are animal nutrition; animal reproduction, breeding and genetics; and animal health. Animal production and health problems are identified and solutions developed by strategically applied isotopic, nuclear, nuclear-based and nuclear-derived tools, in conjunction with conventional technologies to:

- Characterize and optimally utilize the nutritional value of locally available feed and feed resources to enhance energy conversion while protecting the environment and minimizing greenhouse gas 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; and
- Assess and reduce the risk of transboundary animal and zoonotic diseases to livestock and livestock owners 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 for the development, validation and dissemination of technologies, know-how and expertise worldwide.

Vaccines against COVID-19 are now available and indeed there is light at the end of the tunnel. It will, however, take several months to get back to some sense of 'normal' and in the meantime, we want to stress that people take care and do not become complacent.

All the best.



Gerrit Viljoen

Head, Animal Production and Health Section

International Symposium on Sustainable Animal Production and Health: Current Status and Way Forward (Virtual Event) 28 June – 2 July 2021

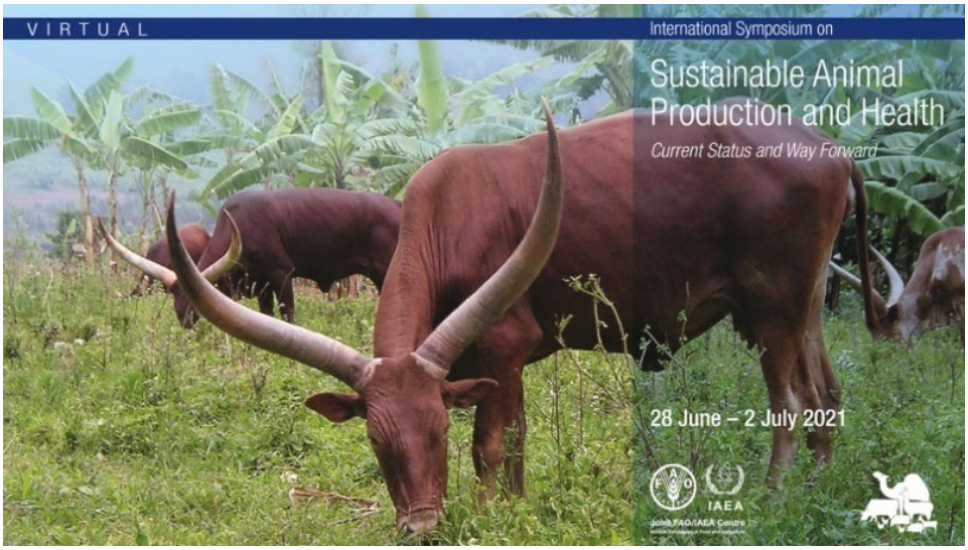
The main objectives of the symposium were to provide information and share knowledge on modern and novel technologies in animal production and health, and their application to support sustainable livestock production systems; identify capacity and research needs and address gaps and new opportunities 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. Meeting participants had the opportunity to discuss the constraints, opportunities and advantages for the effective transfer of laboratory technologies and farm practices.

The Symposium was highlighted by opening remarks of IAEA Director General Rafael Mariano Grossi, FAO Director General QU Dongyu and OIE Director General Monique Eloit. The three Directors General also participated in the commemoration of the 10th anniversary of rinderpest eradication that took place in the afternoon in a special session of the symposium organized by FAO.

The symposium was attended by nearly 3000 scientists, academics, research managers, laboratory personnel, livestock and agriculture policy makers, and donor agencies from national and international organizations, foundations and trusts. Participants were from 169 countries (30.6% from Africa, 28.1% from Asia & the Pacific, 24.6% from Europe and 16.7% from the Americas). The event was composed of nine sessions over five days with technical presentations given by 53 recognized world leading experts which shared their knowledge and expertise on the latest technological aspects in the areas of animal production (especially on nutrition, reproduction and breeding) as well as in the prevention, control and diagnosis of transboundary animal and zoonotic diseases. Of particular interest were the sessions on advances in vaccinology, zoonotic diseases, challenges for better livestock production and molecular tools for animal production and health. In addition, the Book of Synopses containing more than 150 extended abstracts submitted by participants was greatly appreciated.

Presentations highlighted the way forward and what the world can expect from science for the improvement of livestock production in response to the increasing demand for meat and milk; others underscored the challenges and possible strategies for controlling emerging and re-emerging diseases that, in many cases, are driven by the effects of climate change, increased urbanism and globalization. We must consider that the world demands more and healthier animals and animal products produced in an "environmentally safe, clean and ethical" way. We need to coordinate international efforts in order to meet the demands animal-sourced food; to improve the efficiency of production systems in more than 20 livestock species reared under various environment and climatic conditions; and to provide sound disease surveillance that includes national control and eradication programmes for transboundary and zoonotic diseases. The symposium also showed close and solid partnerships between the IAEA with other international organizations like FAO, the World Health Organization (WHO) and the Organisation for Animal Health (OIE) to achieve our goals and objectives, contributing to One Health and the 2030 Agenda for Sustainable Development.

The Animal Production and Health Section and the Scientific Committee of the Symposium thank speakers and participants for their valuable contribution to the event.



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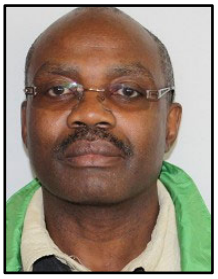
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Joint FAO/IAEA Programme

VETLAB Network Bulletin

02/2021

In this issue:

VETLAB is an initiative of the Joint FAO/IAEA Division

To the readers

In the current pandemic time, veterinary laboratories are working under extreme challenging conditions. In fact, the COVID-19 spread, and the related health control measures and restrictions have a serious impact on the daily work of the laboratories. At the same time, other emerging or re-emerging animal and zoonotic diseases demand for intensified surveillance and control efforts. As highlighted in recent issues of the bulletin, transboundary animal and zoonotic diseases, for example avian influenza, African swine fever, lumpy skin disease, are currently threatening livestock economies and public health around the world. Today more than ever, working together, sharing experiences and expertise are key elements to strengthen our skills and preparedness and successfully go through this difficult period.

It is extremely difficult to exactly predict what the next disease outbreak would be and where it will emerge. What we in the laboratory can - and must - do is to improve our preparedness and make sure we have the proper diagnostic and surveillance tools available and operational. The VETLAB Network is constantly working to assist and support partners in their daily work to fulfil their mission at the highest possible standards.

The team of the APH Subprogramme would like to take this occasion to thank all the VETLAB partners for their contributions to and participation in the International Symposium on Sustainable Animal Production and Health organized by IAEA and held from June 28 to July 2 2021. It has been a great success, with nearly 3000 participants from 169 countries attending the event which included nine sessions on animal health and production policies, transboundary animal and zoonotic diseases, animal genetics, vaccinology and diagnostics. The Proceedings of the symposium will soon be available.

VETLAB Highlights

Support to Capripoxvirus characterization in newly affected and endemic countries

APHL, through VETLAB, has assisted several LSD affected countries in the characterization of local capripoxviruses isolates. Recent examples were the characterization of LSDV in Nepal, Bhutan, Myanmar for newly affected countries, and Uganda and Nigeria for endemic countries. Further, the results of earlier works for the characterization of LSDV in cattle in Bangladesh, wildlife (eland) in Namibia, cattle in East Africa (Ethiopia, Kenya, and Sudan), and GTPV in sheep and goats were published through peer review journals (Ref: 1,2,3).

Detection of highly pathogenic influenza H5N1 in Senegal and Mauritania

In late December 2020 and beginning of 2021, Senegal experienced events of mortality in poultry and wild birds. The Laboratoire National d'Elevage et de Recherches Vétérinaires LNERV (Dakar), thanks to the emergency support of the Agency's VETLAB initiative, analysed the clinical samples using PCR and sequencing, and confirmed H5N1, enabling the veterinary authorities to notify the outbreaks officially to OIE. LNERV also supported Mauritania following episodes of mortality in wild birds in early 2021.

Molecular characterization of ASF in newly affected and endemic countries

The VETLAB supported Burkina Faso and Nigeria to characterize ASFV isolates from recent outbreaks (2019-2021). The two countries experienced at least one major ASFV outbreak in 2020 and 201. Analysis of the recent isolates would help to understand if these outbreaks involved different ASFV strains, as they appeared to have been more significant than the previous ones. The results of similar support provided to Mongolia and Indonesia led to two peer-review publications in international journals (Ref: 4,5).

Comparison of commercially available molecular diagnostic RT-PCR reagents and kits for SARS-CoV-2 detection

Mitigation of SARS-CoV-2 infection requires the availability of accurate and sensitive detection methods. Several commercial molecular diagnostic kits are available in the market. To facilitate selecting alternative options for molecular detection of SARS-CoV-2 in resource-limited settings, APHL, in collaboration with the AGES Laboratory in Mödling, Austria, evaluated 11 assays (eight commercial PCR master mixes and three ad hoc molecular diagnostic kits). The results were recently published (Ref: 6).

02/2021

VETLAB Network Bulletin

VETLAB Capacity Building Initiatives

On-line Preparatory Course on Proficiency tests organisation and management

The VETLAB Network, in collaboration with the Botswana National Veterinary Laboratory (BNVL) in Gaborone, the Central Veterinary Laboratory in Harare-Zimbabwe, and the Enhancing Research for Africa Network (ERFAN), has supported this on-line training. Eighty-one participants from 20 countries attended this on-line training consisting of once-a-week training sessions from 27th April to 8th June 2021.

Training Course for VETLAB Partners on Sequencing and Bioinformatics

The purpose of the training is to strengthen the capacity of the VETLAB partner laboratories in using conventional and new sequencing technologies and the relevant bioinformatic tools for the accurate identification of pathogens. This training specifically aims to introduce the basics of next generation sequencing (NGS) data analysis and phylogenetic reconstructions. The event will be held virtually in November 2021. The detailed format and agenda will follow soon.

VETLAB Networking Activities

Interlaboratory test for the diagnosis of PPR

The yearly interlaboratory comparison (ILC) exercise to assess countries' diagnostic capacity for the accurate detection of PPR has been conducted and panel shipped to 37 veterinary laboratories. Of these, 28 laboratories in Africa, Asia and Europe have provided the results so far. The ILC serology panel was developed in collaboration with IZSAM in Teramo (Italy), which is here gratefully acknowledged.

Coordination Meeting of the Veterinary Diagnostic Laboratory (VETLAB) Network with Directors of African and Asian Veterinary Laboratories

The purpose of the meeting is to update partners on the activities of the VETLAB Network and to discuss the main challenges and gaps in implementing animal and zoonotic diseases diagnosis. The meeting will take place from 11 to 15 October 2021. The venue of the meeting and/or mode of delivery (virtual or face-to-face) will be confirmed based on prevailing COVID-19 conditions.

VETLAB Network Laboratories:

Indonesian Research Centre for Veterinary Science (IRCVS) in Bogor

The IRCVS is a research institute under the Indonesian Ministry of Agriculture which has task and function to conduct research on animal diseases, detection, and control of emerging and re-emerging zoonotic and transboundary animal diseases. It is also designated as the national reference laboratory for animal diseases in Indonesia. For this purpose, IRCVS actively collaborates with other national and international agencies such as IAEA.

With the support of the IAEA-coordinated VETLAB Network, IRCVS successfully detected the African swine fever (ASF) from the clinical cases in North Sumatra and West Java provinces, and isolated the virus in primary cell culture. Sequence analysis indicated that Indonesian isolates belonged to Genotype II, serogroup 8 (Ref: 6). A serological PPR survey was conducted in local goats and sheep by using the ELISA test, and the data are under analysis. Considering the spread of lumpy skin disease (LSD) in neighboring countries, LSD laboratory tests were conducted on clinical suspected samples received from East Java District Livestock Service and Disease Investigation Center Regional Bukit Tinggi (North Sumatra). Samples tested negative by classic and real time PCR. In collaboration with IAEA and the VETLAB Network, the establishment of these methods in IRCVS is helping the laboratory to fulfill its mandates of reference laboratory and supports the government to reduce the risk of introduction in Indonesia of exotic animal diseases.

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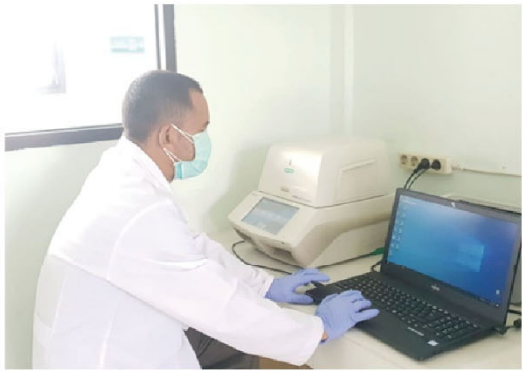

Real time PCR test run at the IRCVS

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Forthcoming Events

Consultancy Meetings on Development of Tools for the Mining, Monitoring and Tracing of Zoonotic Pathogens (Virtual Events)

- in the Americas and the Caribbean Region
- in Asia and Pacific
- in Europe and Central Asia

Charles Lamien and Gerrit Viljoen

Experts on zoonotic diseases will meet virtually in August 2021 to share experience and advise the Joint FAO/IAEA Centre on the implementation of the Zoonotic Disease Integrates Action (ZODIAC) all around the globe.

The purpose of the meeting is to explore the priority disease pathogens and the tools needed for their mining, monitoring, tracing and characterization in order to perform comprehensive field validation of the assays in multiple competent laboratories and prepare standardized operating procedures (SOPs).

Fifth Research Coordination Meeting on Early Detection of Transboundary Animal Diseases to Facilitate Prevention and Control through a Veterinary Diagnostic Laboratory (VETLAB) Network (D32032) (Virtual Event)

Ivancho Naletoski and Charles Lamien

The fifth coordination meeting of CRP D32032 was planned together with the annual meeting of the Directors of the VETLAB Network laboratories. However, it had to be postponed until October 2021 due to the COVID-19 related travel restrictions.

Participants should discuss the methodologies for verification of the produced quality control standards, the dissemination of the multi-pathogen detection platforms, the support from iVetNet in sharing of validated standard operational procedures and the sequencing service of APH among partner laboratories of the VETLAB Network and wider. A presentation with the project summary was prepared for the International Symposium on Sustainable Animal Production and Health, to present the project achievements and future plans to the wider veterinary audience. The meeting is scheduled to be held virtually from 11 to 15 October 2021.

Coordination Meeting of the Veterinary Diagnostic Laboratory Network with Directors of African and Asian Veterinary Laboratories (Virtual Event)

Charles Lamien and Giovanni Cattoli

The purpose of the meeting is to update partners on the activities of the Veterinary Diagnostic Laboratory (VETLAB) Network and to discuss the main challenges and gaps in implementing animal and zoonotic diseases diagnosis.

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

The meeting will be held in parallel with the Fifth Research Coordination Meeting of the VETLAB Coordinated Research Project (CRP) D32032 to allow interactions between the laboratory Directors and the CRP experts and their critical assessment of the CRP's progress.

Owing to the COVID-19 pandemic, the meeting planned to take place in Vienna, Austria, from 22 to 26 June 2020 was postponed. Now it is scheduled to be held virtually from 11 to 15 October 2021.

Technical Meeting on the Use of Laboratory Techniques to Support the Peste des Petits Ruminants Global Eradication Programme (Virtual Event)

Charles Lamien and Giovanni Cattoli

The purpose of the event is to identify and select laboratory diagnostic tests that will contribute to the implementation of the Peste des Petits Ruminants Global Eradication Programme and to enable partners of the Veterinary Diagnostic Laboratory (VETLAB) Network and beyond to discuss diagnosis gaps and collaborative research opportunities.

This meeting was originally planned to take place in Vienna, Austria, in September 2020. Owing to the COVID-19 pandemic, it will now be held virtual in September 2021.

Training Course for Veterinary Diagnostic Laboratory Network Partners on Sequencing and Bioinformatics

Charles Lamien and Giovanni Cattoli

The purpose of the training is to strengthen the capacity of the Veterinary Diagnostic Laboratory (VETLAB) Network partner laboratories in using conventional and next generation sequencing (NGS) technologies and the relevant bioinformatic tools for the accurate identification of pathogens causing transboundary animal and zoonotic diseases.

Owing to the COVID-19 pandemic, the meeting planned to take place in Seibersdorf, Austria, from 22 June to 3 July 2020 was postponed. This training aims to introduce the basics of NGS data analysis and phylogenetic reconstructions. The event will be held virtually from 25 October to 5 November 2021. The detailed format and agenda will follow soon.

RER/5/025 and RAS/5/085: Series of virtual training courses on bio-risk management in veterinary laboratories

Counterparts of the ongoing Regional European (RER/5/025) and the Regional Asian (RAS/5/085) projects, during June and July 2021 will attend a series of eleven training courses on bio-risk management in veterinary laboratories. The organizer of the courses is the European Biosafety association. The courses will cover the following topics: i) Biological Risk Assessment – How safe are we in our labs if we apply the risk based approach according to the new WHO Biosafety Manual?; ii) ISO 35001 – An introduction to the biorisk management standard; iii) Gene Drives: technologies, applications & biosafety challenges; iv) Introduction to vaccinology; v) Auditing for maximum impact – an introduction; vi) Disinfection and Sterilization; vii) Bloodborne viruses and pathogens; viii) Biosafety Culture; ix) Epidemiology - what is it and what do these numbers really mean?; x) Personal Protective Equipment; and xi) Blended learning. Twenty- five participants, mainly biosafety officers, from each of the two projects, will attend the course series.

Past Events

Online Preparatory Course on Proficiency Tests Organization and Management

In collaboration with the Enhancing Research for Africa Network (ERFAN), the VETLAB Network has supported the online preparatory course on organization and management of proficiency tests co-organized by the Botswana National Veterinary Laboratory (BNVL) in Gaborone and the Central Veterinary Laboratory in Harare-Zimbabwe. Ninety-four participants from 24 countries attended this on-line training consisting of once-a-week training sessions from 27th April to 8th June 2021.

The scope of the course was to introduce the basic concepts and terminology as well as ISO standards and international guidelines to organize and conduct interlaboratory comparison and prepare participants to the on-site course expected to be held as soon as COVID-19 restrictions allow. The final aim is to enable participants to acquire laboratory competences linked to the organization of Ring test for serological, bacteriological and molecular assays.

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

Hermann Unger and Gerrit Viljoen

The third and final Research Coordination Meeting on Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers took place virtually from 21 to 23 April 2021. It started with a presentation from the Seibersdorf laboratory of the APH, focusing on the application of irradiated lactobacilli as immune inducers and showcased the favourable effects of such additives when added to vaccines.

The next day the Research Contract Holders (RCHs) gave their presentations. Starting with Ms F. Motamedi-Sedeh on irradiated Influenza H9N2; irradiation with 30 kGy was sufficient to inactivate the virus. A prime / boost application by injection or nasal route proved to protect chicken and the immune profile of the response was tested with several ELISA based assays. This was followed by Mr E.H. Chowdhury on irradiation of avian *Mycoplasma gallisepticum*. The pathogen was successfully isolated and irradiation trials initiated. Unfortunately, the COVID-19 pandemic halted all further work. Next, Ms S. Ahmed presented her work on *Mannheimia haemolytica* (MH) and *Pasteurella multocida*. The irradiated MH. vaccine (20 kGy, Trehalose) was applied to rabbits in parallel with formaldehyde inactivated vaccine (traditional). Results indicate a good immune stimulation compared to traditional approach and showed safety of the product. As the second step, in a sheep trial, the animals were challenged after vaccination. None of the vaccinated animals showed any clinical symptom while non-vaccinated animals suffered the classical symptoms of the disease. For *Pasteurella multocida*, the initial irradiation experiments have been completed.

Next day, Mr T. Eguale presented his work on irradiated Fowl Typhoid (caused by *Salmonella gallinarum*; SG). Irradiation at 2600 Gy caused complete inactivation of SG whereas SG exposed to 2400 Gy showed a better immunogenicity and was safe for chickens. On post-mortem samples taken from chickens that survived the challenge infection until the end of the experiment, SG could not be isolated from the liver, spleen or feces. Protection was only 50% of that of the commercial vaccine, so further attempts to improve the vaccination protocol will be made to determine whether a higher immunity can be achieved. Mr J. Rajapakse presented his results on the irradiated Goat Haemonchosis. The irradiated vaccine worked perfectly and delivered nearly 100% protection. Further work on stability of the vaccine and a large-scale field trial have been initiated. Finally, Mr Sebastian Ulbert gave a presentation on application of E-beam technology on vaccine development.

Second Research Coordination Meeting on Improvement of Diagnostic and Vaccine Tools for Emerging and Re-emerging Animal Health Threats (D32035)

Hermann Unger and Viskam Wijewardana

The Second Research Coordination Meeting on Improvement of Diagnostic and Vaccine Tools for Emerging and Re-emerging Animal Health Threats was held virtually from 19 to 21 April 2021. The purposes of the meeting were to review and update participants on the activities done thus far, and streamline future research activities. The meeting started with two presentations from the Seibersdorf laboratory of the APH, on (i) practical aspects of the tools to measure immunity in general and (ii) specifically on the QPCR immune marker panel. This was followed by presentations on vaccine development by research contract holders: Mr Martin Mwiringi on Sheep & Goat Pox, Ms Diddi Aisatou and Mr Muhhamad Shah on PPR, Mr Molagene Bitew on fowl cholera and Ms Boky Tuasikal on mastitis. Mr Darmawan Darwis from Indonesia presented data on chitosan, an immune stimulator which

could be used as a vaccine adjuvant during mucosal delivery. Finally, Mr Francesco Bonfarnt, a technical contract holder, gave a presentation on an irradiated vaccine developed against low pathogenic avian influenza. His data show the irradiated vaccine is as effective a traditional formalin inactivated vaccine when delivered through the sub-cutaneous route and performs better than the traditional vaccine when delivered through the mucosal route. Next and final RCM for this project will take place in 2023.

Third Research Coordination Meeting on Application of Nuclear and Genomic Tools to Enable the Selection of Animals with Enhanced Productivity Traits (D31028)

Victor Tsuma and Mario Garcia Podesta

The meeting was held as a virtual event from 7 to 11 June 2021. Ten research contract holders (RCH), one technical contract holder (TCH), four agreement holders (AH), four observers and eight staff from the Joint FAO/IAEA Centre attended the meeting.
Research and technical contract holders presented results of individual contracts, discussed the way forward, and agreed to implement the remaining activities originally planned for the CRP. AH and staff from the Animal Production and Health Laboratories of the Joint FAO/IAEA Centre made presentations on the state of the art on application of genomics in the genetic evaluation and breeding of cattle.
Important conclusions of the meeting were:

- Most RCHs had achieved good results in relation to the workplans of the CRP;
- Challenges related to the COVID-19 pandemic affected data collection and DNA sample processing for some countries; and
- It was agreed to (i) complete phenotype record collection and DNA extraction for affected countries by end of September 2021; (ii) increase sample size for genetic wide association studies (GWAS);(iii) complete admixture analysis and assessment of heterosis effects in crossbred cattle; (iv) estimate local genomic estimated breeding values (gEBVs) for the sampled animals and compare with gEBVs estimated on the US or European scale; and (v) disseminate results from the CRP.

The final report for the CRP is expected by the end of December 2021.

Group photo of the participants at the Third Research Coordination Meeting of the CRP D32028

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

Victor Tsuma and Mario Garcia Podesta

The meeting was held as a virtual event from 14 to 18 June 2021. Eight research contract holders (RCH), two technical contract holders (TCH), one agreement holder (AH), seven observers and eight staff from the Joint FAO/IAEA Centre attended the meeting.
RCH and TCH presented results of individual contracts, discussed the way forward discussed, and agreed to implement of the planned CRP activities that were yet to be completed. AH made presentations on the state of the art for quantification of intake and diet selection of grazing ruminants, and reference and near infrared reflectance spectroscopy (NIRS) analysis of feed and faecal material.
Important conclusions of the meeting were:

- Challenges related to the COVID-19 pandemic delayed conducting of the second experiments. Notwithstanding, all experiments were completed, and samples sent to TCHs for n-alkane and NIRS analysis.
- N-alkane and NIRS sample analyses have been completed by the TCHs for most of the countries and results have been sent to some of RCHs; and
- It was agreed that any pending samples be sent to the TCHs, and that remaining n-alkane and NIRS analyses will be done by the TCHs by March 2022.

The final report of the CRP is expected by the end of April 2022.

Group photo of the participants at the Third Research Coordination Meeting of the CRP D32029

Consultancy Meeting on Defining Possible Research Work Incorporating Nuclear and Nuclear-related Techniques to Improve Livestock Feeding in Developing Countries for Sustainable Productivity and Reduced Greenhouse Gas Emissions

Victor Tsuma and Mario Garcia Podesta

The meeting was held as a virtual event from 24 to 28 May 2021. Five consultants from Brazil, Finland, Mexico, Tunisia, and USA and staff from the Joint FAO/IAEA Centre attended.

The objectives of the meeting were to:

- Review the current state of knowledge/advances and research needs in livestock nutrition in the context of climate change;
- Identify sustainable livestock feeding/nutrition strategies to apply in developing countries; and
- Formulate a new coordinated research project (CRP) to enable Member States to use nuclear and related technologies and resources to optimize livestock feeding practices that reduce greenhouse gas (GHG) emissions and help mitigate climate change.

The current knowledge and information available on livestock nutrition in the context of climate change was reviewed. Worldwide, there is renewed interest in greenhouse gas (GHG) emission mitigation from livestock. Many opportunities exist to reduce GHG emissions per unit of product from ruminant livestock, including nutrition and feeding strategies. The meeting agreed that a CRP on "Optimizing nitrogen and energy in cattle feeding for reduced GHG emissions" be formulated. The aim of the CRP would be to enable Member States, especially developing countries, to use nuclear and related technologies and resources to optimize livestock feeding practices that reduce GHG emissions and help mitigate climate change.

First "ZODIAC Senior Consultant Meeting"

Ivancho Naletoski and Gerrit Viljoen

The first "ZODIAC Senior Consultant Meeting" was held online on 22, 24 and 26 February 2021 and focused on the development of the Coordinated Research Projects (CRPs) in pillar 2 ("Developing and making novel technologies available for the detection and monitoring of zoonotic diseases"). The goal of the meeting was to provide scientific and research advice, and guidance that will form the basis for the applied research for the development and validation of the needed laboratory tools to mine, detect and survey zoonotic pathogens at the animal-human interface. The meeting participants have reviewed the activities included in the ZODIAC workplan in order to best reflect the needs for response to zoonotic challenges. This included the identification and selection of advanced research veterinary centres, veterinary institutes, and veterinary laboratories to conduct specific activities of the ZODIAC CRPs.

Twenty-seven experts from national laboratories or international research organizations (Africa Centres for Disease Control and Prevention (African CDC), AU PANVAC, AU IBAR, OIE, FAO, Enhancing Research for Africa Network (ERFAN), European Center for Disease Control (ECDC), WHO, the United States Centers for Disease Control and Prevention (CDC), National Center for Scientific and Technical Research (CNRST)- Morocco, National Institute for Communicable Diseases (NICD) from South Africa, National School of Veterinary Medicine of Sidi Thabet (ENMV) from Tunisia, Commonwealth Scientific and Industrial Research Organisation (CSIRO)-Australia and Central Veterinary Laboratory (LANADA) from Bingerville, Ivory Coast) attended the meeting. Comprehensive recommendations for the research topics were generated at the meeting.

First Consultancy Meeting on Development of Tools for the Mining, Monitoring and Tracing of Zoonotic Pathogens in Africa

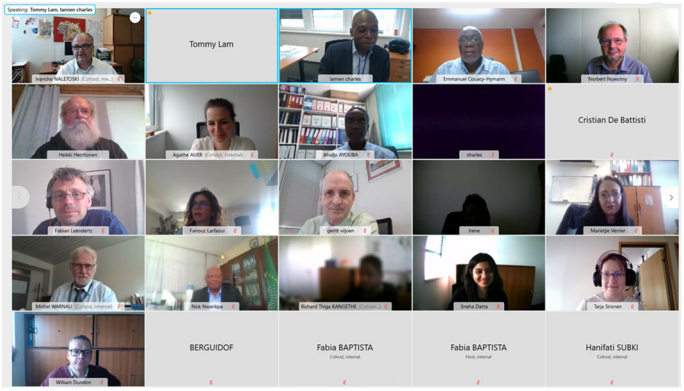
Charles Lamien and Gerrit Viljoen

The first consultancy meeting on the development of tools for the mining, monitoring and tracing of zoonotic pathogens in Africa was held on 31 May, 02 and 04 June 2021. Sixteen experts from Hong Kong, PR of China, France, Germany, Ivory Coast, South Africa, Finland, Norway, UK, and international and Pan African organization (FAO, OIE, AU-IBAR, and AU-PANAVAC)

with scientific experience in the development of tools for detection and characterization of zoonotic pathogens attended the meeting. Based on their experience working on the African continent, the meeting participants have identified the main zoonoses and targeted species as well as the pathogen screening, surveillance, and mining strategies to focus on, in the framework of the CRP, for creating and validating disease detection and surveillance tools. They have also identified several research gaps in diagnostics and surveillance that the ZODIAC CRP could address.
The participants have agreed that the CRP should focus on the following specific objectives:

- Mining/tracing strategies and practical approaches;
- Sample targeting, collection, and submission; and
- Novel approaches or innovative technologies.

The topics of the meeting were mainly focused on the advanced molecular technologies for detection and discovery of pathogens in wildlife carriers, domestic animals as well as arthropod vectors. Number of ideas and recommendations were presented to define the research topics. The team of the Animal Production and Health Subprogramme will collaborate closely with the expert teams during the development and implementation of the coordinated research project for the African region.



Group photo for the participants at the first virtual consultancy meeting on the development of tools for the mining, monitoring and tracing of zoonotic pathogens in Africa

The Second Research Coordination Meeting on the 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 Second Research Coordination Meeting on the 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) took place virtually from 16 June to 18 June 2021.

The purpose of the meeting was to review the activities of the project partners, as well as the samples collected for determination of the presence of the avian influenza in wild migratory birds.
Feather samples were collected from each positive bird in order to assign the geographical origin of the birds / determine their migration pathways. All partners attended the meeting (Canada, Germany, Iran, Republic of Korea, Nigeria, Romania, Russian Federation and United Kingdom) and presented their results. Multiple avian influenza viruses were detected and typed, predominantly of the H5N8 and N5N1 subtypes. Part of the detected viruses were also sequenced using Sanger sequencing or whole genome sequencing. The results of the project were published in national or international journals (Rev Rom Med Vet 2020 30 | 3 : 27 30; Veterinary Today, 2020 . N 2 33), p. 83 89) and at the Annual EU NRL meeting for Avian Influenza and New Castle Disease of poultry at IZSVe, Padova, Italy). Feather samples will be sent for further processing for stable isotope analysis.

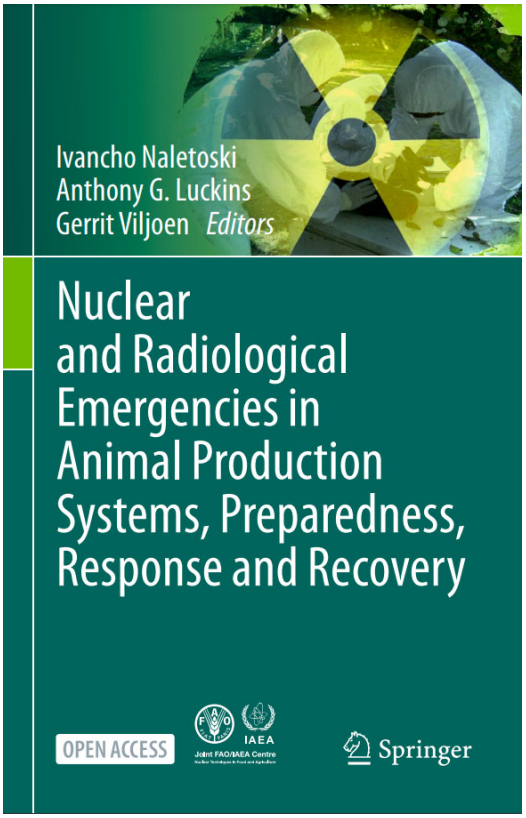
Group photo of the participants at the Second Research Coordination Meeting of the CRP D32034

The manuscript “Nuclear and Radiological Emergencies in Animal Production Systems, Preparedness, Response and Recovery” published as an open source material on the Springer Web site

Animal Production and Health Subprogramme has recently published a manuscript entitled: “Nuclear and Radiological Emergencies in Animal Production Systems, Preparedness, Response and Recovery”. The manuscript covers important aspects of the preparedness, response and recovery; before, during and after nuclear or radiological emergencies in animal production systems. It also contains two annexes with detailed “Datasheets on the Management Options” (actually detailed SOPs of the applicable management

options), as well as "Worked Examples to Illustrate Decision-Aiding Framework". The manuscript is available as an open access publication (free of charge) on the Springer web site: https://link.springer.com/book/10.1007/978-3-662-63021-1.

Authors of the chapters in the manuscript are experienced experts in emergency preparedness for, and response to nuclear and radiological emergencies. We hope that the manuscript will be an important source of professional information for the responsible veterinary operators in Member States.



Cover page of the manuscript: "Nuclear and Radiological Emergencies in Animal Production Systems, Preparedness, Response and Recovery"

Stories

COVID-19 / Grossi Praises IAEA's 'Historic Response' To Pandemic

The International Atomic Energy Agency's "historic response" to COVID-19 in 2020 saw it support 285 national laboratories in 127 countries and territories, and process 1,950 orders for RT-PCR (real time reverse transcription-polymerase chain reaction) equipment and kits, IAEA Director-General Rafael Grossi said in opening remarks to the Board of Governors this June. Member states provided extrabudgetary assistance to the value of €26.3m.

Mr Grossi said "This effort, as the pandemic itself, has not finished," Countries still need help urgently. So far, 128 countries have received IAEA shipments and 274 RT-PCR units had reached the countries of their destination. RT-PCR, is a nuclear-derived technique that can be used in the detection and characterization of viruses.

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Zoonosis: What is a Zoonotic Disease?

Zoonotic diseases are infectious diseases that are transmitted from animals to humans, like COVID-19, bird flu, malaria or Ebola. Zoonotic diseases affect around 2.6 billion people every year. Some zoonotic diseases, such as rabies, only spread from direct animals-to-human contact. Others, more dangerously, start with animals and are capable of causing widespread human-to-human transmission. Nuclear-derived techniques can be used to track pathogens as they move from animals to humans and thus help the world respond better to any future outbreaks.

Click here to watch the video

Experts to Discuss Health and Sustainability of Animal Production at the Symposium

The IAEA International Symposium on Sustainable Animal Production and Health – Current Status and Way Forward organized by the Joint FAO/IAEA Centre of Nuclear Techniques in Food and Agriculture will start on Monday 28 June 2021 and it will draw on lessons learned and current best practices to provide a roadmap for the sustainable improvement of animal production whilst protecting people's health and the environment. The focus of the symposium will be on the contributions and impact of nuclear technologies and applications.

"It is more apparent today than at any other time, that the care we give livestock and the animals we live with, has a direct impact on each and every one of us," said Najat Mokhtar, IAEA Deputy Director General and Head of the Department of Nuclear Sciences and Applications. "At this symposium, we are gathering experts in animal health from across nations, to discuss how nuclear applications can not only improve animal rearing and health, but also enhance human livelihoods."

The International Symposium on Sustainable Animal Production and Health was last held in 2009, and the 12-year gap has left a lot of room for innovation in this year's edition. A completely virtual event and unrestrained by physical limits, the symposium will attract over 2200 experts and attendees from more than 150 countries. An event app allowing participants to engage with one another and the sessions they are attending, will help keep the gathering connected.

This year's symposium will be of interest to animal and public health scientists and other professionals, policy makers, experts from international organizations and associations that deal with animal health, field veterinarians, livestock keepers, veterinary students, and professors in the animal production and health fields. Over five days, the symposium will share knowledge on modern and novel technologies in animal production and health, and their application to support sustainable livestock production systems. Discussions will focus on constraints, opportunities, and advantages for the effective transfer of nuclear and nuclear-derived or related technologies between countries. With more than 50 speakers, leading experts will share their experiences and progress, and identify capacity and research needs to address gaps and opportunities for transferring technologies and building capacities for solving factors that limit livestock productivity and increase animal disease outbreaks, particularly in developing countries.

"As societies develop, so does their demand for animal products," said Gerrit Viljoen, Head of the IAEA's Animal Production and Health Section. The Food and Agriculture

Organization of the United Nations (FAO) projects world meat production to double by 2050, most of which is expected to come from developing countries. “Meeting this demand in a way that’s sustainable, moral, and limits the potential for diseases moving between animals and from animals to humans will require innovation and new technologies. Nuclear techniques and our understanding of how they can be applied to animal production and health have made great strides over the last decades and we’ll be exploring all of these aspects at the symposium,” Viljoen explained.

Covering recent developments in technologies applied to animal production and health, the symposium will dive into topical areas, including immunological and molecular tools for animal production and health; advances in vaccinology; emergency preparedness and response; zoonotic diseases, including COVID-19 and the IAEA ZODIAC initiative; enhancing livestock’s contribution to One Health and the SDGs; challenges for better livestock production in the developing world; advances in biotechnologies for improving livestock breeding and feeding; and the application of improved technologies for sustainable livestock productivity.

Click here to read more

Improving zoonotic disease prevention and livestock production through nuclear-derived techniques

A symposium organized by the Food and Agriculture Organization of the United Nations (FAO) and the International Atomic Energy Agency (IAEA) opened on Monday 28 June 2021 focusing on the prevention of animal disease outbreaks that can cause human pandemics like COVID-19, as well as ways to boost sustainable animal production to feed growing populations.

The week-long International Symposium on Sustainable Animal Production and Health virtually brings together over 2600 international experts in veterinary medicine, genetics and biochemistry, among other scientific fields, to discuss topics such as emergency preparedness and response to outbreaks, advances in animal disease vaccine development and the latest biochemistry tools to improve livestock production, breeding and feed.

"Sustainable animal production and animal health systems are essential to attain the Four Betters – better production, better nutrition, a better environment, and a better life, leaving no one behind," said FAO Director General QU Dongyu. "Protecting animal health under the One Health framework is at the core of our work. This Symposium is an excellent platform to discuss progress, but more importantly, to envisage the future," he added.

"The IAEA's mandate to promote nuclear technologies and their peaceful applications is especially important in health, food and agriculture," IAEA Director General Rafael Mariano Grossi said.

The IAEA Animal Production and Health Laboratory, which is part of the Joint FAO/IAEA Centre of Nuclear Techniques in Food and Agriculture, has carried out vital research and development work to help countries tackle animal and zoonotic diseases such as Avian flu, African swine fever, Zika and Ebola in the past decade. More recently, it has been at the centre of the IAEA's Euro 26 million assistance to 130 countries to use nuclear-derived RT-PCR tests in efforts to contain the spread of COVID-19.

Building on this experience, last year, the IAEA launched the Zoonotic Disease Integrated Action (ZODIAC) initiative to support countries in the use of nuclear and nuclear-derived techniques for the timely detection and control of pathogens at the animal-human interface. "The IAEA is staying present and offering this platform with a nuclear-specific component," Director General Grossi said while recalling past Agency assistance to the international community in battles against significant outbreaks.

"The world is looking to us to produce synergies and provide leadership for a One Health approach that prevents future pandemics originating from animal sources," said FAO Director General QU Dongyu.

The Symposium marks ten years since the successful eradication of rinderpest - the second viral disease to have been defeated globally after smallpox was eliminated in 1980. For centuries, the cattle and wild animal plague seriously threatened food security, especially in Africa and Asia. Its eradication was declared in 2011 by the Paris-based World Organisation for Animal Health (OIE), following an international effort that benefited from FAO and IAEA support to develop tools to quickly detect and efficiently monitor rinderpest cases in the field.

"The eradication of rinderpest is a perfect example of the effectiveness of well-built partnerships," OIE Director General Monique Eloit said in her opening remarks. Surveillance, she added, is an essential component of disease prevention along with vaccines, and the Joint FAO/IAEA Centre plays a key role in this regard,

supporting the overarching goals of the OIE and FAO. Although rinderpest no longer occurs in livestock, the rinderpest virus-containing material is being stored in laboratories of more than 20 countries where it poses a risk through inadvertent or malicious release. FAO is leading the process of reducing the number of laboratories keeping the virus by advocating for and offering assistance to destroy or relocate it to highly secure FAO-OIE rinderpest holding facilities.

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Nuclear-Derived Techniques Help Farmers Combat Cattle Disease Outbreaks in Asia

A disease once limited to the hot and humid climates of Africa has over the last four years appeared for the first time in various parts of Asia and Europe. Afflicting cattle with a debilitating and sometimes deadly condition, the rapid spread of lumpy skin disease is as mysterious as it is alarming. Laboratories of the IAEA and the Food and Agriculture Organization of the United Nations (FAO) are supporting Asian countries in diagnosing lumpy skin disease and identifying different genetic strains of the disease. This is a prerequisite to launching an effective response to this animal disease pandemic.

According to the FAO, the disease causes significant economic losses for farmers, while creating costs to authorities in preventive and control measures. The FAO estimates that the lumpy skin disease outbreak in the Balkans in 2016–2017 directly cost affected countries more than EUR 20 million in vaccinations, compensation for culled and dead animals and aerial fumigation. The economic impact of this now much wider Asian outbreak is yet to be quantified.

“We need to carefully re-look at what the vectors are for this disease. We have some understanding of how it has spread in Africa. We have a weak idea of what the vectors are in Europe, but we are at a total loss at what vectors are causing its spread in Asia,” said Giovanni Cattoli, Head of the Animal Production and Health Laboratory of the Joint FAO/IAEA Centre of Nuclear Techniques in Food and Agriculture. The laboratory team is in direct contact with laboratories, researchers and veterinary authorities in Bangladesh, Bhutan, Cambodia, Indonesia, Nepal, Mongolia, Myanmar, Sri Lanka, Thailand and Viet Nam, trying to help them understand the origins and spread of the virus using the nuclear-derived polymerase chain reaction technique — the same method that is used worldwide to identify and analyse COVID-19.

“The virus is spreading very fast — much faster than could be expected. It emerged in China for the first time in 2019 and after just a year, we already have more than one strain of the virus in Asia,” explained Cattoli.

Global travel restrictions imposed under the pandemic, however, have affected the IAEA’s ability to support the response to the outbreak. When lumpy skin disease first appeared in Bulgaria in 2016, experts were able to travel there to conduct outbreak investigations. This time, Cattoli and his team collaborate online with counterparts in affected Asian countries to give emergency support in the form of equipment, reagents and consumables, and to investigate the outbreaks and analyse the samples submitted to the FAO/IAEA laboratory in Austria. Through the Veterinary Diagnostic Laboratory (VETLAB) network, laboratories in Asia can share data and results in near real-time, and the FAO/IAEA laboratory team can provide comments and suggestions immediately on how countries can improve their procedures.

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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)

Testing and validation of Bactrian SNP panel in the multi-species camelid microarray

In 2020, a multi-species camelid DNA chip was developed by Animal Production and Health Laboratory (APHL) in collaboration with Veterinary Medical University (Austria) and International Camel Genome Consortium. The chip consisted of ~200K markers with >60K from each of dromedary, Bactrian (Figure 1) and new world camelid species. To validate the 60K Bactrian SNP (single nucleotide polymorphism) panel, 96 samples were tested to generate library files that can convert signal data into genotypes. The validation process was successful with extraction of genotypes at more than 51 000 marker loci and a success rate of 86.2%. The thresholds for quality control parameters were set high with DQC>0.82, SNP QC call rate >97%, average call rate for passing samples ≥ 98.5 and percent passing samples ≥ 95. About 67.32% of genotyped markers were classified under PolyHigh Resolution (presence of both homozygotes and heterozygotes), 10.52% under NoMinor Homozygotes (absence of minor allele homozygotes) and 8.36% under MonoHigh Resolution (monomorphic) categories.

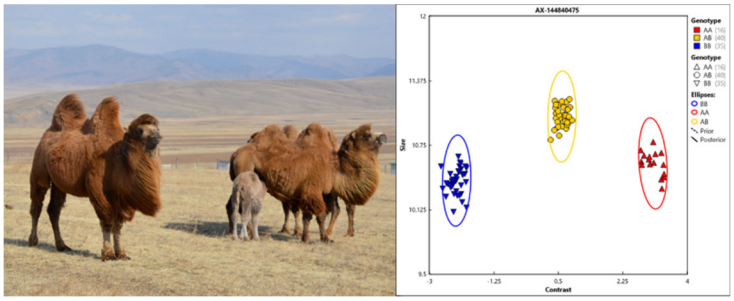

Figure 1. Double humped Bactrian camel of Mongolia (left) and a sample SNP cluster-plot from multi-species camelid microarray (right)

When dromedary specific library files were used to genotype Bactrian camel samples, ~79.8% of markers were successful, of which 6.7% were classified under PolyHigh Resolution and 37% under NoMinor Homozygotes category. This approach can help in generating data on 25K additional markers for Bactrian camels. The successful validation of 60K Bactrian SNPs has now enabled genetic and genome wide evaluation of double humped camel populations. The process of validation and field testing will be continued for other camelid species (Alpaca, Llama and Vicugna) for the use of camel breeders across Africa, Asia and Latin America.

Identification of novel gene markers associated with host resistance and resilience against gastro-intestinal parasites in Argentinian sheep

Selective breeding of genetically resistant animals is considered a promising strategy to face the problem of nematode resistance to anthelmintics and mitigate concerns about the presence of chemical residues in animal food products and the environment. Gastrointestinal nematode resistance is a complex, multifactorial trait related to host immunity. However, the mechanisms underlying host resistance and response to infection remain to be fully elucidated. In this context, a genetic association study was initiated by Instituto Nacional de Tecnología Agropecuaria (INTA), Argentina in collaboration with APHL, Seibersdorf. The objective of this study was to provide insight into the chromosomal regions determining nematode resistance and resilience in Corriedale and Pampinta sheep breeds. A total of 170 single nucleotide polymorphic (SNP) markers from 76 candidate genes related to immune response were discovered by APHL using targeted resequencing approach. Real time PCR based genotyping assays for each of these markers were developed by APHL and transferred to INTA for genotyping and further analysis.

A total of 624 Corriedale and 304 Pampinta lambs underwent artificial or natural challenges with infective larvae mainly from *Haemonchus contortus*. Fecal egg counts, estimated breeding values for fecal egg counts, and rate of packed cell volume change and FAMACHA© score change over the challenge were used, when available, as indicators of host parasite resistance or resilience. Phenotype-genotype association studies were conducted and significance values obtained were adjusted for multiple testing errors. Eight SNPs, located on OARs 3, 6, 12, and 20, reached significance in Corriedale sheep under artificial challenge. Those SNPs represent allelic variants from the MHC-Ovine Lymphocyte Antigen-DRA, two C-type lectin domain families, the Interleukin 2 receptor β, the Toll-like receptor 10, the Mannan binding lectin serine peptidase 2, and the NLR family, CARD domain containing 4 genes. In Pampinta lambs under natural challenge, three significant SNPs located in the TIMP metallopeptidase inhibitor 3, the FBJ murine osteosarcoma viral oncogene homolog, and the Interleukin 20 receptor alpha genes, on OARs 3, 7, and 8, respectively were identified. The novel DNA markers identified in the study can potentially be used for future marker assisted sheep breeding programs in Argentina.

Impact of crossbreeding on host resistance against nematode parasites in Sri Lankan local goats

Uncontrolled crossbreeding of indigenous tropical goats with exotic breeds to increase genetic potentials for growth and body size is a common strategy for genetic improvement. However, in most cases, goats of all genotypes are managed under similar extensive or semi-intensive conditions without corresponding improvements in feeding or health care. Fitness characters such as parasite resistance are believed to be present in indigenous goats and important under typical field conditions but potentially diluted by uncontrolled crossbreeding. A study was therefore designed by University of Peradeniya, Sri Lanka with technical support from APHL, Seibersdorf to compare the characteristics of resistance to gastrointestinal (GI) strongyle parasites in 279 Sri Lankan Indigenous (SLI) and 243 Jamnapari crossbred (JCB) kids. Kids were 4 to 6 months old and maintained under similar management on 68 farms in the Eastern Province of Sri Lanka. Body weight (BW), packed cell volumes (PCV), and fecal egg count (FEC) were determined for each goat at the start of the study and again 7 days later. The sum of FEC across the two measurement times was 1123 ± 333 eggs per gram of feces (epg) for JCB kids and 488 ± 107 epg for SLI kids (P = 0.03), and the breed difference in FEC was larger for females than for males (P = 0.04 for breed x sex interaction). The proportion of goats with FEC > 3000 epg was greater for JCB goats (7.2%) than for SLI goats (1.3%). These results clearly showed that, when infected, the crossbred (JCB) goats were more severely affected than purebred indigenous (SLI) goats.

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

In continuation of Joint FAO/IAEA efforts towards implementing the Global Plan of Action Plan for Animal Genetic Resources (AnGR), APHL supported Member States in at least three major strategic priority areas: characterization, sustainable use and development, and capacity building.

Meta-analysis of cattle from Asia, Africa and Europe (Figure 2): As part of developing baseline reference data and global assessment of native zebu cattle of Asia and Africa, several indigenous breeds were genotyped and sequenced under various coordinated research and technical cooperation projects. Data on short tandem repeat (STR) genotypes, genome-wide single nucleotide polymorphism (SNP) genotypes and mitochondrial control region sequences were generated to evaluate biodiversity, population structure, genetic admixture and genetic relationship of Asian, African and European cattle breeds. As a first step, STR data on >2700 cattle from 73 breeds/populations located across 17 countries were compiled on a master data file. Preliminary analysis on diversity and genetic relationship has been completed and further analysis on population structure and genetic admixture is in progress.

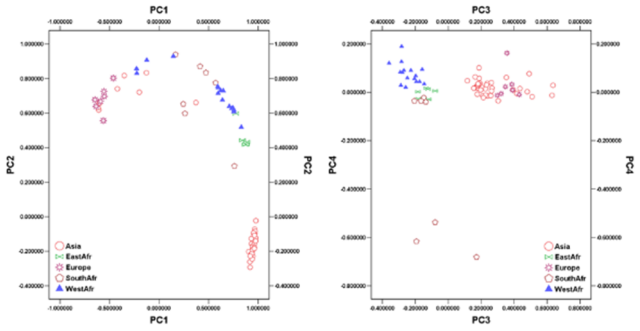

Figure 2. Preliminary results on genetic relationship of cattle from Asia, Africa and Europe

Population structure of endangered indigenous Jaffna sheep of Sri Lanka

Genetic diversity is an essential component in the fitness of a population, for it to survive and adapt to the changing environmental conditions. Baseline information on molecular genetic diversity provides insights into breed history and helps in formulating strategies for conservation and genetic improvement of precious germplasm. Jaffna Local is an isolated and endangered indigenous sheep population in the island country of Sri Lanka. The native tract of Jaffna sheep has close geographic proximity with South Indian sheep breeds. Little or no information is available on diversity, population structure, historic admixture and South Indian ancestry in Jaffna sheep. Hence, University of Peradeniya, Sri Lanka initiated a study on Jaffna sheep in collaboration with Animal Production and Health Laboratory of the Joint FAO/IAEA Centre. The study was aimed (i) to evaluate genetic diversity of Jaffna Local sheep using short tandem repeat markers; (ii) to estimate genetic relationships between Jaffna Local and South Indian sheep breeds; (iii) to assess population structure and genetic admixture in Jaffna Local sheep; and (iv) to evaluate phylogenetic evolution and phylogeography of Jaffna local sheep based on mitochondrial DNA control region variations (Figure 3). A total of 235 sheep were genotyped at 19 short tandem repeat marker loci and sequenced for mitochondrial DNA (mtDNA) control region to assess population structure, genetic admixture and phylogeography.

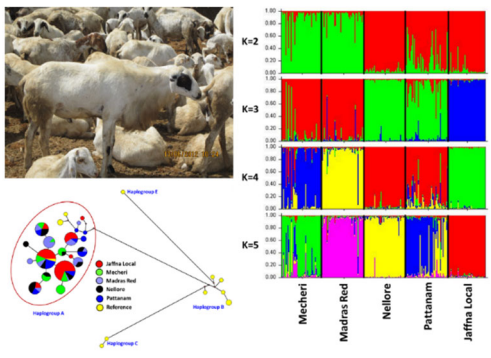

Figure 3. Population structure and phylogeography of endangered Jaffna sheep of Sri Lanka

The results revealed Jaffna Local sheep having relatively low diversity and a high estimated inbreeding coefficient as compared to major South Indian breeds. The pairwise FST showed Jaffna Local sheep having the lowest genetic divergence with Pattanam sheep from South India and the greatest divergence from Mecheri and Madras Red sheep breeds. Bayesian clustering analysis with no prior population information also indicated very little gene flow from South Indian sheep into the Jaffna Local population. Analysis of the mtDNA control region revealed 16 haplotypes in Jaffna Local sheep, of which 10 were observed to be singletons and specific to this breed. The maternal lineages of all the Jaffna Local and South Indian sheep belonged to haplogroup A (HPG-A). The study clearly showed the genetic uniqueness of Jaffna Local sheep. It is imperative to implement selective breeding programmes in the native tract to prevent genetic dilution and foster conservation of this important indigenous genetic resource of Sri Lanka.

Strengthening laboratory infrastructure

APHL continued its efforts to improve the laboratory capacity of Member States and enable implementation of advanced DNA-based technologies for efficient management of locally available AnGR. Equipment required to perform molecular genetic characterization was successfully installed, tested and validated at Seibersdorf laboratory. All the equipment was subsequently shipped to animal genetic laboratories located in four countries viz. Cameroon, Eritrea, Mongolia and Indonesia.

Animal Health

Development of in-vitro assays that measure vaccine immunogenicity in Avian spleenocytes through the use of qPCR (a nuclear related technique)

Measuring immune responses is the key determinant in vaccine efficacy during vaccine development and vaccine quality assessment during the production stage. To assist vaccine research projects in chicken, APHL has undertaken the development of experimental protocols that will measure the expression of Avian immune markers during animal trials and when carrying out in vitro assays using chicken spleenocytes (APHL Newsletter 70, July 2019). The panel under development at Seibersdorf uses cytokine expression analysis by qPCR to add to the existing panels of ruminants and pigs that are available at APHL (Sassu et al., 2020; APHL Newsletter 73, December 2020). Thirty-two targets including cytokines, pathogen pattern receptors, cell surface markers and three calibrator genes were selected from published reports and through communication with collaborators for optimization using spleenocytes that had been stimulated with phorbol 12-myristate 13-acetate (PMA) and Ionomycin in vitro for interleukin production. Control spleenocyte cultures were maintained in the same conditions as treated cells. RNA was subsequently extracted from cells and used in a qPCR assay using SYBR green that measures the targeted interleukins. The conditions for PCR cycling were adopted from previous work (Adams et al., 2009). Activated spleenocytes displayed consistent signature melt curves for 22 of the chosen 32 targets including two calibrator genes (Figure 4). This assay has subsequently been used for measuring immune marker expression in Chicken samples from vaccine animal trials (Avian Influenza and *E. coli*) and will be utilized for future chicken vaccine experiments.

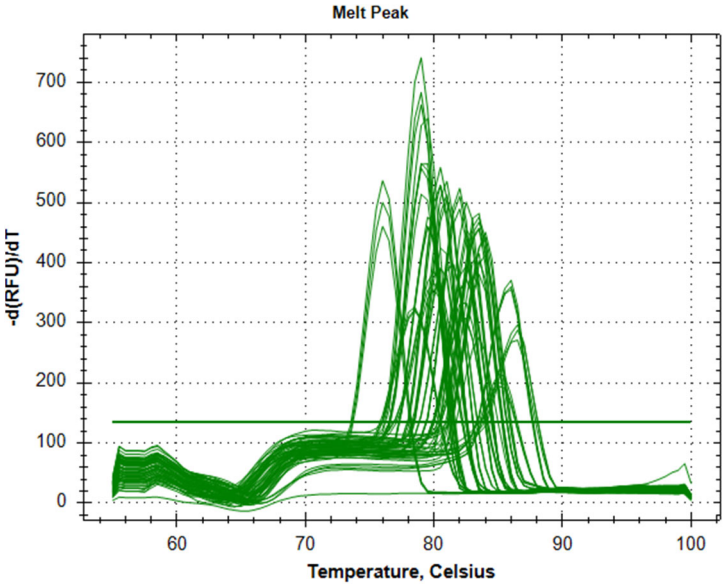

Figure 4. Melt curve analysis of 20 Avian interleukin and surface marker targets along with beta-actin and GAPDH from a qPCR reaction carried out on chicken spleenocytes stimulated using PMA and Ionomycin. The single peaks confirmed that the designed primers amplified specific single products

References:

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2. Adams SC, Xing Z, Li J, Cardona CJ. Immune-related gene expression in response to H11N9 low pathogenic avian influenza virus infection in chicken and Pekin duck peripheral blood mononuclear cells. Mol Immunol. 2009 ;46(8-9):1744-9. https://doi.org/10.1016/j.molimm.2009.01.025.
3. APHL Newsletter 70, July 2019. Page 22.
4. APHL Newsletter 73, December 2020. Page 21.

In-vitro studies of virus host interaction provides further understanding of ASF irradiated vaccine

Over the past decade, African swine fever (ASF) has spread to several European and Asian countries and is still moving further, putting the pig industry and the connected value chains at stake. Different vaccine strategies for ASF have been evaluated in the past decades. Inactivated vaccines, DNA vaccines, subunit vaccines and adenovirus-vectored vaccines have been tested and proved to be unsuccessful. Some gene deleted ASFVs have shown potential as live attenuated vaccines, but it is not known if they could convert to more virulent strains during their replication in pigs.

Despite inactivated vaccines being very efficient at inducing antibodies, on occasion capable of blocking the virus in fluids, they are not very efficient at inducing specific cytotoxic CD8+ T cells (CTLs), crucial for elimination of virus-infected cells. Incorporation of new adjuvant formulations and novel, and more innocuous, inactivation procedures might contribute to designing efficient inactivated vaccines in the future.

A scientific collaboration has been established between the research group at the "Friedrich Loeffler Institut" (FLI), Germany, and APHL for testing what dose of Cobalt60-Gamma-irradiation is required to inactivate African Swine Fever Virus, preserving antigenicity and thus the ability to elicit immune response. The main goal of this collaboration is to test whether an irradiated-inactivated African Swine Fever Virus can be efficient, as a vaccine, in protecting domestic pigs from lethal infection.

From the literature we know that humoral immunity elicited by any type of vaccine against ASF is not enough to confer protection against lethal challenge; in fact, there is still a debate on whether the production of neutralizing antibodies occurs at all. Instead, there is continually more evidence that cell-mediated immunity plays a key role in this regard. Nevertheless, more research is needed to identify which cytokines or patterns are crucial to halt the viral replication or to suppress the immunopathology caused by ASF.

To better understand these aspects, two batches of WT-Estonia/2014 ASF strain were inactivated by two methods: irradiation (30 kGy of gamma-radiation) and chemical inactivation (BEI). The inactivation procedure was validated by standard methods (hemadsorption assay and immune fluorescence analysis). The aim was to test in vitro how irradiated inactivated, chemical inactivated and live ASFV can differently up- or down-regulate a set of immune markers in Swine Peripheral Blood Mononuclear Cells (PBMCs). This set of 32 markers for qPCR analysis include primers targeting genes responsible for innate immunity, adaptive immunity, pathogen-recognition receptors and signal transduction pathway.

From the early results, (Figure 5) gene expression examined through a heatmap analysis, it is evident, by hierarchical clustering, that irradiated ASFV behaves more similarly to the live virus compared to the chemically inactivated one. These in-vitro data suggest that the irradiated virus is immunologically recognized more like a live virus than a killed virus, although it does not cause any active replication. As many irradiated vaccine experiments are conducted in an empirical manner, the methods that are described here could be adopted to analyse irradiated vaccine virus candidates in vitro prior to conducting in-vivo studies.

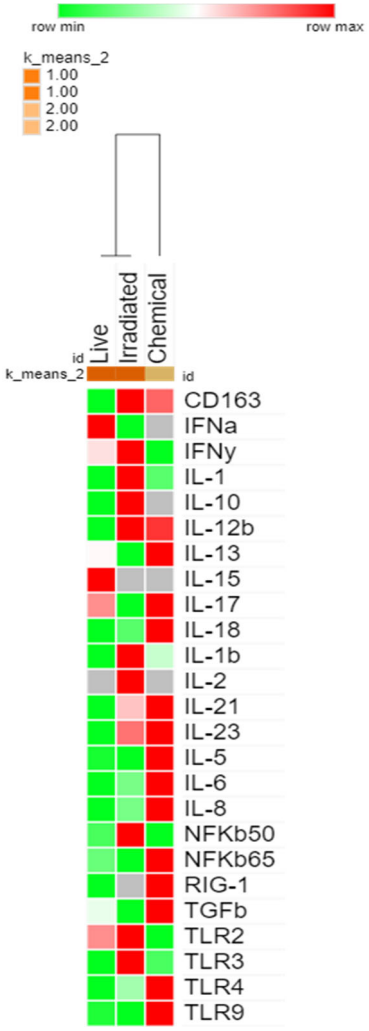

Figure 5. Immune marker gene expression induced by live, irradiated- and chemical-inactivated ASF on Swine PBMCs. Following 2 days of incubation with various stimulants, PBMCs were harvested, total RNA was extracted, cDNA was generated, and gene expression was quantified by qPCR in relation to a house keeping gene. Gene expression was then standardized to non-treated PBMCs and analysed using a heatmap (Morpheus by Broad Institute)

Molecular characterization of LSDV and other capripoxviruses in domestic animal and wildlife

Lumpy skin disease (LSD) is a contagious viral disease of cattle caused by the Lumpy skin disease virus (LSDV), a member of the Capripox virus genus. APHL has continuously supported the diagnosis of capripoxviruses by developing and transferring molecular detection and characterization tools to Member States affected or at risk. In addition, APHL is providing continuous support to all

LSDV-infected countries to characterize their local strains in domestic animals and wildlife and facilitate the differentiation with the vaccinal viruses.

In the last few months, APHL supported the characterization of capripoxviruses collected in several countries in Asia, including Bangladesh in 2019-2020, followed by Viet Nam, Nepal, Myanmar and Bhutan. While analyses are still ongoing in most countries, the initial findings showed the circulation of an LSDV KSGP-0240/NI-2490-like virus in Bangladesh (BMC Vet Res 17, 61, 2021, https://doi.org/10.1186/s12917-021-02751-x), fully characterized by the whole genome sequencing of the virus genome (manuscript in preparation).

APHL also supported the characterization of LSDV from endemic countries in Africa, including both recent (Namibia, Uganda and Nigeria) and archived samples (Kenya, Sudan, Ethiopia). This led to the identification of LSDV in wildlife (Eland, Taurotragus oryx) in Namibia (accepted for publication in the Journal of Wildlife Diseases, https://doi.org/10.7589/JWD-D-20-00181)
(Figure 6). Besides, the uniqueness of a vaccine-like pathogenic LSDV detected in a sample collected in Kenya in 2011, before LSDV expansion to the Middle East and Europe, was further confirmed by targeted next-generation sequencing and published (Microorganisms 2021 , 9(6),1142; https://doi.org/10.3390/microorganisms9061142).

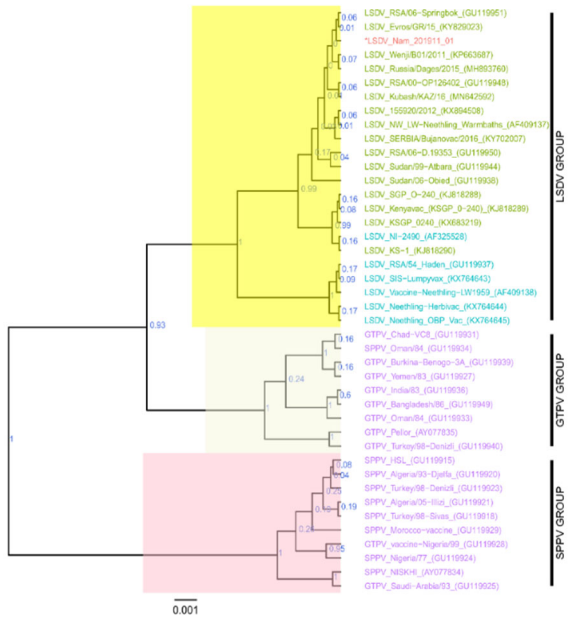

Figure 6. Maximum clade credibility (MCC) tree based on the complete RPO30 gene sequences of capripoxviruses. The posterior probabilities are plotted as respective node labels. The Namibian LSDV sequence from eland is highlighted in red and reference sequences are represented with their accession numbers

Investigations of African Swine Fever virus molecular epidemiology and outbreaks

African swine fever (ASF) is a severe hemorrhagic disease of domestic and wild pigs caused by the African swine fever virus (ASFV). In recent years, ASF has steadily spread towards new geographical areas, reaching Europe and Asia. APHL supported several VETLAB partners in African and Asia in ASFV diagnosis and characterization. Of interest, APHL analysed retrospectively ASFV collected in three West African countries to better understand the virus's evolution.

Hence, ASFV samples collected between 1989 and 2016 in Burkina Faso, Mali and Senegal were analysed and compared to publicly available ASFV sequences. The phylogenetic analysis showed that all viruses belong to genotype I, with the ASFVs from Burkina Faso and Mali grouping with genotype Ia and Senegal genotype Ib (Figure 7). There were four CVR tetrameric tandem repeat sequences (TRS) variants in Burkina Faso, two in Senegal and one in Mali. The three countries did not share any common TRS, and all CVRs of this study differed from previously reported CVRs in West Africa, except for Senegal. Three of the five isolates from Senegal fully matched with the CVR, p72, and p54 sequences from ASFV IC96 collected during the 1996 ASF outbreak in Ivory Coast, suggesting the spread of the same ASFV strains across countries. This study highlights the importance of monitoring ASFV genetic variability at the regional level in West Africa.

Besides the characterization of ASFV in archived samples, APHL also recently supported Burkina Faso and Nigeria to characterize their local strains for 2020 and 2021 outbreaks.

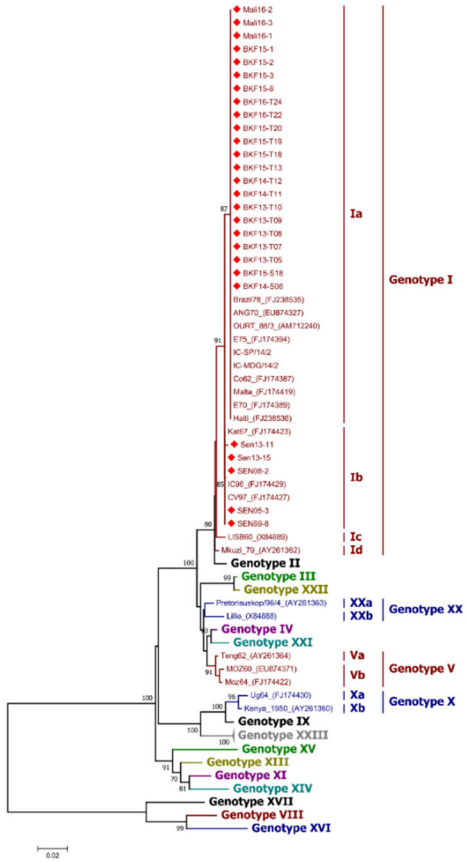

Figure 7. Minimum Evolution tree based on the full p54 gene, depicting the genetic relationships of Burkina Faso, Mali and Senegal isolates with representatives of the known ASFV genotypes. The evolutionary distances were computed using the p-distance method. Only the bootstrap values higher than 70% are shown. The isolates of this study are highlighted with red filled squares before the name

APHL also assisted several Asian countries (Mongolia, Indonesia, Lao PDR and Viet Nam) detect and characterize their local isolates.

After entering China in 2018, ASF reached Mongolia, Viet Nam, Lao PDR and Indonesia (Figure 8) in 2019. The findings on the molecular characterization of ASFVs were recently published for Indonesia (Transbound Emerg Dis. 2021 Mar 16. doi: 10.1111/tbed.14070) and Mongolia (Transbound Emerg Dis. 2021 Apr 5. doi: 10.1111/tbed.14095).

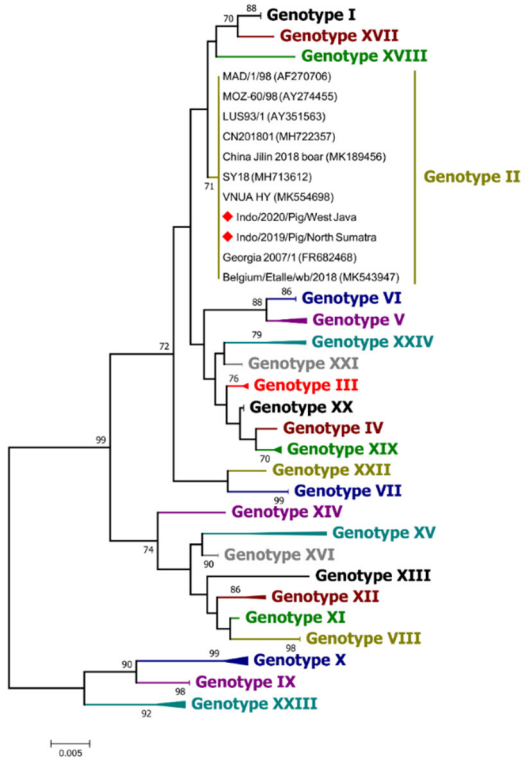

Figure 8. Neighbour-joining tree of the partial p72 gene sequences. The tree shows the genetic relationships of the Indonesian 2019 and 2020 ASFVs with representatives of the 24 known ASFV genotypes. The evolutionary distances were computed using the Maximum Composite Likelihood method. Bootstrap values greater than 70% are shown. The isolates from this study are highlighted with red diamonds

Full genome sequencing of Highly pathogenic influenza H5N6 from Mongolia

In April 2020, following a report on mortalities in whooper swan and swan goose in the Bayankhongor Province and Khovd Province in Mongolia, the State Central Veterinary Laboratory in Ulaanbaatar confirmed Influenza A H5N6 in the samples and requested the VETLAB Network support for the full characterization of the local viruses, which was carried out by APHL.

The eight viral segments were amplified in four samples, using PCR, and the amplicons were used to prepare a sequencing library. The Ion S5 platform was used for sequencing. Following the cleaning ofraw sequences remove low quality and short reads, using FASTQ-MEF (ea-utils), we assessed the quality of the remaining reads with FastQC (v. 011.5). De Novo Assemblies were performed using Spades and CAP3 to produce larger fragments. Using the Denovo assembly's contigs, BLAST searches were performed to determine sequences belonging to an H5N6 isolate from Mongolia (MT872354 to MT87236) as the most relevant references. After mapping the cleaned raw reads against the reference sequences using BWA, we used SAMtools to generate pileup files and performed variant calling using BCFtools. The consensus sequences were produced with vcfutils.pl and seqtk.

A maximum likelihood tree of the HA gene was constructed using MEGA 7, to analyse the HA from the Mongolia AI samples in relation with publicly available sequences.
The result shows that the Mongolian viruses belong to subclade 2.3.4.4h.

Syndromic surveillance of zoonotic abortifacient agents in ruminants

Abortions cause significant economic losses in livestock. Among abortifacient agents are bacteria such as *Brucella* and *Leptospira spp. Listeria monocytogenes* and *Coxiella burnetii* are common agents affecting ruminants, presenting public health importance because of their zoonotic potential. To facilitate the early and accurate detection of these important zoonotic abortifacient agents, a multiplex HRM assay for detecting these four bacteria was developed and tested. This is collaborative research involving APHL and the Botswana National Veterinary Laboratory (BNVL). Primers designed on well-established targets for the PCR detection of *Brucella spp* (IS711), *Coxiella burnetiid* (IS1111), *Leptospira spp* (LiPL32) and L*isteria spp* (ssrA) were used to produce PCR amplicons of different sizes and different GC content. In-silico simulation was performed using the uMelt software (https://www.dna.utah.edu/umelt/umelt.html) to predict the expected PCR amplicon's melting temperatures (Tm) and avoid overlapping or similar Tm between the targeted pathogens. The results showed that each bacterium is detected at a specific and distinct Tm: *Leptospira spp* (75.6-75.8), *Listeria spp* (77.4-77.6), *Coxiella spp* (80.40-80.60), and *Brucella spp* (83.0-83.2). The assay did not detect pathogens such as campylobacter, *Staphilococcus aureus*, Trichomonas, Pseudomonas, Salmonella, Pasteurella, and E. Coli, confirming the specificity.

The assay is undergoing field validation at the BNVL and has also been transferred for evaluation at the Research Center for Veterinary Science, Bogor, Indonesia.

Fellows, Interns and Consultants

Ms Sneha Datta joined the APHL as a Molecular Biologist on 1 December 2020. She is working with the APHL team to create innovative metagenomics approaches for pathogen detection and discovery, including user-friendly data analysis pipelines. Her previous experience in Next-Generation Sequencing, including whole-genome sequencing, RNA sequencing, exome sequencing, shell scripting, and Python programming, will help her to prepare easy-to-go and automated recipes for MS scientists in the framework of the PUI and ZODIAC projects.

Ms Hanifati Subki joined the APHL as an intern on 1 October 2020. She is working on integrating the APHL activities and workflows with the iVetNet platform. Her activities will be essential to the record-keeping and implementing of a quality management system at the APHL for better serving MS.

Mr Federico Verly joined the APH as an intern on 2 November 2020. Holding a Master's degree in International Relations, Mr Verly has assisted in the organization of the International Symposium on Sustainable Animal Production and Health – Current Status and Way Forward by liaising with the contributors, formatting documents, compiling information and updating the Section's database.

Mr Norbert Nowotny joined the Joint FAO/IAEA Centre of Nuclear Techniques in Food and Agriculture as a consultant on 5 October 2020. He supports the team in developing and fine-tuning the ZODIAC project. He comes from the University of Veterinary Medicine, Vienna, where he is the head the Viral Zoonoses, Emerging and Vector-Borne Infections Group.

Ms Irene Mek joined the APHL as a consultant (Laboratory Scientist - Diagnostician) on 6 January 2021. She works on designing and evaluating molecular, serological and virological techniques/tools for detection and surveillance of zoonotic diseases at the wildlife/livestock interface in the framework of the PUI and ZODIAC projects. She also provides technical and scientific support the Veterinary Diagnostic Laboratory (VETLAB) network activities.

Ms Agathe Auer joined the APHL as laboratory specialist and is responsible for the quality control of SOPs and diagnostic laboratory equipment, reagents and consumables for emergency preparedness and response missions for zoonotic infectious diseases in Member States. She has a PhD in Infection Biology and is a certified Quality Manager according to DIN ISO 17025. She supports the APHL team by conducting laboratory testing for the adaptation and re-validation of protocols and comparability of assays.

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
D31029	Quantification of Intake and Diet Selection of Ruminants Grazing Heterogeneous Pasture Using Compound Specific Stable Isotopes	V. Tsuma M. Garcia
D32032	Early Detection of Transboundary Animal Diseases (TADs) to Facilitate Prevention and Control through a Veterinary Diagnostic Laboratory Network (VETLAB Network)	I. Naletoski C. Lamien
D32033	Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers	H. Unger G. 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.Viljoen
D32035	Improvement of Diagnostic and Vaccine Tools for Emerging and Re-emerging Animal Health Threats	H. Unger V. Wijewardana
D31030	Improving Efficiency of Animal Breeding Programs using Nuclear Related Genomic Information – Practical Applications in Developing Countries	V. Tsuma M. Garcia
D32036	Application of Advanced Molecular Characterization Technologies Through the Veterinary Diagnostic Laboratory Network (VETLAB Network)	I. Naletoski C. Lamien
D32037	Novel Test Approaches to Determine Efficacy and Potency of Irradiated and Other Vaccines	V. Wijewardana G.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 project 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 being undertaken, 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 (PTA) of sires under local conditions, which will be correlated with genomic PTAs of sires in their country of origin.

Most research contract holders have completed the work 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 of those animals. The technical contract holder on early pregnancy diagnosis has completed the laboratory work and identified candidate conceptus-derived proteins. Regarding the technical contracts on sequencing dromedary whole genomes by using radiation hybrid (RH) technology, DNA has been extracted from 95 selected hamster-dromedary RH clones and is being sequenced using next generation techniques.

The third research coordination meeting scheduled to take place in Vienna, Austria, from 22 to 26 June 2020, had to be postponed owing to the COVID-19 pandemic and was held virtually from 7 to 11 June 2021.

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

Victor Tsuma and Mario Garcia Podesta

The project aims at developing a practical method to estimate pasture intake of ruminants grazing heterogeneous pastures and rangeland by using stable isotopes, which will provide tools for better grassland management that enhance animal productivity and reduce impact on the 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 agreement holders 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/yaks grazing on pasture/natural grasslands by 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 third research coordination meeting was held from 14 to 18 June 2021.

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 and 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 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 project targets the establishment of such standards for use in serological and molecular diagnostic techniques and aims to 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) Procedures for easy access, free-of-charge genetic sequencing services for pathogens of the priority diseases among the partners of the VETLAB Network; and

v) An information platform for integrated information collection, geo-visualization, analysis and decision making.

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

The fifth research coordination meeting was planned for the week from 22 to 26 June 2020 in Vienna, Austria, however, due to the movement restrictions imposed by the COVID-19 outbreaks, the meeting is tentatively rescheduled for the second half of 2021.

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

Hermann Unger and Gerrit Viljoen

This coordinated research project (CRP) kicked off in early 2017 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 shortcoming of the initial CRP was the lack of proper immunological tools to define the immune response elicited. This issue was addressed by establishing immunology research and development at the APHL 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 project has delivered considerable outputs thus far. Among them, an irradiated vaccine against Goat

Haemonchosis now has reached field level testing in Sri Lanka while irradiated influenza vaccine against for chickens is now being tested in long-term studies in Iran. More details are available under Research Coordination Meeting on Irradiation of Transboundary Animal Disease (TAD) Pathogens as Vaccines and Immune Inducers. This CRP will end in June 2022 and it is anticipated that all counterparts will be able to deliver an experimental irradiated vaccine with the proof of protection and a proper formulation.

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 project 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 - Use of Stable Isotopes to Trace Bird Migrations and Molecular Nuclear Techniques to Investigate the Epidemiology and Ecology of the Highly Pathogenic Avian Influenza) 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, has attracted the attention of the scientific community; however, this technique can also be used in epidemiological studies that target long-range transmission 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 SI ratios in the environment (especially open waters).

During the first phase of the CRP, several important steps in the linking of SI ratios of feather samples (bird migrations) with the epidemiology of AI were established.

Achievements of project D32030 have shown not only that the isotope assignment works but have delivered a full package of techniques that will strengthen 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: (i) detecting birds that carry avian influenza viruses and eventually other dangerous pathogens; and (ii) 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 was planned for the week from 22 to 26 June 2020 in Vienna, Austria, however, due to the movement restrictions imposed by the COVID-19 outbreaks, the meeting is tentatively rescheduled for the second half of 2021.

Novel Animal Vaccine Formulations Enhancing Mucosal Immunity (D32035)

Hermann Unger and Viskam Wijewardana

Background:

Vaccination has proven to be the best preventive measure against infectious diseases. Despite significant successes, there are several limitations to the currently practiced approaches. In veterinary medicine, the application of vaccines by injection limits their use for small ruminants in rural areas. This practice requires well-trained staff taking care to practice the utmost hygiene and maintain a cold chain for the vaccines. It is also not easy to inject individual birds in poultry rearing. Additionally, injected vaccines rarely induce production of specific mucosal antibodies (IgA) covering the mucosal tissues in the nose, mouth and lungs, which are the primary site of multiplication for bacteria or viruses before they provoke a systemic infection. Such IgA antibodies can efficiently be induced by 'mucosal' vaccines, i.e. formulations that are applied to the nose, mouth or eyes. These mucosal vaccines, especially eye drop vaccines, have the big advantage in requiring small volumes as the vaccine dose. Therefore, the application can be done by village vaccinators and the cold chain will be relatively easy to maintain. Recent experiments on formulating such mucosal vaccines for ruminants have presented a number of challenges: low viscosity leading to spills, unsuitable components for freeze drying or the process of formulating the components appropriately. Additionally, the measurement of IgA is still done by a 'research tool' and existing general laboratory tools must be adapted to allow their measurement in standard laboratories.

The expected outcome of this project is the development of several different mucosal vaccine formulations against viral diseases like peste des petits ruminants or influenza or against bacterial diseases like Mycoplasmas or Pasteurella. In parallel, the tools to measure specific IgA induced in the

mucosae will be developed and applied. Experimental combinations of live attenuated viruses together with killed bacterial preparations will be tested to evaluate an enhancing effect of such combinations. The Second Research Coordination Meeting of this CRP was held virtually from 19 to 21 April 2021. Among highlights from this project include the considerable protection induced by application of an irradiated vaccine against low pathogenic avian influenza (H9N2) vaccine through mucosal route in chicken which was done by the technical contract holder in Italian health authority and research organization for animal health and food safety (IZSVe).

Improving Efficiency of Animal Breeding Programs using Nuclear Related Genomic Information – Practical Applications in Developing Countries (D31030)

Victor Tsuma and Mario Garcia Podesta

A new CRP has been launched, whose aim is to enable use of nuclear and related genomic technologies in Member States to enhance the efficiency of national breeding programs for increased milk productivity and dairy animal adaptability to the production environment. Specifically, the CRP aims to (i) develop nuclear and related genomic tools/resources such as radiation hybrid maps and DNA microarrays for tropical dairy species, (ii) identify genomic regions of importance for milk and adaptability traits in local dairy animal populations, (iii) establish strategies to incorporate genomic information for selection and breeding of dairy animals, and (iv) develop and validate radiolabelled biomarker assay for early pregnancy diagnosis in cattle. Three major dairy animal species viz. cattle, buffalo and camel will be targeted.

Up to 10 research contracts will be awarded to institutes submitting appropriate research proposals. Institutions willing to participate in the CRP must have access to national cattle breeding and artificial insemination programs, access to dairy farms/smallholder households and basic laboratory facilities for DNA analysis, and preferably be recipients of collateral financial support from national, bilateral, or international sources. The research contract (RC) will last for five years and individual RC holders (RCH) will receive about €9000 per year to cover costs of local expenses, minor equipment and DNA analysis based on the availability of satisfactory progress report. Four Research Agreements will be awarded to institutes that have expertise in specific areas of importance to the CRP.

To join the CRP, submit your Proposal for Research Contract or Agreement directly to the IAEA's Research Contracts Administration Section (email: research.contracts@iaea.org), using the form templates (http://cra.iaea.org/cra/forms.html) on the CRA web site. For further details please feel free to contact project officers (Victor Tsuma, V.Tsuma@iaea.org or Mario Garcia Podesta M.Garcia-Podesta@iaea.org).

Application of Advanced Molecular Characterization Technologies Through the Veterinary Diagnostic Laboratory Network (VETLAB Network) D32036

Ivancho Naletoski and Charles Lamien

The Animal Production and Health Section (APH) of the Joint FAO/IAEA Centre has established a free-of-charge Sanger sequencing service for all counterparts of the subprogramme. So far, over 4000 samples have been submitted for Sanger sequencing by 30 counterpart laboratories (mainly partners in the VETLAB Network) and the results were published in 27 articles in peer reviewed journals.

APH intends to upgrade this service with additional workflows which should enable counterparts' access to service-based Whole Genome Sequencing (WGS) including the possibility for metagenomic analysis.

Such workflows need to be validated, primarily for biological inactivation of the field samples prior to submission, as well as regarding the quality of the DNA / RNA extracted from the field samples. Additionally, standardized bio-informatic package for processing of the raw data and further phylogenetic analysis needs to be validated and verified for use by the counterpart community. In order to perform these activities, a new Coordinated Research Project (CRP) was developed and approved by the management of IAEA. Priority targets for this CRP will be the established users of the Sanger sequencing service of APH. However, the final objective of the CRP is to further disseminate the validated workflows to the wider counterparts' community.

Novel Test Approaches to Determine Efficacy and Potency of Irradiated and Other Vaccines (D32037)

Herman Unger and Viskam Wijewardana

This new Coordinated Research Project (CRP) should complete the effort in evaluating irradiated and other novel vaccines and their application with innovative tools to determine their immune response and design immunological tools for quality control and efficacy. Thus, applicants must have an ongoing vaccine production and a full-blown and active tissue culture lab to be able to take part in this effort.

The overall expected outcomes are (i) new in vitro procedures for vaccine efficacy testing omitting animal challenge trials based on in vitro stimulation assays employing irradiated antigens; (ii) evaluation of immune marker cytokine specific mRNA qPCR's; (iii) cytokine assays like ELISPOTS or ELISA; and (iv) lymphocyte staining for fluorescent epitope quantification assays. This CRP will not be able to support development of technical capacities but needs the inputs of each participant to understand the immune response delivered by the specific vaccine and the basic methods of their evaluation. It is expected that these new procedures will help vaccine producing labs in future to better perform quality control of their products with a higher confidence in the results due to a more technical approach. The main techniques involved will be lymphocyte tissue culture and stimulation and qPCR of mRNA's. This CRP will start in January 2022.

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 following URL:

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

Technical Cooperation Projects

Country TC Number	Description	Technical Officer(s)
Albania ALB5008	Improving and Enhancing National Capabilities for Early Detection of Vector Borne Diseases through the Application of Conventional and Molecular Methods	I. Naletoski
Angola ANG5016	Recovering the Vaccine Production Unit and Monitoring Active Animal Immunity	V. Wijewardana
Burundi BDI5002	Improving Animal Production Through Enhanced Application of Nuclear and Related Techniques	I. Naletoski V. Tsuma
Burkina Faso BKF5021	Improving Local Poultry Production Through Incorporation of Nutraceuticals in Feeds and Genetic Characterization	V. Tsuma
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 Trans-boundary Animal and Zoonotic Diseases	G. Viljoen
Botswana BOT5018	Reducing the Incidence and Impact of Transboundary Animal and Zoonotic Diseases	C. Lamien
Botswana BOT5021	Improving Reproductive and Productive Performance of Crossbred Dairy Cattle	G. Viljoen K. Periasamy
Bulgaria BUL5017	Enhancing the National Diagnostic Capabilities for Detection of Hepatitis E Virus in Pigs and Pig Products	I. Naletoski
Belize BZE5010	Strengthening national capacities to control animal diseases	G. Viljoen
Chad CHD5008	Improving Bovine Productivity Using Artificial Insemination	V. Tsuma
Cameroon CMR5024	Improving Goat and Sheep Productivity in Rural Areas Using Nuclear-Derived Techniques for Genetic Marker Identification, Reproduction Harnessing and Feed Analysis	V. Tsuma
People's Republic of China CPR5025	Developing Integrated Strategies to Improve Nitrogen Utilization and Production Efficiency in Dairy Cows	G. Viljoen

Country TC Number	Description	Technical Officer(s)
El Salvador ELS5014	Strengthening National Capacities for the Control of Brucellosis	I. Naletoski
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	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 R. Cardoso Pereira J. Bouyer
INT5157	Supporting National and Regional Capacity in Integrated Action for Control of Zoonotic Diseases	I. Naletoski
Côte d'Ivoire IVC5037	Enhancing Diagnostic Capacity for HPAI H5N1 Avian Influenza, using nuclear-derived technique	I. Naletoski
Côte d'Ivoire IVC5038	Studying Small Ruminant Respiratory Diseases	C. Lamien
Cambodia KAM5003	Supporting Sustainable Livestock Production	M. Garcia
Kenya KEN5038	Using Nuclear Techniques to Evaluate and Improve the Impact of Mutated Forages on the Performance of Smallholder Dairy Cows	M. Garcia I. Bimpong
Kyrgyzstan KIG5001	Establishing Effective Testing and Systematic Monitoring of Residues and Food Contaminants and of Transboundary Animal Diseases	I. Naletoski J. Sasanya
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 M. Zaman
Lao P.D.R. LAO5005	Reducing the Incidence and Impact of Transboundary Animal and Zoonotic 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
Lesotho LES5010	Using Nuclear and Molecular Technology to Improve Livestock Production and Health	G. Viljoen
Madagascar MAG5020	Improving Stockbreeding Productivity Through the Application of Nuclear and Related Techniques for Reducing Rural Poverty	I. Naletoski
Madagascar MAG5024	Applying Nuclear and DNA-Based Techniques to Improve Productivity of Local Livestock	V. Tsuma
Mauritania MAU5007	Supporting Genetic Improvement of Local Cattle Breeds and Strengthening the Control of Cross-Border Diseases - Phase II	M. Garcia
Mali MLI5026	Improving the Diagnosis of Livestock Diseases	I. Naletoski

Country TC Number	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
Mali MLI5029	Upgrading Capacities to Differentiate Priority Animal and Zoonotic Diseases Using Nuclear Related Molecular Techniques	I. Naletoski
Mongolia MON5023	Enhancing Livestock Production Through the Improved Diagnosis and Prevention of Transboundary Animal Diseases	G. Viljoen
Mongolia MON5025	Improving Breed Characterization of Cashmere Goats to Facilitate the Establishment of Strategic Breeding Programmes	G. Viljoen
Morocco MOR5037	Enhancing Control of Chemical Food and Feed Contaminants, Animal Disease Diagnosis and Trade in Fresh Fruits	I. Naletoski J. Sasanya C. Blackburn
Mozambique MOZ5008	Strengthening National Capacity for the Application of Nuclear and Related Techniques to Improve Animal Health and Production	G. Viljoen
Mozambique MOZ5009	Strengthening National Capacity to Control the Incidence and Impact of Transboundary Animal and Zoonotic Diseases	G. Viljoen
Myanmar MYA5026	Improving the Livelihoods of Smallholder Livestock Farmers by Developing Animal Feeding Strategies for Enhanced Food Security	G. Viljoen
Myanmar MYA5028	Reducing the Incidence and Impact of Transboundary Animal and Zoonotic Diseases	G. Viljoen
Namibia NAM5018	Strengthening Animal Health and Food Safety Control Systems	G. Viljoen J. Sasanya
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
Nigeria NIR5041	Improving Livestock Productivity through Enhanced Nutrition and Reproduction Using Nuclear and Molecular Techniques	V. Tsuma
Pakistan PAK5052	Improving Livestock Productivity Using Nuclear and Related Techniques by Exploiting Indigenous Feed Resources while Reducing Enteric Greenhouse Gas Emissions	M. Garcia
Palestine PAL5007	Upgrading Animal Feeding Laboratory in Terms of Human Capacity Building and Infrastructure	I. Naletoski
Papua New Guinea PAP5003	Enhancing Genetic Characterization and Improving Productivity of Cattle by Enhanced Reproduction and Better Feeding - PHASE-II	V. Tsuma
Paraguay PAR5011	Improving the Conservation of Germplasm of High Performance Livestock and Native Cattle	M. Garcia
RAF0042	Promoting the Sustainability and Networking of National Nuclear Institutions for Development	I. Naletoski T. Pascual
RAF0051	Supporting Specific Needs in the African Region Due to Emergencies	I. Naletoski G. Viljoen
RAF5068	Improving Livestock Productivity through Strengthened Transboundary Animal Disease Control using Nuclear Technologies to Promote Food Security (AFRA)	C. Lamien

Country TC Number	Description	Technical Officer(s)
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	I. Naletoski
RAF5082	Enhancing Veterinary Diagnostic Laboratory Biosafety and Biosecurity Capacities to Address Threats from Zoonotic and Transboundary Animal Diseases (AFRA)	I. Naletoski
RAS5069	Complementing Conventional Approaches with Nuclear Techniques towards Flood Risk Mitigation and Post-Flood Rehabilitation Efforts in Asia	I. Naletoski L. Heng L. Jankuloski E. Fulajtar
RAS5078	Enhancing Food Safety Laboratory Capabilities and Establishing a Network in Asia to Control Veterinary Drug Residues and Related Chemical Contaminants	G. Viljoen J. Sasanya
RAS5085	Using Nuclear Derived Techniques in the Early and Rapid Detection of Priority Animal and Zoonotic Diseases with Focus on Avian Influenza	I. Naletoski
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 J. Bouyer
RER5025	Improving Early Detection and Rapid Response to Potential Outbreaks of Priority Animal and Zoonotic Diseases	I. Naletoski
RER9137	Enhancing National Capabilities for Response to Nuclear and Radiological Emergencies	I. Naletoski K. Kotus K. Motomitsu
RLA5071	Decreasing the Parasite Infestation Rate of Sheep (ARCAL CXLIV)	M. Garcia
RLA5084	Developing Human Resources and Building Capacity of Member States in the Application of Nuclear Technology to Agriculture	J. Adu-Gyamfi I. Naletoski J. Sasanya W. Enkerlin Hoeflich S. Sivasankar
Senegal SEN5042	Using Nuclear and Related Techniques in Improving the Productivity of Domestic Ruminants	V. Tsuma
Seychelles SEY5008	Building Capacity for Diagnosis of Animal Diseases using Nuclear and related Techniques (Phase I)	G. Viljoen
Serbia SRB5004	Strengthening of National Reference Laboratories Capacities for Early Detection, Epidemiological Surveillance and Control of Transboundary Animal Diseases in Emergency Situations	I. Naletoski
Sri Lanka SRL5042	Applying Molecular Diagnostics to Zoonotic Diseases	C. Lamien
Sri Lanka SRL5045	Establishing a National Centre for Nuclear Agriculture	C. Lamien F. Sarsu M. Zaman
Sri Lanka SRL5046	Improving Livelihoods Through Dairy Cattle Production: Women Farmers' Empowerment	M. Garcia
Sri Lanka SRL5049	Supporting Control of Stomach Worm Infection in Goats	V.Wijewardana
Kingdom of Eswatini SWA5001	Reducing the Incidence and Impact of Transboundary Animal and Zoonotic Diseases	G. Viljoen

Country TC Number	Description	Technical Officer(s)
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	M. Garcia
Tajikistan TAD5006	Applying Nuclear and Molecular Techniques for Diagnosis and Control of Transboundary Animal Diseases	I. Naletoski
Togo TOG5001	Improving and Promoting Bovine Milk Production through Artificial Insemination	V. Tsuma
Togo TOG5003	Improving Livestock Production and Milk Quality Using Artificial Insemination	V. Tsuma
Tunisia TUN5030	Enhancing Feed and Food Safety by Appropriate Management of Livestock Feed Resources for Safer Products	M. Garcia
U.R. of Tanzania URT5031	Improving Indigenous Cattle Breeds through Enhanced Artificial Insemination Service Delivery in Coastal Areas	V. Tsuma
U.R. of Tanzania URT5036	Enhancing Artificial Insemination Services and Application of Radioimmunoassay Techniques to Improve Dairy Cattle Productivity	V. Tsuma
Vietnam VIE5023	Reducing the Incidence and Impact of Transboundary Animal and Zoonotic Diseases	G. Viljoen
Zimbabwe ZIM5024	Establishing an Artificial Insemination Center to Enhance the Rebuilding of the National Herd	V. Tsuma

Publications

Books and Book Chapters

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Metwally S, Viljoen G, El Idrissi A. 2021. Veterinary Vaccines: Principles and Applications. Wiley-Blackwell

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VETLAB Network

The Veterinary Diagnostic Laboratory (VETLAB) Network is a global network of national veterinary laboratories 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). To date, the network comprises 71 laboratories in 45 African and 19 Asian countries and is now working to expand to Central and Eastern Europe, the Caribbean and Latin America. The laboratories work with each other and experts from the Joint FAO/IAEA Centre to use nuclear, nuclear-derived and other methods for monitoring, early detection, diagnosis and control of diseases.

Since the onset of the COVID-19 pandemic, the VETLAB Network has been instrumental to technically supporting laboratories in affected countries. Indeed, in some of these countries, VETLAB laboratories are supporting medical laboratories for SARS-CoV-2 RT-PCR testing. Through the network, emergency support was delivered to the national veterinary laboratories appointed by the respective national health authorities to conduct COVID-19 testing. The emergency packages delivered to these laboratories contained all needed reagents, reference material, consumables as well as major equipment to safely run recommended diagnostic tests based on RT-PCR, including biosafety cabinets and real time PCR platforms. Support was also provided to MS laboratories for the detailed and rapid characterization of SARS-CoV2 isolates by genetic sequencing.

In addition, transboundary animal diseases continue to cause severe losses, such as African Swine Fever (ASF) and Lumpy Skin Disease (LSD) epidemics in Asia. Efforts concentrated on procuring reference material such as positive controls, equipment and reagents for the rapid implementation and expansion of ASF diagnosis and confirmation. Every year the VETLAB Network organizes ring trials, training courses and one meeting of the Directors of African and Asian laboratories. In 2020 and in the first semester of 2021, due to the COVID-19 pandemic and related sanitary restrictions, some of these events had to be postponed and implemented in a virtual format. More information can be found in other sections of this newsletter. We hope to fully resume all VETLAB training activities as soon as possible. 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 issue.

Impressum

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