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*This picture shows the LAMP laboratory at the RCM in Zanzibar*

## To Our Readers

Dear Colleagues,

In this newsletter I want to highlight the biggest event on the animal calendar – Rinderpest is no longer a threat to livestock farmers' world wide. It is expected that FAO and OIE will jointly declare the world to be free from Rinderpest in 2011. In commemoration of this, I want to pay tribute to the members that made this possible such as regional organizations (EC, AU/IBAR CG-Centres etc), international organizations (FAO, OIE, IAEA etc), individual countries (France, Japan, The Netherlands, United Kingdom, United States of America, Italy etc) and the Member States that suffered from this disease and worked towards its eradication. Together with all the role players, the Joint FAO/IAEA Division and the IAEA Technical Cooperation Department's contribution to the development, evaluation and validation of nuclear and nuclear related immunological and molecular diagnostic technologies was a niche and critical area.

The International Atomic Energy Agency (IAEA) has two mechanisms of technical support to Member States - the development, evaluation and validation of nuclear and nuclear related technologies through the Coordinated Research Project (CRP) mechanism, and the transfer and sustainable implementation of the CRP developed technologies through the IAEA's Technical Cooperation Project (TCP) mechanism. The development of nuclear and nuclear related immunological and molecular diagnostic technologies were jointly developed between CRPs of the Joint FAO/IAEA Division and TCPs of the Technical Cooperation Department.

### **The IAEA's nuclear and nuclear-related contribution: the early and rapid diagnoses of transboundary animal diseases**

Central to the success of the rinderpest vaccination campaign was the use of an internationally validated and standardized FAO/IAEA ELISA kit supplied with all the necessary controls, from the FAO rinderpest reference laboratory, the Institute of Animal Health at Pirbright, UK and the FAO/IAEA laboratories in Seibersdorf, Austria. Although vaccination was at the heart of eradication efforts, it would not have been as successful without the development and deployment of diagnostic tests to determine where the disease was, where it was spreading to, which animals were infected and/or at risk and, most importantly, to monitor the efficiency of the vaccination campaigns. Traditionally, animal immune responses to rinderpest vaccination had been assessed by the virus neutralization test. Unfortunately this test is not easily standardized, is relatively expensive and time consuming and was difficult to implement in many veterinary laboratories. It was therefore considered unsuitable both for detecting antibodies to the virus in the many thousands of blood samples required for monitoring vaccination campaigns, for use in epidemiological studies and for detecting the virus itself.

The IAEA through the APH subprogramme of the Joint Division and the TC Department responded to these unmet needs by introducing ELISAs which had all the required attributes to support initially, PARC and eventually throughout all GREP countries. These nuclear related serological tests were initially developed using radio-isotope labelling and tracing techniques but were replaced with enzyme labelling and tracing to circumvent the disadvantages of the short half-life and higher technical proficiency requirements of radioisotopes. Additionally, the new generation ELISA tests could be used in relatively simple veterinary laboratories. It was clear from the outset that the ELISA was an ideal tool to meet the needs of GREP for effective serological surveillance to confirm that sufficient animals were being protected by vaccination to ensure elimination of the virus from national herds.

Once the assay met these international requirements, it was adapted to a kit format linked to a quality assurance programme and provided with a standardized set of laboratory equipment for its routine use. In close collaboration with the developer, these kits (competitive ELISA for the detection of antibodies to rinderpest and immunocapture ELISA for the detection of antigen of rinderpest virus and PPR virus) were developed by the World Reference Laboratory for Rinderpest, the Institute for Animal Health at Pirbright, UK and the World Reference Laboratory for peste des petits ruminants (PPR) at the Institut d'élevage et de Médecine Vétérinaire des Pays Tropicaux (IEMVT), Montpellier, France and standardized and validated by the APH subprogramme of the Joint Division and its associated laboratory at Seibersdorf. These assays were then subsequently disseminated and implemented in Member State laboratories. These kits contained all the required reagents and a standardized protocol. The competitive ELISA for rinderpest antibody detection became the recommended prescribed test by the OIE for international trade. Additionally, agreement was reached on the interpretation of the results and how these could be used to determine the effectiveness of vaccination. A key component of this involved the computerization of the data collected and the analysis of the results. Technical diagnostic test support to the laboratories and scientists working in rinderpest affected countries was provided principally through FAO/IAEA Coordinated Research Projects (CRPs) and IAEA Technical Cooperation Projects and are summarized below:

#### **Contribution of IAEA to Rinderpest Eradication**

The IAEA through the Joint FAO/IAEA Division and the TC Department has supported the programme to eradicate rinderpest for 25 years. The chart below highlights a few specific events over this time-scale. The Joint Division's contribution to the global effort included: developing a network of laboratories to diagnose the disease, organizing training workshops, supplying diagnostic kits and manuals, provision of technical backstopping, producing international guidelines and developing regional TC Projects in Africa and Asia.

- 1986: Animal Production and Health Section of the Joint FAO/IAEA Division incorporates nuclear and related diagnostic techniques (ELISA) into its Coordinated Research Programme (CRP) on animal health. Technical Cooperation support for the programme to eradicate rinderpest commences: in total 20 national TC projects and seven regional projects in Africa and Asia were funded over the next 25 years.
- 1987: First research coordination meeting of the CRP on disease diagnosis using ELISA included six projects on rinderpest. Pan African Rinderpest

Campaign (PARC) initiated; a central requirement was the need for a suitable low-cost diagnostic technique to monitor large numbers of samples for protection monitoring.

- 1987-1990: A CRP involving 14 countries, 'The Sero-monitoring of Rinderpest in Africa', using ELISA is initiated by FAO/IAEA in conjunction with PARC, and with financial assistance from the Swedish International Development Authority (SIDA).
- 1991-1994: A CRP, 'Sero-surveillance of rinderpest and other diseases in Africa using immunoassay techniques, Phase II', initiated by FAO/IAEA/SIDA/PARC establishes antibody monitoring facilities in 21 laboratories.
- 1997-2000: A CRP, 'Rinderpest sero-monitoring and surveillance in Africa using immunoassay technologies' under FAO/IAEA/PARC established in 20 African countries.
- 2001: Technical Information Document, 'Performance Indicators for Rinderpest Surveillance'.
- 2003: Technical Information Document, 'Definition of Technical Guidelines and Standard Operating Procedures for the Surveillance and Testing of Rinderpest as part of GREP'.
- 2010 onwards: Rinderpest support to FAO and OIE (where appropriate and needed), support to Member States regarding Rinderpest Virus sequestration, support to veterinary diagnostic laboratories, support to veterinary and public health

Concerning other news from the subprogramme, we would like to welcome Ivanko Naletoski as the new technical officer in the Animal Production and Health Section. Ivanko brings into the subprogramme his

expertise in the control of animal diseases, in particular the development and use of molecular and serological tools for the study, diagnosis and epidemiology of livestock diseases. Ivanko was the head of the Serological and Molecular Diagnosis Laboratory of the Faculty for Veterinary Medicine of the University of the Former Yugoslav Republic of Macedonia in Skopje. We bid farewell to Len Dimailig who moved to the Department of Nuclear Safety and Security. We thank Len for being there for us when we needed her the most – she was a pillar of strength in the subprogramme.

Both past and future activities are described in detail in this Newsletter and are also accessible at our website (<http://www-naweb.iaea.org/nafa/aph/index.html>); I thus need not mention them in this section. Please contact us if you have any further ideas, comments, concerns or questions. As discussed in previous newsletters, the Animal Production and Health subprogramme will continue to move progressively forward and in pace with developments within the livestock field, to optimally serve our Member States.

Finally, I wish you all and your families a happy, healthy and safe New Year.



Gerrit Viljoen,  
Head, Animal Production and Health Section

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The Animal Production and Health Laboratory, Seibersdorf, is a collaborating Centre for ELISA and molecular technologies in animal disease diagnosis for the OIE.

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## Forthcoming Events

### **Research Coordination Meeting on the Development of Molecular and Nuclear Technologies to Foot-and-Mouth Disease (FMD)**

Technical Officer: Gerrit Viljoen

The first meeting of the Coordinated Research Project (CRP) on the development of molecular and nuclear technologies to foot-and-mouth disease (FMD) will be held from 10 to 14 January 2011 at FAO Headquarters in Rome Italy.

The meeting aims to (i) evaluate, discuss and finalize the details of standardized work plans and protocols of work for the first 18 months of the CRP, (ii) discuss the current situation with regard to *in vitro* testing of animals and assessment of NSP contamination linked to NSP antibody testing, (iii) discuss general activities for the whole life of the CRP including allocation of research tasks and management scheme for each set of tasks to allow reporting and formulate coordination plans, and to (iv) discuss FMD research and formulate coordination plans.

### **Research Coordination Meeting on the Use of Enzymes and Nuclear Technologies to Improve the Utilization of Fibrous Feeds and Reduce Greenhouse Gas Emission from Livestock**

Technical Officer: Nicholas Odongo

The first research coordination meeting on the use of enzymes and nuclear technologies to improve the utilization of fibrous feeds and reduce greenhouse gas emission from livestock will take place at the Agriculture and Agri-Food Canada's Lethbridge Research Centre, Alberta, Canada, from 7 to 11 February 2011.

The meeting aims to (i) evaluate, discuss and finalize the details of standardized work plans and protocols of work for the first 18 months of the CRP, and (ii) to discuss general activities for the whole life of the CRP including allocation of research tasks and management scheme for each set of tasks to allow reporting and formulate coordination plans. During the meeting, presentations on enzyme activity assays (SOP to be implemented on the CRP), batch cultures incubations, enzyme handling and storage, enzyme application methods, *in vitro* methane measurements, microbial protein synthesis using <sup>15</sup>N and purine derivatives, volatile fatty acid measurements, SF<sub>6</sub> for measuring methane production *in vivo*, safety aspects and experimental designs will also be given.

### **Research Coordination Meeting on the Genetic Variation on the Control of Resistance to Infectious Diseases in Small Ruminants for Improving Animal Productivity**

Technical Officer: Mario Garcia

The first research coordination meeting on genetic variation on the control of resistance to infectious diseases in small ruminants for improving animal productivity will take place at the IAEA Headquarters in Vienna, Austria, from 21 to 25 February 2011.

The meeting aims to revise and update work plans of individual Research Contracts and get acquainted with the small ruminant production systems in the participating countries and existing information related to infectious disease resistance. In addition, Agreement holders will present up-to-date laboratory techniques, methodologies, and available sources for identifying, evaluating, and monitoring gene markers, and the Technical Officer will present the existing resources (optimized genes, DNA-markers, sequencing procedures, and nuclear techniques) that have been developed or optimized at Seibersorf's Laboratories. The RCM will focus on evaluating and agreeing on the details of standardized work plans and protocols of work for the next 18 months, and on general activities for the whole period of the CRP; including criteria for selecting 'resistant' and 'susceptible' populations, phenotypic data collection and parasite monitoring.

### **Coordination Meeting of the AFRA Project RAF5057 on Strengthening the Capacities for the Diagnosis and Control of Transboundary Animal Diseases in Africa**

Technical Officer: Hermann Unger

This meeting will be held from 28 March to 1 April 2011 in Uganda

The meeting aims at reviewing the work done so far in the Member States and the adjustments necessary for the future. The development of the laboratory management software 'vet-LIMS' has progressed very well so far and will be presented during the meeting. It should be released shortly afterwards. A special focus will also be on the experience gained in the diagnostics and epidemiology training courses to come up with a decision on which further topics have to be addressed. There are currently 31 African countries participating with 1 more expected to join in 2011.

## Past Events

### Research Coordination Meeting on the Early and Sensitive Diagnosis and Control of Peste des Petits Ruminants

Technical Officers: Diallo Adama; Hermann Unger

The second meeting of the CRP on Peste des Petits Ruminants (PPR) was held in Ouagadougou, Burkina Faso, from 19 to 22 July 2010.

The meeting was attended by three Agreement holders and nine Contract holders. One could not attend but he sent his activities report.



All contract holders reported on PPR outbreaks in their respective countries in the past 2 years, 163 in 2008 in Cameroon, with mortality rate being sometimes as high as 90%. The serosurveillance studies that were carried out showed PPR antibodies prevalence varying between 25 to 76%, the mean being at around 40-46%. Interestingly, in the study area in Pakistan about 60% of cattle and 67% of buffaloes were found positive to PPR antibodies. However no clinical cases were reported in those animals. However in Sudan, PPR virus seems to cause disease in camels. Indeed some pathological tissues collected during respiratory syndrome outbreak in camels were tested positive by gene amplification and also by detection of the PPRV antigen by immunocapture test. One of these positive tissues was ground and the supernatant, inoculated to sheep and goats, caused typical PPR in those animals. At the moment where FAO, the African Union and some countries in Asia are calling for a global control of PPR, it becomes highly important that studies be carried out to evaluate the potential role of buffaloes, cattle and camels in the epidemiology of PPR, results to be considered in the strategies for the control of this disease.

Two partners have reported on the development of an immunochemistry technique for the identification of PPRV in formaldehyde fixed specimen and a LAMP assay for PPR diagnosis. The LAMP, not only being available to be read by naked eye it seems also to be more sensitive than classical reverse transcription assay.

### Consultants Meeting to Develop a Roadmap for the Implementation of Modern OIE Principles and Methods of Diagnostic Test Validation

Technical Officer: Gerrit Viljoen

The meeting was held from 6 to 9 September 2010 in Vienna, Austria.



Diagnostic tests are an essential tool for the early detection and the control of animal diseases. To be confident in the test results, the diagnostic tests carried out need to have been validated fit for the specific purpose(s) for which they are used, in veterinary diagnostic laboratories applying a quality management system.

Following the WAVLD (*World Association of Veterinary Laboratory Diagnosticians*) Pre-Symposium meeting held in Melbourne in November 2007 on validation of diagnostic assays: 'from theory to practice', OIE established an OIE *ad hoc* group on validation of diagnostic assays. This OIE *ad hoc* group had the mandate to revise and to update all the OIE documents related to the validation of diagnostic assays, taking into account the discussions and conclusions of this pre-symposium meeting. The *ad hoc* group met three times between 2008 and 2010. As a result of these meetings, the two chapters on validation of diagnostic assays of the OIE Manuals for Terrestrial and Aquatic Animals were combined into one. The resulting introductory chapter provides concise, comprehensive and

complete general principles and methods on validation of the diagnostic assays for infectious diseases (including immunological and molecular assays). Seven appendices providing useful best practices in areas related to the validation are being finalized to complete this general chapter.

An approach is needed to promote and achieve a common, worldwide understanding of these modern principles and methods on validation of diagnostic assays that were developed in the framework of the OIE ad hoc group and then endorsed by the OIE Biological Standards Commission and finally adopted by the World General Assembly of the OIE Delegates in May 2009.

While it is recognized that all three organizations have their individual training programmes, the network of the Joint FAO/IAEA Division with their focus on building the capacities of diagnostic laboratories could be used. Given this available expertise, training workshops to increase the knowledge and application of the OIE validation principles and methods could be conducted in the immediate future with the global understanding that developed and developing countries should be involved.

As an OIE Collaborating Centre for ELISA and Molecular Techniques in Animal Disease Diagnosis, the Joint division decided to organize jointly with the OIE a consultants meeting for developing a roadmap for the implementation of these principles and methods on validation of diagnostic assays, with a specific focus on developing an approach and framework for training workshops.

## Objectives

The general objective of the consultants meeting was to discuss technical issues for the implementation of the OIE Principles and methods of diagnostic test validation with the more specific objectives:

- 1) To develop a module-type course manual based on the OIE concept, e.g. chapter and annexes including a layout for an answer and question section, which can be used to assess the quality of the course and pass/fail criteria for individual participants.
- 2) To develop an implementation plan for regional training courses and workshops including possible funding mechanisms.
- 3) To identify regional laboratories and individuals, who could serve as trainers, and individuals, who can be trained as trainers.

## Results

A framework for a training manual with key elements was produced. This included a description of the target

audience, identification of 10 training modules and a proposal to conduct these training courses.

The participants agreed that the target audience for the training course should be laboratory and professional staff actively involved in diagnostic test validation or application. This would include staff of the National Reference Laboratories or other responsible institutions which play a strategic role in their country. The tutorial material which would be developed could also interest the diagnostic kits manufacturers.

The participants identified 10 training modules which would cover relevant diagnostic applications. They could be selected all together or just some of them depending of the particular needs of the people trained. The learning content would be a mixture of theoretical information which would be illustrated by practical examples. As much as possible real case scenarios and datasets would apply. OIE validation chapter, best practice appendixes, template and guidelines should be the reference documents.

The 10 modules would be the following:

**Module 1** would introduce the training course pointing out the need for modern diagnostic test validation. It would set the scene and assist in understanding and setting context of diagnostic test validation. Important external relations of test validation such as the link to international quality standards, the relation to IAEA, OIE and FAO and contingency plan requirements would be explained. A glossary of terms and literature/internet references would also be provided.

**Module 2** would describe the six OIE fitness for purpose categories and how these impact on the choice of the method.

**Module 3** would describe the early development and optimization aspects of diagnostic tests.

**Module 4** would describe how to determine early performance characteristics, such as analytical sensitivity and specificity and criteria for potential provisional assay recognition.

**Module 5** would describe advanced criteria for test validation, such as diagnostic sensitivity and specificity and considerations for cut-off selection. It would include epidemiological issues and how these are related to diagnostic test validation.

**Module 6** would describe the importance of reference panel testing to determine the reproducibility of a diagnostic test. It would include the selection of a reference panel, selection of laboratories and analysis of results.

**Module 7** would describe how to assess the usefulness of an assay when applied in other laboratories and for a particular program for which it was designed, e.g. disease eradication campaign. It would include considerations of positive and negative predictive value, prevalence and stage of disease and interpretation of

results. Also included would be operational characteristics of a test such as costs, throughput, turn around time, etc. and integration of the new test into the existing diagnostic repertoire.

**Module 8** would describe the need and how to monitor assay performance. This would be particularly important in the context of end user responsibilities and needs. It would include the use and analysis of internal quality control samples (IQC) and proficiency testing.

**Module 9** would describe test application from the laboratory perspective. It would provide information to understand the status of validation of the test method which would be used in a laboratory and the impact of modifications of diagnostic tests, e.g. change of reagents or change of species on their performance characteristics. Methods to assess the equivalence of different methods would also be described.

**Module 10** would contain a set of statistical applications relevant to the validation process. This would be a specialist module targeted more to statisticians and might include more advanced topics not covered explicitly in other modules.

The participants discussed the potential organisation of these training courses and proposed the following approach. The training courses should be organized in the different regions of the world and be based on a physical meeting supported by a web tutorial and a DVD distributed to the trainees. The OIE collaborating centres could be the preferred venue for these training courses. A first training course could be for example organized with 15-20 participants in Seibersdorf or Teramo for 1-2 weeks depending on the audience proficiency and the number of modules. Experience from this first pilot training course would help to improve the training courses subsequently organized then in each region. There would be a need to include an examination at the end of the course to evaluate the quality of the learning progress.

### Conclusions and recommendations

The participants endorsed the OIE principles and methods for validation of diagnostic assays for infectious disease as these promote a science-based, unified and global approach to test validation and application, provide transparency and support customers such as risk assessors and decision makers.

They also endorsed that appropriate tests for diagnosis require an integrated approach and that it is ultimately the responsibility of the end user to correctly apply diagnostic tests within a laboratory quality management system and to ensure that they are fit for use for the populations in which the tests will be applied.

There was a consensus among participants representing multiple sectors (academia, industry, international organizations, laboratory diagnosticians, national

authorities and research institutes) that the proposed course framework provides a good starting point for broadening the understanding of the principles of validation of diagnostic assays through training courses and other educational activities.

For the benefit of animal disease diagnosis in all countries, the participants proposed to develop training courses based on the first 9 modules described above. A web based application of the OIE validation principles which is currently being developed by the NAHLN network could be used as basis for the development of these course modules. The course plan should include identification of individuals to deliver the first course, a location and date, appropriate attendees from different regions with breadth of expertise (e.g. supervisors of laboratories coming from the different regions of the world), and a course assessment tool. A pilot training course should be organized first. The preparation and organization of these courses would require identification of appropriate and sustainable funding sources.

They proposed that FAO, OIE and IAEA continue to collaborate in promoting the principles of the OIE validation pathway.

Given the increased importance of zoonotic diseases worldwide, the participants emphasized the need to consider future involvement of other stakeholders in the framework of One World One Health.

## Research Coordination Meeting on the Control of Contagious Bovine Pleuropneumonia (CBPP)

Technical Officer: Hermann Unger

The final research coordinated meeting on control of contagious bovine pleuropneumonia (CBPP) took place from 13 to 17 September 2010 in Zanzibar, the United Republic of Tanzania.

This meeting brought together 10 Agreement holders and 5 experts to review the work done during the last 4 years and come up with the results achieved and recommendations on future research work. The scientists from Angola, Botswana, Burkina Faso, Ivory Coast, Kenya, Mali, Namibia, Uganda, Zambia and Zimbabwe presented the summaries of their results obtained in the sero-monitoring of CBPP and the problems encountered. Basically the CBPP c-ELISA was seen as a good test, but on a number of occasions problems with the reference sera were encountered, which had consequences to the quality control of the test. It is regarded as a good epidemiological test for prevalence studies but does not identify vaccinated cattle as positive in most experiments. The LPPQ ELISA was seen as the better test due to its ease of handling, but misses out chronic infections. The standard test for CBPP, the

Complement Fixation Test, was only good in recent infections and it could be shown in post mortem trials, that it misses out on chronic carriers. A newly developed Card Agglutination Test was never compared to a large sample number and so it was decided to evaluate this test for its suitability as a field confirmation test. The molecular diagnosis of CBPP was attempted in a number of laboratories, but due to lack of positive samples requiring the rather cumbersome culture of mycoplasmas, this procedure was not seen as an ideal tool for pathogen detection. The new isothermic LAMP test for CBPP was demonstrated and the participants agreed to test this procedure in their laboratories in order to come up with a standard protocol for the sample preparation and the LAMP procedure.

The expert presentations then shed light on a number of problems still encountered with the control of CBPP. So far nobody had attempted to analyse the presence of IgA after infection or vaccination and its value for assessing protection is unknown. The use of multiple expression antigens as the basis for an indirect ELISA was recently published, but so far no clear cut identification of specific antigens is described for field samples. The identification of specific antigens could help to develop a lateral flow device for field diagnosis. The protective immune response to vaccination is still unclear and it is questionable if a mucosal vaccine could bring major improvements.

Finally two different disease notification systems using mobile phones were presented and the application for CBPP control discussed. This rather rapid information technology might soon find its place in the veterinary field service, to at least document the suspected cases and allow for an appropriate response.

The meeting concluded that this CRP had helped not only to validate some test procedures and create a network of scientists exchanging information, but also created a wealth of scientific data, which should support further research in immunology and vaccinology and finally the control of CBPP.

### **Regional (AFRA) Training Course on Surveillance Technology, ELISA, GIS (RAF5057 005)**

Technical Officer: Hermann Unger

The regional training course took place from 20 to 24 September 2010 in Accra, Ghana.

This final epidemiology training course focused on the interpretation and data management of ELISA results and the reporting of these including the integration of GIS information. After lectures on test methods and test validation procedures, practical sessions on ELISA results, GIS and disease mapping were held. During the course tools to depict spatial distribution of disease

events and meaningful reporting were covered. The interpretation and analysis of serological data including QA was done using Excel spreadsheet functions. Two experts from South Africa trained 23 participants from Burkina Faso, Botswana, Chad, Cameroon, the democratic Rep. of the Congo, Egypt, Ghana, Cote d'Ivoire, Kenya, Libyan Arab Jamahiriya, Mauritania, Mali, Morocco, Niger, South Africa, Sudan, Uganda, Zambia, and Zimbabwe.

### **Regional Training Course on Animal health in Molecular Diagnosis, Genotyping and Phylogenetic Analyses of Avian Influenza (Bird Flu) and Other Mammalian Influenza A Subtypes (RER5015)**

Technical Officer: Adama Diallo

The regional training course took place from 20 September to 1 October 2010 at the Animal Production and Health Laboratory in Seibersdorf, Austria.

The course aimed at enhancing knowledge on early diagnosis and epidemiology tools for highly pathogenic avian influenza (AI) and other mammalian influenza A subtypes (involving the use of nuclear and nuclear-related and molecular technologies), and bioinformatics tools that are required to analyse the data.

Thirteen participants from Bulgaria, Croatia, Greece, Hungary, Kyrgistan, the Former Yugoslav Republic of Macedonia, Romania, the Russian Federation, Serbia and Turkey attended the course and are now partners trained on early and rapid diagnosis, genotyping and phylogenetics of AI and other mammalian influenza A subtypes; the ability to use molecular and nuclear-related tools for the diagnosis and differentiation of AI and other mammalian influenza A subtypes and the capacity of the partners to troubleshoot and interpret their results are strengthened; as well as enhanced capacity of animal disease diagnosis in participating laboratories. The lectures were provided by experts from Austria, Italy, Sweden and Switzerland.

### **EU FMD Week 2010**

Technical Officer: Gerrit Viljoen

The EUFMD Week 2010 was held from 27 September to 1 October in Vienna, Austria.

The FMD Week 2010 brought together around 200 persons involved in FMD science and control issues, and will be used for side meetings of 4 other Projects and Networks. It was also held 'back to back' with the OIE/FAO Annual FMD Reference Labs Network meeting (at Pirbright, UK) to maximize the opportunity and cross-over to bring international surveillance experts together. A side event on 28 September covered the new CRP on FMD. Official EuFMD-Website:

[www.fao.org/ag/againfo/commissions/en/eufmd/eufmd.html](http://www.fao.org/ag/againfo/commissions/en/eufmd/eufmd.html)

## First Research Coordination Meeting on the Use of Irradiated Vaccines in the Control of Infectious Transboundary Diseases of Livestock

Technical Officers: Antony Luckins and Adama Diallo

The first research coordination meeting of a coordinated research project (CRP) on the use of irradiated vaccines in the control of infectious transboundary diseases of livestock was held from 11 to 15 October 2010 in the Vienna International Centre, Vienna, Austria.

It was attended by nine Research Contract holders and three research agreement holders who have extensive experience in vaccine development for helminth and protozoan parasites.



The purpose of the meeting was to allow the participants to present their ideas and approaches for developing attenuated anti-microbial and anti-parasite vaccines by irradiation technology and to discuss with the Research Agreement holders how they could modify and improve the work programmes for the next two years. There was a wide-ranging focus targeting diseases that were particularly problematic for smallholder farmers. The CRP includes studies on *Trypanosoma evansi*, *Theileria annulata*, *Ichthyophthirius multifiliis*, *Brucella abortus*, *B. melitensis*, *Fasciola hepatica*, *F. gigantica* and *Haemonchus contortus*.

The research agreement holders shared their extensive knowledge of radiation attenuation and vaccine production to provide a comprehensive outline of the current status of the technology. Although molecular methods have been used for many years in attempts to create vaccines from defined, genetically engineered antigens, there has been little success, thus, for instance, with helminth parasites, radiation attenuation is still the gold standard for vaccine efficacy. The relative advantages of different forms of irradiation were discussed in relation to the requirements of attenuation.

It was generally agreed that the criteria for attenuation needed to be critically re-examined with each of the pathogens under investigation. A major requirement of this meeting therefore was the need to create work plans that would deliver the basic information required to establish the optimum conditions for attenuation, to devise methods to assess the degree to which the pathogens had been inactivated and to validate their effectiveness as immunogens in protecting animals against infection. These conditions need to be met before it will be possible to develop the technology and apply it under field conditions.

The value of the present CRP lies in bringing together scientists who will be working on different pathogens, thereby providing the means for cross-fertilization of ideas as well as increasing the amount of information acquired.

The CRP will therefore take a leading role in establishing standard operating procedures for the various processes required to develop radiation attenuated vaccines.

### Conclusions

1. The Animal Production and Health Laboratory, Seibersdorf will play a lead role in developing standard operating procedures for attenuation of *T. evansi*. This will involve *in vitro* culture of trypansomes and determination of their metabolic activity following different doses of irradiation.
2. *In vivo* studies on *T. evansi* will take place in Kenya, where initially isolates of the strains known as *T. evansi* A and B will be adapted to *in vitro* culture to provide standard material for infection and attenuation. The early stages of infection in rabbits and goats will be monitored by using sequential biopsies of skin from the site of inoculation and examination of the draining lymph nodes. The effect of different doses of irradiation on the process of establishment in the host will then be studied.
3. Two laboratories, in Georgia and Argentina, will be working in collaboration on *Brucella abortus*. The counterpart in Argentina will work with the current attenuated vaccine strain while in Georgia a strain isolated from the field will be used, for which a macrophage culture system will be established. Initially, irradiation sources will be compared ( $^{60}\text{Co}$ , X-rays and E-beam) along with development of methods to assess the metabolic activity of irradiated bacteria as a means of distinguishing between killed organisms and those that are still competent metabolically. Cell cultures will be prepared from irradiated *Brucella* and eventually, animal testing will be carried out to measure antibody and interleukin production in vaccinated animals, and their resistance to challenge infection.

4. In Sri Lanka, the objectives in the next year will be to determine dose of irradiation that arrests the development of *Haemonchus* larvae in the abomasum wall, assess the potency of irradiated larvae in inducing an immune response in goats and determine resistance to challenge. Among the parameters monitored will be egg counts, haematology, pepsinogen levels and antibody response to ES antigens.
5. There are few reports of irradiated vaccines for *F. hepatica*, so considerable developmental work will be required in Ethiopia. This will entail identifying the optimum attenuating irradiation dose for the larval metacercariae, determining the number of irradiated metacercariae required for oral vaccination and the effect of different vaccination schedules in inducing protective immune responses. An essential component of the research is the production of infective metacercariae using laboratory reared snails. Parameters measured will include antibody responses, cytokine level and *post mortem* findings of fluke development.
6. A similar programme of work is envisaged for *F. gigantica* studies in Sudan. Basic facilities for production of metacercariae will be established, irradiation dosage determined and efficacy of vaccination assessed by measuring antibody levels, cellular immune responses together with liver enzyme changes and associated pathology.
7. The available vaccines for *I. multifiliis* prepared by chemical treatment or freeze thawing are not effective in protecting fish. It is planned to prepare trophont and theront culture facilities to enable generation of material for irradiation. The metabolic activity of the parasites before and after irradiation will be monitored by Q RT-PCR in order to identify the most effective dosage. Fish will be vaccinated with irradiation attenuated parasites and the immune response in serum and at the skin surface compared with that induced by a formalin-killed vaccine. Fish will be challenged to determine efficacy of protection.
8. Complementary studies on *Th. annulata* will be done in Turkey and China. In Turkey, the vaccine source will be schizont infected cultures, while in China piroplasm-infected red cells. In both instances preliminary trials will determine the most effective dose of irradiation to inhibit the development of the parasites prior to their use in vaccination trial in animals.
9. The participants endeavour to keep in contact with the Technical Officer and the research agreement holders to inform on the progress of their work, and should submit a report to the IAEA at the end of the first year.

Participants:

Research Contract holders: Ms Rosemary Bateta, Kenya, Mr Tadesse Eguale, Ethiopia, Ms Ibtisam A.

Goreish, Sudan, Mr Levan Makaradze, Georgia, Ms Gülay Vural, Turkey, Ms Marzieh Heidarieh, Islamic Republic of Iran, Mr Guang-yuan Liu, China, Mr Jayanthe Rajapakse, Sri Lanka, Mr Luis Ernesto Samartino, Argentina.

Research Agreement holders: Mr Alan Wilson, UK, Mr Zhao Rong Lun, China, Mr Palmy Jesudhasan, USA

Other participants: Mr Muhammet Aksin, Turkey, Mr Marika Ramis

## Consultants Meeting on the Use of Stable Isotopes in the Tracing of Wild bird Movements and Correlation with Occurrence of Avian Flu

Technical Officer: Antony Luckins

The meeting was held from 8 to 10 November 2010 in Vienna, Austria.

Influenza is one of the most common infectious diseases in animals and man. There are three genera, two of which, Influenza Types B and C, are predominantly human pathogens, while the third, Influenza A occur in domesticated animal species including pigs, horses and poultry and a wide variety of wild birds as well as humans. Over 100 species of wild migratory birds, particularly ducks, swans, geese and various wading birds, harbour avian influenza (AI) viruses. Infections are transmitted amongst wild birds by shedding of the virus and faecal contamination of water. Most of the influenza strains found in wild birds are of low pathogenicity producing only mild disease in domesticated birds, whilst wild birds are unlikely to become sick. Of considerable concern is the emergence in birds of a highly pathogenic strain of Influenza A virus, Highly Pathogenic Avian Influenza (HPAI) of the H5N1 subtype (HPAI, H5N1). Animal influenza viruses threaten animal health, livestock productivity and food security in poor countries, but they can also evolve into dangerous human pathogens. This has been seen with HPAI. Its main impact has been on domesticated poultry, but a number of fatal human infections have also occurred. The International Scientific Task Force on Avian Influenza and Wild Birds reported that waning attention to HPAI was reducing opportunities for surveillance and research, thereby affecting efforts to understand the epidemiology of the disease. Nonetheless, the disease continues to be a major problem in Egypt and parts of Asia and recently outbreaks have occurred in poultry in Romania and in wild birds in the Russian Federation, China and Mongolia. There is a need to improve knowledge of the role wild bird populations might play in the dissemination of infection. Tracing the movements of wild birds in relation to where they originated as well as their stopover points in their migration between breeding and non-breeding grounds is a challenging task. There has been some progress in this field by using satellite tracking, geo-

locators or external markers of wild birds, but these procedures have only been applied to relatively few species and individuals, and it will be necessary to utilize methods that can be used on a larger scale and not biased to the initial capture location if we are to fully understand the role of migratory birds in the spread of avian influenza. A technique that has already been used to trace migrants is based on the stable isotope signatures of tissues of birds, especially those in feathers. Stable isotopes are involved in important biological and ecological processes and there is a strong correlation between levels of these isotopes in the environment and the concentration of the same isotopes in avian tissues. Of most interest are deuterium ( $\delta D$ ) ratios in tissues that reflect those in surface (lakes, rivers, oceans) and ground waters. Since hydrogen isotope composition of environmental water varies spatially across the globe in a predictable manner and its presence relayed to animal tissues,  $\delta D$  analyses of animal tissues provides a way of linking isotope data on water isoscapes with those in biological tissues such as feathers. These isotope data have been shown to successfully reveal migration patterns and enable identification of the breeding areas of birds sampled at non-breeding grounds and disease outbreak sites.

The measurement of naturally occurring stable isotopes in avian tissues to provide information on origins of individuals is based on matching knowledge of spatial isotopic patterns in foodwebs (isoscapes) with those in tissues such as feathers. In North America and Europe, deuterium feather isotope measurements reflect those expected from the long-term IAEA GNIP dataset. By combining stable isotopic measurements of bird tissues and assays of HPAI H5N1 and other pathogens, it will be possible to investigate spatial patterns of transmission. For Africa and Asia, correlations between the GNIP precipitation deuterium isoscape and avian tissues have not been established. It will be necessary to calibrate the feather-precipitation  $\delta D$  relationship for these two continents for waterfowl and provide a critical tool in the use of an isotopic forensic tool to investigate HPAI transmission involving wild birds. This can be accomplished by measuring waterfowl tissues known to be grown at specific sites along their flyway.

In order to relate bird movements with outbreaks of HPAI it will be necessary to: - evaluate the potential of precipitation deuterium isoscapes based on the IAEA GNIP program for determining origin of waterfowl in Africa/Asia by calibrating this isoscape with feathers grown at known origins; utilize species-specific telemetry and other marking data to identify waterfowl flyways including stopover, breeding, non-breeding and moult locations in Africa/Asia and use these data to inform isotopic sampling strategies and evaluate tissues from waterfowl (feather and blood) samples previously collected and identify key samples for isotope analysis to support the first two objectives. For historical anal-

yses of samples, analysis of 35 feathers from the same bird species, over the time period when they are arriving to their overwintering site, would be the goal. Analyses need to focus on  $\delta D$  measurements of feathers of known moult chronology and, potentially, of claws.

The selection of countries that would be ideally included in a future CRP is: Mongolia, China, Bangladesh, Myanmar, India, Kyrgyzstan, Tajikistan, the Republic of Korea, Japan, Egypt, Kenya, Niger, Nigeria, Mali, and Senegal. Standard operating procedures for collection and handling of samples should be provided in advance of any further work in the field.

Given that global wildlife surveillance for HPAI H5N1 has shown only a handful of positive results among the 750,000 samples collected to date, it is recommended that AI surveillance should not be limited to HPAI H5N1 but should entail complete screening for likely H and N subtypes. Furthermore, in the context of the next pandemic risk, the next influenza virus that will impact livestock and livelihoods, it is likely not to be an H5N1 virus but another subtype.

The consultants recommended that the Bar-headed Goose be used as a focal species for a CRP as this species was significantly impacted by HPAI H5N1 and as a result, significant migration ecology work has been conducted to gain insight into the details of migratory connectivity. Other waterfowl species known to be reservoirs of influenza viruses should also be prioritized in a future CRP.

Isotope analyses may provide a valuable tool for studying movement and transmission of other transboundary animal diseases, in addition to HPAI H5N1. For example, FMD and Mongolian gazelle, Pasteurellosis and Saiga antelope, RVF and cattle or buffalo, Henipah and rabies viruses and bats. Other examples might include evaluation of pathogen transmission routes through bushmeat or wildlife trade, poultry trade, livestock trade, etc. The diagnostic capacity of other tissues in addition to feathers should be explored. This would include the use of hair, toe nails, hooves, blood, muscle, etc.

In general, the use of deuterium isotope measurements in keratinous tissues has been used to understand the origin of wildlife. However, it may be of considerable use to also conduct analyses for other stable isotopes (strontium, lead, sulphur, etc.), heavy metals, trace elements, or even genetic markers to hone in on a single specific site and the unique signature of that site. This approach should be guided by knowledge of corresponding isoscapes or elemental patterns across the landscape or could be utilized effectively in situations where birds are known to use just a few important sites that can be fingerprinted in this way.

There are already clearly defined working partnerships (Asia-Pacific working group on migratory waterbirds and AI), MOU's, multilateral environmental agreements (UNEP, CMS, AEWA) including the East Asian Australasian Flyway partnership which provide connectivity and frameworks for field work, coordination, and project implementation. Both Wetlands International and FAO are extensively connected to these partnerships and networks therefore any future CRP should include partners and colleagues associated with these existing frameworks and working relationships.

### **Consultants Meetings on the Effect of Climatic Change on Animal Production and Health – the Way Forward and Role of livestock on climate change and mitigation strategies**

Technical Officers: Mario García Podestá and E. Nicholas Odongo

These meetings took place in Vienna, from 17 to 19 November 2010.

The purpose of the two joint meetings was to review the challenges and opportunities of the intersection between livestock production systems and global climate change, to discuss how livestock production systems can adapt to climate change and to identify strategies to mitigate greenhouse gases emissions from livestock, and to demystify some of the myths and misconceptions about livestock production and global warming.

The meeting was attended by two FAO staff from the Animal Production and Health Division and 10 experts in sustainable animal production, rumen fermentation, methanogen genomics and methanogenesis, water and land use in relation to climate change, heat stress on livestock fertility, management of animal manure, climate change and animal diseases, mitigation and adaptation strategies for livestock systems, and modelling climate changes and livestock productivity, from Australia, Austria, Canada, Italy, Netherlands, USA, UK, and a representative of ILRI-Ethiopia.

Evidence from the Intergovernmental Panel on Climate Change (IPCC, 2007) is showing that climate change is real and that its effects will get worse. The world's poorest people, some one billion mostly in Africa and Asia, depend on livestock for their day-to-day livelihood. The IPCC report further shows that these poorest and most vulnerable people will be the worst affected by climate change. Climate change will have far-reaching consequences for dairy, meat and wool production mainly via impacts on feed, grass and range productivity seriously exacerbating the main constraint to livestock development in many developing countries which is the scarcity and fluctuating quantity and

quality of available feed resources, nutrient imbalance in many native pastures and crop residues and/or lack of or limited usage of commercial concentrate feeds.

Moreover, as the world gets hotter and drier, glaciers will melt, and the amount of arable land will shrink even further. The increased concentration of greenhouse gases e.g. methane in the troposphere has been implicated in the consistent increase in atmospheric temperature and global warming. Emissions from animals account for just over half of all agricultural emissions, or about 18% of total 'anthropogenic' (generated by human) emissions. Methane (CH<sub>4</sub>) is produced as part of normal digestive processes in ruminants. Methane production from ruminants fed highly fibrous diets is higher than those from animals fed better quality forages.

Globalization and climate change is having an unprecedented worldwide impact on emerging and re-emerging animal diseases and zoonoses. Climate change is disrupting natural ecosystems by providing more suitable environments for infectious diseases allowing disease-causing bacteria, viruses, and fungi to move into new areas where they may harm wild life and domestic species, as well as humans. Diseases that were previously limited only to tropical areas are now spreading to other previously cooler areas e.g. malaria. Pathogens that were restricted by seasonal weather patterns can invade new areas and find new susceptible species as the climate warms and/or the winters get milder.

Currently, one billion people face hunger and are 'food insecure' due to the adverse effects of climatic changes on crop and livestock productivity, increased demands from emerging markets, use of grains in the production of biofuels, and reduction of arable land due to increasing urbanization. Presentations in the meeting revealed that human population will increase by 38% (9.1 billions) by 2050; however, population in developed countries will only increase by 7% whereas in developing countries by 54%; and therefore, there is an urgent need in increase food production. Unfortunately, the global warming and climate change may have a dramatic impact on sustainability of livestock production systems. On the other hand, there is a bias in perception of livestock that does not recognize its multiple values which has been exacerbated by recent publications. They have created several misconceptions and the wrong image of the role of livestock as a source of food for the world and livelihood for millions of farmers around the world which is important to address and correct.

Discussions focused on sustainable animal productivity in a changing world, considering the various livestock systems and geographical conditions, the impact of climate change on emerging and re-emerging animal diseases and those of zoonotic nature as well as the

response of pathogens to climatic variations. It was clear that rise of average temperature seriously affects feed intake, growth rate and fertility due to heat stress, so actions should be taken to reduce the vulnerability of farming communities. In some cases, especially under tropical conditions, housing facilities can be adapted to sustain milk production and reproductive efficiency while keeping high producing dairy cows, a situation that has proved to be profitable under specific conditions in some countries. Nevertheless, other less sophisticated and less input demanding solutions are necessary to evaluate in order to mitigate the effect of climate change and climate variations on livestock productivity.

The group of experts agreed on publishing a book providing state-of-the-art information on climate change and livestock production and health, especially considering that there are plenty of myths, misconceptions and information without scientific support that is wrongly used by politicians and the media that is affecting the demand for food of animal origin and the future of livestock in developing countries. The outline of the book includes a situation analysis to put livestock into context, and then present the effects of climate change on livestock considering production and health issues, the effects of livestock on climate changes considering land use and non-CO<sub>2</sub> emissions. An important component of the book will be solutions and responses to the problem in terms of mitigation and adaptation options, to end up with conclusions and way

forward. The timeline for preparing and peer reviewing the book is one year.

The list of conclusions and recommendations of the meeting is available from the Animal Production and Health Section.

### **Regional Training Course on Animal Genetics in Bioinformatics Tools and Microsatellite Analyses and Sequencing (RER5015)**

Technical Officer: Adama Diallo

The regional training course took place from 22 November to 3 December 2010 at the Animal Production and Health Laboratory in Seibersdorf, Austria and aimed at enhancing knowledge on highly pathogenic avian influenza (advanced molecular genetics tools by use of nuclear and nuclear related and molecular technologies), in bioinformatics tools and microsatellite analyses and sequencing. The ultimate goal was to train partners in molecular genetic analyses.

The course was attended by participants from Albania, Bosnia and Herzegovina, Bulgaria, Croatia, Greece, Hungary, Montenegro, Poland, Romania, the Russian Federation and Turkey. The training was provided by lecturers from Austria, China, Finland, Germany, Italy, and Kenya.

## Stories

### IAEA helps to improve the productivity of cattle, camels and yaks in Mongolia through better nutrition and reproductive management

The livestock sector is the main pillar of the economy in Mongolia which, in addition to providing export products, provides food, clothing and shelter. The livestock sector employs 30 per cent of Mongolians and is a core survival strategy for nomadic families that rely entirely on pastureland livestock herding. In 2009 there were 43.4 million heads of livestock in Mongolia 0.3 million camels, 2.2 million horses, 2.5 million cattle, 18.4 million sheep and 20 million goats. However, there is large variation in animal numbers from year to year because of the death of millions of animals due to harsh climatic conditions. For example, the Dzud of 2009/2010, with temperatures plunging to  $-50^{\circ}\text{C}$ , resulted in huge livestock losses. By the end of April 2010, thick snow continued to blanket 60% of Mongolia making the animals unable to graze for months with a resultant 8 million head of livestock (20% of the animal population) reported to have died by mid June 2010. Furthermore, drought occurs in summer coupled with severe cold and snow in winter, both of which affect plant growth and feed availability as well as animal survival. The animals are raised primarily for their meat, although goats are valued for their hair which can be used to produce cashmere.

From the very first project implemented in 1986 i.e. 'Uses of nuclear energy achievement in animal husbandry' to the latest project MON/5/016 'Improving productivity of cattle, camels and yaks through better nutrition and reproductive management' (2007-2008), with the assistance of IAEA, specialized laboratories on radioimmunology to monitor reproductive efficiency and isotopic nutrition tracing and labelling technologies to evaluate nutritive value of feed have been established at the Mongolian State University of Agriculture and Research Institute of Animal Husbandry.



Studies have been undertaken for the determination of the nutritional value of various feed resources, the use of urea-molasses blocks, and interspecies cross-

breeding using semen from yak into cattle cows to increase beef and milk production. The main achievements to date include the production of Agency supported concentrated feeds (urea molasses, medicated

and or nutrient and element concentrated block). Additionally, improved nutrition management has decreased the input costs for farmers by almost 67% and the production of adapted soil and water animal feed crops together with an animal feeding programme have increased the quality of animals during winter including protecting animal lives (this was demonstrated in selected farms and this practice should now be taken up by all farmers and herders). The host institute and the project team have also improved their knowledge and skills on the use of artificial insemination (AI) in yaks and cattle and in feed evaluation and use of alternative feeds including evaluation of the nutritional value of feed resources available in different zones of Mongolia, including identification of toxic plants and plants containing bioactive compounds of industrial applications. Below are a few examples of previous work:

1. The relationships between chemical constituents of plant materials and methane production: The relationships between chemical constituents of 17 plant materials and *in vitro* methane production were determined at 24 h of incubation using the Hohenheim gas method. The methane production reduction potential (MRP) was calculated by assuming net methane concentration for the control hay was 100%, the MRP of *Bergenia crassifolia* leaves and roots, and *Peltiphyllum peltatum* leaves, was >40%. Amongst the chemical constituents, neutral detergent fibre had a high correlation ( $r = 0.86$ ) with methane concentration. There was a negative relationship between total phenol, total tannins or tannin activity and methane concentration. The results showed that leaves of *Rheum undulatum*, *Vaccinium vitis-idaea*, *B. crassifolia*, *Rhus typhina* and *P. peltatum*, and roots of *B. crassifolia* had considerable potential to decrease (i.e., >25%) enteric methane production from ruminants (Animal Feed Science and Technology, (2009) 150: 230-237).



2. Use of indigenous plants for treatment of animals infected with helminth parasites: Herbal infusions of prairie sagewort (*Artemisia frigida*, family *Asteraceae*), tansy (*Tanacetum vulgare*) and garlic (*Allium sativum*) were tested against ovine internal parasites *Nematodirus* and *Haemonchus* spp with the objective to evaluate their antihelminthic activity against common helminths of ruminants. Twenty five local Mongolian sheep, infected naturally with *Nematodirus oiratianus* and *Haemonchus contortus*, were grazed on pasture in Taryalan soum county of Khuvsgul aimag province for the duration of the experiment. The efficiency of the infusions (30%) was measured by the percentage reduction in the number of eggs produced by *N. oiratianus*. The egg count was reduced by 13.3% using prairie sagewort, 15.8% with tansy and 11.3% using garlic. The number of eggs produced by *H. contortus* was reduced by 22, 20.5 and 17.8%, respectively 5 days after administration of the plant infusions. In untreated (control) sheep infected with *N. oiratianus* and *H. contortus* there was little change in egg counts during this period, the levels remaining at 97.8 to 100% of the initial egg counts. The results indicated that infusions of 30% prairie sagewort, tansy or garlic to sheep had significant anthelmintic activity against *N. oiratianus* and *H. contortus* leading to a reduction in egg shedding.
3. Urea molasses multinutrient blocks have been found to be highly beneficial in several member states (MS). The use of solid urea-molasses blocks, i.e. solid blocks made with urea, molasses, vitamins and minerals, as a supplement for the nutrients deficient in the main feed, offers several advantages, namely, ease of transport, storage and use. It has reduced risks of toxicity compared with other approaches, such as giving a small amount of urea in the drinking water, or sprinkling a urea solution on fibrous feeds before they are fed to animals.

A mineral block feed production unit is functional in Ikh Tamir sum and Arkhangai aimag. A 1 kg mineral block contains 900 g of salt, 80 g of sodium sulphate, 10 g of wheat flour, 5 g of cement, 0.00409 mg of magnesium, 0.00186 mg of magnesium, 0.00153 mg of iron, 0.00145 mg of zinc, 0.0004 mg of iodine, 0.0003 mg of cobalt, 0.0002 mg of copper, 0.00017 mg of selenium. In order to promote the mineral blocks,



initially they were distributed free of charge to 12 herders, in Khunui, Bort, Khokh nuur rivers and the lake basins. This resulted in decreased number of abortions in the local cattle population. A herder's man from the Khan-Ondor communi-

ty of Ikhtamir sum, Arkhangai aimag bought 70 kg of the blocks but although he concluded they were effective, he considered that the blocks were expensive. This station is planning to produce concentrated feed used with straw, urtica and other grains.

Some case histories: Farmer Tsogtbuyan, from Bayanchandmani sum of Tov aimag has about 200 cows. He has 35 hectares of improved pasture land which he has owned for 60 years. Half of the pasture was cultivated and planted with perennial grasses and legumes and the other half he drilled and spread seed for botanical composition improvement. A fenced-off area of the pasture is used for herding milking cows in spring and late autumn to stabilize milk production. This farmer milks his cows twice daily and the average milk production is 14 L. Every year his cows are fed two tons of mineral blocks. In 2009 he obtained trace mineral cattle boluses from an FAO project and did not feed any mineral blocks. Tsogtbuyan prepares hay and silage and bought-in concentrate feed from the Altantaria Company for 350000 tugrik (1 euro = 1680 tugrik). He also prepares his own concentrate which contains 70 per cent wheat grain and 30 per cent straw and costs 350000 tugrik. Milk production was stable after replacing the Altantaria company feed concentrate with a mixture of ground wheat grain and chopped straw. By using the cattle bolus and own feed concentrate, milk production was stable. Transportation and purchase of grain reduced the costs by 60000-70000 tugrik which is to the benefit of the farmer.

Erdenee has been farming dairy cattle for three past two years. He has 30 milking cows and his farm uses hay and concentrated feed, purchased from a distributor working for a Russian company. He milks his cows twice daily with an average milk yield of 9.5 L. Last year, he started using mineral blocks and milk production increased to 10 L per day because of the blocks. Furthermore, when blocks were used, the cows' fertility increased and the number of abortions was lower.

The results of the comparative economic efficiency survey carried out in 2009 amongst three farmers using different feeding conditions are available online.

In future, in order to improve the productivity and sustainability of the farming systems it will be essential to ensure integration of crop and livestock systems. Owing to the harsh winters and the long and dry summer and autumn, inadequate nutrition will continue to be a major constraint to livestock production. Feed conservation and preservation and use of appropriate feeding practices, including feed supplementation strategies for increased animal productivity and enhanced reproductive performance, will continue to be a high priority for the sub-sector and should therefore continue to be supported under dedicated national projects, although some of the activities can also be

delivered through RCA or regional projects. Expected outcomes include (i) increased capacity of the National Agriculture Research Systems to optimally use locally available fibrous feed resources, (ii) reduction in livestock losses due to increased availability of locally

## The Agency supports portable diagnostic devices to enhance ‘at-source’ control of transboundary animal diseases

Molecular genetic testing plays a vital role in safeguarding public health – from diagnosing disease to monitoring for pathogens with pandemic potential; from detecting potential bioterrorism threats to safeguarding the food supply via crop and farm animal surveillance. Meanwhile, government agencies like the US Public Health Service and CDC, WHO and medical, emergency response and agriculture professionals are struggling to keep up with increasing demands of testing to ascertain safety and security. Finite financial resources, a shortfall in qualified technical personnel and time-consuming test technologies restricted to specially equipped laboratories have created an environment of need poorly served by the solutions currently available.

As a result of the FAO/IAEA Joint Division funding from the CRP on the early and rapid diagnosis of transboundary diseases (D3.20.25), DxNA has completed as of 26-30 April 2010 the validation of its GeneSTAT system and H5N1 assay at the National Institute of Veterinary Research (NIVR) in Hanoi. DxNA LLC (See [www.dxna.com](http://www.dxna.com)) was established in June 2008 as the successor company to Dx Nucleic Analytics, which was founded in 2005. DxNA manufactures and markets a proprietary PCR-based disease detection platform which consists of a unique, rapid and portable PCR system which consists of a PCR analyzer and accompanying closed-system testing cartridge under the trademark GeneSTAT®. The system consists of a 4.5 Kg thermocycler and cartridges which receives a sample, processes the sample, automatically performs amplification and detection, and provides a reportable result within 70 minutes.



The testing was done under the direction of Dr. Nguyen Tien Dung, Head of the Virology Department at NIVR. Assistants within the NIVR who had actual hands-on experience in running samples on the DxNA device

were Dao Thanh Van, Dao Duy Tung, and Bui Ngoc Anh. Dr. Mark Rosenfeld from DxNA was in Hanoi to observe the work. He provided about 15 minutes of demonstration on how to use the GeneSTAT system, and then turned it over to ‘a remarkable group of technicians’ to do the work. We were fortunate to

available feed resources for cold season feeding and (iii) improved survival rates, improved productive and reproductive efficiency, better growth rates, higher milk yields and improved farmer livelihood.

have skilled laboratory people working with us in Vietnam. However, the GeneSTAT system is designed to be able to be used by unskilled workers with minimal training. As we say, ‘Anytime. Anyplace. By anyone.’

The official report of the Vietnam results was given in Rome at the FAO/IAEA Meeting on Rapid Reporting of Transboundary Diseases held between 10 and 14 May 2010. The experience of actually using these rapid, portable devices is best described by Ms. Dao, who said, “It was very easy to use! Sample preparation took about two minutes. We all learned how to use the GeneSTAT within a few minutes.”

When asked what she liked best about the GeneSTAT, she said “it was easy to understand how to use it. I put the sample into the cartridge, entered my information into the computer, pressed a button, and it processed the sample and gave us a result while we had some tea. It was wonderful.”



Muscovy Duck Tracheal on GeneSTAT 25-04-2010

Ms. Dao was asked if there were problems. She said, “We had four GeneSTAT machines running at once, all hooked to a computer. On one set

of runs, we did not start the computer program for one device. We were worried, but it was no problem. GeneSTAT stored the result, and we were able to download it later. We also could see the result on the GeneSTAT device whether it was hooked up to a computer or not.



Craig Mosman of DxNA, who was also in Hanoi for the testing, thanked the FAO/IAEA Joint Division for the funding received to develop the GeneSTAT portable device and H5N1 test: “Our contract with the

FAO/IAEA has been critical to the development of our GeneSTAT system.

As a result of that funding, we have proven our platform at three different respected laboratories. In addition to the validated H5N1 test, in 2009 DxNA received

FDA Emergency Use Authorization for its H1N1 test, and is continuously developing new tests in both the human and veterinary markets to serve our customers. We are now making GeneSTAT available to member states, and look forward to a continued working relationship with the FAO/IAEA Joint Division.”

## The Artificial Insemination Centre in Cameroon – A success story

The production of high quality livestock and livestock products depends largely on the type of animals, management practices and environment. The North West region of Cameroon is located at 1300 to 2500 m above sea level and is suitable for cattle farming as the climatic conditions are fair, the annual rainfall of 2000 mm is regular and the region is not tsetse fly infested. Farmers were interested in producing milk and dairy products and therefore Heifer International initiated a project in the 1970s to increase milk production in the area. During the start-up phase of the project, Holstein and Jersey bulls were used for mating with the local breeds and farmers agreed to return the first-born calves to Heifer International for further distribution to other farmers. Unfortunately, the advent of brucellosis coupled with poor reproductive performance constrained the natural mating approach used by the farmers and the programme required more effectively targeted inputs, resulting in the involvement in the project of the Bambui Cattle Centre, one of several government owned breeding stations.



Fig. 1. Traditional milking in Cameroon  
Photo: Dr. Henri Bayemi

The Bambui Cattle Centre was historically equipped with a disease diagnosis laboratory, a natural breeding/mating system, housing for livestock and rotational grazing paddocks. The IAEA through a TC Project expanded the capacity of the Centre to strengthen its capability for semen processing, artificial insemination (AI), and reproductive disease diagnostics. A radioimmunoassay laboratory was established for the determination of various reproductive hormones – a test for the early detection of non-pregnancy and a diagnostic test for brucellosis. Dr. Henri Bayemi and his team successfully collected and processed semen from local bulls and trained technicians on heat detection, oestrus synchronization and on the AI technique. At this stage, the first inseminations were only carried out at the

Bambui cattle farm. Later on, two Friesian bulls were donated by Heifer International and the team was able to initiate an AI field service in the vicinity of Bamenda, the hometown of the AI Centre.



Fig. 2. Semen Collection at Bambui Cattle Centre  
Photo: Dr. Henri Bayemi

In 2007, following the approval of IAEA TCP CMR/5/015, the project team had already managed to support 200 farms on aspects related to cattle management, feeding, preventive health care, and AI services. Subsequently, the Bambui cattle centre has performed more than 500 inseminations with nearly a 70% conception rate using oestrous synchronization. Furthermore, in the last 4 years, the prevalence of Brucellosis has been reduced drastically as a result of the establishment of a control programme, use of AI and culling of infected animals.

One of the main constraints for AI in some African countries such as Cameroon is the availability and cost of liquid nitrogen for transporting semen, especially in rural areas, a key element to preserve semen for prolonged periods. The Bambui team overcame this by developing a chilled semen processing methodology using egg-yolk and coconut water in which sperm can survive for up to seven days. The initial average motility is around 75% but decreases to 60% but is still sufficient for insemination purposes. What is crucial is to keep the semen ampoules in boxes with ice once the semen is transported from the AI centre! Certainly, the sooner the cow is inseminated the better but this methodology opens great opportunities to inseminate cows in rural areas without requiring the use of expensive and sometimes scarce liquid nitrogen.



Fig. 3. Holstein x White Fulani bull at Bambui  
Photo: Dr. Henri Bayem

Currently, AI is being conducted when farmers report cows in heat, but in the initial phase of the project, most cows were inseminated after oestrus synchronization using Prostaglandin F2a whilst more recently the Ovsynch protocol in anoestrous cows has been followed where, cows receive two injections of Gonadotrophin Releasing Hormone (GnRH) on days 0 and 9 and one injection of Prostaglandin F2a on day 7 and the insemination is done on day 9. Pregnancy in cattle is now routinely and regularly monitored using a human progesterone enzyme immunoassay kit, which was validated for cattle (Bayemi et al., 2007, *Trop Anim Hlth Prod* 39; 335-338).

In addition to the AI service farmers also receive technical advice on improved feeds and feeding and instruction on processing milk to produce yoghurt and ambient temperature- matured cheese as well as better access to veterinary services i.e. an integrated and sound technical package for improving livestock productivity and farmer livelihoods. Farmers are gradually accepting the proposed technology, especially AI using heat synchro-

nization, and in consequence, the expenses/income ratio is increasing favourably.

Field days have been organized for technology transfer and to promote AI, non-pregnancy diagnosis through the analysis of progesterone in blood samples and disease diagnosis using nuclear and nuclear-related techniques, such as radioimmunoassay and ELISA. The increased capacity and technology and expertise acquired by the Centre has enabled staff to train foreign veterinarians and AI technicians from Botswana, Burkina Faso, Madagascar, and Central African Republic and also to conduct technical consultancies in neighbouring countries.

Recently, the AI Centre has obtained a mobile ultrasound diagnostic device to facilitate pregnancy diagnosis and detection of reproductive disorders affecting the functionality of the ovaries. The laboratory has also been renovated by the Government and modern equipment installed to ensure semen quality, especially in terms of transmissible diseases. The project team is now preparing to enlarge the AI coverage by including other areas in the region as well as other regions of the country. Support to the farming community will be further enhanced as, in parallel, the laboratory will also expand its disease diagnostic services to include other livestock diseases as well as carrying out milk testing and quality control.

This successful project exemplifies the efforts of the IAEA in assisting member states to improve livestock productivity and food security and reduce poverty.

These stories as well as other articles are also available under 'Highlights' on our Homepage  
<http://www-naweb.iaea.org/nafa/aph/index.html>

# Coordinated Research Projects

## Peste des Petits Ruminants (PPR)

Technical Officers: Adama Diallo, Hermann Unger

This CRP has been running for three years. The overall objective is to develop, validate and transfer to Member States sensitive, specific and rapid tests for the diagnosis of PPR to help them better manage and control this transboundary animal disease. The activity reports received from the different research contract holders indicate the widespread prevalence of PPR in the different countries. Progress towards achieving the project objectives is satisfactory:

- 1) The evaluation of the current competitive ELISA has started and first results were taken into consideration to improve the robustness of the assay,
- 2) A ring test was organized in June between different laboratories to evaluate the use of classical gene amplification for the diagnosis of PPR.
- 3) Interesting epidemiological data are being accumulated, data to be considered in PPR control strategies namely the findings to the potential involvement of peste des petits ruminants in a camel respiratory disease. In some countries the seroprevalence PPR antibodies in cattle was high although no disease could be linked to this infection.
- 4) New tests were developed (realtime RT-PCR, LAMP, cell for virus isolation). Those new tests are under evaluation.

## The Early and Rapid Diagnosis of Transboundary Animal Diseases: Phase I - Avian Influenza

Technical Officer: Gerrit Viljoen

This CRP focuses on the early and rapid diagnosis and control of avian influenza (as technological target) through the advantageous use of nuclear, nuclear associated and nuclear related technologies, in conjunction with non-nuclear technologies. In particular, the rapid, sensitive and specific detection of disease agent nucleic acids using molecular technologies (e.g. RT-PCR and PCR sequencing), and the use of isotopes (P32/33, S35 and S35Met) to label or trace virus nucleic acid or proteins during development and comparative phases of research, and for the evaluation or characterization of targeted genes.

The overall objective is to develop, evaluate and validate early and rapid detection technologies to provide Member States (MS) with the capacity to detect, moni-

tor, contain and control TADs. The CRP is supporting the build up of competence in the use of modern biotechnology, including molecular and serological methods, to provide systems and technologies to be used in the field as well as in laboratories. A major target for diagnostic systems will be the highly pathogenic avian influenza viruses, but such systems are pertinent to all other TADs since the technologies addressed in this CRP form part of an early response diagnostic capability platform. The IAEA is supporting Member States in their efforts to control diseases of importance. This, amongst others, involves the development, evaluation and validation of the appropriate nuclear, nuclear associated and nuclear related technologies and the harmonization and dissemination of protocols and procedures. Technical advice is therefore given to Member States (or any other party) as to the diagnosis of a disease, the best 'fitness for purpose' tools and quality assured procedures, including prophylactic measures (e.g. vaccines), to use in close collaboration and consultation with experts in the field. In the case of avian influenza, it is important for the rapid and differential diagnosis to classify isolates as highly pathogenic or not, in order to activate appropriate control measures - this is seen as the bottleneck activity for most developing countries.

The final RCM took place from 10 to 14 May 2010 in Rome, Italy.

## Control of Contagious Bovine Pleuropneumonia (CBPP)

Technical Officer: Hermann Unger

This CRP is now in its last year. The validation of the CBPP c-ELISA is completed and publications on the findings are underway. This test is certainly superior to the CFT, in terms of quality control, but needs a careful definition of the purpose as it detects infected animals up to 2 years post infection. As it basically does not detect vaccinated animals, this test is perfect for prevalence studies. The LPPQ ELISA was rated as the easiest test, and gave very reliable results for active CBPP infections. Unfortunately this test is not produced anymore. ILRI is currently working on a new indirect ELISA for CBPP testing panels of expression antigens. By the end of 2010 the report is expected and we hope to be able to test the new ELISA in a number of countries. There is no test available to detect 'protection' after vaccination, which makes it difficult to evaluate the efficiency of vaccination campaigns.

In order to allow for the quick confirmation of a potentially CBPP infected animal in the field a newly developed Card agglutination test will be compared to existing methods for suitability and reliability.

As the culture of Mycoplasmas is rather difficult, partly due to the fact that nasal swabs contain too many pathogens resistant to the antibiotics in the culture media and post mortem samples are rarely taken, the use of molecular methods was tested. PCR is established in a number of labs, but here as well the transport of the samples and the successful DNA extraction present major obstacles. A newly developed isothermal diagnostic technique, Loop mediated amplification (LAMP) was distributed to the participants during the last RCM. The reading devices were also sent and it is hoped that this method can be established, adapted and validated in the counterpart laboratories.

During the last RCM in Zanzibar in September 2010 the results obtained so far were discussed and the remaining questions regarding CBPP summarized. There is still plenty of fundamental research to be done, before a scientifically sound approach for the control of this disease appears on the horizon.

## Veterinary Surveillance of Rift Valley Fever

Technical Officers: Gerrit Viljoen; Hermann Unger

Rift Valley Fever (RVF) is a zoonosis caused by a bunya virus inflicting great economic losses from reduced productivity, abortions in pregnant animals and high mortality in animals and humans. RVF is defined as one of the haemorrhagic fever viruses in the emerging diseases group. It was first isolated in 1930 in the Rift Valley of Kenya from sheep and is endemic in sub Saharan Africa. Periodic disease has been recorded in animals and humans with major outbreaks in Egypt, South Mauritania, Madagascar, Northern Kenya, South Africa, Sudan and Somalia. In September 2000 RVF was reported outside of the African Continent for the first time in Saudi Arabia and Yemen. These outbreaks lead to more than 2000 human cases killing nearly 300 people and 20 000 abortions in livestock in Yemen. This expansion in epidemic area to the Arabian Peninsula raises the possibility of threat of RVF to other parts of Asia and Europe.

Transmission of RVF is by mosquitoes or by contact. Many different species of mosquitoes are known to be vectors. There is, therefore, the potential for epizootics and associated human epidemics following the introduction of the virus into new areas.

RVF-vaccines for veterinary use are available, but live-attenuated vaccines have been shown to produce birth defects and abortions, while inactivated vaccines induce only short lived and incomplete protection. A live-

attenuated vaccine for humans is under development and not yet commercially available. The diagnosis of RVF depends nowadays on serology. The existing enzyme linked immuno sorbent assay (ELISA) is widely used but lacks specificity and is produced from virus culture, potentially transporting the germ. Direct virus diagnosis demands high security labs not available in most countries.

The Polymerase Chain Reaction (PCR) is a quick, reliable and safe alternative molecular tool providing high sensitivity but is not yet a frequently used method in most laboratories. A competitive ELISA for RVF would have the additional advantage of being species specific and supporting research in the potential hosts of this disease.

The target of this CRP is to support countries at risk of major RVF outbreaks to gain the capacity for a quick and reliable diagnosis of this disease and by the evaluation of epidemiological patterns allow an early warning.

### Specific research objectives

- Evaluation of RT-PCR and PCR sequencing for early detection of virus and its use in molecular epidemiology
- Evaluation, validation and use of the existing and new ELISA's in serological surveys
- Evaluation of recombinant antigens for use in indirect and competition ELISA's (rC-ELISA).
- Harmonization of SOP's and introduction of quality assurance procedures for RVF-ELISA and RT-PCR.
- Set up of an epidemiological database supporting risk assessment for RVF outbreaks.

### Expected research outputs:

- Validated diagnostic tools and descriptions of RVF tests based on fitness for purpose.
- Standard diagnostic procedures for surveillance and early diagnosis using PCR and ELISA, defined reference material available.
- An rC-ELISA developed to measure antibodies against RVF from all species (including non-ruminant species).
- An epidemiological databank established.

The CRP will draw to a close in Dec 2010 and a follow-up CRP will be proposed for 2011.

## The Use of Enzymes and Nuclear Technologies to Improve the utilization of Fibrous Feeds and Reduce Greenhouse Gas Emissions from Livestock

Technical Officer: Nicholas Odongo

The world's poorest people, some one billion, depend on livestock for their day-to-day livelihood: food, fibre,

manure, draught power, transport, ready source of cash, etc. However, livestock production in many developing countries is constrained because of poor nutrition. Because of climatic conditions, animal feeds are in short supply and what is available is of poor quality. The problem is particularly critical during the dry season when farmers may suffer great animal losses. Furthermore, there is a lack of and/or limited use of commercial concentrate feeds, e.g. soybean, cottonseed and groundnut meals, etc because the resource poor farmers cannot afford them. The problem is also being exacerbated by the decreasing availability of arable land because of the rapidly increasing human population, soil/land degradation, urbanization and effects of global warming.

Furthermore, methane production from ruminants fed poor quality diets such as straw and stover is higher than those from animals fed better quality forages. The increased concentration of greenhouse gases (e.g. methane) in the troposphere has been implicated in climate change and global warming. Methane production is negatively correlated with energy utilization and it can range from two to 12% of the gross energy intake, thus, reduction of methane production through the use of enzymes and rechanneling the hydrogen to short-chain fatty acids and microbial mass is desirable. Reducing methane emission from ruminant animals has implications not only for global environmental protection but also for efficient animal production.

Recent research is showing that supplementing livestock diets with fibre degrading enzymes can improve the efficiency of feed utilization, resulting in improved animal performance and a reduction of methane emissions. For sustainable development of the livestock sector it is essential to secure sufficient supply of balanced feeds from resources that do not compete with human food – production of grain in developing countries is mostly for human consumption. Novel approaches through the utilization of tree leaves, agro-industrial by-products, feed additives and aquatic sources are required to bridge the gap between supply and demand of feeds.

This CRP will:

- a) Determine the effects of supplementing livestock diets with enzymes on (i) fibre degradation *in vitro*, *in situ* and *in vivo*, (ii) feed intake and digestibility, (iii) ruminal fermentation and microbial protein synthesis and (iv) on milk production and composition and/or growth performance.
- b) Determine the mode of action, the critical enzymic activities and application method and rates needed to elicit the desired response.
- c) Determine the effects of supplementing livestock diets with enzymes on animal performance, enteric methane production and cost-benefit analysis.

- d) Build capacity in developing countries on the use of nuclear and related technologies to improve livestock productivity and to create opportunities for research collaboration internationally.

The First RCM for the CRP is scheduled for 7-11 February 2011 in Lethbridge, Alberta, Canada.

## The Use of Irradiated Vaccines in the Control of Infectious Transboundary Diseases of Livestock

Technical Officer: Antony George Luckins

The livestock sector is an important source of income for small holder farmers in the developing world. The growing demand for livestock products driven by population growth will provide the rural poor additional opportunities to increase livelihoods. Paramount to meeting this demand will be the need for governments to improve animal health, particularly where it relates to control of infectious diseases since they are a major constraint on livestock productivity and there is an urgent need to tackle the problem in order to ensure food security. Although diseases caused by viruses and bacteria are of major concern, often causing serious epidemics and compromising international trade, parasitic diseases caused by helminths such as *Fasciola* and *Schistosoma* or protozoa like *Trypanosoma* and *Theileria* exert a persistent, debilitating effect on livestock productivity throughout Africa, Asia and South America. With one or two exceptions their control relies on chemotherapy, however, this approach has a number of disadvantages. Firstly, even though animals are cleared of infection rapidly after treatment, it often fails to prevent re-infection thereby requiring frequent administration of the drug. Secondly, parasites are able to adapt genetically to the action of the drugs, resulting in the development of drug resistance – a common cause of overuse or inaccurate administration. This latter problem is compounded by the widespread use of fake products in many MS. Also, long term treatment brings with it the accumulation of drug residues in meat and milk, a situation that could compromise export of livestock products to the developed economies.

While this is a strong reason to develop vaccines against parasitic diseases, it will be necessary to apply innovative techniques to achieve this aim since in the past there has been only limited success in producing effective vaccines. Although a few attenuated, live vaccines are available they have a limited shelf life and it can be difficult to select appropriate, genetically attenuated organisms. Recombinant vaccines have also failed to live up to their promise, and there are a number of reasons for this. Parasites are complex organisms, comprising thousands of proteins, and identifying a single protective antigen is difficult, if not impossible since the immune response is multifaceted, requiring activation of several different immune pathways and it

is synergy between different antigens that enables this to occur.

The way in which this can be achieved is through the use of gamma radiation attenuated organisms where there is strong evidence that both cellular and humoral immune responses are activated, simulating the response that occurs when live organisms are introduced into the host. Irradiation of whole organisms obviates the need to identify specific antigenic components required for subunit vaccines, or the time and resources to create genetically attenuated organisms. Moreover, although radiation attenuated organisms are metabolically active and follow a similar migration route to non-irradiated organisms in the host they fail to develop into a mature infection. Gamma irradiation is also practicable for use with those bacteria or viruses where there are currently no effective vaccines available – irradiation will more efficiently preserve the antigenic and adjuvant structures destroyed by conventional chemical or heat treatment. Effective storage and delivery of vaccines is an essential part of a strategy for control of animal diseases and developments in cryobiology enabling lyophilization of whole cells will make it possible freeze-dry vaccines, even those prepared from helminths or protozoa. Radiation attenuation would also expedite rapid emergency vaccine production during epidemic outbreaks of microbial diseases and freeze-drying would enable such vaccines to be stored and transported without need for a cold chain, thereby benefiting resource-poor MS.

The CRP will:

- a) Develop techniques for irradiation attenuation of, for example, *Trypanosoma*, *Theileria*, *Fasciola*, *Schistosoma* and RVF and FMD virus
- b) Use nuclear techniques to assess metabolic activity of irradiated organisms and follow their migration and establishment in the host from site of injection
- c) Test irradiated vaccines in experimental animals to determine protective dose and monitor level and duration of immunity
- d) Develop flow-through irradiation techniques to enable fast preparation of vaccines.

The first research coordination meeting took place from 11 to 15 October 2010 in Vienna Austria and a detailed report can be found in the 'past events' of this newsletter.

## Development of Molecular and Nuclear Technologies for the Control of Foot-and-Mouth Disease (FMD)

Technical Officer: Gerrit Viljoen

Foot-and-mouth disease (FMD) is one of the most important livestock diseases known to man due to its high infection rate (ease of spread) and its effect on the limitation of livestock movement and trade. An out-

break of FMD will have a devastating effect on a country's food security with direct impact on national and international trade. The confirmatory diagnosis of FMD and its effective control through prophylactic, quarantine or slaughter-out procedures are therefore of paramount importance as it has financial and trade implications. Vaccination with inactivated FMD virus is undertaken to control FMD in endemic countries or countries at risk. Vaccines, whilst widely available but which should match (i.e. should be of homologous serotype and strain isolate) with virulent FMD viruses circulating in the region of vaccine use, are of variable quality, not from the homologous outbreak serotype/strain isolate, and are often stored under inadequate temperature conditions and therefore might be not as effective in the field as determined in animal experiments. Due to insufficient knowledge on vaccine strength and antigenic match (antigenic cartography) between vaccine strain and outbreak virus, it is often not possible to pinpoint the weakness of the vaccination strategy and to take action on this weakness.

Vaccine effectiveness can be determined by animal challenge, but this is both costly and difficult. *In-vitro* systems have been developed in different countries since the 1980s, but these are not standardized for international use. Many countries now produce FMD vaccines but often without effective consideration of their effectiveness. This CRP will investigate methods and possibly provide internationally acceptable guidelines for procedures which test a vaccine's ability to induce the production of protective antibodies in cattle without the need for animal challenge experiments.

In many developing countries, vaccination will continue to be an essential component for the progressive control of FMD. Maximising the effectiveness of current vaccines and supporting research to improve the effectiveness and quality of those and or new vaccines will be critical. Countries using locally produced vaccines need to assure trade partners that they are using quality assured vaccines in order to overcome the restrictive effects of endemic FMD. The provision of internationally accepted guidelines for quality assurance and alternatives to the present need for animal challenge vaccine trials would be a significant step forward. It is likely that control and eventual eradication in endemic areas with a low level resource base (much of Africa, parts of Asia and Latin America) will require the use of quality assured vaccine preparations, correct vaccine formulations (i.e. homologous strain or isolate vaccine to protect against outbreak, new generation vaccines with a broader protection base (i.e. cross protection between different strains and isolates) or alternative formulations of existing vaccines.

The CRP will:

- a) Establish methods and develop internationally agreed protocols for measuring the potency of FMD vaccines using *in vitro* methods;
- b) Establish guidelines for optimum population vaccination intervals based on *in vitro* measurements of potency and duration of the antibody response to structural proteins, after vaccination of cattle and small ruminants with commercially available FMD vaccines, including evaluation of reduced dose options such as intradermal administration of FMD vaccine;
- c) Establish protocols and guidelines for application and interpretation of vaccine matching methods (antigenic cartography) to identify the extent of expected cross-protection of type A or SAT viruses; and,
- d) Provide further global coordination of current research into FMD vaccines for use in endemic settings.

The first research coordination meeting is scheduled to be held early in 2011 in Vienna, Austria.

## Genetic Variation on the Control of Resistance to Infectious Diseases in Small Ruminants for Improving Animal Productivity

Technical Officer: Mario García Podestá

Farmers in developing countries, due to the pressure for higher animal output and to the 'advantages' of small

number of highly specialized breeds from the developed world have been replacing or crossbreeding their local breeds with exotic animals for many years. The genetic improvement has been quite successful in many places; however, neglecting or upgrading indigenous animals with exotic breeds is leading to a deterioration in genetic diversity.

Much of the genetic biodiversity controls advantageous traits influencing adaptability to harsh environments, productivity, or disease resistance. However, these indigenous animals are underutilized in conventional breeding programmes, due to a lack of knowledge and failure to identify breeds and animals carrying the most advantageous traits. There are indigenous breeds with some degree of enhanced resistance as compared to exotic ones reared in the same environment, especially for gastrointestinal nematode infections. Therefore, the present CRP is aiming, through genomic studies using radiolabeled nucleotides in DNA hybridization, DNA characterization, and hybrid mapping procedures for identifying molecular markers of economic interest which will open possibilities in the future to select and breed animals for enhanced resistance to diseases. The CRP will also aim to develop capacity in developing countries in the use of molecular and related technologies and create opportunities for international research collaboration.

The first research coordination meeting is scheduled to be held in Vienna, Austria, from 21 to 25 February 2011.

## General information applicable to all Coordinated Research Projects

### Submission of Proposals

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

<http://www-crp.iaea.org/html/forms.html>

Such proposals need to be countersigned by the Head of the Institutions and sent directly to the IAEA. They do not need to be routed through other official channels unless local regulations require otherwise.

### Complementary FAO/IAEA Support

IAEA has a programme of support through national Technical Cooperation (TC) Projects. Such support is available to IAEA Member States and can include additional support such as equipment, specialized training through IAEA training fellowships and the provision of technical assistance through visits by IAEA experts for periods of up to one month. Full details of the TC Programme and information on how to prepare a project proposal are available at the URL <http://pcmf.iaea.org/>

For further information contact Roswitha Reiter ([r.reiter@iaea.org](mailto:r.reiter@iaea.org))

# Activities of the Animal Production and Health Laboratory

## Radiation Hybrid Mapping for Goat: Construction of a goat (*Capra hircus*) whole-genome radiation hybrid panel

The goat (*Capra hircus*) is an important agricultural species worldwide with centuries of phenotypic observations, trait selection, and breed differentiation. However, the understanding of the goat at the genomic level lags behind other livestock species, such as cattle, pig, chicken, and sheep. To improve our understanding of the genetic components of traits related to goat health, production and biology, there is an urgent need to develop a detailed goat genome map. The most recent genetic map for the goat was published twelve years ago. This map was an upgrade of the previous genetic map through the addition of microsatellite markers and genes from bovine, ovine, and human maps. It is an important resource, but it is seriously lacking in marker density for effective gene discovery through comparative genomics. One of the primary limiting factors in construction of the genetic map is the need for genetic polymorphic markers. This can be overcome with the use of a radiation hybrid (RH) map as polymorphisms are not necessary for marker placement, and allows for use of non-polymorphic markers, such as those developed from EST and BAC sequences.

Radiation hybrid (RH) mapping is a method for producing high-resolution maps, that can be used for integrating linkage maps and also serve as a link across species for comparative mapping. Therefore, it is of critical importance to construct a RH panel providing a resource for rapid and large-scale physical mapping of the goat genome. This will facilitate the resolution of the genetic and physical distances prior to designing strategies for positional candidate cloning of the gene(s) that are involved in economically important traits.

The Animal Production and Health Laboratory (APHL) has participated in development and optimization of different Nuclear and Nuclear related technique in the past few years. One of the success stories in this area is the involvement of APHL in collaboration with other institute in a project for the development and characterization of a goat (*Capra hircus*) whole-genome radiation hybrid panel (Goat RH5000). Our aim is to (1) develop and characterize a whole-genome radiation hybrid panel (GoatRH5000) in the goat; (2) develop an initial radiation hybrid map for the goat using SNP markers; (3) develop a goat RH mapping server allowing the user to map goat markers relative to a frame-

work of previously mapped markers; (4) provide a unique tool for the study of goat genomics and potentially to utilize important traits in the genetic improvement the goat; (5) train researchers, graduate students and technicians to conduct genetic research in the goat.

### *Develop and characterize a whole-genome radiation hybrid panel (GoatRH5000)*

For the construction of *Capra hircus* radiation hybrid panel, cells from a goat donor were irradiated with a cobalt 60 source for a total dose of 5000 rad. Then those cells were hybridized with the recipient Chinese hamster TK- cells (A23) and this generates 130 RH colonies. Some of these hybrids will be used for further genome mapping studies.

### *Develop an initial radiation hybrid map for the goat using SNP markers*

Different types of maps can be created using the RH panel. Maps can be based on microsatellite, EST-based markers or single nucleotide polymorphism (SNP) panels. Mapping based on microsatellite and EST markers is a relatively slow process, but it can be well suited to comparative mapping activities. A rapid method for developing a RH map is the use of SNP panels. The cattle panel has approximately 62,000 SNPs and the sheep panel has approximately 49,000 SNPs on the panel. Thus, approximately 31,000 SNP locations could be used for development of an RH map from the cattle SNP panel, and approximately 44,100 SNPs could be used for the development of a RH map from the sheep SNP panel. Currently APHL is involved in a collaborative project which aims at developing such a map using SNP chips.

### *Rationale and significance*

The RH map project is designed to develop a fundamental tool for genomic research in the goat. Additionally, this tool will be useful for the entire research community through the comparative genomic analyses with other species. The development of a RH map for the goat will allow researchers that discover a phenotype of interest to use as a model for comparative analysis and gene discovery. Because the goat has adapted to virtually every type of environment and has many biomedical conditions similar to humans and other ruminants, it will be a valuable resource for this comparative genomic approach.

### *Appropriateness to CRP for small ruminant*

As the phenotypic characterization is finalized, it is important to begin development of the tools for the genetic characterization of goat breeds. The RH panel

and map will be a valuable tool used for this purpose. Additionally, the CRP on 'Genetic Variation on the Control of Resistance to Infectious Diseases in Small Ruminants for Improving Animal Productivity' will continue the genetic characterization of the goat breeds, specifically to associate the genetic and phenotypic characterization. The RH panel and subsequent map will be necessary to conduct the comparative genomics in order to utilize the genotype-phenotype associations.

## Genetic characterization of indigenous chicken breeds in search for the unique properties of the immune-related genes

Domestic chicken (*Gallus gallus domesticus*) is one of the most important animal species worldwide as it provides a large proportion of animal protein in human diet and also serves for entertainment and decorative purposes. The contemporary chicken was most probably developed from its main wild ancestor, red jungle fowl (*Gallus gallus*), after the domestication event(s) in Southeast Asia in 3,200 BC. Since that time chicken evolved from the wild form to the multiple contemporary layers, broilers, bantams, game and fancy breeds not to mention indigenous village chicken. At first sight the diversity within domestic chicken is huge, which should provide an excellent base for breeding animals that are well adapted to the local environmental conditions. However, the industrialization and globalization of the animal production in the 20<sup>th</sup> century severely affected the distribution of chicken genetic resources worldwide, practically limiting the breed composition to commercial stocks of broilers and egg-type hens. Consequently, many chicken breeds have already become extinct or are seriously endangered with extinction.

Currently, the world poultry market is facing recurrent outbreaks of contagious diseases, which provide a serious health and economic threat. Commercially bred animals that stand for most genetic resources nowadays are likely to lose the genetic resistance through a long-term process of one-sided selection for production traits. Besides, the real health status of the stocks is unknown to the breeders due to the extensive vaccination programs and veterinary drug abuse. Combined with large flock sizes of the industrial farms amounting to thousands of animals, the pathogen transmission on the farm is immediate. In case of the emergence of the highly contagious disease like the highly pathogenic avian influenza (HPAI), the whole stock of the given area can be endangered with infection and following culling of the animals. This is threatening the livelihood of millions of livestock farmers, jeopardizing commercial poultry production, and seriously impeding regional and international trade. Last but not least, the epidemic diseases breaking out in commercial flocks of domestic animals pose a serious hazard to human

health, especially in many of the developing countries where the food hygiene needs to be drastically improved.

Enhancing the immunocompetence in chicken can be provided by use of already existing rich sources of genetic variation, i.e. such gene variants (alleles) that are associated with the genetic resistance to diseases. But, the main obstacle preventing from direct application of modern molecular techniques in practical animal breeding is lack of knowledge on which genes determine the fitness and robustness of the individuals. This task is not easy given complexity of the immune system and multiple animal-pathogen interactions as well as not yet fully understood genomic determination of the physiological processes. However, the evolutionary processes that followed the event of chicken domestication and its distribution to every corner of the world contributed to the adaptation of the animals to different environments. Since the process of adaptation of the indigenous chicken to local, often harsh and extreme environmental conditions, demanded positive selection towards enhanced immune resistance, it is likely that those animals are now carrying fixed alleles determining their better immunocompetence. Therefore, search for the beneficial mutations in the genomic DNA is often based on local, unselected breeds. This approach may finally lead to the discovery of evolutionary processes that resulted in adaptation and consecutively better immune resistance of indigenous chicken.

However, the first step to select a breed that can be useful for further study on genetic resistance is to provide comprehensive characterization, including the breed history, population size and distribution, physiological properties of the animals, as well as the genetic polymorphism within the loci that are specifically associated with interesting traits, such as disease resistance. Since there is still a number of unexplored chicken breeds that can potentially carry interesting mutations within immune-related genes, a project was initiated by Animal Production and Health Laboratory in the FAO/IAEA Agriculture and Biotechnology Laboratory with the objective to provide the genetic characterization of the indigenous chicken breeds. For the study, chicken DNA samples were received from many regions. Examples of breed included in the study are (i) the Green-legged Partridge-like, an indigenous Polish breed with characteristic green shanks that lays eggs with lower cholesterol content and (ii) the Kadaknath – an Indian fowl with black coloured plumage and meat that is a rich source of iron.

Since the immune system developed many divergent pathways to fight pathogens, the genetic characterization of the indigenous chicken analysed here included polymorphism within a few loci engaged in different immune mechanisms. Primarily, the major histocompatibility complex (MHC) was investigated. Resistance

to some contagious diseases (e.g. Marek's disease) is strongly associated with the MHC variant. Secondly, the single nucleotide polymorphisms (SNPs) within some key genes of the immune response were also studied. One of them was the Myxovirus resistance gene (*Mx*) that was reported to confer resistance to avian influenza virus. The other was the interleukine 2 gene (*Il-2*), which is an immunoregulatory cytokine produced exclusively by Th1 cells (T helper 1 lymphocytes) and driving the immune system towards macrophage activation and antibody production. Finally, the third sequenced gene fragment included Toll-like Receptor 7 (*TLR7*), a member of a specialized receptors family, which recognize the invasions of pathogens and subsequently trigger the immune response. The product of the *TLR7* gene analysed here can sense the single-stranded RNA fragments characteristic to several viruses like Avian Influenza Virus.

The results of the study carried out by IAEA to provide genetic characterization of the indigenous chicken breeds, will contribute to a better recognition of those animals. Even though local breeds are not considered highly productive and well suited for industrial farms, they possess many desired features due to their adaptation to the local environments where they derived from. Therefore, it's worthwhile to characterize the genetic resources of indigenous breeds for better understanding of their uniqueness, which might be of help for the future implementation in animal breeding.



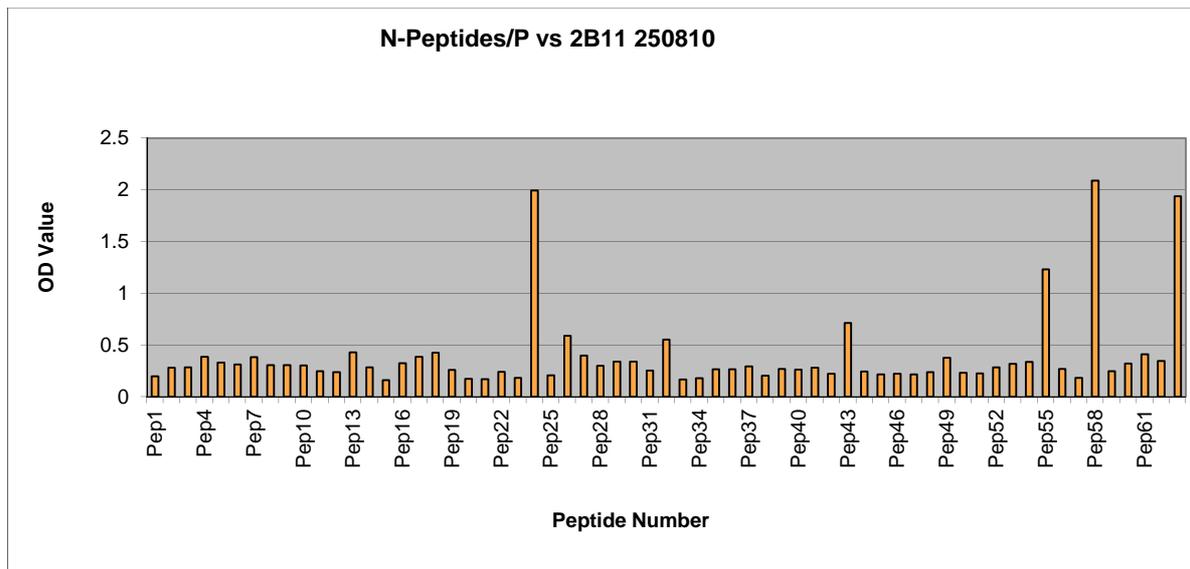
A hen of Green-legged Partridgelike from Poland  
Photo K. Surmiński ([www.produktregionalny.pl](http://www.produktregionalny.pl)).

## Study of the PPRV Protein-Protein interaction study: mapping the phosphoprotein (P) binding site on the nucleoprotein (N).

In 2003, the APHL embarked on an EC and Wellcome Trust supported collaborative project to develop a PPR marker vaccine. Within that project, APHL was in charge of studying the virus protein-protein interactions to identify on the nucleoprotein (N) zone (s) not essential for the virus to be used as the marker insertion site and also for the development of the marker vaccine companion test. In previous Newsletters, we reported on the results generated by the nucleoprotein mapping study by identifying the N-N interaction sites and also the binding site of the matrix protein (M) and the phosphoprotein (P) onto N. The N self interactions lead to the formation of the virus nucleocapsid. N and M interactions are essential for the virus particle maturation while N and P interactions are necessary for the RNA transcription and replication. The N-N and N-M interactions were studied by the use of deleted protein and also by peptide mapping. For N-P interactions, only deleted mutants were used. This year, the N-P interactions were studied by peptide mapping to identify N-peptide in the binding of P.

Both P and proteins are produced as recombinant proteins in insect cells infected by the baculovirus into which their genes have inserted. Prior to the N-P interactions study, the recombinant P protein was semi-purified and inoculated to mice to produce antibody anti P. A spleen of one of the inoculated mice was used to produce hybridomas. One of these cells monoclonal antibody anti P, the mAb 2B11, was selected and grown. The binding site of this mAb onto P was mapped by the use of both serial deleted recombinant P protein, immunoprecipitation and polyacrylamide gel electrophoresis, and peptide P, peptide ELISA. Using the combination of the two approaches, it was found that the epitope for the mAb 2B11 correspond to the following amino acid sequence <sup>261</sup>ESSERNASVGSVPSARS<sup>280</sup> on the P protein.

This monoclonal being well characterized it was used in the peptide mapping by detecting the P protein bound to N-peptide. Using the monoclonal has the advantage over a polyclonal serum by inducing less background. Preliminary results using this antibody indicated the possible involvement of four N peptides (see the figure here under): one in the middle of N protein and three in the c-terminal. These results have yet to be confirmed by other tests.



### Development of tools for molecular epidemiology of Capripoxviruses and search for suitable targets for their differentiation.

Sheep poxvirus (SPPV), goat poxvirus (GTPV) and lumpy skin disease virus (LSDV) are the only three members of the Capripoxvirus (CaPV) genus of the Poxviridae family. They are responsible for economically important disease in sheep, goats and cattle respectively.

Sheep pox and goat pox diseases are present in Africa (mainly north of the equator), in the Middle East, in Turkey, in India and other Asian countries from Central Asia to China. Sporadic incursions have also been reported in Greece. The endemic region of lumpy skin disease (LSD) is limited mainly to African countries (including Madagascar), with sporadic outbreaks occurring in the Middle East.

CaPVs are thought to be host specific; however, the host affinity remains complex, mainly in the case of sheep pox and goat pox. In addition, there is an increasing amount of evidence for wildlife harbouring CaPVs (mainly, LSDV). Because of their close relationship, LSDV, GTPV and SPPV possess common immunogenic properties; with the consequence that serological tests cannot be used for their differentiation.

Interestingly, the sequencing of the full genome of few CaPV isolates has proven that a molecular based method could represent a good alternative for strain identification and molecular-based epidemiology studies. Indeed, it has been shown using their full genetic information that these viruses are distinct and could be classified as three groups composed of LSDV, GTPV and SPPV. However, due to their relatively large size

(as compared to others viruses), the full genome sequencing of each CaPV isolate cannot be systematically undertaken, because of the cost and also the time required for the analysis. Therefore, the search for suitable host range genes and genotyping targets will offer more realistic tools, as the sequencing of such genes can be efficiently implemented in several laboratories.

In order to contribute to a better understanding of CaPVs molecular epidemiology, the APHL has undertaken a collaborative research work to identify genes that can be used for their genotyping and could carry species specific signatures for the development of differential diagnostic tests. This work was financially supported by the French Ministry of Foreign Affairs (FMFA) fund through the FSP-LABOVET project (Strengthening of five veterinary research laboratories to monitor and control animal diseases in Africa) that run from 2005 to 2009. The main collaborators were, CIRAD (the French Institute which was one of the two European partners of the FSP-LABOVET project, with the APHL), the Institute for Veterinary Disease Control (Austria), Onderstepoort Veterinary Institute (South Africa), Pendik Veterinary Control and Research Institute (Turkey), the Institute for Animal Health, Pirbright Laboratory (UK) and the Institut National de la Médecine Vétérinaire (Algeria).

More than 50 CaPV isolates from different geographical origins were provided by these partners to APHL or CIRAD for sequencing.

Two genes, the CaPV G-protein coupled chemokine receptor (GPCR) and the 30 kDa RNA polymerase subunit (RPO30) genes were identified and sequenced for all isolates. The obtained sequences were compared together with those of 8 others isolates that were retrieved from the GeneBank.

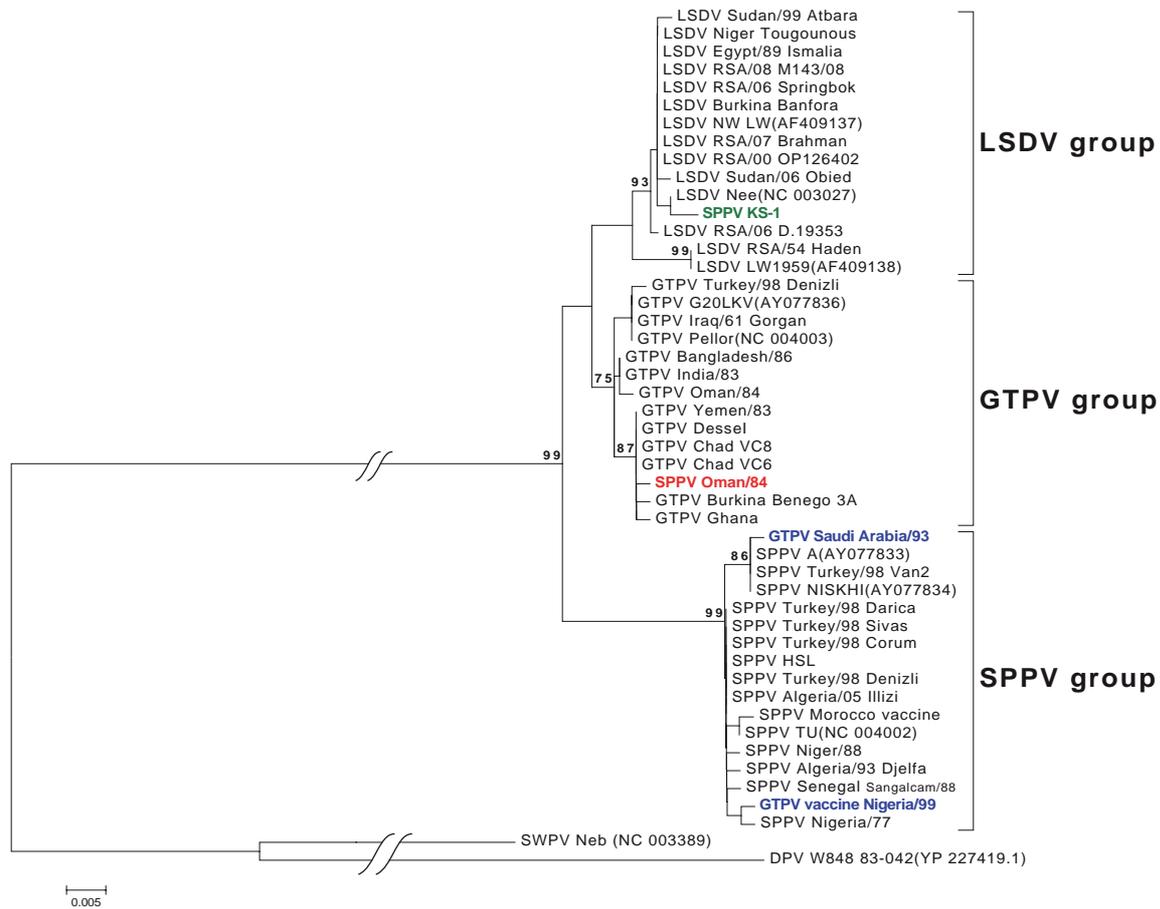


Figure 1. Phylogenetic analysis of 46 capripoxviruses including 8 retrieved from GenBank. The consensus tree was constructed based on the nucleotide sequence alignments of the RPO30 gene homologue using the Neighbor-Joining method and the MEGA4 software. The percentages of bootstrap scores above 70 % (out of 1000 replicates) are indicated on the branches. The scale bar represents genetic distance. The homologue gene sequences from one *Deer poxvirus* isolate and one *Swine poxvirus* isolate retrieved from GenBank were used as out-groups.

The data analysis revealed that the genetic information within each of these genes could be used to produce a phylogenetic tree with a similar topology to what was obtained with the full genome information. As for the full genome tree, 3 clusters composed of LSDV, GTPV and SPPV were found (see example of the RPO30 gene in Figure 1). Another important finding is the presence of isolates that relocate outside the group corresponding to their host. This provides evidence of cross infection with CaPVs such as SPPV infecting a goat or GTPV infecting a sheep. Consequently, this shows the weakness for the actual way of naming and classifying CaPVs that is solely based on the host species from which the virus was first isolated. Therefore, these results strengthen the need to create molecular based tools for CaPVs classification, such as those developed by the APHL and its collaborators. These studies have also allowed identification of species specific signatures in both of these genes that could be used to design simple CaPVs genotyping tools that can be used to screen large number of samples during outbreak. The

most interesting signatures were the presence of a 21-nucleotide deletion on the RPO30 gene of all and exclusively SPPV group members, which could be used to differentiate them from LSDV and GTPV, and the presence of species specific signatures on the CaPVs GPCR that could be used as a target to allow genotyping with hybridisation probes.

These results were disseminated to the scientific community through two peer review publications (Journal of General Virology, 2009, 90, 1967-1977 and Veterinary Microbiology (2010, doi:10.1016/j.vetmic.2010.09.038).

The identification of these two genes as potential genotyping target for CaPVs represents a great contribution that will facilitate understanding CaPVs epidemiology. Some species specific signatures that were found within each of these genes were targeted by the APHL to design simple and cost effective molecular based procedures for CaPVs strains identification which will not require any sequencing work.

# IAEA Collaborating Centre on Animal Genomics and Bioinformatics

The IAEA Collaborating Centre is composed by laboratories from three world class research and teaching Brazilian institutions [Animal Biochemistry and Molecular Biology Laboratory (LBBMA), São Paulo State University, UNESP, Araçatuba; Laboratory of Computational and Systems Biology (LCSB), Instituto Oswaldo Cruz - FIOCRUZ (Oswaldo Cruz Foundation), Rio de Janeiro; and Animal Biotechnology Laboratory (ABL), Animal Sciences Department, Escola Superior de Agricultura Luiz de Queiroz, University of São Paulo, Piracicaba. The Liaison Officer of the Centre is Dr. Jose Fernando Garcia (UNESP).

The Centre is developing a web-based platform to link its various laboratories with the Animal Production and Health Section and Seibersdorf's Laboratories.

The web-based platform will provide updated tools and information to IAEA and FAO Member States, research laboratories and to the international scientific community in general on the fields of genomics and bioinformatics such as laboratory protocols, standard operating procedures (SOPs), nuclear and nuclear related techniques, detailed genome search and analysis tools, radiation hybrid map information, livestock molecular markers database and discussion forums.

The application will initially be more focused on the needs of the Coordinated Research Project 'Genetic variation on the control of resistance to infectious diseases in small ruminants for improving animal productivity'. The IAEA Collaborating Centre, in conjunction with A4 Marketing & Publicidade Ltda has started its construction. It has been named 'Science Satellite' and its logotype is as follows:



The link is: <http://www.a4pe.com.br/ss/index2.php>. The application will include features like Facebook, Twitter, searches, blog, news, and useful information for general users, and will be available in English and Spanish. The programming is based on a PHP platform, using CS and Tableless technology in order to make website access and use faster and easily indexed. The scientific information will be provided by the three laboratories of the IAEA Collaborating Centre and IAEA project counterparts, plus contributions from other interested laboratories.

In the medium term, the implementation of the platform is expected to get:

- Increased capability of MS in the use of livestock genomic resources for the implementation of sustainable breeding programmes.
- International network on the use of the GRB-db for the exchange of information in relation to blood/DNA sequences/primers.
- International network on the use of ProtozoaDB as a database for genomics of protozoan species affecting livestock.
- Increased transfer of technologies (i.e. harmonized and validated procedures, training courses, databases).
- Increased dissemination of IAEA activities in the field of genomics to MS, to the scientific community and to the general public.

The launch of the web tool will be in February 2011.



# Technical Cooperation Projects

TC Project	Description	Technical Officer(s)
BEN/5/003	Veterinary Drug Residue Monitoring Programme <b>Objective:</b> To develop a capacity for veterinary drug residue monitoring in livestock products.	Unger / Diallo
BEN/5/006	Improving Animal Health and Productivity <b>Objective:</b> To strengthen, diagnose, and control African swine fever, and increase animal productivity.	Unger / Diallo
BKF/5/006	Establishment of Feeding Tables for Feedstuffs that are Locally Available to Stockholders in Burkina Faso <b>Objective:</b> To improve the reproductive performance of local livestock bred through food supplementation strategies, develop feeding table for locally available food resources, characterize genetic types of cattle used for milk production, improve the effectiveness of artificial insemination on local cattle breeds, and train a qualified team on animal production (nutrition, feeding, reproduction and genetics).	Garcia Podesta / Odongo
BKF/5/008	Strengthening the Development of Small Ruminant Production <b>Objective:</b> To combat poverty in the rural environment in Burkina Faso by improving production by evaluating the productivity of different genetic types of small ruminants, improving productivity and reproduction performance of local small ruminants through improved feeding and management practises, and evaluating the impact of gastrointestinal and reproductive diseases in small ruminants and the effectiveness of the medicinal plants commonly used by breeders.	Garcia Podesta / Unger
BOL/5/019	Implementing Molecular Techniques to Upgrade the Diagnostic Facilities of National Animal Health Programmes <b>Objective:</b> To strengthen the diagnostic capacity of the animal health laboratories supporting programmes for the control and eradication of animal diseases in Bolivia through the use of molecular diagnostic techniques and training of staff in the use of the techniques; to provide rapid and precise diagnosis of animal diseases to allow better control of economically important diseases of livestock.	Luckins / Schaten
BOT/5/005	Improving Diagnosis of Animal Diseases <b>Objective:</b> To employ nuclear molecular diagnostic techniques for improved diagnosis of trans-boundary animal diseases, such as foot and mouth disease, contagious bovine pleuropneumonia, and avian influenza.	Viljoen
BUL/5/012	Developing and Validating Molecular Nuclear Technologies for Rapid Diagnostics of Foot and Mouth Disease and Genotyping of Indigenous Cattle Breeds <b>Objective:</b> To improve livestock by rapid diagnosis and effective control of foot and mouth disease, and genotyping of indigenous cattle breeds through development and validation of molecular nuclear methodologies.	Viljoen
BZE/5/004	Strengthening the Veterinary Diagnostic Laboratory with Capacities in Polymerase Chain Reaction Diagnosis (Not funded) <b>Objective:</b> To ensure food security through early detection of H5/H7 avian influenza, and other exotic diseases, and to ensure the capacity for quick response to disease outbreaks with epidemiological surveillance.	Viljoen
CAF/5/004	Improving Livestock Production Through Disease Control and Artificial Insemination <b>Objective:</b> To improve animal production in the Central African Republic through livestock disease control and improved breeding by use of artificial insemination.	Unger / Garcia Podesta
CMR/5/015	Use of Nuclear Techniques for Improving Ruminant Productivity & Disease Control <b>Objective:</b> Develop capability for improved breeding by disease control and artificial insemination.	Garcia Podesta / Unger

TC Project	Description	Technical Officer(s)
CMR/5/017	Improving Animal Productivity and Health <b>Objective:</b> To strengthen capacity and outreach regarding artificial insemination in ruminants, and to control livestock diseases impeding reproduction and productivity.	Garcia Podesta / Unger
ERI/5/005	Zoonotic (diseases that can be transmitted from animals to humans) Disease Control and Analysis of Veterinary Residues in Foods <b>Objective:</b> The objective of the project is to determine: 1. The epidemiological prevalence of brucellosis and tuberculosis in the major dairy producing areas; 2. Baseline data on veterinary drug residues in milk and meat products.	Cannavan / Unger / Patel
ERI/5/006	Controlling Major Epizootic Diseases and Other Mycoplasma Infections of Livestock <b>Objective:</b> To improve the control of transboundary animal diseases, and continue the eradication of tuberculosis and brucellosis.	Unger / Luckins
ETH/5/012	Integrating Sterile Insect Techniques for Tsetse Eradication <b>Objective:</b> To eradicate the tsetse fly from the southern Rift Valley, thereby creating an environment conducive to livestock development and improved agricultural production.	Feldman / Parker / Viljoen
ETH/5/014	Monitoring and Control of Major Animal Diseases <b>Objective:</b> To strengthen the diagnostic capacity of the National Veterinary Institute to monitor and control trans-boundary diseases, particularly foot and mouth disease and contagious bovine pleuropneumonia.	Viljoen
GAB/5/002	Diagnosis and Control of Animal Diseases <b>Objective:</b> To aid identification and control of livestock diseases.	Luckins / Unger
HON/5/004	Improving the Nutrition and Health Conditions of Livestock in Order to Increase Productivity and Reproductivity (Phase II) <b>Objective:</b> To strengthen and improve livestock production in Honduras.	Garcia Podesta / Odongo / Viljoen
HON/5/005	Improving the Nutrition and Health Conditions of Livestock in Order to Increase Productivity and Reproductivity (Phase II) <b>Objective:</b> To strengthen and improve livestock production in Honduras.	Garcia Podesta / Odongo / Viljoen
INS/5/034	Development of Environmentally Sound Livestock and Agricultural Production <b>Objective:</b> To improve livestock productivity without adversely affecting the environment through improved feed supplementation strategies, managing nutrient waste on farms and reducing methane emissions.	Odongo
IVC/5/030	Assessing the Genetic Profile for Improved Livestock Production <b>Objective:</b> To assess the genetic profile of livestock for the effective revival of stockbreeding in Côte d'Ivoire.	Garcia Podesta / Unger
KEN/5/027	Assessment of Local Feed Resources for Enhancing Fertility and Productivity of Smallholder Dairy Cattle <b>Objective:</b> To assess the potential of local feed resources for enhancing the fertility and productivity of smallholder dairy cattle in the Nakuru District of Kenya.	Odongo / Garcia Podesta
KEN/5/028	Applying Nuclear Based Techniques to Control Animal diseases <b>Objective:</b> To improve the capacity to diagnose and carry out surveillance of Contagious Bovine Pleuro-Pneumonia (CBPP), Brucellosis, Rift Valley Fever (RVF), Peste Des Petits Ruminantes (PPR) and Highly Pathogenic Avian Influenza (HPAI) using nuclear and related technologies.	Unger
MAG/5/016	Applying Nuclear Techniques to Optimize Animal Production <b>Objective:</b> To increase animal production through the improvement of animal health and control reproduction in the Amoron'i Mania region.	Garcia Podesta / Odongo / Luckins

TC Project	Description	Technical Officer(s)
MAU/5/002	<p>Improving the National Capacity in Diagnostics for Animal Diseases (Infection and Parasitic Diseases)</p> <p><b>Objective:</b> To strengthen the diagnostic capacity of the Centre National D'Elevage et de Recherches Veterinaires (CNERV) to monitor and control trans-boundary animal diseases, particularly foot and mouth disease and contagious bovine pleuropneumonia.</p>	Luckins / Schaten
MAU/5/003	<p>Improving the National Capacity in Diagnostics for Animal Diseases (Infection and Parasitic Diseases)</p> <p><b>Objective:</b> To strengthen the diagnostic capacity of the Centre National D'Elevage et de Recherches Veterinaires (CNERV) to monitor and control trans-boundary animal diseases, particularly foot and mouth disease and contagious bovine pleuropneumonia.</p>	Unger / Schaten
MLI/5/023	<p>Improving National Capabilities for Characterization of Serotypes of Major Animal Diseases Using Molecular Biology Techniques</p> <p><b>Objective:</b> To identify various serotypes present in Mali in order to improve animal health and increase productivity in milk and meat through increased capabilities for diagnosis and control of foot and mouth disease, trypanosomes and tuberculosis.</p>	Unger / Viljoen / Schaten
MON/5/013	<p>Diagnosis and Surveillance of Transboundary Animal Diseases and Production of Diagnostic Reagents</p> <p><b>Objective:</b> To obtain international recognition of freedom from several transboundary animal diseases, to develop a capacity for the local production, standardization and validation of diagnostic reagents and diagnostic kits, and to establish a quality system for diagnosis of transboundary animal diseases using the local produced diagnostic kits.</p>	Luckins / Viljoen
MON/5/016	<p>Improving Productivity of Cattle, Camels and Yaks Through Better Nutrition and Reproductive Management</p> <p><b>Objective:</b> To increase milk, meat and wool production of yaks, cattle and camels by improving the quality and quantity of feed with high nutritional value and tolerance to low temperature and improving the genetic potential using artificial insemination coupled with radio immunoassay for progesterone.</p>	Odongo / Garcia Podesta
MON/5/017	<p>Supporting the Sustainable Production and Supply of Vaccines and Diagnostic Kits for Transboundary Animal Diseases</p> <p><b>Objective:</b> To produce vaccines and diagnostic kits for transboundary animal diseases.</p>	Viljoen / Luckins
MOR/5/030	<p>Improving Sheep and Goat Production in Morocco through Genomic and Reproductive Physiology Characterization with the Help of Radio-immunoassay and Molecular Techniques (Not yet funded)</p> <p><b>Objective:</b> Increase sheep and goats for consumption and producers' revenue while preserving natural resources.</p>	Garcia Podesta / Malek
MOZ/5/002	<p>Promoting sustainable Animal Health, Reproduction and Productivity Through the Use of Nuclear and Related Techniques</p> <p><b>Objective:</b> To obtain sustainable improvement in animal reproduction and breeding and animal health through the use of nuclear and nuclear related technologies.</p>	Viljoen
MYA/5/013	<p>Integrated Approach for Enhancing Cattle Productivity</p> <p><b>Objective:</b> To improve smallholder dairy cattle production in Yangon and Mandalay regions.</p>	Garcia Podesta / Odongo
MYA/5/015	<p>Strengthening the National Capacity for the Production of Veterinary Vaccines</p> <p><b>Objective:</b> To enhance the national capacity for quality vaccine production to support efforts to control infectious diseases in livestock production, particularly FMD.</p>	Unger / Diallo

TC Project	Description	Technical Officer(s)
MYA/5/018	<p>Enhancing the Lifetime Health and Performance of Offspring and Improving the Profitability of Livestock Production Systems Through Selective Breeding and Management of the Maternal Environment</p> <p><b>Objective:</b> To improve livestock production and thereby increase profitability through improved management of the maternal environment and health care programmes; b) To train technicians in advanced technologies in the field of research and development, breeding, reproduction, dairy production, nutrition and waste management and train technical staff in livestock data analysis and data processing.</p>	Garcia Podesta / Diallo / Unger
NER/5/013	<p>An Integrated Approach for Improvement of Livestock Productivity</p> <p><b>Objective:</b> To increase the productivity of livestock through implementation of an integrated programme dealing with nutrition and reproduction.</p>	Odongo / Garcia Podesta / Diallo
PER/5/029	<p>Genomics of the Alpaca: Identification of Expressed Genes and Genetic Markers Associated with Productivity and Embryonic Mortality</p> <p><b>Objective:</b> To identify and characterize the factors associated with embryonic mortality in alpacas.</p>	Garcia Podesta / Malek
RAF/5/055	<p>Support to African Union's Regional Programmes for Control and Eradication of Major Epizootics</p> <p><b>Objective:</b> To support within the framework of a strategic partnership with the African Union, the global effort of control and eradication of major trans-boundary animal diseases affecting livestock in the region led by the Inter-African Bureau for Animal Resources (AU/IBAR). This programme will aim at helping African countries to improve and produce livestock to ensure their role and participation in international markets that will lead to poverty alleviation and increased livelihoods. The specific objectives of the project are (i) to provide support to selected national veterinary laboratories to implement a quality assured disease control programme; (ii) to transfer appropriate and state-of-the-art technology to support diagnostic, surveillance and epidemiological activities relating to the control of major livestock diseases; and (iii) to support the establishment of a regional centre in Africa (Pan African Veterinary Vaccine Centre [PANVAC]) that would be responsible for (a) the production, assembly and distribution of diagnostic kits; (b) evaluating and monitoring the development of quality assured animal vaccines and (c) advising on the use of vaccines and vaccine strategies.</p>	Viljoen / Lelenta
RAF/5/057	<p>Strengthening Capacities for the Diagnosis and Control of Transboundary Animal Diseases in Africa (AFRA)</p> <p><b>Objective:</b> To strengthen the diagnostic capacity of national veterinary services to monitor and control major transboundary animal diseases, particularly foot and mouth disease, peste des petits ruminants and contagious bovine pleuropneumonia.</p>	Unger / Diallo
RER/5/015	<p>Supporting Early Warning and Surveillance of Avian Influenza Infection in Wild and Domestic Birds and Assessing Genetic Markers for Bird Resistance</p> <p><b>Objective:</b> To establish early bird flu diagnosis and assessment of genetic markers for AI resistance with nuclear molecular methods in the region of Bosnia and Herzegovina, Bulgaria, Croatia, the Former Yugoslav Republic of Macedonia, Montenegro, Serbia, Turkey, Uzbekistan, Kyrgyzstan and the Russian Federation.</p>	Viljoen / Diallo
RLA/5/049	<p>Integrated Control of Fascioliasis in Latin America (in support of National Programmes)</p>	Viljoen / Schaten
SIL/5/010	<p>Improving the Productivity of Ndama Cattle In Sierra Leone</p> <p><b>Objective:</b> To strengthen the diagnostic capacity to monitor and control animal diseases affecting cattle, (ii) to apply feeding strategies and supplementation packages, and (iii) to produce hybrids with greater potential for increased growth rate and milk yields.</p>	Garcia Podesta / Odongo / Viljoen
SIL/5/011	<p>Controlling Economically Important Livestock Diseases</p> <p><b>Objective:</b> To design epidemiological surveys and adopt appropriate rapid laboratory techniques for the diagnosis of PPR and NCD in small ruminants and local chickens.</p>	Unger

TC Project	Description	Technical Officer(s)
SRL/5/041	<p>Maximizing Productivity on Goat Farms through Cost-Cutting and DNA-Based Technology in Selection for Breeding</p> <p><b>Objective:</b> To improve the productivity of goats of small-holder farmers in Sri Lanka, by introducing new strategies such as supplementary feeding, improved management practices and disease control and by transferring genetic technologies to assist in proper selection of superior breeding animals.</p>	Garcia Podesta / Odongo / Viljoen / Malek
SRL/5/042	<p>Applying Molecular Diagnostics to Zoonotic Diseases</p> <p><b>Objective:</b> To enhance the long-term epidemic preparedness by developing competence in molecular diagnosis and surveillance of zoonotic infections.</p>	Kashyap (NAHU) / Unger
SUD/5/031	<p>Setting up a National Network for the Control of Livestock Diseases that affect Exports</p> <p><b>Objective:</b> To establish capacity to diagnose Brucellosis in ruminants to improve food safety and secure animal exports.</p>	Unger
UGA/5/028	<p>Improving the Capacity for Diagnostic of Animal Diseases</p> <p><b>Objective:</b> To strengthen the diagnostic capacity of the Diagnostics and Epidemiology Laboratory of the Ministry of Agriculture, Animal Industry and fisheries to monitor and control transboundary animal diseases of importance (e.g. CBPP, FMD, AI, Rabies, Brucellosis and RVF) to Uganda.</p>	Viljoen / Unger
UGA/5/030	<p>Improving the Diagnostic Capacity in Animal Diseases (Phase II)</p> <p><b>Objective:</b> To strengthen the diagnostic capacity of the National Animal Diseases Diagnostics and Epidemiology Laboratory in the detection of animal disease and food-borne pathogens including drug residues.</p>	Unger / Luckins
URU/5/026	<p>Increasing the Profitability of Dairy Producers by Improving Reproduction Efficiency, Rational Sustainable Use of Genetic Resources</p> <p><b>Objective:</b> To implement integrated management strategies to improve the profitability of medium size grazing dairy farms by means of (a) integrated nutritional strategies; (b) strategic reproductive interventions; and (c) marker-assisted selection.</p>	Garcia Podesta / Odongo
ZAM/5/025	<p>Development of Feeding Strategies for Smallholder Dairy Animals in Njolwe and Palabana Dairy Tenant Schemes</p> <p><b>Objective:</b> To improve household food security and income generation among small scale farmers through increased production and marketing of livestock by developing sustainable feeding and breeding strategies based on increased use of locally available resources.</p>	Garcia Podesta / Odongo

# Publications

## Water requirements for livestock production: a global perspective

*A.C. Schlink, M.-L. Nguyen & G.J. Viljoen.*

**Rev. sci. tech. Off. int. Epiz., 2010, 29 (3) In Press**

Water is a vital but poorly studied component of livestock production. It is estimated that livestock industries consume 8% of the global water supply, with most of that water being used for intensive, feed-based production. This study takes a broad perspective of livestock production as a component of the human food chain, and considers the efficiency of its water use. Global models are in the early stages of development and do not distinguish between developing and developed countries, or the production systems within them. However, preliminary indications are that, when protein production is adjusted for biological value in the human diet, no plant protein is significantly more efficient at using water than protein produced from eggs, and only soybean is more water efficient than milk and goat and chicken meat.

In some regions, especially developing countries, animals are not used solely for food production but also provide draught power and fertiliser for crops, as well as using fibre and crop by-products that would otherwise go to waste.

The livestock sector is the fastest-growing agricultural sector, which has led to increasing industrialisation and, in some cases, reduced environmental constraints. In emerging economies, increasing involvement in livestock is related to improving rural wealth and increasing consumption of animal protein. Water usage for livestock production should be considered an integral part of agricultural water resource management, taking into account the type of production system (e.g. grain-fed or mixed crop-livestock) and scale (intensive or extensive), the species and breeds of livestock, and the social and cultural aspects of livestock farming in various countries.

Limited availability of water or the presence of contaminants in the supply have a significant impact on animal health and productivity. The water demands of livestock may also compete with those of the human population and water required for crop production. Crops can make direct use of rainfall or stored water through irrigation, whereas animals consume crops or pastures, leading to potential reductions in water efficiencies for food production. This water must be added to the water directly consumed by the animals to maintain life as

well as to the water used during product processing. This apparent 'inefficiency' of water use has been highlighted in recent accounting models of global water use.

This paper considers the role of livestock production and the efficiency of its water usage in producing protein for human consumption. The issues of water efficiency and the role of livestock in environmental pollution have been used to question the continued role of livestock as a human food source. Models of water efficiency are in an early stage of development, compared to those of livestock pollution, but water efficiency issues have the potential to gravely affect livestock production. The current models imply that livestock production is an efficient source of human food. However, unfavourable perceptions could lead to reduced demand by consumers and policy planners, who may believe that the negatives of livestock production far outweigh the positives, in both the developed and developing worlds. If this attitude towards livestock production is allowed to go unchallenged, it will have severe long-term implications for livestock producers and professionals, such as farmers, veterinarians and production specialists. Veterinarians have long played a role in ensuring that livestock have access to clean and adequate water supplies but, to date, have been reluctant to enter the broader debates of water competition between different production systems and efficiency of water use by livestock.

This paper highlights the background to the role of livestock production in the global economy and provides a broad overview of water usage by livestock. The authors propose an alternative way of assessing the efficiency of water use by livestock: through the concept of human, dietary utilisable protein. This approach has not been considered by previously published models.

All the global models are in the early stages of development and do not specifically address the issues of developed and developing countries, or their various production systems. However, the models are starting to highlight the role of global trade in effective water transfer between countries. At this point, they are not appropriate for considering the inherent complexity of livestock usage of water, which varies significantly between regions, due to historical differences, as well as differences in production systems and species. The authors recognise that such considerations must be included in more detailed studies of water use in the future.

## Virus-like particles expressing the nucleocapsid gene as an efficient vaccine against Rift Valley fever virus.

*Pichlmair A, Habjan M, Unger H, and Weber F.*

**Vector Borne Zoonotic Dis., 10: 701-703**

Rift Valley fever virus (RVFV), a member of the family Bunyaviridae, regularly accounts for large and severe outbreaks among humans and livestock in Africa and Arabia. Therefore, safe and efficient vaccines are highly needed. Here, we report the production of recombinant virus-like particles (VLPs) that, in addition to their similarity to RVFV particles, are able to express the viral nucleocapsid (N) gene. A single inoculation of  $1 \times 10^6$  of these N-VLPs was sufficient to protect 100% of mice from infection with a lethal dose of  $1 \times 10^5$  PFU of RVFV. Our study demonstrates that N-VLPs can be considered as a safe and efficient vaccine against the emerging pathogen RVFV, and that VLPs that actively produce a viral antigen may be considered a strategy to improve the immunogenicity of VLPs in general.

## Use of the Capripoxvirus homologue of Vaccinia virus 30 kD RNA polymerase subunit (RPO30) gene as a novel diagnostic and genotyping target: development of a classical PCR method to differentiate Goat poxvirus from Sheep poxvirus.

*Charles Euloge Lamiena, Christian Le Goff, Roland Silber, David B. Wallace, Vely Gulyaz, Eeva Tuppurainen, Hafsa Madani, Philippe Caufour, Tajelser Adam, Mehdi El Harrak, Antony George Luckins, Emmanuel Albina and Adama Diallo*

**Veterinary Microbiology. (2010) (in press)**

*Sheep poxvirus* (SPPV), *Goat poxvirus* (GTPV) and *Lumpy skin disease virus* (LSDV) are capripoxviruses (CaPVs) responsible for causing severe poxvirus disease in sheep, goats and cattle, respectively. Serological differentiation of CaPVs is not possible and strain identification has relied on the implicitly accepted hypothesis that the viruses show well defined host specificity. However, it is now known that cross infections can occur and authentication of identity based on the host animal species from which the strain was first isolated, is not valid and should be replaced with molecular techniques to allow unequivocal strain differentiation.

To identify a diagnostic target for strain genotyping, the CaPV homologue of the *Vaccinia virus* E4L gene which encodes the 30 kD DNA-dependant RNA polymerase subunit, RPO30 was analyzed. Forty six isolates from different hosts and geographical origins were included. Most CaPVs fit into one of the three different groups according to their host origins: the SPPV, the GTPV and the LSDV group. A unique 21-nucleotide deletion was found in all SPPV isolates which was exploited to develop a RPO30-based classical PCR test to differentiate SPPV from GTPV that will allow rapid differential diagnosis of disease during CaPV outbreaks in small ruminants.

## Real time PCR Method for Simultaneous Detection, Quantitation and Differentiation of Capripoxviruses

*Charles Euloge Lamiena, Mamadou Lelemta, Wilfried Gogerb Roland Silber, Eeva Tuppurainen, Mirta Matijevice, Antony George Luckinsa and Adama Diallo.*

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The genus *Capripoxvirus* (CaPV) comprises three members namely, *sheep poxvirus* (SPPV), *goat poxvirus* (GTPV) and *lumpy skin disease virus* (LSDV) affecting sheep, goats and cattle respectively. CaPV infections produce similar symptoms in sheep and goats, and the three viruses cannot be distinguished serologically. Since there are conflicting opinions regarding the host specificity of CaPVs, particularly for goatpox and sheeppox viruses, the development of rapid genotyping tools will facilitate more accurate disease diagnosis and surveillance for better management of capripox outbreaks.

This paper describes a species-specific, real time Polymerase Chain Reaction (PCR), based on unique molecular markers that were found in the G-protein-coupled chemokine receptor (GPCR) gene sequences of CaPVs, that uses dual hybridization probes for their simultaneous detection, quantitation and genotyping.

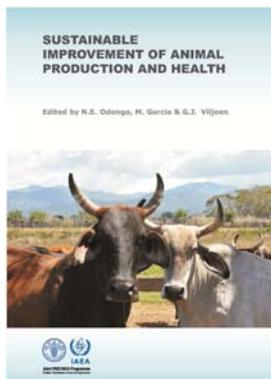
The assay can differentiate between CaPV strains based on differences in the melting point temperature ( $T_m$ ) obtained after fluorescence melting curve analysis (FMCA). It is highly sensitive and presents low intra- and inter-run variation.

This real time PCR assay will make a significant contribution to CaPV diagnosis and to the better understanding of the epidemiology of CaPVs by enabling rapid genotyping and gene-based classification of viral strains and unequivocal identification of isolates.

**Key words:** Capripoxvirus, sheep pox, goat pox, lumpy skin disease, G-protein-coupled chemokine receptor, genotyping.

## Recently Published

### Sustainable Improvement of Animal Production and Health



The growing world population is vulnerable to limitations in the production of agricultural products and to any change, be it climatic realities and/or variations or civil strife that upset the delicate balance of providing affordable food for all. It is alarming that the world's poorest people, some one billion living mostly in Africa and Asia, depend on

livestock for their day-to-day livelihood. To reduce poverty, fight hunger and ensure global food security, there is an urgent need to increase livestock production in sustainable ways. An International symposium on 'Sustainable Improvement of Animal Production and Health' was organized by the APH subprogramme of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture in cooperation with the Animal Production and Health Division of the Food and Agriculture Organization of the United Nations in 2009 to address the animal husbandry and public health issues that threaten global food security and safety.

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### CD-ROMs

A CD-ROM is available dealing with training material for the diagnosis of rinderpest and for the preparation for the OIE pathway. It was produced under an IAEA Technical Cooperation project RAF/0/013 ICT based training to strengthen LDC capacity. Contact Gerrit Viljoen at [g.j.viljoen@iaea.org](mailto:g.j.viljoen@iaea.org) for further information.

A new batch of CDs with a training package to help artificial insemination (AI) technicians to improve the performance of AI and field services provided to farmers was produced for users with a slow internet connection and is now available through the APHS. It is also accessible from the AP&H Section website:

<http://www-naweb.iaea.org/nafa/aph/index.html>

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<http://www-naweb.iaea.org/nafa/aph/index.html>

## Impressum

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