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Bos taurus x Bos indicus crosses on the slopes of the Andes in the tropics of Peru

To Our Readers

Dear Colleagues,

It is with great enthusiasm that I address you in a new era – the era of a rinderpest free world. The OIE and FAO have declared the world-wide eradication of Rinderpest recently – but more about this later. The first part of this year has been a busy time for all personnel in the subprogramme. Apart from our regular Coordinated Research Project (CRP) activities and our technical support given to national and regional Technical Cooperation projects (TC), we were involved in the technical planning of projects for the new TC projects by Member States for the 2012/2013 biennial project cycle. We were also occupied with finalizing the IAEA's 2012/2013 Work and Budget Programme. It is hoped that our inputs will serve the best interests of our Member States. Please look at our web site and our Animal Production and Health Newsletter to familiarize yourselves with all the activities of the subprogramme.

IAEA International Atomic Energy Agency

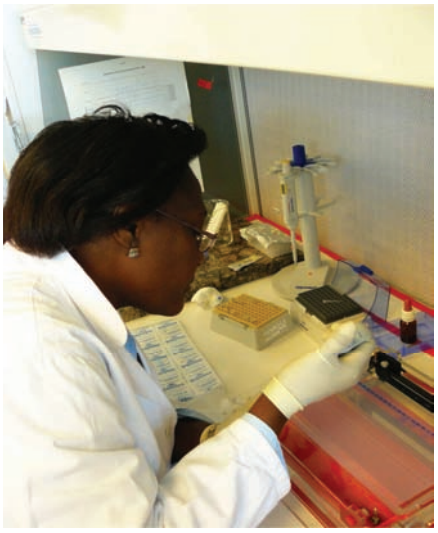
Rinderpest is arguably the most devastating disease of cattle and for centuries a major cause of famine and poverty - but not anymore. This great achievement comes some 30 years after smallpox, the only other virus to be eliminated from the world, was declared eradicated by the World Health Organization. Rinderpest was first described by Prof. Bernardino Ramazzini in 1712. It caused major livestock losses in Northern Italy until Dr. Lancisi, on request by Pope Clement XI, suggested the 'stamping-out' policy or the slaughter of all infected and exposed animals for the control of this disease. This remained the procedure of choice for keeping rinderpest under control over the next three centuries. However, the advent of steam powered ships enabled greater international trade in livestock and accelerated the spread of infectious livestock diseases such as rinderpest. The most devastating effects of rinderpest were experienced in Africa, where infected cattle imported from India into Eritrea in the 1880's spread the disease rapidly into different parts of the continent, killing millions of cattle and wild animals and causing widespread starvation. Once again, the stamping-out policy eventually controlled the disease.

The concept of preventing rinderpest by inducing immunity was first attempted in 1774 in the Netherlands using a rudimentary form of vaccination. The breakthrough, however, came in 1928 when a vaccine developed in India in goats was found to confer immunity in cattle. This vaccine was used extensively in Asia and Africa but occasionally caused disease. Advances in virology led in 1957 to a safer tissue culture version of vaccine developed by Prof. Walter Plowright whilst working in Kenya. This vaccine, giving life-long immunity through a single injection, formed the basis of the global campaign to eradicate rinderpest. The development in Niger of a heat stable version of the vaccine in 1990 improved the efficiency of the vaccination campaign especially in remote areas. In the 1930s, country specific mass vaccination campaigns were employed to reduce the incidence of rinderpest. The Inter-African Bureau of Epizootic Diseases was founded in 1950 to coordinate control efforts among different countries and eliminate rinderpest from Africa. The first internationally backed campaign was the Joint Programme 15 (JP 15) led by the Organization of African Unity (OAU, the present African Union) which ran from 1962 to 1976 and was aimed to control rinderpest in 22 African countries. Although JP15 was highly successful in controlling outbreaks, vaccination was stopped prematurely, leaving two foci of infection which led to a resurgence of the disease in the late 1970s and early 1980s. Funded principally by the European Community, the Pan African Rinderpest Campaign (PARC) was initiated by OAU in 1987 in 34 African countries to attempt to eradicate the disease from Africa. In order to ensure success, it was agreed to seek clear scientific based evidence that national vaccination programmes were indeed effective and that sufficient animals were immune to ensure elimination of the virus. Similar concerted efforts under the aegis of FAO were organized in South and West Asia. Due to the success of these programmes in eliminating the virus, FAO launched the Global Rinderpest Eradication Programme (GREP) in 1994 in close association with OIE, IAEA and other partners to coordinate the global eradication of rinderpest and its declaration by 2011.

Although vaccination against rinderpest was at the heart of the eradication efforts, vaccination alone would not have done the job without the development and deployment of diagnostic tests to monitor the success or otherwise of vaccination campaigns. Initially, animal immune responses to vaccination were assessed mainly by the virus neutralization test. Unfortunately this test could not be standardized and implemented in many veterinary laboratories, and it was therefore unsuitable both for detecting antibodies to the virus in the many thousands of blood samples required for monitoring vaccination campaigns and for detecting the virus itself. Besides, without on-the-spot training and a system in place to identify and resolve problems faced by national veterinary services in their use, these tests would also have been insufficient. The Joint Division responded to these unmet needs by introducing a radio immunoassay (RIA) based nuclear related diagnostic technique, the enzyme linked immunosorbent assay (ELISA), which had all the required attributes to support PARC. These tests were produced as kits to simplify transport and use and distributed to all countries within PARC and later, worldwide along with the necessary training and support. It was clear from the outset that the ELISA was an ideal tool to meet the needs of PARC for effective serological surveillance to confirm that indeed sufficient animals were being vaccinated to ensure elimination of the virus from a population. Working closely with the Pirbright Institute for Animal Health (the developer of the rinderpest specific ELISA platform), the Animal Production and Health Section in Vienna and its associated laboratory at Seibersdorf set about developing, standardizing and validating an ELISA for the detection of antibodies to rinderpest virus in cattle that would meet international standards established by the OIE. Once the assay met these international requirements, it was adapted to a kit format, linked to a quality assurance programme for use of the kit and matched against a standardized set of laboratory equipment for its routine use. Over and above this, agreement was reached on the interpretation of the results and how these could be used to determine the effectiveness of vaccination.

Critical at that stage was now to ensure the sustainable transfer of the ELISA technology, the associated equipment and kits and the submission of results at both the national and international level. A key component of this involved the computerization of data collection and result analysis. Support to the laboratories and scientists undertaking the work in rinderpest affected countries was provided principally in two ways: through FAO/IAEA Coordinated Research Programmes (CRPs) and IAEA

Technical Cooperation Projects. The CRPs provided research contracts to institutions in Member States thereby creating a network of laboratories and individuals that could test and assist in refining and validating the ELISAs under different conditions and work towards standardizing the tests.



These projects also included research agreement holders from the World Reference Laboratory for Rinderpest, Pirbright in the 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) in France to provide advanced technical support. The first CRP which was initiated in 1986 with generous financial assistance from the Swedish International Development Authority (SIDA) enabled the Joint Division to introduce the rinderpest ELISA technique developed by the Animal Health Institute, UK into 21 national veterinary laboratories in 19 African countries and to have the test for detecting antibodies fully validated and standardized.

A second CRP, also funded by SIDA saw the introduction of a test that was both more sensitive and that could distinguish between antibodies to rinderpest and those against the closely related PPR virus – attributes that meant it could be used as a surveillance tool within national rinderpest campaigns. Quality assurance systems were also put in place, standardized sampling frames were designed and computer software was developed for data management and analysis. Support by SIDA ended in 1993, having successfully established the capacity to carry out surveillance for rinderpest in 17 African countries. Thereafter, further European Community funding to mainly OAU/IBAR established the PARC epidemiology project commencing in 1996. The laboratory component was fundamental to this endeavour and the Joint Division therefore implemented a third Rinderpest CRP with FAO and PARC, aiming to consolidate the laboratory network and the technology base to perform all monitoring activities. When this last CRP was completed the surveillance capability for Rinderpest was available in 22 African countries and another 16 countries in the Asian region.

The more than 60 National and regional TCPs funded through the IAEA's Department of Technical Cooperation also played a vital role in supporting PARC and GREP by supporting the establishment and strengthening of national laboratory infrastructure and human resources through the provision of equipment and training fellowships and courses in Africa and Asia. In planning and implementing these projects, the high level of teamwork that characterised the interactions between counterparts and staff within TC and the Joint Division staff ensured that inputs of hardware, reagents and guidance on technical issues advice were both appropriate and provided in a timely manner.

The declaration of rinderpest eradication will not end the commitment of international bodies to promote continued vigilance by veterinary authorities concerning this disease. As vaccination is abandoned, laboratories will be requested to sequester their remaining virus stocks in order to reduce risks of laboratory 'outbreaks'. At the same time laboratories have to be identified where viruses, sera and vaccine banks can be maintained or that can serve as virus depositories. Also, a monitoring strategy has to be devised to ensure that early warning systems are in place and laboratory capacities are sufficient to continue surveillance. The OIE, FAO and IAEA (through the Joint FAO/IAEA Division) will assist these efforts by providing advice on further research and the storage of potentially hazardous biological materials. With this I want to give credit to all the past and present Joint FAO/IAEA Division Staff that so effortlessly worked towards the aim of ridding the world from Rinderpest.

Lastly, some news from the subprogramme - Kathrin Schaten returned to Germany where she is planning to finish her Master's degree in Public Health and Epidemiology at the Berlin University. Later in the year she plans to take up a PhD position in the University of Edinburgh; Anna Slawinska completed her consultancy in the Animal Production and Health Laboratory at Seibersdorf and has resumed her position as Assistant Professor in the University of Technology and Life Sciences in Bydgoszcz (Poland). She is assigned undergraduate and graduate courses to which she lectures on different aspects of life sciences. She is also active in applying for external funds and collaboration. At present she coordinates a state research project to determine if the administration of synbiotics *in ovo* has an advantageous impact on the development of the chicken immune system. In the near future Mrs Slawinska and collaborators plan to

adopt new genomic technologies in their laboratory (Illumina BeadChip, Roche 454 sequencing) for immune-related gene analysis in poultry and waterfowl. We wish them both well. Mr. Rudolf Pichler from Austria joined APHL in March 2011 as Laboratory Technician to work on animal genetics. He has 20 years' experience on molecular biological technologies. Before joining the APHL, Mr. Pichler worked at the Austrian Institute of Technology where he was involved in the development of an IVD test kit for detection of human pathogens. He has also participated in identifying methylation-related tumour markers for cancer detection. His field of expertise is PCR, real time PCR and DNA- and protein-microarrays. At AHPL he will be involved in detection of single nucleotide polymorphism (SNP) for markers associated with disease resistance in animals using new technical methods (KASP- and microarray-technology). Looking back at the activities of the past six months, we had several workshops, training courses, research co-ordination meetings (RCMs) and consultants meetings. Activities scheduled for the next half-year include project review meetings, RCM's, inter-regional training courses and regional workshops. Both past and future activities are discussed in further detail in this Newsletter and are also accessible at our website. Let us know if you have any ideas, comments, concerns or questions. We thank all those who have responded to our request to update their contact and mailing address details and urge those who haven't to please do so by replying to S.Piedra-Cordero@iaea.org. This will ensure that the next copy of our newsletter will be received. By also sending us the addresses of unsubscribed colleagues we will be able to widen our network.

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Forthcoming Events

National Training Course on Basic Epidemiology and the Surveillance of Important Livestock Diseases (BOL5019)

Technical Officer: Ivancho Naletoski

The National Training Course will be held in cooperation with the Government of Bolivia through the Laboratorios de Investigacion y Diagnostico Veterinario (LIDIVECO) from 13 to 24 June 2011 in Cochabamba, Bolivia. Fourteen participants from 3 main Bolivian laboratories and 1 participant from the veterinary authority will attend the training. Two international experts will lead the course.

The course will include theoretical and practical exercises in the following fields: i) Application of Epidemiology in Veterinary Practice; ii) Introduction to Study Designs; iii) Evaluation of Diagnostic Tests; iv) Measures of Disease Frequency; v) Outbreak Investigation and vi) Establishing Causation. The use of free Internet software for data analysis (Win Episcope or similar) will also be included.

Regional Training Course on Transboundary and Zoonotic Animal Diseases: Early Detection, Surveillance and Epidemiology

Technical Officer: Charles Lamien

The Regional Training Course on Transboundary and Zoonotic Animal diseases: Early detection, Surveillance and Epidemiology will be held from 20 June to 1 July 2011 in Uganda. The purpose of the course is to enhance knowledge and practical application of early detection and surveillance of important transboundary and Zoonotic animal diseases that have high incidence in the Congo basin region of Africa.

Workshop on Biosafety, Virus Sequestration and Risk Analysis for Laboratories holding Rinderpest Virus Infective Material (FAO funded)

Technical Officers: Adama Diallo and Hermann Unger

The Training workshop on biosafety, virus sequestration and risk analysis for laboratories holding rinderpest virus infective material will be held from 4 to 7 July 2011 at AU-PANVAC in Debre Zeit, Ethiopia.

The purpose of this workshop is to prepare the future activities after the declaration of Rinderpest eradication, specifically the research needs implying the use of whole virus, and the bio-containment requirements, the decontamination of and sequestration of virus containing stocks.

Regional Training Course on Advanced Bioinformatics and Laboratory Data Management for Enhanced Quality Assurance and Quality Control RER/5/015

Technical Officer: Adama Diallo

The Regional Training Course on Advanced Bioinformatics and Laboratory Data Management for Enhanced Quality Assurance and Quality Control will be held from 11 to 22 July 2011 at the APH Laboratory in Seibersdorf.

The Regional Training Course (RTC) aims at enhancing knowledge and practical application of advanced bioinformatics and laboratory data management to improve quality assurance (QA) and quality control (QC) through good laboratory practices in analysing, storing and retrieving genomic information and managing biobank resources.

Regional (AFRA) Training Course on Epidemiological Surveillance Targeting PPR and CBPP (RAF5057)

Technical Officer: Hermann Unger

The Regional (AFRA) Training Course on Epidemiological SurveillanceTargeting PPR and CBPP will be held from 11 to 22 July 2011 in Lusaka, Zambia.

The purpose of the training course is to update the knowledge on the Epidemiology of PPR and CBPP, present and discuss the potential approaches to perform disease and sero-surveillance, perform serological tests for PPR and CBPP, data interpretation and trouble shouting, and perform rapid disease diagnosis by isothermal PCR methods, their interpretation and trouble shooting. This AFRA training course is for veterinarians working on the control and surveillance of PPR and CBPP in Africa. The participants must be directly involved in performing the different diagnostic tests as well as the planning of surveillance activities.

Mid-term Coordination Meeting (RAF5057/9002/01)

Technical Officer: Hermann Unger

This meeting will be held from 10 to 14 October 2011 in Entebbe, Uganda under Technical Cooperation Project RAF5057 on Strengthening Capacities for the Diagnosis and Control of Transboundary Animal Diseases in Africa (AFRA)

The objective of the meeting is to review/update the project document with emphasis on the project workplan to evaluate the project and to identify key actions to be taken by each participating country to achieve the project output.

Training Course on Classical and Molecular Veterinary Virology

Technical Officer: Adama Diallo

The Training Course on Classical and molecular Veterinary Virology will be held from 28 November to 9 December 2011 in Austria.

The first week is dedicated to Molecular Virology-Multiple viral pathogens detection and will be held at the Animal Production and Health Laboratory in Seibersdorf. The second week will cover Classical Virology and will be held at the Clinical Virology, University of Veterinary Medicine, Veterinaerplatz 1, 1210 Vienna, Austria

Past 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) was held from 10 to 14 January 2011 at FAO Headquarters in Rome, Italy, in collaboration with FAO and EU-FMD. It was attended by all, but one, Research Contract holders and Agreement holders as well as several observers from EU-FMD and FAO and Foot-and-Mouth (FMD) vaccine and diagnostic manufacturers and producers.

Discussions were focused on (1) the status of FMD in the participating counterpart's respective countries (eg. FMD free vs FMD free zone with or without vaccination vs FMD endemic) with respect to the risks and threats; (2) what are currently being done in terms of vaccine matching; (3) what criteria are being used to choose FMD vaccines and how they are being applied; (4) how are vaccine potency being determined and utilized; (5) how are post-vaccination monitoring and surveillance being performed; (6) the status of counterpart's vaccine laboratory quality assurance and FMD laboratory analysis and diagnoses (i.e. their analysis and/or diagnostic laboratory proficiencies and capacities both for routine testing and research, laboratory infrastructure and procedures). The workplans of all the Research Contract holders (RCH) and the Agreement holders (AH) were developed and discussed and all the agreement holders will supervise (based on their respective expertise) identified aspects of the workplans. The next RCM will take place in the second quarter of 2012 in Zambia.

Background:

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 outbreak 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 1980's, but these are not standardized for international use. Many countries now produce FMD vaccines but often without effective consideration of their effectiveness.

In many developing countries, vaccination will continue to be an essential component for the progressive control of FMD. Maximizing 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.

This CRP investigates vaccine matching procedures, vaccine potency testing methods and guidelines, and procedures by which an FMD vaccine's ability to induce the production of protective antibodies in cattle without the need for animal challenge experiments can be evaluated.

Conclusions:
All the counterparts developed their workplans such that, individually and or collectively, they work towards generating solutions set by the objectives of the FMD CRP.

It is important to:

- Establish methods and develop internationally agreed protocols for measuring the potency of FMD vaccines using *in vitro* methods.
- 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;
- 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
- Provide further global co-ordination of current research into FMD vaccines for use in endemic settings and to cooperate with other FMD institutions such as EU-FMD and PANAFTOSA
- To evaluate and standardize:
  - Virus neutralization (VN) tests
  - Early and rapid lateral flow and dip-site technologies and their application and use
  - Antigenic cartography (at IAH and OVI) in relation to virus neutralization tests (VN).

Research Coordination Meeting on the Use of Enzymes and Nuclear Technologies to Improve the Utilization of Fibrous Feeds and Reduce Green-house Gas Emission from Livestock

Technical Officer: Nicholas Odongo

The first RCM was held in Lethbridge, Alberta, Canada from 7 to 11 February 2011. It was attended by all 15 Contract (Research and Technical) and Research Agreement holders, one IAEA staff member (Mr Nicholas Odongo, the Scientific Secretary) and one observer. The meeting was formally opened by Dr Brian Freeze, Research Manager of the Lethbridge Research Centre followed by a presentation by the Scientific Secretary on the activities of the Animal Production and Health Section, the objectives of the CRP and the objectives of the RCM. The CRP was initiated in September 2010 with the award of eleven (11) research contracts, three (3) research agreements and one (1) technical contract.

The meeting commenced with the research contract holders presenting the work to be undertaken during the CRP – background information, constraints and issues and proposed mitigation strategies. The agreement holders presented their current research on the use of enzymes for ruminant production to set the scene for future work and the technical contract holder presented *in vitro* screening approaches for commercial enzymes. This was followed by a plenary session when each research contract holder's workplan was thoroughly discussed to harmonize and focus the workplans and future activities in the context of the objectives of the CRP and the agreement holders and technical contract holder presentations.

This was followed by practical sessions in the laboratory to demonstrate use of *in vitro* techniques to measure effects of enzymes on fibre digestion and methane production; characterization of enzyme products, including measurement of enzyme activities in exogenous products; measurement of enteric methane from cattle using the tracer gas technique; use of whole animal chambers for measurement of enteric methane from cattle and use of life cycle analysis to quantify the effects on methane mitigation strategies on whole farm greenhouse gas emissions.

Working with the agreement holders and/or the technical contract holder in small groups, each research contract holder revised and refined their individual workplans and formulated milestones. A clear step-by-step screening protocol to assay, characterize and formulate enzymes/enzyme preparations for ruminant diets was developed. Details of standardized workplans and protocols of work for the first 18 months of the CRP of each research contract was then presented in the plenary and re-evaluated, thoroughly discussed and finalized.

The 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 were then discussed and coordination plans formulated. Finally, conclusions from the meeting were drawn and recommendations made, discussed and adopted. The research

agreement holders and scientific secretary moderated and facilitated the discussions.

Conclusions and Recommendations

1. Use of fibrolytic enzymes in ruminant feeding is not common, its mode of action not well elucidated and available results across studies are highly variable.
2. A clear step-by-step screening protocol to assay, characterize and formulate enzymes/enzyme preparations for ruminant diets was developed. It was agreed that all those just starting to work with enzymes should adapt the standardized screening protocol for both the measurement of enzyme activity and for in vitro screening to ensure reproduceability of results.
3. It was recognized that the mode of application or delivery of enzymes especially in extensive grazing systems will be a challenge. Suitable modes of delivery will need to be investigated.
4. It was also recognized that availability and consistent quality of the enzymes might be a challenge.
5. These challenges notwithstanding, the participants endorsed the objectives of the CRP and there was consensus that the activities and expected outputs described in the project document were satisfactory.
6. Furthermore, although the potential benefits of using enzymes are expected to be moderate, the use of enzymes offer an important advancement in the nutrition of livestock in developing countries. Enzymes also offer a safe alternative to hazardous chemical treatments to improve the nutritive value of the low quality feeds used in most of the developing world.

The CRP will increase our understanding of the nutritive value of local feeds in the different countries, increase local technical capacity to use nuclear and related technologies to improve livestock productivity and increase south-south and south-north research collaboration and sharing of knowledge.

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 of a coordinated research project (CRP) on Genetic Variation on the Control of Resistance to Infectious Diseases in Small Ruminants for Improving Animal Productivity was held from 21 to 25 February 2011 at the Vienna International Centre, Vienna, Austria.

The aim of the meeting was to discuss current information related to infectious disease resistance, revise and update workplans of individual research contract holders, and to standardize workplans and protocols of work for the next 18 months, including criteria for selecting 'resistant' and 'susceptible' populations, phenotypic data collection and parasite monitoring. The meeting was attended by 13 research contract holders, four agreement holders, two IAEA and one FAO staff members, and four observers. Mr Mario Garcia acted as scientific secretary of the meeting. The meeting was opened by Mr Herman Unger, acting Section Head of the Animal Production and Health Section (APHS) emphasizing the relevance of gene mapping and gene sequencing in the identification of productive traits of economic importance. Mr Garcia presented the role of the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, the activities of the Animal Production and Health Section (APHS), and the objectives and main points of discussion of the RCM.

In the first two days of the meeting, each Research Contract holder presented a description of indigenous and adapted goat or sheep breeds in their countries, including the current knowledge on gastrointestinal parasite resistance, and details of the two main research trials that were designed for the first phase of the CRP. Agreement holders presented up-to-date information related to molecular markers, genome wide analysis, genomic selection and parasite resistance in breeding programmes. In addition, a full day was devoted for lectures by IAEA and FAO staff, agreement holders and two the invited observers on procedures for data collection, use of FAMACHA, DNA extraction and quality control, sample identification and storage, experimental design, and collaborative work. These presentations generated fruitful discussions that clarified steps and activities to be carried out during the first phase of the project.

Two major research trials will be conducted during the first phase of the project. The technical details, experimental designs, and sampling protocols were fully discussed and agreed during the meeting. A summary of the objectives and methodology of each trial is as follows:

Artificial Challenge: The main objective of this trial is to quantify the relative resistance to gastrointestinal parasites of two or more breeds using an artificial challenge protocol.
For this purpose, two sheep or goat breeds will be selected, one being known to be more resistant and one more susceptible to gastrointestinal (GI) nematode parasitism. A minimum of 20 animals per breed will be randomly selected from different flocks but within a common region or village, ear tagged, moved to an experimental site and housed in pens or drylots without access to parasite-contaminated feeds. Animals should be 4-6 months old at the time of challenge and can be of both sexes if they are equally distributed.
Animals will be dewormed and 4–6 weeks later, after confirming the absence of parasite eggs in faeces, will be individually weighed, a blood sample for DNA extraction collected, and a dose of 5,000 infective L3 of *Haemonchus contortus* larvae cultured administered to each animal. Sentinel animals (without L3 experimental infection) would be included to monitor the effectiveness of the system. Body weight, faecal egg counts (FEC), packed cell volume (PCV), and FAMACHA scores among other measurements will be taken at 28, 35, and 42 days after artificial infection. It should be ensured that animals must be fed to continue growing at rates typical of the species and location during the entire trial.
Field Trial: The main objective of this trial is to quantify the relative resistance to GI parasites in one or more breeds under field challenge and provide data suitable for genomic association studies.
Depending on the characteristics of each project, a minimum of 500 animals of one breed (the resistant breed) or a minimum of 200 animals of two breeds (one susceptible and one resistant) will be used in the study. Animals have to be ear tagged, grazed on common areas, and must be 4 to 6 months of age at the time of sampling; however, data can be accumulated across several farms and over two years. Initial sampling should be done at a time when parasite challenge is present. If deworming is needed, all animals must be allowed to graze together for at least 28 days without deworming before the start of sample collection.
Body weight, FEC, PCV, and FAMACHA scores will be taken twice, one week apart, and a blood sample for DNA extraction should be collected on the occasion of the first sampling.

Project counterparts will send DNA sample subsets to the Animal Production and Health Laboratory of the IAEA Seibersdorf Laboratories for quality control of DNA purity.

Conclusions and Recommendations

- The objectives of the meeting were accomplished.
- Individual workplans were revised, standardized, and updated to ensure proper implementation and fulfilment of the overall objectives of the CRP.
  - research contract holders (RCH) should follow the revised workplans and in case of the need of any possible deviations due to unexpected reasons should be discussed with agreement holders and communicated to the technical officer (TO) for advice and acceptance.
- Key elements of the experimental designs and protocols were highlighted, discussed, and clarified.
  - Detailed field and laboratory protocols should be provided to RCH.
  - All events and data collected should be fully registered to allow proper interpretation of results.
  - Methods and procedures for DNA extraction and purification should be documented and submitted to the TO.
- The repository bank for DNA material of the IAEA Seibersdorf Laboratories can store back-up DNA samples if needed, and also conduct quality control of DNA samples to improve genotyping output.
  - RCH should send a subset of DNA samples to the IAEA for quality control.
  - Laboratories with high risk of electricity failures should consider back-up DNA samples in alternative safer places.
- The Challenge Trial and at least 50% of the Field Trial have to be completed by the end of 2012 and data presented at the 2nd RCM.
  - RCH should frequently contact Agreement holders and TO on progress and constraints.
  - Take advantage of this experimental set up in parallel experiments.
  - Measure other traits for improving the quality of results and for facilitating data interpretation.
  - Selection of animals should consider age of the mother and single/triplets to avoid differences.

National Training Course on Serology; ELISA and Molecular Disease Diagnostics and Analysis of the Diagnostic DATA (ERI5006)

Technical Officer: Hermann Unger

This training course was held from 14 March to 1 April 2011 at the Central Laboratory, Asmara, Eritrea.
Ten local experts received training by Mr Hamadou, Cameroon and Mr Alhassan, Ghana.

Experts' Meeting to support efforts towards vaccine development and FMD control in Mongolia (MON5017/9001/01)

Technical Officer: Hermann Unger

This meeting was held from 18 to 21 April 2011 at the Vienna International Centre, Vienna.

The experts proposed that in order to support the effort to control FMD in Mongolia, campaigns should be planned to facilitate disease surveillance and sero-surveillance for FMD antibodies post-vaccination. It would also be of great value to screen the wildlife population using NSP testing to determine FMD prevalence in wild ruminants and possible implication in the dissemination of FMD. Appropriate assistance in establishing proper surveillance strategies will be given. However, effective control of FMD in Mongolia should be seen in the context of a regional approach.

Although the veterinary authorities have installed capacity to diagnose FMD with a variety of diagnostic procedures no FMD culture for strain typing has so far been attempted, due to space limitations in the BSL3 laboratory. In order to allow for a rapid vaccine strain selection in cases of outbreaks this capacity should be provided. Cooperation with regional FMD laboratories will allow the training of staff in appropriate techniques and the transfer of technology to enable the installation of this capacity in Mongolia. The provision of specific equipment and training needs will be addressed through IAEA TC projects. The official channels for the typing of FMDV isolates will be continued as before (regional, OIE and FAO reference laboratories).

In order to ensure rapid reporting of outbreaks in the field and to aid control efforts, appropriate technology should be established to allow for rapid disease diagnosis and reporting in the course of the TC projects. An internet-based system for exchange of information on the prevalent FMDV strains should be established. This should be addressed in the context of a regional project.

A local capacity to formulate different strains of FMD vaccine should be available in order to respond rapidly to outbreaks. Therefore, the existing infrastructure of BIOCOMBINAT should be inspected for determing its ability to address the specific needs for an oil-in-water vaccine formulation process. Bulk concentrated FMD vaccine should be purchased by national authorities with the prerequisite that the eventual supplier will have to train staff in the formulation process and support trial runs in Mongolia. The necessary freezer capacity for the bulk storage as well as the training and the adaptations to the formulation process shall be provided via a TC project.

The meeting came to the conclusion that, thus far, proof of principle for irradiated vaccines has only been established for gamma irradiated pathogens, but can be assumed to work also for electron beam (EB) radiation, based on available data. An irradiation procedure does not only cater for a rapid production process (important in an outbreak situation) but potentially allows for a lower dose of immunogen and for different application routes. electron beam irradiation therefore should be evaluated for its immunogenic capacities for viral diseases to allow for the optimum specification in the bidding process envisaged for the installation of a pilot plant in Mongolia. In order to allow a technically sound decision on the installation of an EB irradiation facility it will be required to compare cobalt-60 and EB-irradiated vaccines for influenza and alpha viruses for their sterility and immunogenicity in a mouse model and compare cobalt- and EB-irradiated vaccines for sterility and induction of immunity.

Upon completion of the trials a decision on the appropriate radiation technology to be installed (the Mongolian pilot facility) will be taken. The two options are: installation of EB equipment in the IVM (i.e. full installation) and installation of all aspects of the EB equipment without the EB machine (full installation without the EB machine – this will be done upon completion of the irradiation trials. At this point a decision will be made by the Mongolian counterparts to proceed with a cobalt gamma or EB irradiation source.

In order to determine the efficacy of an irradiated FMD vaccine a number of tests should be carried out. Firstly, to irradiate FMDV with Cobalt-60, assess the degree of inactivation and immunize mice (including controls vaccinated with conventional FMD vaccine) and evaluate the immune response to both preparations for specific antibodies. If the Co-60 irradiated vaccines show immune induction, repeat the vaccination with pigs, by the intra-dermal and intra-nasal routes and prove immune induction by testing for specific IgG responses and Th1 (IFN) induction; in addition evidence for any cross protection against other strains of FMD should be tested by VNT. Following successful completion of these experiments irradiation of FMDV should be attempted by EB. If these experiments are promising, the vaccine should be tested in ruminants with challenge after 4 weeks post vaccination (booster). In parallel, QC and QA parameters will have to be developed for defining the irradiation process and the respective test methods. This will include

testing of the vaccine efficacy under different dosage regimes.

National Training Course on Epidemiology of Transboundary Animal Diseases (ERI5006)

Technical Officer: Hermann Unger

This training course was held from 25 April to 13 May 2011 at the Central Laboratory, Asmara, Eritrea.
Thirteen participants received practical training on veterinary epidemiology including: repeating basic principles of veterinary epidemiology; designing of sampling frames with respect to the nature of a given disease; sampling strategies; surveillance and monitoring; analysis of diagnostic test results to determine the prevalence; calculations of QC data for ELISA results (ROC analysis); reporting of disease studies. Ms Amulen and Ms Nantima from Uganda provided the training.

Consultants Meeting on Upcoming Technologies for Early and Rapid Diagnosis of Infectious Diseases

Technical Officer: Ivancho Naletoski

This meeting was held from 18 to 20 May 2011 at the Vienna International Centre, Vienna.

Twelve speakers from research laboratories in USA, Germany, UK and Italy presented their research in the field of novel diagnostic technologies (multiplex diagnostic platforms for protein or DNA/RNA detection). Observers from FAO, OIE and the countries of the region were also present.

The main purpose of the meeting was to present the latest technologies for detection of transboundary animal diseases (including zoonoses), experience exchange and opinions on the current level of development, as well as to discuss the level of their validation and harmonization. Implementation of the novel technologies into the whole diagnostic process was discussed, such as sampling protocols, testing and reporting. Development or update of the information platforms for uptake, storage and data analysis was also discussed. The specific objective was to establish approaches on how to implement these technologies in Member States, in order to improve the animal health component of the food security chain.

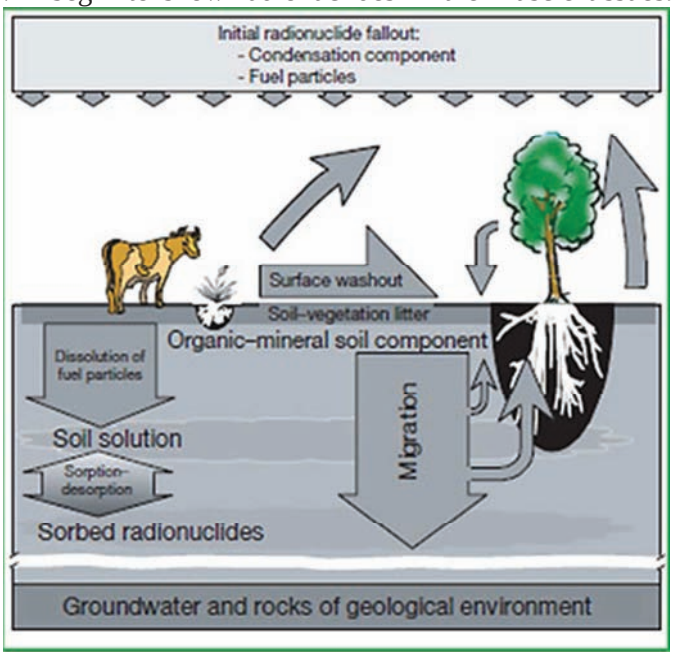
Participating Institutions were the University of California; Center for Grain and Animal Health Research, USDA/ARS-USA; Center for Infectious Immunity, Mailman School of Public Health, Columbia University-USA; representatives of three commercial companies and one independent consultant from USA, Friedrich-Loeffler Institute for Animal Health, Germany, Island Riems, Germany; Aminal Health and Veterinary Laboratories Agency, Weybridge, UK; and Instituto G. Caporale, Teramo, Italy.

Stories

Radioactive Contamination and Animal Production and Health

The recent nuclear crisis in Japan, which took place in the aftermath of a serious earthquake and tsunami, has focused the attention of the world on the problems that arise if there is a major release of radionuclides into the atmosphere. Of first concern is the human population that is exposed to contamination in the immediate vicinity of the nuclear power plant, and the need for rapid action to prevent their suffering ill effects. However, there are potentially longer term problems due to the ecological impact from contamination with radionuclides, on pastures and water courses that are used for rearing livestock. These problems are of concern to the animal health professional, firstly because of the possible ill effects to animals that have been raised on contaminated land and secondly because of the risk to humans from consumption of meat and milk products from livestock that have eaten contaminated feed.

Reports from the Government of Japan indicate that several radionuclides of consequence to human health have been found in the soil, vegetation and in animals, or their products. These include iodine-131 and caesium-137 that have both been found in the soil, in milk and in leaf vegetables such as spring onions and spinach. Some of the samples have been reported to be above the levels allowed by the Japanese food hygiene law for emergency monitoring criteria for intake of vegetables. Where these radionuclides have contaminated grazing land, milk from livestock is affected, so it is possible that any beef cattle will begin to show radionuclides in the muscle tissues.



No data are yet available on contamination in standing water such as lakes, reservoirs and fish ponds.
Investigations that were conducted in the aftermath of the Chernobyl accident have shown that radiation doses to livestock that became contaminated with radionuclides of iodine or caesium were significantly below the levels that cause clinical damage, and it was unlikely that there would be any detrimental effect on their health. This is because the risk to their welfare from genetic defects or cancer caused by the accumulative somatic damage from radiation over a long period was unlikely to occur, given their short life span. Nonetheless, products from the animals still present a risk to humans, and in the interest of food safety, monitoring for radioactive contamination is essential. For instance, in the UK, following the Chernobyl accident in 1986, restrictions were placed on the marketing of sheep on 10,000 farms owing to contamination with caesium-137. Twenty years later those restrictions were still in place on 400 farms and 220,000 sheep were being monitored. In Scotland, these measures were finally relaxed on the last farm still undergoing monitoring in June 2010. Similar restrictions were put in place in other countries affected.

What are Radionuclides and why are they Hazardous to Animal and Human Health?

Radionuclides are radioactive atoms either man-made or naturally occurring. All elements have radioactive forms so there are many radionuclides. At Chernobyl, some 60 radionuclides were emitted from the reactor, but only a few were considered to present serious health hazards to humans and animals, namely iodine-131, caesium-137, caesium-134, strontium-89, strontium-90 and plutonium-239. iodine-131 is a volatile radionuclide that emits beta and gamma rays and combines easily with organic materials and soil minerals. Water, grass, vegetables and animal fodder become contaminated. The half-life is 8 days, but in the thyroid it can last for 100 days potentially causing malignant tumors. Caesium-137 spreads readily in the environment in soil, water and in the air. It can be ingested or inhaled and locates in muscle tissue, bones and fat. It has a half-life of 30 years and is extremely toxic. Strontium-89 (half-life 50 days) and Strontium-90 (half-life approximately 30 years) occur in the soil, food and water. These elements can be deposited in bones and remain in the body for long periods. Plutonium-239 is an alpha ray emitter with an extremely long half-life. It can enter the body by ingestion or inhalation, and although some is eliminated it remains in bones and the liver for years, where it can cause tumour development.

Environmental Impacts of Radionuclide Contamination

The Chernobyl nuclear accident caused ecological problems in the Ukraine and Belarus as well as further afield in Europe, in the higher northern latitudes. Contamination over this long distance was primarily by caesium and iodine. The transfer of radionuclides in the environment depends on the particular ecosystem, thus for the most important element, caesium, transfer is higher in the natural environment than in agricultural ecosystems. This is due to the physicochemical behaviour of the soils; in natural systems where there is a lack of nutrients there is no competition between caesium and potassium, leading to higher transfer rates of caesium. Transfer is low where there has been intensive agriculture and the soils have a high nutrient status and a high proportion of clay materials. In forest there is a multilayered structure consisting of a mineral layer low in clay and a layer rich in organic matter. Radionuclides migrate down through the soil so that eventually they are no longer present in the root-containing zone. Migration is lower in peaty soils than in highly organic soils. In forests, there is initially a filtering of contaminants by the tree canopy then, following leaf/needle fall and rain run-off, the soil on forest floor becomes the main repository for contamination with radionuclides. However, trees and plants continue to become contaminated through root uptake. Radiocaesium can be recycled in trees through uptake and regular leaf/needle fall, and stored in the long-term in the trunks of the tree. Fruits and fungi present in the forest become contaminated, with very high levels of caesium-137 being found in mushrooms. Surface water systems are also directly affected by nuclear fallout but persistence of radionuclides in catchment soils and river and lake sediments is important in determining their distribution. In rivers, due to the constant flow of water there is less contamination in the longer term, since bottom sediments tend to be replaced, particularly in flood conditions. In contrast, in lakes where there is little or no water exchange, contamination in bottom sediments can be high.

In the immediate vicinity of Chernobyl, the 23.000 km^2 of land heavily contaminated and this area, known as the exclusion zone, was subject to restriction of agricultural activities. Animals living in this area were exposed to high levels of radionuclides via food, water and air and levels in some were many hundreds of times higher than in unaffected populations - many animals that remained in this zone died from radiation induced illnesses. Today, mammals, birds, fish and amphibians show morphological deformities, and genetic disorders. The greatest impact from radioactive contamination was in Ukraine, Belarus and Russia, but radionuclides from Chernobyl were carried in the atmosphere into other countries in Europe and Scandinavia including Austria, Bulgaria, Croatia, Czech Republic, Finland, Germany, Greece, Hungary, Italy, Moldova, Norway, Poland, Romania, Slovenia, Sweden, Switzerland and the United Kingdom. Other affected territories were in Asia (including Turkey, Georgia, Armenia, United Arab Emirates and China), northern Africa, and North America. The major issue in many of these countries was contamination of food products (milk and meat) from domesticated livestock that had ingested radionuclides that were then introduced into the human food chain. The most common source of this contamination came from ruminants, including wild animals that grazed in natural or semi-natural ecosystems that were minimally managed by man. These were in areas such as mountain pastures, marshlands and tundra.

Transfer of Radionuclides to Livestock Grazing on Contaminated Pastures

Animals grazing in natural, semi-natural, or forest habitats are more prone to acquire high levels of radionuclides than those on agricultural land. In Austria, the alpine regions were among the most heavily contaminated areas. Cattle are moved to mountain pastures in the summer and within 10 days their levels of radiocaesium began to rise. Levels reached two orders of magnitude greater than when they were grazed on agricultural land. Sheep in Wales, England and Scotland are often reared on unimproved land, and levels of radiocaesium reached 4000 Bq/kg in the immediate aftermath of Chernobyl. In forest regions, the species involved are likely to be wild ruminants such as deer or wild boar. Caesium-137 levels are high in these animals because they ingest fungi that are known to take up large quantities of the radionuclide. In contrast, wild ruminants grazing on agricultural land have lower levels of radiocaesium. In Croatia in 2002, radiocaesium levels were high, but not sufficient to cause concern to human health, except in cases where there might be high consumption such as hunters. In southern Germany, caesium-137 levels in meat often exceed several thousand Bq/kg; again ingestion of fungi is the reason for this. Since radiocaesium remains in the top 10-15 cm of soil in these forest zones where fungal hyphae grow, it is likely that the wild boar meat will continue to be contaminated for the foreseeable future. There is a financial penalty for this, as the German government compensates hunters who cannot eat meat that is too highly contamined. Fish in enclosed lakes or ponds often have high levels of radiocaesium; fish in Finnish lakes had levels varying from 16-6400 Bq/kg in 2003.

Sampling for Measurement of Radioactivity

Various procedures can be used for monitoring levels of radionuclides in milk and meat, and although there are no standardized methods it is essential that surveys take into account all aspects of contamination in relation to the environment, likely exposure, the animal species and their foraging habits. The following list indicates some of the procedures used to monitor animals exposed to radiation.

Live animals - cattle, sheep, goat, poultry

- Live sampling with a hand held, battery operated monitor, e.g. Canberra 10, calibrated for use with live sheep (as used in UK, Norway etc.). For cattle - place monitor on hindquarters for one minute. Small birds can be monitored by whole body measurement.

Meat:

- Meat - slaughter animals - samples 1.5×1.5×1.5 cm taken from muscle of two legs. Measurements of muscle samples from different parts of the same animal does not differ by more than 10%
- Fresh meat samples from cattle of 20-100g
- Fresh muscle samples of 20-25g from wild deer
- Samples of 250-500g lean caribou meat collected and air dried in the sun, or by infra-red before analysis
- Muscle sample from wild boar of at least 500g; samples frozen after collection

Milk:

- Milk - bulk milk samples collected daily to average out physiological differences in the dietary habits of individual cows
- In affected and sensitive areas - daily monitoring of milk for Sr-90/Cs-137/I-131
- In areas at risk but not contaminated - sample as often as possible, but not less than 14 day intervals

Figure 2: Disposal of contaminated milk.

Mitigating the Effects of Radionuclide Contamination

Contamination can be mitigated by taking measures to transfer radioactive pollutants. It is important to reduce exposure wherever possible, especially in the immediate aftermath of contamination, i.e. by bringing livestock in from pasture and confining them to pens to prevent their grazing on contaminated pasture. Animals should be fed with uncontaminated feed as soon as possible. Changing land use is effective in reducing transfer to man. A switch from milk production to beef or pigs can reduce radionuclide transfer by 5-fold. To reduce radiocaesium in milk cattle can be supplied with a caesium-binding compound such as ammonium ferric cyanoferrate (or AFCF, 'Prussian Blue') as a bolus into the rumen, in compounded concentrate feed, in salt licks, or simply sprinkled on the diet. AFCF reacts with consumed radiocaesium in the intestine to form a complex that is eliminated in the faeces. In the case of meat-producing animals, moving to uncontaminated pastures and feeding uncontaminated feed may only be necessary close to the time of slaughter since the biological half-life of radiocaesium, for example, is of the order of two to four weeks, depending on the species. In the case of wild boar meat, brining in sodium chloride and potassium nitrate can reduce caesium-137 levels by >70%.

Figure 3: Cattle are kept under cover (i.e. in a barn) to protect them from radiation fallout. They are provided with uncontaminated fodder and water.

The most salutary lesson learned in the past 25 years has been the need for the regulatory authorities in countries affected by contamination to take a much broader view of the environmental consequences and adopt a more holistic approach in addressing the situation. Thus, the international scientific community has a more fundamental understanding and greater insight into the way in which different ecosystems are affected by nuclear contamination, which will provide the basis for predicting the risk to, and likely impact on, agriculture in the Fukushima incident.

Development of Feeding Strategies for Improved Meat and Milk Production on Smallholder Dairy Farms in Zambia

Introduction

Livestock rearing is one of the leading farming activities practiced by rural communities in Zambia. The animals kept include cattle, goats, sheep, pigs and various species of poultry of which chickens are the most common. Rearing of animals in rural areas is mostly done on a subsistence level where the emphasis is to produce for own household consumption with very little left for sale to generate income. However, in recent years, there has been an increased effort by the Zambian government to

encourage farmers to improve animal productivity as a way of creating employment and income generation.

These efforts have, however, been limited by the inherent low productivity of the animals kept on traditional small-scale farms which has been attributed to the extensive management system practiced in rural areas whereby animals are left to scavenge on their own in search of feed and water. Additionally, the low productivity has been attributed to inferior quality of the local breeds that is characterized with long calving intervals, poor reproductive efficiency and slow growth rates. Because of poor feeding practices and lack of veterinary services, animals on traditional small-scale farms are also susceptible to a wide range of diseases that exacerbates the problem resulting in increased mortality rates. Small-scale farmers also lack knowledge and technical skills for better management of animals to improve productivity. To address these issues, a technical cooperation project was designed with financial assistance from the International Atomic Energy Agency (IAEA) to develop dairy management strategies for improved production of meat and milk in Palabana and Njolwe dairy tenant schemes based on increased use of locally available resources. The resources under consideration included use of indigenous livestock breeds and locally adapted feed crops that have potential as energy and protein supplements for milking animals.

Livestock Production - Constraints and Opportunities

The study started with a baseline survey to highlight constraints limiting increased production of meat and milk in the project area. This was followed by documentation of existing opportunities for increased production and marketing of meat and milk. Among the major constraints identified to be limiting animal productivity was lack of proper breeds to foster increased meat and milk production (Figure 1).

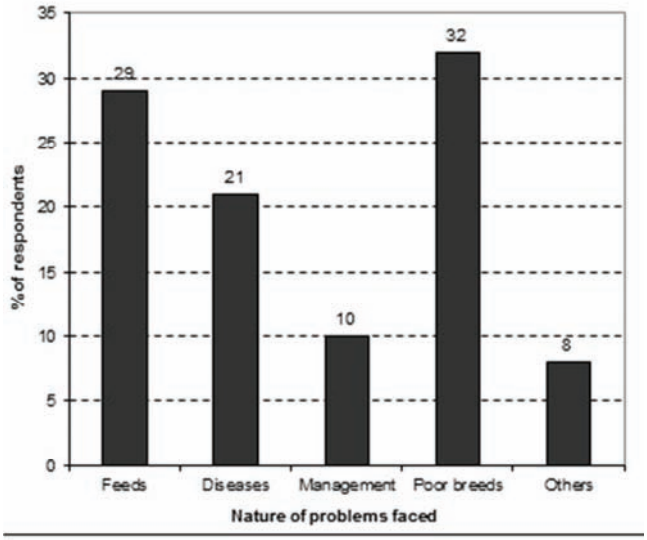


Figure 1. Major production constraints documented to be limiting increased milk and meat production in the proposed study area.

Most of the farmers in the project area were found to be using crosses between the Holstein Friesian dairy breed with indigenous cattle that had no capacity for increased milk production. In addition to inferior milking animals, there was also limited supply of quality feeds for animals to warrant increased meat and milk production. Despite local farmers having undergone basic training in Pasture and Fodder production and in animal feeding techniques, there were simply no efforts on their part to produce feed for their animals. Most of them simply depended on natural grazing and use of fibrous crop residues. Existing opportunities for increased production and marketing of milk in the project area included the farmers themselves having been organized into a dairy cooperative that had acquired facilities for milk cooling, storage and delivery to processing companies. There were also a number of Government and Non-Government Organizations (NGOs) working in the area to offer extension services and facilitate supply of dairying inputs including veterinary and artificial insemination (AI) services.

Production and Nutritional Evaluation of Locally Adapted Feed Crops

In response to the dairy production constraints and opportunities highlighted in the baseline survey, the first task for the project was to improve the quality and availability of feeds for milking animals in the area. Due to limited availability of farming inputs and the local farmers' inability to produce pasture and fodder crops, it was felt that animal feeding strategies should be based on the use of locally adapted crops including natural grasses, tree forages, fodder and grain legumes. This saw establishment of a nursery of pasture and fodder crops at the University of Zambia School of Agricultural Sciences field station (Table 1).

Figure 2. Some of the contact farmers were able to produce enough pasture and fodder crops (Velvet beans) to warrant start of on-farm feeding experiments (photo: Dr.J. Simbaya).

The purpose of the nursery was to grow materials for seed production and produce materials for feed nutritional evaluations. Based on productivity and easy of establishment, a number of feed crops were selected to be established on-farm during the following season as a way of introducing farmers to pasture and fodder production on their farms. Even though the response was not as expected with some farmers, a number of them grew

adequate amounts to warrant initiation of on-farm feeding experiments (Figure 2). However, before commencement of on-farm pasture production and feeding trials, part of the fodder crops produced on-station was harvested and subjected to chemical composition nutritional evaluations in the laboratory (Table 2).

Table 1. List of pasture and fodder crops established at the university nursery for seed production and nutritional evaluations.

Grasses	Fodder legumes	Grain legumes	Fodder trees
Rhodes grass – Boma	Lablab beans	Velvet Beans	Mulberry trees
Rhodes grass – Callide	Red sunhemp	Jack beans	Leaucaena leucocephala
Rhodes grass Mbabara	Black sunhemp	Green gram	Cassava leaves
Buffel grass – Molopo	Archer dolichos	Pigeon peas	Moringa olifera
Panicum maximum-green panic	Stylo graham	Cow peas	Sesbania sesban
Napier grass	Silverleaf desmodium		
Bana grass	Siratro		
Nile grass			
Topedo grass			
Green gold			

Table 2. Chemical composition of locally adapted pasture and fodder crops established at the University nursery.

Feedstuff	Dry Matter	Protein	Fat	Ash	calcium	Seed size
Velvet beans	93.6	23.5	10.6	3.57	0.34	0.83
Cow peas	95.2	28.1	9.5	4.03	0.30	0.12
Green gram	96.9	28.6	10.3	4.20	0.36	0.03
Jack beans	94.6	24.0	12.0	3.64	0.31	0.94
Pigeon peas	96.4	22.2	9.1	3.91	0.43	0.14

On-station Feed Supplementation Trial

The chemical composition analyses were followed by on-station feeding trial to evaluate the use of velvet beans (*Mucuna purensis*) as a protein supplement for milking animals on smallholder dairy farms. The feeding trial was aimed at evaluating Velvet beans processing methods to maximize their utilization by dairy animals. The study was in response to a number of farmers who had been introduced to growing velvet beans by local NGOs but had no idea how to use them as a feed resource. Most farmers deemed the grain poisonous and were only grazing the crop to animals in situ. The processing methods evaluated included grinding of the grain, the pods and pods together with the vines. The prepared velvet beans materials were used to formulate supplemental rations for milking animals as a replacement to purchased dairy concentrates. Some of the results of on-station feeding trial at the University of Zambia showed that animals fed velvet beans based diets were capable of maintaining milk production throughout the study period. This was despite the fact the study was done during the months of September and October when there is a critical shortage of feed and water and the milk production levels are expected to be at their lowest. The on-station feeding trial was repeated on-farm with selected contact farmers. The results on feed intake and milk yield showed that the animals had no problem in accepting the velvet beans based diets and maintained milk production levels. This was despite some earlier cases of diarrhoea at the beginning of the feeding trial. The on-farm trials also showed positive response as the animals fed experimental diets had increased milk production levels throughout the study period.

Evaluation of Herbaceous Legumes and Protein Supplements –still on-going

The next stage of study has concentrated on the evaluation of fodder and grain legumes as protein supplements for milking dairy animals. The protein sources are divided into three groups consisting of grain legumes (velvet beans, jack beans, pigeon peas and mindolo beans), herbaceous legumes (velvet beans and lablab beans) and tree fodder forages (Moringa, mulberry leaves, Leucaena leucocephala). This study is still on-going starting with proximate analysis to determine nutrient composition in selected locally adapted pasture and fodder crops (Table 3). The next stage will be to determine in sacco nutrient degradation in fistulated dairy steers using the nylon bag technique. Nutrient degradation evaluations will be followed by on-station feeding trials *to determine acceptability and effect on milk production by milking animals on smallholder dairy farms*. The on-station feeding trials will be followed by on-farm trials that will evaluate rations selected from the on-station trials. These will then be taken to stakeholders for eventual training of farmers and publication of results to stakeholders.

Table 3. Chemical composition of selected fodder legumes/trees for evaluation as potential of being used as protein supplements for milking dairy animals.

Forage	Dry matter	Protein	Ether extract	Ash	calcium
Kapeta nsufu	92.7	24.8	12.1	3.63	0.75
Paulina Foliata	89.7	18.9	-	2.63	0.63
Sesbania sesban	91.8	21.1	9.6	3.54	1.44
Lablab beans	91.7	24.5	9.4	3.51	1.31
Stylo	91.6	22.7	12.9	3.38	1.45
Pigeon peas	92.7	24.1	12.5	3.77	1.02
Siratro	91.1	18.1	-	3.36	0.66

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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 involvment 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, monitor, contain and control TADs. The CRP is supporting the buildup 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 Pleuro-Pneumonia (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 under way. 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 devel-

oped 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 was held 7-11 February 2011 in Lethbridge, Alberta, Canada and the report can be found under Past Events in 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 outbreak 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 they have 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 was held in January 2011 in Rome, Italy and the report appears under Past Events of this newsletter.

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 numbers 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 to identify molecular markers of economic interest which will open possibilities in the future to select and breed animals for enhanced resistance to diseases. The CRP also aims to develop capacity in developing countries in the use of molecular and related technologies and create opportunities for international research collaboration.

The utilization of modern molecular and breeding technologies in animal genetics is an important task. With next-generation sequencing technique, availability of more than 400K SNP (single nucleotide polymorphisms) array at reasonable cost, maturity of methods such as microarray and expression QTL, GWAS (genome wide association study) and knowledge of existing copy of number of variation of genome (CV), one should be able to use the state-of-art and take advantage of such an opportunity to help Member States better and more efficient.

An objective for several current and planned TC and CRPs in the Animal Production and Health sub-Programme is to transfer the technical capacity for DNA analysis and marker assisted selection to Member States. In line with this objective, the APH laboratory in Seibersdorf has developed different assays to identify host candidate genes for parasite resistance in small ruminants and detect them. Comparative mapping was used to fine-map a chromosomal region and improve the ability to identify genes responsible for parasite resistance. A total of 809 genes were selected based on the cattle BTA5 region sharing synteny with markers on the OAR3 QTL region associated with parasite resistance on previous scans. In addition, 100 genes involved in immune response pathways were added to study. Arrangements were made to acquire samples of blood and DNA from representative breeds from different populations and each SNP verified in different populations.

Data analysis from a total of 909 genes allowed identification and selection of 100 SNPs on different genes. As a follow-up strategy to the laboratory's development of DNA-based markers, a pilot study was initiated to integrate KASP genotyping system in its activities. The patented KASP SNP genotyping system is a homogeneous fluorescent resonance energy transfer (FRET) based system which allows for the detection of SNP's without the need for a separation step. Coupled with the power of competitive allele specific PCR, the KASP system offers a good system for determination of SNP or insertion/deletion genotypes in the laboratory. It offers the simplest and most cost-effective way to determine SNP genotypes which will allow the laboratory to run assay for up to 5000 samples for each of selected SNP after completion of the pilot study.

The first research coordination meeting was held in Vienna, Austria, from 21 to 25 February 2011 and the report appears under Past Events in this newsletter.

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 Svetlana Piedra Cordero (s.piedra-cordero@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 these characteristics at the genomic level lags behind that known in 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.

Organization of a Proficiency Ring Test for the Diagnosis of Peste des Petits Ruminants (PPR) by Classical RT-PCR (CRP D32026: The Early and Sensitive Diagnosis and Control of Peste des Petits Ruminants)

In 2007, the IAEA launched a Coordinated Research Project (CRP) on diagnosis of Peste des Petits Ruminants (PPR). Entitled Early and Sensitive Diagnosis and Control of Peste des Petits Ruminants**,** this CRP has the overall aim of developing, validating and transferring to Member States sensitive, specific and rapid tests for the diagnosis of PPR to help them better manage and control this transboundary animal disease (TAD) which is threatening small ruminant production in many developing countries. Indeed, in Asia and Africa, PPR is the main killer of sheep and goats and the major threat to the livelihood of poor farmers because these animals are the livestock species they rely on. This is the reason why an animal disease consultancy which was carried out in the early 2000s singled out PPR as one of the most important animal diseases to be taken into consideration when determining poverty alleviation policies. Because of the high negative economic impact in countries affected by PPR, this disease is one of the priorities of the FAO Emergency Preventive System (EMPRES) programme. At the recent global rinderpest eradication (GREP) meeting which was held in Rome from13 to 14 October 2010, and taking into consideration the risk that PPR may pose in a 'rinderpest-free world', experts requested FAO to develop strategies in collaboration with other partner institutions for the progressive control of PPR, or even its eradication as has been achieved for rinderpest. The African Union is now seriously considering working on the possibility of embarking on an African continental control programme for PPR, a disease which is expanding in both the North and South of the continent. This strategy would require the availability of validated, highly sensitive and specific diagnostic tests for rapid identification of the disease.

One specific objective of the current IAEA CRP on PPR is to evaluate and validate the classical reverse transcriptase-PCR (RT-PCR) methods currently in use for the diagnosis of this disease. Among the 13 contract holders, 12 are currently using the classical RT-PCR assay for PPR diagnosis, although they are not all using the same techniques. Indeed, there are several different methodologies for RNA extraction and different sets of primers targeting different regions of the PPRV genome, etc. For consistency of approach it is important that these methodologies are standardized, compared, validated and harmonized. In 2010, the Animal Production and Health Laboratory (APHL), Seibersdorf organized an inter laboratories proficiency test for the application of the classical RT-PCR to PPR diagnosis, the results of which were obtained and analysed in early 2011.

The panel of samples was composed of pathological specimens from field samples of PPRV-infected and non-infected animals in a lysis buffer (to inactivate pathogens), and positive control sample of PPRV RNA. This panel was designed in order to provide additional and effective quality control for the classical PCR. The positive control RNA was the target of the set of primers NP3/NP4 on the PPRV N gene but with an internal deletion (*Couacy-Hymann, E., Roger, F., Hurard, C., Guillou, J.P., Libeau, G. and Diallo A. Rapid and sensitive detection of peste des petits ruminants virus by a polymerase chain reaction assay. J. Virol. Methods, 2002,* ***100****, 17-25*). It was used to spike both negative and positive samples. However, all of the infected pathological specimens included in the panel will be tested with other sets of primers described in the literature for PPR diagnosis using RT-PCR and qRT-PCR assays.

Although the panel was sent to all the 13 Contract holders, only 9 of them have participated effectively in the ring test by sending back their data. Their findings were analysed and the results are summarized the table. They were then transmitted to the participants, each of them recognizing only their own results.

Three out of the 9 participants obtained the expected results and one of them even specified the presence of a deleted amplicon. One counterpart incorrectly identified all the samples as being negative for PPRV. Each of the participants was contacted and helped to understand how the test was implemented and given guidance on how they might solve the problems that were encountered. A second ring test will be organized in 2011.

Lab. Nr.	Shipped samples			Pos tested Pos	Pos tested Neg	Neg tested Neg	Neg tested Pos	Correct (Absolute)	Correct (%)
		Pos	Neg						
1	13	12	1	8	4	0	1	8	**62**
2	13	12	1	6	6	1	0	7	**54**
3	13	12	1						
4	13	12	1	9	3	1	0	10	**77**
5	13	12	1	**12**	**0**	**1**	**0**	**13**	**100**
6	13	12	1	**12**	**0**	**1**	**0**	**13**	**100**
7	13	12	1	**12**	**0**	**1**	**0**	**13**	**100**
8	13	12	1	6	6	0	1	6	**46**
9	13	12	1						
10	13	12	1	0	12	1	0	1	**8**
11	13	12	1	6	6	1	0	7	**54**

Tools for genotyping capripoxvirus.

Efficient control of transboundary animal diseases relies primarily on early detection to allow an adequate response to disease outbreaks. More specifically, it allows a rapid implementation of control measures, such as the restriction of animal movements or the vaccination of herds that are considered to be at risk, to confine the disease in a limited area within a country or a region. For diseases such as capripox disease, where different group of strains, sheep pox virus (SPPV), goat pox (GTPV) or lumpy skin disease virus (LSDV) can cross infect different animal species (sheep, goats or cattle). The accurate identification of the genotype of the strain involved in an outbreak is crucial for the selection of the best vaccine strain to be used, and unequivocal classification of the viral strain is essential, especially when wildlife are involved. Usually, genotype determination is achieved by analysis of the restriction fragments or the gene sequencing followed by analysis of the data. These approaches require expertise and equipment that are not available in most of the laboratories of developing Member States (MS) of the IAEA and FAO Simple tools such as those based on PCR-based genotyping without the need of gene sequencing might be more accessible technologies for MS, especially since they can be used for screening a large number of samples. In the past 5 years, the APHL has been developing such tools for rapid, specific and highly sensitive detection of several pathogens responsible for transboundary animal diseases. An example is the developing of nucleic-acid amplification-based assay for genotyping capripoxviruses, a research activity that was part of a project supported by the French Ministry for Foreign Affairs to strengthen veterinary diagnostic laboratories in Africa.

Within the framework of this project, APHL and collaborators have identified two genes for capripox virus (CaPV) strain discrimination. The sequencing of these genes and subsequent phylogenetic analysis makes it possible to place the strains to one of the 3 groups within the CaPV genus: SPPV, LSDV or GTPV. Furthermore, two species-specific signature markers that were identified in this work were exploited to develop simple methods for CaPV genotyping without the need for gene sequencing.

The first relies on a 21-nucleotide deletion that was found in the RPO30 gene of only SPPV group members. This deletion was exploited to develop a classical PCR method to discriminate between SPPV and GTPV, in small ruminants with capripox disease (see Figure 1). The

analytical performances of this method are described in Lamien et al., (*Vet. Microbiol., 2011, **149**, 30–9*).

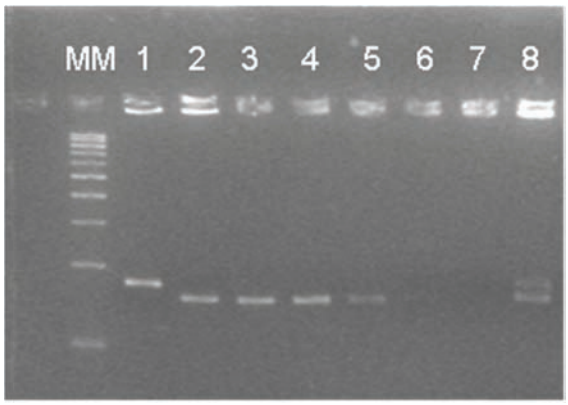


Figure 1Figure 1. Simultaneous detection and genotyping of Algerian Capripoxviruses by classical PCR: MM = Marker, 1 = GTPV control, 2 = SPPV_Djelfa, 3 = SPPV Illizi, 4 = lot 07/09, 5 = lot 10/09, 6 = W, 7 = Cell control, 8 = SPPV + GTPV

A second genotyping tool, based on real time PCR with hybridization probe was published in 2011 *(J. Virol. Methods, 2011, **171**, 134–40*). This method exploits species-specific signatures, found in CaPVs GPCR gene, which allows discriminating between the 3 member species of CaPVs, SPPV, GTPV and LSDV. These two methods were proven to be usable for direct genotyping using clinical specimens and were used recently to determine the genotype of field CaPV isolates of Ethiopia and Algeria (see example for real time PCR in Figure 2). APHL is promoting the transfer of these technologies to selected MS through field supportive missions and Fellowship training courses. It is expected that the effective usage of these genotyping methods will contribute significantly in enhancing the control of CaPVs in MS through a better selection of vaccine strain and monitoring of capripox disease in wildlife.

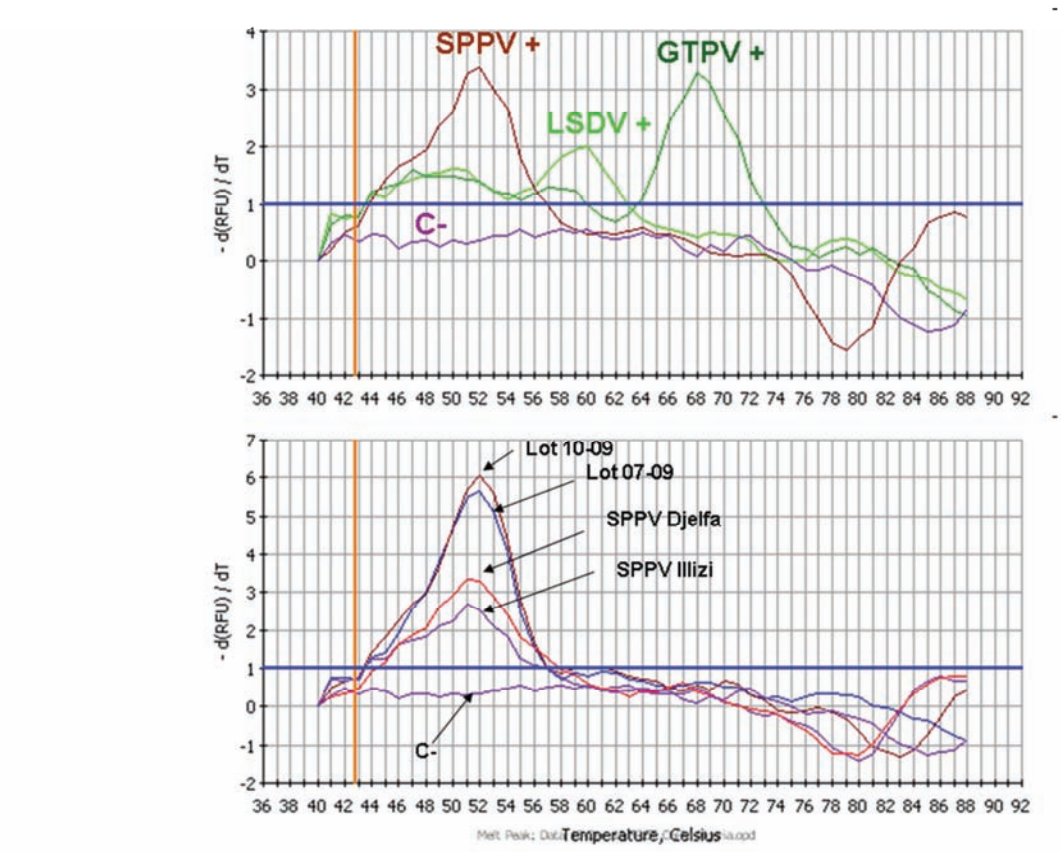


Figure 2. Determination of Algerian capripoxvirus genotypes by real time PCR: SPPV + = sheep poxvirus positive conrol; LSDV + = lumpy skin disease positive control; GTPV + = goat poxvirus positive control; C- = negative control.

Fellows

Mr Kebadire Tlotleng, from Botswana, with a fellowship provided by the IAEA TC department, TC project BOT/11001, spent 3 months (March-May, 2011) in APHL for training on peste des petits ruminants diagnosis including cell culture and virus isolation, classical RT-PCR and qRT-PCR.

Consultant

Esayas Gelaye Leykun, a scientist from the National Veterinary Institute (NVI), Debre Zeit, Ethiopia, joined APHL in May for one year in connection with the IAEA PUI project with the support of the US government.

Technical Cooperation Projects

TC Project	Description	Technical Officer(s)
BEN/5/006	Improving Animal Health and Productivity **Objective**: To strengthen, diagnose, and control African swine fever, and increase animal productivity.	Unger / Diallo
BKF/5/008	Strengthening the Development of Small Ruminant Production **Objective**: 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 **Objective**: 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 / Naletoski
BOT/5/005	Improving Diagnosis of Animal Diseases **Objective**: 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 **Objective**: 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.	Naletoski / Viljoen
BZE/5/004	Strengthening the Veterinary Diagnostic Laboratory with Capacities in Polymerase Chain Reaction Diagnosis (Not funded) **Objective**: 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 **Objective**: To improve animal production in the Central African Republic through livestock disease control and improved breeding by use of artificial insemination.	Naletoski / Garcia Podesta
CMR/5/017	Improving Animal Productivity and Health **Objective**: To strengthen capacity and outreach regarding artificial insemination in ruminants, and to control livestock diseases impeding reproduction and productivity.	Unger / Garcia Podesta
ERI/5/006	Controlling Major Epizootic Diseases and Other Mycoplasma Infections of Livestock **Objective**: To improve the control of transboundary animal diseases, and continue the eradication of tuberculosis and brucellosis.	Unger / Naletoski
ETH/5/012	Integrating Sterile Insect Techniques for Tsetse Eradication **Objective**: 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

TC Project	Description	Technical Officer(s)
HON/5/004	Improving the Nutrition and Health Conditions of Livestock in Order to Increase Productivity and Reproductivity (Phase II) **Objective**: 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) **Objective:** To strengthen and improve livestock production in Honduras.	Garcia Podesta / Odongo / Viljoen
IVC/5/030	Assessing the Genetic Profile for Improved Livestock Production **Objective**: 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 **Objective**: 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 **Objective**: 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 **Objective:** To increase animal production through the improvement of animal health and control reproduction in the Amoron'i Mania region.	Garcia Podesta / Odongo / Naletoski
MAU/5/003	Improving the National Capacity in Diagnostics for Animal Diseases (Infection and Parasitic Diseases) **Objective**: 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.	Unger / Naletoski
MLI/5/023	Improving National Capabilities for Characterization of Serotypes of Major Animal Diseases Using Molecular Biology Techniques **Objective**: 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.	Unger / Naletoski / Viljoen
MON/5/017	Supporting the Sustainable Production and Supply of Vaccines and Diagnostic Kits for Transboundary Animal Diseases **Objective**: To produce vaccines and diagnostic kits for transboundary animal diseases.	Viljoen / Luckins
MOR/5/030	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) **Objective**: Increase sheep and goats for consumption and producers' revenue while preserving natural resources.	Garcia Podesta / Malek
MOZ/5/002	Promoting sustainable Animal Health, Reproduction and Productivity Through the Use of Nuclear and Related Techniques **Objective**: To obtain sustainable improvement in animal reproduction and breeding and animal health through the use of nuclear and nuclear related technologies.	Viljoen

TC Project	Description	Technical Officer(s)
MYA/5/018	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 **Objective**: 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.	Garcia Podesta / Diallo / Unger
NER/5/013	An Integrated Approach for Improvement of Livestock Productivity **Objective**: To increase the productivity of livestock through implementation of an integrated programme dealing with nutrition and reproduction.	Odongo / Garcia Podesta / Diallo
PER/5/029	Genomics of the Alpaca: Identification of Expressed Genes and Genetic Markers Associated with Productivity and Embryonic Mortality **Objective**: To identify and characterize the factors associated with embryonic mortality in alpacas.	Garcia Podesta / Malek
RAF/5/057	Strengthening Capacities for the Diagnosis and Control of Transboundary Animal Diseases in Africa (AFRA) **Objective**: 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.	Unger / Diallo
RER/5/015	Supporting Early Warning and Surveillance of Avian Influenza Infection in Wild and Domestic Birds and Assessing Genetic Markers for Bird Resistance **Objective**: 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.	Naletoski / Diallo
RLA/5/049	Integrated Control of Fascioliasis in Latin America (in support of National Programmes	Viljoen / Naletoski
SIL/5/011	Controlling Economically Important Livestock Diseases **Objective**: To design epidemiological surveys and adopt appropriate rapid laboratory techniques for the diagnosis of PPR and NCD in small ruminants and local chickens.	Unger / Naletoski
SRL/5/041	Maximizing Productivity on Goat Farms through Cost-Cutting and DNA-Based Technology in Selection for Breeding **Objective**: To improve the productivity of goats of smallholder 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.	Garcia Podesta / Odongo / Viljoen / Malek
SRL/5/042	Applying Molecular Diagnostics to Zoonotic Diseases **Objective**: To enhance the long-term epidemic preparedness by developing competence in molecular diagnosis and surveillance of zoonotic infections.	Kashyap (NAHU) / Unger
UGA/5/030	Improving the Diagnostic Capacity in Animal Diseases (Phase II) **Objective**: 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.	Unger / Luckins / Naletoski
URU/5/026	Increasing the Profitability of Dairy Producers by Improving Reproduction Efficiency, Rational Sustainable Use of Genetic Resources **Objective**: 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.	Garcia Podesta / Odongo

TC Project	Description	Technical Officer(s)
ZAM/5/025	Development of Feeding Strategies for Smallholder Dairy Animals in Njolwe and Palabana Dairy Tenant Schemes **Objective**: 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.	Garcia Podesta / Odongo

Publications

Identification of quantitative trait loci affecting resistance to gastrointestinal parasites in a double backcross population of Red Maasai and Dorper sheep

M. V. B. Silva, T. S. Sonstegard, O. Hanotte, J. M. Mugambi, J. F. Garcia, S. Nagda, J. P. Gibson, F. A. Iraqi, A. E. McClintock, S. J. Kemp, P. J. Boettcher, M. Malek, C. P. Van Tassell and R. L. Baker

Animal Genetics, doi: 10.1111/j.1365-2052.2011.02202.x

A genome-wide scan for quantitative trait loci (QTL) affecting gastrointestinal nematode resistance in sheep was completed using a double backcross population derived from Red Maasai and Dorper ewes bred to F1 rams. This design provided an opportunity to map potentially unique genetic variation associated with a parasite-tolerant breed like Red Maasai, a breed developed to survive East African grazing conditions. Parasite indicator phenotypes (blood packed cell volume – PCV and faecal egg count – FEC) were collected on a weekly basis from 1064 lambs during a single 3-month post-weaning grazing challenge on infected pastures. The averages of last measurements for FEC (AVFEC) and PCV (AVPCV), along with decline in PCV from challenge start to end (PCVD), were used to select lambs (N = 371) for genotyping that represented the tails (10% threshold) of the phenotypic distributions. Marker genotypes for 172 microsatellite loci covering 25 of 26 autosomes (1560.7 cM) were scored and corrected by GENOPROB prior to QXPAK analysis that included Box–Cox transformed AVFEC and arcsine transformed PCV statistics. Significant QTL for AVFEC and AVPCV were detected on four chromosomes, and this included a novel AVFEC QTL on chromosome 6 that would have remained undetected without Box–Cox transformation methods. The most significant P-values for AVFEC, AVPCV and PCVD overlapped the same marker interval on chromosome 22, suggesting the potential for a single causative mutation, which remains unknown. In all cases, the favourable QTL allele was always contributed from Red Maasai, providing support for the idea that future marker-assisted selection for genetic improvement of production in East Africa will rely on markers in linkage disequilibrium with these QTL.

Influence of exogenous enzymes ensiled with orange pulp on digestion and growth performance in lambs

H.M. Gado, A.Z.M. Salem, N.E. Odongo and B.E. Borhami

Animal Feed Science and Technology 165: 131–136 (2011)

Twenty-four Ossimi male lambs were used to evaluate effects of inclusion of ensiled orange pulp (EOP) in lamb diets either with or without addition of exogenous enzymes (ENZ) of ZADO® on digestion and growth performance. Lambs (21.1±1.01 kg body weight (BW)) were assigned to one of three groups of 8 animals/group in a randomized complete block design being: Control (basal diet with 0 g/kg EOP), EOP (Control with 150 g/kg EOP) or EOP + ENZ (EOP with 5 g/kg of ZADO®) in a 90-day experiment. Ensiling the orange pulp increased the crude protein, ether extract and metabolizable energy of the silage by 29, 46 and 8%, respectively, and reduced the secondary metabolites, such as total phenolics, saponins and alkaloids. Silage lactic acid and ethanol were increased by35%and 54%, respectively for EOP and EOP with ENZ, but all silage quality parameters were in the normal range. Concentration of NH3–N before feeding was decreased ($P < 0.05$) by 11 and 13% in EOP and EOP + ENZ, respectively, whereas at 3 and 6 h after feeding ruminal VFA concentration was increased ($P < 0.05$) by 23 and 9% respectively, only in EOP + ENZ lambs. NDFom intake was increased ($P = 0.036$) by 52 and 59%, whereas the ADFom increased ($P = 0.032$) by 8 and 11% in EOP and EOP + ENZ lambs, respectively. Nutrients digestion were higher ($P < 0.05$) in EOP + ENZ than EOP lambs. Digestible DM was increased by 18%, whereas the fibre fractions (NDFom and ADFom) were increased by 93 and 47% with similar EOP + ENZ. DM intake among groups, whereas feed efficiency was higher ($P = 0.042$) by 19 and 31% in EOP and EOP + ENZ lambs compared to control diet. Live weight gain increased ($P = 0.038$) by 92% in EOP + ENZ lambs whereas it increased by 54% in EOP lambs. Addition of EOP to the diet improved feed efficiency and live weight gain suggesting a good quality feed, which could probably be used to replace a part of the concentrate in ruminant diets.

Trypanosoma brucei brucei: A comparison of gene expression in the liver and spleen of infected mice utilizing cDNA microarray technology

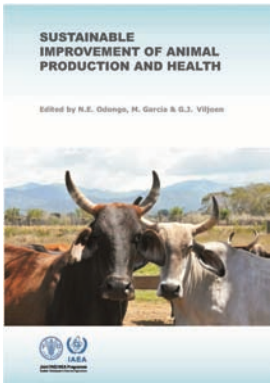
San-Qiang Li, A. Luckins, Zhao-Rong Lun

Experimental Parasitology 128: 256–264 (2011)

Trypanosoma brucei brucei, the infectious agent of the disease known as Nagana, is a pathogenic trypanosome occurring in Africa, where it causes significant economic loss to domesticated livestock. Although many studies on the histopathology of organs of mice infected with *T. b. brucei* have been reported, little work has been done regarding gene expression in these organs in infected mice. In this paper, we describe the use of cDNA microarray to determine gene expression profiles in the liver and spleen of mice infected with *T. b. brucei* (STIB 920) at peak parasitaemia (12 days after infection). Our results showed that a total of 123 genes in the liver and 389 genes in the spleen were expressed differentially in *T. b. brucei* infected mice. In contrast, however, in an acute infection in mice caused by *Trypanosoma brucei evansi*, a species genetically related to *T. b. brucei*, 336 genes in the liver and 190 genes in the spleen were expressed, differentially, indicating that the liver of mice was more affected by the acute *T. b. evansi* infection whilst the spleen was more affected by the subacute *T. b.brucei* infection. Our results provide a number of possible reasons why mice infected with *T. b. evansi* die sooner than those infected with *T. b. brucei*: (1) mice infected with *T. b. evansi* may need more stress response proteins to help them pass through the infection and these are probably excessively consumed; (2) proliferating cell nuclear antigen was more down-regulated in the liver of mice infected with *T. b. evansi*, which indicated that the inhibition of proliferation of hepatocytes in mice infected with *T. b. evansi* might be more severe than that in *T. b. brucei* infection; (3) more hepatocyte apoptosis occurred in the mice infected with *T. b. evansi* and this might be probably the most important reason why mice died sooner than those infected with *T. b. brucei*. Studies of the changes in the gene expression profile in the liver and spleen of mice infected with *T. b. brucei* may be helpful in understanding the mechanisms of pathogenesis in Nagana disease at the molecular level. By comparing the gene profiles of the liver and spleen of mice infected with *T. b. brucei* with *T. b. evansi*, we have identified a number of factors that could explain the differences in pathogenesis in mice infected with these two African trypanosomes.

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.

http://www-naweb.iaea.org/nafa/aph/public/aph-sustainable-improvement.html

Freedom from Rinderpest

Rinderpest (also known as cattle plague), a highly contagious viral disease of cattle, buffalo, yak and several wildlife species is no more. Countries that had suffered from the ravages of rinderpest were officially recognized as disease free by the World Organisation for Animal Health (OIE) in May 2011 and by the Food and Agriculture Organization of the United Nations (FAO) in June 2011, when those organizations declared that rinderpest had been eradicated worldwide. The IAEA (together with FAO, OIE and AU) made a significant technical contribution over a period of almost 20 years through the development, evaluation, validation and distribution of immunological and molecular nuclear and nuclear related technologies for the diagnosis and control of rinderpest. This EMPRES special issue on 'Rinderpest' highlights the contribution of the different organizations to its eradication.

http://www.fao.org/docrep/014/i2259e/i2259e00.pdf

IAEA-TECDOCS are available electronically on:
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Email: sales.publications@iaea.org
http://www-pub.iaea.org/MTCD/publications/publications.asp

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 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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Websites

- The AP&H Section website is being updated on a regular basis. Please feel free to look at it and make comments.
  http://www-naweb.iaea.org/nafa/aph/index.html

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