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Milk collection point by a processing plant Sabga, North West province Cameroon

To Our Readers

Dear Colleagues,

Once again the year is at end and a new exciting year lies ahead of us. The past year has been a busy time for all staff in the subprogramme. Apart from our regular Coordinated Research Project (CRP) activities and our technical support given to national and regional Technical Cooperation (TC) projects, we were involved in the technical evaluation of applications for new TC projects by Member States for the 2007/2008 biennial project cycle. We have also prepared the IAEA's 2008/2009 Work and Budget Programme. It is hoped that our inputs will serve the best interests of our Member States.

As with previous newsletters, I want to introduce a topic to hopefully stimulate discussion and debate and encourage interactions between all. It has been well discussed in previous newsletters that the trend towards intensification of livestock production in developing countries presents both opportunities and risks. The potential opportunities are the flow-on benefits to the local economy and producers and the potential risks are the flow-on costs to the environment, livestock health and welfare and human health, through increased chemical and nutrient pollution, disease transmission and centralization of feed resources. Understanding nutrition is one of the keys to taking advantage of the opportunities and minimizing the risks.

In developed countries it has been well accepted that intensification of livestock production can lead to higher levels of greenhouse gas emissions and a localization or concentration of nutrient wastes and pollution of waterways, increased chemical and drug use to overcome the increased risk of disease transmission and can also put pressure on the local feed and reproduction management systems. There is now a global pressure from consumers to have sustainable production systems, where high quality and safe products are produced efficiently with minimal impact on the environment and human health, which will not miss the new developments in livestock production in developing countries. Farmers in these communities will be under similar pressures as producers in developed countries, for example, limiting the input of, and finding 'natural' alternatives to, chemicals; exploring alternative sources of feed; reducing methane emissions and nutrient pollution, and addressing animal health and welfare issues; in other words increasing productivity from local resources in a clean, safe and ethical way. To achieve this there is an urgent need to improve our understanding of some key areas of nutrition, including identifying plants containing phytochemicals with extra-nutritional attributes; presenting novel mixes of plants to animals that are both profitable and environmentally friendly; making use of an animal's ability to self medicate and select when given a choice; using stable isotope signatures to help record diet selection and changes and, for example, differentiate between intake of browse and tropical grasses; obtaining functional analyses of microbial communities in the rumen; targeted feeding; nutrition of the foetus; and investigating ways to take advantage of compensatory growth. We are planning a series of short articles on each of these areas of nutrition for our newsletters. In this issue, we address the importance of understanding the microbial ecology of the rumen and highlights the exciting advances that are being made in our ability to understand how microbial communities function and respond in the rumen.

With ruminant livestock, we can address many of the opportunities and risks associated with livestock intensification and consumer's demands for clean, safe and ethical production systems by manipulating the rumen, because the efficiency of the rumen and its microbes determines what end-products are produced during fermentation. These end-products determine the amount of pollution expelled or excreted into the environment (e.g. methane and nitrogenous waste), the amount and quality of meat, milk or wool produced, animal health and welfare (e.g. incidence of lactic acidosis and bloat), feed intake and how efficiently feed is utilized. However, the microbial ecology of the rumen is complicated and has not been easy to manipulate or customize to best suit our demands. It is complicated because there are many different types of organisms with many different functions interacting in different ways, for example synergistically or in competition. However, the microbes are the interface between the plant and the animal and we are under

more pressure than ever to manipulate this microbial ecology to help customize ruminant products and reduce pollution. Five key questions have driven investigations of rumen microbiology since concerted efforts to manipulate the microbes in the rumen to improve productivity were initiated over 50 years ago: (1) what microorganisms are present? (2) how many microbes are there? (3) how do they interact? (4) collectively, what do they produce? and (5) how do they respond to external stimuli as a community?

In the early days, the techniques that were available to answer these questions were limited to culture-based and microscopy techniques, which were labour intensive and required some previous knowledge of nutrient and growth requirements. The early researchers and their results provided a solid framework on which to build upon in the 1980s, when molecular tools were first applied to the rumen system and there was movement away from the need to culture microorganisms to study microbial communities. The molecular era can be classified into 3 phases; 1) cloning, sequencing and manipulation; 2) molecular phylogenetics, exploration and microbial ecology; and, most recently, 3) the 'omics' phase. These phases have not been mutually exclusive and, in particular, the developments during phases 2 and 3 together have moved us closer to answering the 5 key questions in the detail necessary to manipulate microbial communities in the rumen. The developments during phase 2 allow us to monitor the types and number of bacteria at a given time and how they change and respond to different environments, without the need to culture organisms. This information can also be used to make inferences about the interactions between organisms in the system. The developments in genomics, transcriptomics and metabolomics during the 'omics' phase (phase 3), including macro and microarray analysis, allow us to examine the entire lifestyle of a single organism in incredibly fine detail at one time. The challenge now is to combine these abilities to provide a true functional description of the microbial ecology in the rumen. This is the exciting phase we have just entered; the phase of metagenomics, which has the potential to provide us with knowledge to understand what we have been trying to manipulate for years.

Metagenomics, in very simple terms, is used to describe the study of genomes in microbial communities. It is a process by which the genetic potential of a microbial community can be accessed and investigated without culturing all the organisms present. Within this field, researchers are starting to link microbial community structure to function using state-of-the-art techniques, for example *in vivo* expression technology (IVET) and techniques involving isotopic labelling like stable isotope probing (SIP). IVET is a technique used to help identify promoters, or on/off switches for genes, that are influenced by environmental stimuli. For example, IVET can be used to identify genes that are induced when a particular substrate is available, or when microorganisms inter-

act with other microbial species, adhere to substrate or colonize the gastrointestinal tract. The techniques where radio or stable isotopes are used enable the tracking of nutrients in microbial communities as well as identifying those organisms in the microbial community that are metabolically active. The technique involves adding labelled substrate to an environmental sample and tracking the label in DNA (other molecules can be used as well, for example RNA, lipids). The DNA from the microbial community that has been involved in metabolizing the substrate will be labelled and can be separated from the rest of the community using density centrifugation.

The molecular era in rumen microbiology has enabled us to establish a much more detailed picture of the number, variety and biochemical function of the systems that were described originally using the early culture- and microscopy-based techniques. However, it is these state-of-the-art techniques under the banner of metagenomics that have the potential to link community description to function and help to design feeding strategies to customize microbial populations in the rumen. We may then be able to address global pressure for sustainable production systems, where high quality and safe products are produced efficiently with minimal pollution and erosion of the environment, minimal impact on human health, minimal chemical inputs and from healthy animals. In news from the sub-programme we have to say goodbye to wish Phil Vercoe, who was with us for a one year sabbatical from his university in Australia (contact email address: pvercoe@animals.uwa.edu.au). Phil was respon-

sible for most of our nutrition activities and performed his duties with pure professionalism and integrated well into the animal production and health team. He was a valuable team-member and under his guidance the animal nutrition activity has received new momentum. I want to thank him for his contributions and want to wish him, Rebecca and children only the best for the future. We will remain in close contact and will continue to make use of his expertise. On a more sunny note, I want to welcome Massoud Malek and Charles Lamien to the sub-programme. Massoud has experience in both statistical/quantitative and molecular genetics and will be responsible for applying molecular and quantitative genetics to increase the efficiency of programmes in animal reproduction. Charles, a post-doctoral fellow, is a veterinary molecular biologist with special interest in reverse genetics. Both will be stationed at our Seibersdorf laboratory.

Finally, I wish you all and your families a happy, healthy and safe new year



Gerrit Viljoen,
Head, Animal Production and Health Section

Staff

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

Forthcoming Events

Second RCM of the CRP on the Veterinary Surveillance of Rift Valley Fever (D3.20.23)

Technical Officer: G. Viljoen

This RCM will be held from 5 to 9 March 2007 in Kenya Nairobi.

The purpose of the Research Coordination Meeting is to review the progress achieved by the Research Contract holders since the last Research Coordination Meeting and plan and agree on the research programme for the following year's activities.

First RCM of the CRP on The Early and Rapid Diagnosis of Transboundary Animal Diseases: Phase I - Avian Influenza

Technical Officer: J. Crowther

The first RCM will take place from 19 to 23 March 2007 at the VIC, Vienna. The purpose of the Research Coordination Meeting is to review the projects and to prepare work plans.

Second RCM of the CRP on the Development and Use for Rumen Molecular Techniques for Predicting and enhancing Livestock Productivity and Training Workshop (D3.10.24)

Technical Officers: Paul Boettcher/Phil Vercoe

This Research Coordination Meeting (RCM) and a Training Workshop on Diversity analysis of rumen microbial communities with associated bioinformatics analysis will take place from 19 to 29 April 2007 at the University of Illinois, Urbana-Champaign, USA.

The purpose of the Research Coordination Meeting is to review the progress achieved by the Research Contract holders since the last Research Coordination Meeting in 2005 and plan and agree on the research programme for the following year's activities. The purpose of the Training Workshop is to provide the Research Contract holders with the theoretical knowledge and technical ability to complete the next stages of research in the CRP, which is to use molecular and nuclear-related technology to analyse microbial communities in the rumen.

Past Events

4th International Veterinary Vaccines and Diagnostics Conference (IVVDC)

Technical Officer: Gerrit Viljoen

The Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture cooperated in the organization of the fourth IVVDC, which was held from 25 to 29 June 2006 in Oslo, Norway.

The conference benefited from good cooperation with the pharmaceutical industry, public institutions and international organizations both economically and scientifically, and the agenda reflected topical problems. The basis of the conference was very wide, from basic to applied research including commercial and regulatory aspects, in order to address current topics faced by researchers. The conference was attended by more than 400 delegates from 50 countries and was divided into 9 sessions based on the various areas of vaccinology and diagnostics. The scientific programme included sessions on molecular diagnostics, immunological memory, vaccines and diagnostics for ruminants, vaccines and diagnostics for fish, vaccines and diagnostics for swine, vaccines and diagnostics for companion animals, vaccines and diagnostics for avian

species, vaccines and diagnostics for equidae, control of livestock diseases in the tropics, regulatory aspects regarding use of genetically modified organisms in veterinary vaccines, Transmissible spongiform encephalopathies, Commercial vaccine and diagnostic development and two special topics; Avian influenza and Transboundary animal diseases. Several of the speakers presented work that was supported by the Agency.

Fifth FAO/IAEA Molecular Diagnostic and PCR Fellowship Training Course

Technical Officer: Gerrit Viljoen

The fifth workshop of this kind was held from 7 August to 1 September 2006 in Pretoria, South Africa in cooperation with Biogenics, PCRbiotech. The course coordinator is Prof. Luis Nel, email: louis.nel@up.ac.za.

The course is intensive and is designed to establish the basic understanding of fundamental molecular biological principles and biotechnological applications. In addition, the intention is that the molecular diagnostic and PCR course will give concrete guidelines and hands-on training on practical aspects of rapid and

sensitive diagnoses. These aspects included good laboratory practice and requirements for setting-up PCR laboratories, standardization of PCR protocols, the implementation of PCR diagnostic tests and good service delivery, awareness of difficulties as well as advantages of the assay, current advances in the PCR procedures and the placement of PCR in relation to other tests. This year, a practical section on extraction of RNA and DNA for uses as templates in PCR was also included as well as DNA sequencing and basic bioinformatics for molecular epidemiology analysis. In general, the course was structured to help those already involved in molecular diagnostics and PCR, with specific emphasis on the use of biotechnology for diagnosticians and scientists in the developing world. Selected protocols which are currently being used as SOP's in individual laboratories as well as detailed training material necessary for PCR technology transfer were given.

The training programme was structured to include the presentation of theoretical principles and methodology as well as 'hands-on' bench work. Approximately 60% of the course was devoted to laboratory instruction and problem solving/quiz/tutorials while 40% was lecture-oriented. Participants were given sufficient time for individual and group discussions as well as performing problem solving activities. Each participant was provided with an extensive manual with protocols and procedures.

The theoretical lectures were followed (where possible) with practical sessions

These lectures included:

- Laboratory setup and Laboratory safety
- Lectures on buffers and general laboratory techniques
- details of the history of PCR and an overview of the technique and its development
- advanced PCR and PCR optimizations
- PCR detection and evaluation techniques
- Limitations of PCR and troubleshooting
- Randomly amplified polymorphic DNA
- Isolation of RNA and DNA
- Vaccinology
- DNA structure, gene expression and gene activity
- Mutation
- Recombinant DNA
- Micro-arrays
- Emerging and re-emerging diseases
- DNA sequence analysis
- Bio-informatics and molecular epidemiology

Alternative Feed Resources: A Key to Livestock Intensification in Developing Countries

Technical Officer: Phil Vercoe

This meeting was held in cooperation with Writtle College and The British Society of Animal Science and

their Ethnoveterinary Medicine Conference Harvesting Knowledge, Pharming Opportunities, at Writtle College, Chelmsford, Essex, UK, on 12 and 13 September 2006.

The two main objectives of the FAO/IAEA co-sponsored meeting were to:

1. Review the opportunities and challenges associated with in vitro screening of plants for bioactive properties and to use feeding behaviour and selection principles for developing systems that integrate novel plants and extracts into feeding systems.

2. Identify opportunities for technical and research projects for increasing the scope of the plant screening activities and developing integrated feeding systems for livestock production that are profitable and environmentally safe. This is with a view to identifying opportunities or ways to establish a global network of researchers screening novel plants for improving animal production and health, and exchanging information.

Participants felt that it was certainly feasible and worthwhile to extend the various screening activities currently being undertaken to a more global scale and that some of the most exciting opportunities lay in developing countries. Presentations during the meeting included lessons learned from large European programmes involving the screening of plants for bioactive properties for improving the health and production of ruminants, as well as a more recent project in Australia, which is based on similar, but slightly broader objectives (including the role of animal behaviour); the role of behaviour when trying to introduce novel plants into livestock production systems and how animals have the ability to self-medicate providing they have choice; using alternative plants/plant by-products to improve animal health and the safety of animal products for human consumption controlling internal parasites (e.g. nematodes) and microbial communities (e.g. *E. coli*); and finally intellectual property and knowledge exchange issues that need to be considered in a programme involving screening local plant resources for bioactive compounds that have the potential to be commercialized.

Various bioactive compounds that we can screen native flora for in vitro were prioritized according to their technical feasibility, likely impact on farmers profitability, likelihood of in vitro effects being found in vivo and the role of nuclear applications in the screening methodology. Based on these criteria, screening for plants with bioactivity against gastrointestinal parasites was ranked highest, as well as efficiency of nitrogen utilization and enhancing fibre digestion.

Project Coordination Meeting of RAF5054 Improvement of Livestock Productivity through an Integrated Application of Technologies

Technical Officer: Paul Boettcher

The project coordination meeting for RAF5054, Improvement of Livestock Productivity through an Integrated Application of Technologies, was held from October 16 to 20 in Tunis, Tunisia and was hosted by the Medicine Veterinary School of Sidi Thabet. The objective of RAF5054 is to develop and facilitate the application of appropriate selection criteria for genetically improved stock; to institute integrated management, nutrition, healthcare and follow-up practices for genetically improved stock; and to use modern reproductive techniques to improve productivity and reproductive efficiency of livestock in the region. RAF5054 is scheduled to run for five years and the objective of the meeting was to develop group and individual work-plans for the duration of the project. Participants from 20 African MS attended the meeting.

First RCM on the Diagnosis and Surveillance of Contagious Bovine Pleuropneumonia (D3.20.24)

Technical Officer: Hermann Unger

This Research Coordination Meeting took place from 30 October to 3 November 2006 in Windhoek, Namibia.

This meeting set the scene for the CRP and individual work plans were coordinated. The 2 serological tests to be evaluated, the LPPQ ELISA and the c-ELISA, were presented and available data discussed. As the robustness of the c-ELISA was reportedly increased, no problems with shipment should now be expected. Lipopolyprotein Q and its pathological background was presented and its potential use in a skin test discussed. Primary results for the detection of *Mycoplasma m.m.SC.* by a new iso-thermic PCR (LAMP-PCR) were shown and this test system was envisaged to be introduced to the collaborating laboratories in 2007. Two scientific presentations explained different approaches to molecular epidemiology of CBPP. One aspect was identifying historic distribution pattern, the alternative approach is on potential pathogenicity factors. While the spatial approach is already published but unfortunately requires sophisticated equipment, the alternative approach was up to now only used for other pathogens and markers have still to be analyzed. This alternative procedure would on the other only demand some PCR equipment, available in most of the cooperating laboratories. Finally a meeting for the counterparts of Zimbabwe and Botswana was arranged together with the local veterinarians in Livingstone, Zambia, to work one week in a CBPP eradication programme, perform serological mass screenings, do clinical examinations in affected herds and to get hands on experience in CBPP post mortems.

Training Course on the Diagnosis of Avian Influenza

Technical Officers: Gerrit Viljoen/ Adama Diallo

An Inter-Regional training course on Molecular Techniques for the Diagnosis of Avian Influenza was held from November 20 to 1 December 2006 at the FAO/IAEA Agriculture and Biotechnology Laboratory in Seibersdorf, Austria. This training course, sponsored by IAEA and FAO was attended by 30 participants from Africa, Asia and Latin America. Its ultimate goal was to contribute to the early detection and early reaction capabilities in Member States. It was an intensive course which included lectures on the epidemiology of highly pathogenic avian influenza, molecular technologies for the diagnosis of avian influenza and practical aspects for RT-PCR, RT-Realtime PCR and gene sequencing and typing of Avian Influenza virus for rapid characterization of the pathogen.

Consultants Meeting on Standards, referencing and validation

Technical Officer: John Crowther

The meeting was held from 21 to 24 November 2006 in Vienna and brought together the institutional and commercial interests to examine the current process and make recommendations for changes/improvements. The areas covered were:

A. Consideration of the progress on kit certification by OIE based on fitness for purpose and defined validation pathways.

B. Needs, definitions, production, storage, distribution costs and use of standards for serological and molecular tests for transboundary diseases of livestock and the definition and roles of reference laboratories/centres.

The IAEA has supported the OIE in the past by funding consultancy meetings and expertise to help improve test/kit validation procedures:

1. Consultants Meeting on OIE Validation and Certification of Diagnostic Assays for Infectious Animal Diseases 18–20 November 2002, Vienna, Austria
2. Consultants Meeting on OIE Validation and Certification of Diagnostic Assays for Infectious Animal Diseases 9–12 December 2003, Vienna, Austria.
3. A consultant and IAEA staff provided a web based application form for the certification of tests by the OIE which were based on fitness for purpose and validation guidelines agreed by the consultants at the previous meetings.

The forms mentioned in (3) are the basis of the certification pathway as shown on OIE web pages. It was concluded that the validation template is of a high standard and extremely useful as a guideline for developing tests. The costs of registration with OIE were believed to be far too high. There was debate over what exactly the OIE registration meant. Producers believed that a single approved registration process for all countries would be a great advantage to relieve them of the process of registering kits in many separate countries. Discussions as to whether OIE should try and establish this accepted registration process were made.

The areas of trade prescribed tests and alternative tests were discussed and it was indicated by some that these

are terms which complicate the validation assessment for countries and that they might be reassessed only in terms of fitness for purpose criteria. It is proposed that a standard be written for validation and certification of tests.

There was debate over the need for referencing in general and more specifically, standards for serological and molecular tests used for diagnosis and surveillance of livestock and human diseases. There can be no doubt that a most damaging factor to obtaining valid data from any studies is the issue of quality of work and the assessment and control of this quality. This can be exemplified by the use of kits for the diagnosis of diseases that are affected by the robustness and formulation of the kit; the skills of the operators; the environmental conditions; the day to day and month to month performance of a test both within the same laboratory and between laboratories; the processing of data and misconceptions about the fitness for purpose of any set of reagents. This situation is further complicated by the lack of consistency in terminology; the assumptions made about status of laboratories; the poor training of staff; the lack of coordination by a variety of funding agencies and the existing rules on testing and emphasis on trade issues. Valid data based on defined sampling strategies was believed to be vital.

It was agreed that central to the quality issue is the production, gathering, characterization, storage and distribution of standard preparations, both for the continual assessment and the development of kits. It was concluded that decisions about standards for any situation are needed through expert advice; applied research and harmonization of efforts, particularly since funding for such efforts is woefully poor.

Discussion points were:

1. The real needs for reference materials in biological situations?
2. Internal quality control aspects.
3. External comparison of tests at the national, regional or International level.
4. Can EQA be achieved made in different ways?
5. Standards for antigen and nucleic acid detection assays.
6. Analytical sensitivity vs diagnostic sensitivity, how do we handle this?

Examples of the use of standards were given in presentations.

A Portal Disease Information Exchange was also discussed. In brief:

1. This could be a Web based, form driven (information has to fit to established easy to fill in template) system.
2. Three data types for three areas are conceived.

- a) Definitive data from validated assays
- b) Less definitive but extensive data from tests
- c) Results where conditions are defined but only on a few samples.

The portal could act as an active, continuously updateable repository for data which can be viewed by anyone. The accumulation of data allows statistics to be applied on a larger data set which will improve validation and allow a different stage of validation to be achieved though increasing the confidence in data.

Management of the PORTAL is required to:

- a) Ensure correct input of data as well as to assign which 'room' the data goes.
- b) To encourage better practice where data does not qualify for inclusion e.g. where IQC is ignored.

Participants: Peter Wright, Canada; Wayne Dimech, Australia; Matthias Greiner, Germany; Valerie Bevan, UK; Barbara M. Martin USA; Malik Merza, Svanova, Sweden; Liesbeth Jacobs, CEDI, Netherlands; Serge Leterme, IDEXX, France; Nigel Ferris, UK; María José Dus Santos, Argentina; Johanna KOOLEN, Netherlands; Terry F. McElwain, USA; Francois Diaz, OIE; Elisabeth Erlacher Vindel; OIE; Keith Perry, UK; Chang Yi Wang, UBI; USA; Hermann Unger, IAEA; Adama Diallo, IAEA; Gerrit Viljoen, IAEA; John R. Crowther, IAEA.

Final RCM of the CRP to Develop Methodologies for Demonstrating Increases in the Productivity of Peri-urban Dairy Cattle using an Integrated Approach to Nutrition, Reproductive Management and Disease Control (D3.10.23)

Technical Officer: Paul Boettcher

This RCM took place from 4 to 8 December 2006 at the University of Edinburgh, UK. This meeting was the final RCM for this CRP. The primary purpose of the RCM was to discuss and evaluate the results of interventions made on the farms participating in the various country projects. The interventions made were designed to increase the quality of diets, reproductive efficiency and general health of the cattle. Specific attention was paid to the economic aspects of the interventions, however, and the costs and benefits of the techniques applied were evaluated. Because this was the final meeting of the project, one session of the programme was devoted to discussion of strategies to ensure sustainability and increased adoption of the most successful interventions.

Ongoing Activities

Education to Improve the Quality of Research in Developing Countries

Technical Officer: John Crowther

The site which holds the education package under development can be visited under URL: researcher-training.org

Two contracts to create new modules for the package have been awarded t

Coordinated Research Projects

Integrated Approach for Improving Small-scale Market Oriented Dairy Systems (D3.10.23)

Technical Officer: Paul Boettcher

This CRP is currently in its fifth and final year and has a full complement of participants, comprising ten Research Contracts, one Technical Contract and four Research Agreements. The final RCM was held in Edinburgh, UK, from 4 to 8 December 2006. The final analyses of data are being undertaken on the costs and benefits of interventions applied to overcome the most serious constraints on farm income as identified by the Economic Opportunity Surveys. Based on preliminary studies, most of the interventions applied were found to be economically beneficial. Nevertheless, encouraging other farmers to adopt them has been recognized as a challenge that will require diligence on the part of the CRP participants.

Development and Use of Rumen Molecular Techniques for Predicting and Enhancing Productivity (D3.10.24)

Technical Officer: Phil Vercoe

There are currently eight Contract holders and five Agreement holders in this CRP. During the last RCM held in Zurich, Switzerland from 12 to 16 September 2005 there were extensive discussions about the need to test the persistency of effects of different plant/plant extracts on rumen fermentation parameters. A technical contract has been prepared to evaluate a method for testing persistency that involves a continuous fermentation system and real time PCR. Contract holders have validated their real time PCR methodologies for quantifying methanogen populations using 2-Bromoethanesulphonic acid and are now starting to screen plant secondary compounds for their effect on *in vitro* methanogenesis, investigating ciliate protozoal activities using a ¹⁴C-labelling technique and screening plants showing promise for initial studies for their persistency and microbial adaptation. The next RCM for this CRP is tentatively scheduled for April 2007.

Gene-based Technologies in Livestock Breeding: Phase 1: Characterization of Small Ruminant Genetic Resources in Asia (D3.10.25)

Technical Officers: Paul Boettcher

The nine counterparts participating in the project have sampled or will sample DNA from more than 2500 individuals from approximately 90 breeds of sheep and goats. The goats have been genotyped for 13 microsatellite markers from the FAO/ISAG panel for genetic characterization. Preliminary analyses indicate that the goats sampled originated from three distinct genetic groups. Results from the characterization were presented at the XIIth Congress of the Asian-Australian Association of Animal Production Societies in Busan, Republic of Korea in September. Most of the contract holders attended and some presented results of their own research. In addition, an informal project planning meeting was held at the Korean Animal Genetics Resource Station of the National Livestock Research Institute in Namwon. The main activities foreseen for 2007 are genotyping of the sheep samples, which will be done at the Joint ILRI/CAAS animal molecular genetics laboratory in Beijing, and phenotypic characterization of the breeds, which will be performed by the counterparts. In addition, the breeds will be genotyped for microsatellites on the Y-chromosome and mitochondrial d-loop and for SNP in several candidate genes. The protocols for the analysis of these markers are being developed through joint activities between the Animal Production Unit in Seibersdorf and the IAEA Collaborating Centre in Animal Genomics and Bioinformatics in Brazil.

Veterinary Surveillance of Rift Valley Fever (D3.20.23)

Technical Officer: Gerrit Viljoen

Rift valley fever (RVF) is a mosquito borne viral disease affecting both livestock and people. In animals it mainly causes abortions while humans show influenza like symptoms leading in a small percentage to death.

The disease is endemic to Africa with sporadic major outbreaks following extreme humid conditions. In 2000, imported RVF infected cattle from Somalia caused an epidemic on the Arabian Peninsula resulting in the death of nearly 300 people and several thousand abortions in ruminants. This expansion in the epidemic area to the Arabian Peninsula raises the possibility of RVF spread to other parts of Asia and Europe, especially since RVF virus (RVFV) can be spread by a wide range of mosquito vectors.

Research Contract holders (C) and Agreement holders (A) are from research institutions in Burkina Faso (C), Guinea (C), Mali (C), Mauritania (C), Senegal (C), Kenya (C), Uganda (C), Yemen (C), Gambia (C), Congo (C) South Africa (A), Germany (A) and France (A).

The Control of Contagious Bovine Pleuro Pneumonia in Sub-Saharan Africa (D3.20.24)

Technical Officer: Hermann Unger

The CRP on control of CBPP in Africa has 10 Research Contract holders from Cote d'Ivoire, Zambia, Botswana, Uganda, Burkina Faso, United Republic of Tanzania, Namibia, Kenya, Zimbabwe, Cameroon and 2 Agreement holders from Switzerland and Austria. The first Research Coordination meeting took place from 30 October to 3 November in Windhoek, Namibia

African Swine Fever Technical Contract 11294 (D3.00.00)

Technical Officer: John Crowther

Indirect ELISA kits are still available from the Institut Sénégalais de Recherches Agricoles ISRA, Laboratoire National de l'Élevage et de Recherches Vétérinaires (LNERV), for the detection of antibodies against ASF. Each kit includes plates, tips and reagents for testing 2800 samples and costs US\$ 2000. Applications for kits should be sent to the Senegal laboratory directly (Dr. Joseph Sarr; Josarr@refer.sn).

Concluded CRPs

The Use of Non-structural Protein of Foot-and-Mouth Disease Virus (FMDV) to Differentiate Between Vaccinated and Infected Animals (D3.20.20)

Technical Officer: John Crowther

The CRP reflects the variability in approach for various countries with respect to diagnosis and control of FMD and the variability in resources and the different epidemiological niches where tests are used. The papers from the different countries can be examined in this light. Ideally the use of tests should be determined solely from the fitness of purpose criteria. This is

reviewed in first paper by the technical officer. Often there is confusion when developing tests and then exploiting them, since the purpose of the test and the validation data to fit that purpose have not been examined properly. This was apparent in the CRP, which was also badly affected since developers' kept changing their reagents and performance criteria. This is a lesson we all have to learn along with a proper understanding of the terms diagnostic sensitivity (DSn) and diagnostic specificity (DSp). Often there is a wish for the most sensitive test without appreciating what the needs for the test are. This is associated closely with the prevalence of the disease or analyte, being studied. In the case of NSP test to determine whether herds are infected, then the prevalence should there be an outbreak is likely to be high (a large percentage of animals might expect to be infected). In this case the diagnostic sensitivity of the test needed can be low. Where there is a need to wheedle out single animals in a large herd, for example if we are looking for positive animals a long time after an outbreak situation, then there is a great need to have both a high diagnostic sensitivity and very high specificity. In between the balance of DSn and DSp come the requirements depending on the likely distribution of FMD. Known history of outbreaks and information on animals cannot be excluded and replaced by testing alone.

Full exploitation of the NSP ELISAs and confirmatory tests has not yet been made. One area is the continuous monitoring of animals to see whether there is a low level of FMD circulating, e.g. testing of animals a long time after an outbreak. This monitoring is an essential feature of the OIE pathway to declaring freedom from infection, but sampling regimens and methods based on statistical criteria need to be devised for each country or region, to indicate the best testing strategies. Continuous surveillance also includes the possibilities of using more mobile forms of NSP tests such as the dipsticks. These have been used successfully in the Republic of Korea in an outbreak situation but are probably highly suitable for supply to local veterinarians who could check animals quickly during transportation. A scheme to use such NSP Dipstick tests is being examined in Bolivia under a Technical Cooperation project.

Availability of kits

Tests utilizing FMDV NSP for use in detecting antibodies are available from commercial sources. These would be the kits supplied from IDEXX (I-ELISA); Svanova (I-ELISA), UBI (peptide NSP I-ELISA) and CEDI Diagnostics kits (C-ELISA). The prices are roughly in line and negotiations as to purchases can be made through contact with the companies. The PANAFTOSA kit is more in house and not available except in South America where there is a mandate to provide kits to laboratories. Dipstick NSP ELISAs are also available.

Performance of kits

The test differed in DSp and DS_n, although this conclusion is mainly based on the cut-off criteria of the manufacturers. In turn this is based on their validation data using limited sera both in numbers and geographical spread as well as epidemiological definition. Where direct comparisons have been made between the kits, using the same sera at the same time, it has been shown that adjustment of the cut-offs allows a far better fit to be made, in terms of DS_n particularly.

Post infected non vaccinated animals

The tests available are suitable for screening cattle and pig sera to show an outbreak and to follow on after outbreaks. Less data is available for sheep and goats however; there is no reason to suspect that the diagnostic potential is lower for these groups. After infection, the expected and measured prevalence is high so that the variation at the herd level does not drastically affect any of the tests and their DS_n. The DSp of the tests for this purpose is also satisfactory.

Differences in performance do confuse results some times (around 4 months) after an outbreak. Here the antibody level in most animals has decreased to near base line (zero antibody levels) so that the variation in testing and analytical sensitivity differences between the tests comes into play which in turn affects the DS_n.

The analytical sensitivities of tests were shown to be different in the CRP by using standard sera and titrated in dilution ranges using a constant serum matrix, reflecting the sample concentration of serum. The cut-off levels set by manufacturer's indicated that the UBI was set at a low sensitivity (consequently is highly specific). This DS_n and DSp relationship for the UBI is also affected by the use of peptide antigens which limit the antigenic target for antibodies.

There were some problems noted from peripheral studies with detecting antibodies against FMDV SAT 2 and 3 using the serotype peptides (O and A) in the test. This reflects the variation noted in sequence data in the NSP region, and this has been addressed more recently. Variation in the ability to detect post-infected cattle SAT sera was also seen using I-ELISA and 3ABC.

Vaccinated animals

The specificity for detecting anti NSP in the face of vaccination was proven for all the tests; however, this is complicated by the specific vaccine used. The quality issue for purification has to be dealt with. In South America this has been a major issue and improving the purification process was shown to drastically reduce the number on animals producing anti-NSP responses after vaccination. The DS_n is drastically affected where there are animals which do convert after vaccination and all vaccines should be assessed in the light of the NSP test being used to assess post infection antibodies to NSP. The work by Braga shows what is possible using certain vaccines. The overall conclusion is that vaccines produced to high quality levels without contaminating

NSP do allow NSP tests to determine antibodies against replicating virus. Here the difference in DSp of the UBI affects greatly the DSp for tests where poor quality vaccines are used. Where this is practised, the UBI test can be recommended since it does not recognize the antibodies produced against processed NSP from vaccines as compared to all tests utilizing 3ABC.

Carrier animals

The risk from carrier animals in spreading disease is probably extremely small. However, the non acceptance of zero risk is behind the alarm and measures taken where FMD infects a none FMD country, making it the leader in feared diseases. The CRP showed that the antibody response to NSP in carriers can be very prolonged (years). This would be a good marker of carrier status; however, some animals do not produce anti-NSP (or much antibody at all) and some produce transient antibodies (in line with antibodies produced against structural antigens). Therefore, the use of anti NSP testing to identify carriers has to bare in mind the data surrounding the system being studied. In a population, for a long time after an outbreak (a year or 18 months) there will be animals, in the absence of clinical disease with antibodies against NSP (carriers) as well as carriers without antibodies to NSP. So it is possible to identify some animals which are carriers, but not all, using NSP tests. Epidemiological assessment of populations in time (suitable random or purposive sampling) through surveillance, can determine the rate of change of populations as to the antibody profile for anti NSP antibodies and antibodies to structural proteins. The decay of both sets of antibodies after an outbreak and any increase in either, indicates residual infection. This is most marked where infection may induce hard to see clinical signs, for example in sheep and goats. It is likely that surveillance of sheep and goats using NSP tests will trigger alarm as to the number of animals with antibodies and hence potential threat (as assumed under zero risk mentality). There is no doubt that after outbreaks involving cattle and sheep that sub clinically infected sheep can be identified and traced for an extended time. This poses great problems with declarations of freedom from FMD virus to the OIE. Data on molecular studies (PCR) and tissue culture isolation, confirms the difficulty of analysing probang samples. The isolation rate for RNA or infectious virus is highly variable and does not correlate to the NSP tests. Although carriers are regarded as being capable of spreading virus, it is amazing that the use of the probang has no parallel in nature. The severe nature of scraping the cells from an animal's throat is never repeated in the wild and no one sees an animal scraping its throat on a bush! The likelihood therefore of virus being disseminated in this way is very low indeed.

Pigs

The CRP indicated that some pigs produce large quantities of antibodies against NSP in the face of very heavy

vaccination and where there is no evidence of infection. Pigs are not regarded as carriers. This means that virus is infecting and circulating in herds without clinical manifestation. This needs to be addressed and the management of pig herds in countries where heavy and efficient vaccination keeps clinically observed disease out of sight.

Animal movement and quarantine control

Where there is an export potential, the screening of animals as they move and in holding stations or strict quarantine, is usefully done using NSP tests. All animals can be assessed reducing the errors of the sampling frame statistic. Ideally the Dipstick test can be used by personnel on the ground and any animals found positive be removed and measures taken to delay others from moving or sending them back to their origin. Because the requirement for vaccination is made, it is widely and erroneously assumed, that animals are immune and non infected. This is not true and animals can become infected as they travel. Vaccination is never the proof of lack of virus (see Myanmar and Thailand data for MTM area of trade). A complete management package for the use of NSP tests has to be worked out to assure animals as negative for FMD at different times during transportation. This also applies to a constant monitoring of any population and methods to test small groups of animals, for example, in trucks by veterinary staff armed with NSP dipsticks, could greatly help monitor infected animals.

Standards

Through the CRP, the IAEA contracted a South African laboratory to produce experimentally infected SAT sera and received these for characterization. The sera will be used to compare tests and provide working standards. This has also been extended to type O, A, C and Asia 1 sera from cattle.

Quality control

All the kits can be improved by addition of a better set of controls. It is imperative that a medium activity control is given for the assays whereby control charts can be plotted to compare the performance of the tests on each plate, from day to day and between laboratories. Ideally samples in the Indirect ELISA should be measured against the medium control and not the strong positive.

Final comments

It was a great pleasure to work with all the counterparts, the Research Contract and Agreement holders. All were enthusiastic and friendly, dedicated and conscientious. No papers from the Agreement holders are presented although they were also enthusiastic and very learned, most of the information they gave is published in peer reviewed journals. Final thanks to the various representatives of the commercial companies of UBI, IDEXX (then Bommeli Diagnostics), CEDI and

SVANOVA, who greatly supported the CRP through supply of kits and attendance at the RCMs. Involvement of commercial companies also accelerated the kit certification process by OIE.

Developing, Validating and Standardizing Methodologies for the Use of PCR and PCR-ELISA in the Diagnosis and Monitoring of Control and Eradication Programmes for Trypanosomosis (D3.20.21)

Technical Officer: John Crowther

The proven ability of PCR amplification methods to provide large amounts of nucleic acid from minute quantities possible in well controlled samples, has led to the generalized opinion that PCR is the method of choice in all situations in providing the most sensitive tool available. Although the theoretical limits of the PCR are high, the true performance of a PCR on field samples is often over estimated and poorly validated.

Laboratory based diagnosis is the process of confirming or clarifying clinical findings through examination of samples from the field. The ability of a test to diagnose as disease accurately (correctly) relies on the statistical basis of sampling, sample condition, handling of the sample before it enters a laboratory environment, the handling of a sample in the laboratory, the methods used to prepare a sample for testing and the test itself in terms of its analytical sensitivity (minimum detection limit for an analyte) and specificity (ability to detect only a specific analytical target). All diagnostic tests are affected by this process. The terms diagnostic sensitivity (DSn) and diagnostic specificity (DSp) refer to the performance of a test with regard to accurately measuring the true status of an animal with regard to the analyte examined from a field situation and with field samples.

The CRP confirmed that the PCR is particularly susceptible to problems DSn and DSp through variation in sample taking, handling and manipulation, as well as extraction of nucleic acid. Great precautions are necessary through the use of protocols that ensure the protection of nucleic acid at all stages; that allow a maximum yield of nucleic acid from samples and that are not affected by too great an amount of nucleic acid or the innate exquisite sensitivity of primers sets. Examples of this are shown in the papers of the CRP. What was also apparent was the need for fastidious management of PCR laboratories in terms of sample handling and amplification steps.

Work in the CRP greatly extended the understanding of the use of universal primers sets to allow diagnosis of Trypanosomes in general. Epidemiological studies have indicated that the PCR protocols examined can detect up to 85% of all Trypanosomes. The universal primers offer a relatively cheap option to diagnosis where they are successful in an area, particularly where large scale

screening is needed, e.g. where interventions such as SIT and drug treatments, are made.

The IAEA is firmly committed to supporting PCR and a newer generation of technologies for PCR e.g. real time PCR systems, offer vastly improved methods for the estimation and differentiation of pathogens. An example of this was given in the CRP. Such technologies will be exploited and more tailored to Member States needs in new CRPs developed to look at the problems of Rift Valley Fever and Contagious Bovine

Pleuropneumonia. A great deal of work was made in the CRP looking at animal and human Trypanosomes and the papers reflect a greater understanding of diagnosis that will allow other researchers a much higher baseline to begin studies in future.

I would like to thank the Research Contract and Agreement holders for their great efforts and wish them every success in their continued research and application of PCR methods in control programmes.

New Coordinated Research Projects

The Early and Rapid Diagnosis of Transboundary Animal Diseases such as Avian Influenza (D3.20.25)

Technical Officer: John Crowther

Applications have been received for research contracts and agreements from 14 Member States. These are being reviewed and contracts will be awarded by December 2006.

Specific Research Objectives of the CRP

The development of sensitive, specific and rapid early detection technologies including penside or hand-held systems to detect and or confirm harmful pathogens present in (1) animals before the onset of disease, (2) animals in the 'disease carrier' status or (3) very low numbers in animals or populations of animals to respond to harmful animal disease events, and those of zoonotic nature, in a timely way. This will involve research and diagnostic laboratory evaluation of hardware and protocols in targeted Member State laboratories in collaboration with private commercial partners. Validation of new tests as well as comparison to conventional tests will be essential. There is also a vital element that commercial and institutional organizations developing tests will act as agreement holders to benefit from validation advantage that the CRP, with its wide range of contract holders from different countries, brings. The Joint FAO/IAEA Programme has main-

tained the technology transfer of PCR and conventional diagnostic methods (e.g. ELISA) for the past 20 years and there is a cadre of well trained personnel to allow the uptake and development of a new system(s) who might be available to the CRP to develop new methods. This will be accomplished by:

1. Agreement on specific systems to be examined and methods reported.
2. To produce harmonized SOPs that are easy to perform, mobile (compact), robust, rugged and where results can be read unambiguously by relatively untrained staff and that are compatible with international databases.
3. Development and evaluation of machines, primers and reagents to allow development of laboratory based tests and validate the tests to OIE stage 1 and 2 by comparison with conventional (accepted tests) using both serum samples and those for direct antigen testing where possible.
4. Modification of laboratory based systems to allow for mobility and simplicity in the field.
5. Validation of mobile tests to OIE stage 1 and 2.
6. Field testing of mobile isothermal devices in a routine diagnostic environment and comparison of sample data to laboratory based testing.

The first Research Coordination Meeting will be held in Vienna in March 2007.

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.iaea.org/programmes/ri/uc.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://www-tc.iaea.org/tcweb/default.as>

For further information contact Roswitha Schellander (r.schellander@iaea.org)

Activities of the Animal Production Unit (APU) at the FAO/IAEA Agriculture and Biotechnology Laboratory

Development of a Marker Vaccine for the Control Peste des Petits Ruminants (PPR).

With other partners from Africa and Europe, the APU is involved in the development of PPR marker vaccine with its companion test. In this project, one of the APU work packages is the mapping of the PPRV nucleoprotein (N) to identify its interaction sites essential for its functions. After having identified two regions involved in the nucleocapsid formation, work was implemented to study the interactions between N and the irus matrix protein, M. For that, M and different N deleted mutants were co-expressed by recombinant baculoviruses in insect cells. The interaction of M with these different N proteins were analysed by both immunoprecipitation and confocal microscopy observation. This study has identified one region which is critical for N-M interaction. This result was confirmed by peptide mapping study which has allowed detecting 3 other possible interaction sites.

Test available for detection of polymorphisms in the Prion Protein Gene

ARMS-Taqman method for PRNP genotyping for sheep is ready to be delivered to counterpart countries for genotyping the 3 codon sites of the prion protein gene.

The ARMS-TaqMan method for PRNP genotyping for sheep is rapid, and could differentiate all genotypes. Reproducibility is achieved as a result of using a quantitative method and quickness because no post-PCR steps are required. Also, costs are lower than the normal TaqMan assay because only one probe is needed to resolve the two alleles. The method established is universally suited for a broad range of typing projects with different requirements. It provides an efficient and inexpensive diagnostic mutation analysis that will improve the quality of PRNP genotyping compared with other low-cost methods. It can be implemented by

most molecular genetic laboratories using standard equipment.

Transfer of Technology

In order to foster the regional diagnostic test kit production, the Animal Production and Health Subprogramme has agreed to transfer to the Onderstepoort Veterinary Institute (OVI) in South Africa the technology for kit production of the ELISA test for trypanosomiasis diagnosis developed at APU.

As part of this agreement, Mr Olivier Matthei, from OVI, with a fellowship from the IAEA Technical Cooperation Department, spent four weeks for training in APU in September 2006. This training covered the following issues: preparation of reagents (trypanosome culture techniques), assay adjustment (antigen titration, serum titration, HRP titration), determination of tentative range of UCL/LCL (ELISA testing, automated data transfer, data analysis), routine use (analysis and interpretation of data, ROC analysis, assay troubleshooting), standardization and quality control aspects (IQC and/or EQC, preparation of standardized technical protocols, preparation of standardized ELISA protocol), logistics (identification of consumables and reagents, identification of recommended equipment, literature review/IAEA publications, assembly/dispatch. After this training, some biological materials were sent to Mr Matthei's laboratory to help starting rapidly the kit preparation. APU, for sometime, will backstop OVI in this kit production.

Training in the APU

Ms Mette Munch, a veterinarian student from Denmark, spent two months, July and August, in APU for training on PCR technique applied to the diagnosis of PPR. She was involved in the development of deleted RNA to be used as internal controls in the amplification reaction.

Technical Cooperation Projects

Projects starting in 2007 and Technical Officers responsible for implementation

TC Project	Description	TO
ANG/5/007	Improvement and Veterinary Assistance to Local Small Stock Breeds (Not yet funded)	Viljoen
BKF/5/006	Establishment of Feeding Tables for Feedstuffs that are Locally Available to Stockholders in Burkina Faso	Boettcher
CAF/5/002	Assistance for Epidemiological Surveillance of Animal Diseases	Viljoen
CMR/5/015	Use of Nuclear Techniques for Improving Ruminant Productivity and Disease Control (Not yet funded)	Unger
ERI/5/005	Zoonotic (diseases that can be transmitted from animals to humans) Disease Control and Analysis of Veterinary Residues in Foods	Byron, Viljoen
ETH/5/014	Monitoring and Control of Major Animal Diseases	Viljoen
GAB/5/002	Diagnosis and Control of Animal Diseases (not yet funded)	Crowther
HON/5/004	Improving the Nutrition and Health Conditions of Livestock in Honduras in Order to Increase Productivity and Reproductivity, Phase II (not yet funded)	Vercoe
INS/5/034	Development of Environmentally Sound Livestock and Agricultural Production (Not Funded)	Vercoe
MAU/5/002	Improving the National Capacity in Diagnostics for Animal Diseases (Infection and Parasitic Diseases)	Viljoen
MON/5/016	Improving Productivity of Cattle, Camels and Yaks Through Better Nutrition and Reproductive Management (not yet funded)	Boettcher
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)	Boettcher
MYA/5/015	Strengthening the National Capacity for the Production of Veterinary Vaccines	Crowther Cannavan
NER/5/013	An Integrated Approach for Improvement of Livestock Productivity	Vercoe
PER/5/029	Genomics of the Alpaca: Identification of Expressed Genes and Genetic Markers Associated with Productivity and Embryonic Mortality (Not yet funded)	Boettcher
SIL/5/010	Improving the Productivity of Ndama Cattle In Sierra Leone	Boettcher Viljoen)
SRL/5/041	Maximizing Productivity on Goat Farms through Cost-Cutting and DNA-Based Technology in Selection for Breeding (Not yet funded)	Boettcher Viljoen
SUD/5/031	Setting up a National Network for the Control of Livestock Diseases that affect Exports	Unger
TAD/5/003	Diagnosis and Control of Brucellosis in Cattle, Sheep and Goats (Not yet-funded)	Crowther
UGA/5/028	Improving the Capacity for Diagnostic of Animal Diseases	Viljoen
URT/5/025	Support for the Delivery of Artificial Insemination services	Boettcher
ZAI/5/015	Upgrading Laboratory Services for Diagnosis of Animal Diseases	Viljoen
ZAM/5/025	Development of Feeding Strategies for Smallholder Dairy Animals in Njolwe and Palabana Dairy Tenant Schemes	Boettcher

Operational Projects and Technical Officers responsible for implementation

ANG/5/004	Monitoring and Control of Transboundary Animal Diseases	Crowther
BEN/5/002	Diagnosis and Control of Animal Diseases	Crowther Viljoen
BEN/5/003	Veterinary Drug Residue Monitoring Programme	Cannavan Byron
BKF/5/002	Development of a Veterinary Medicine to Combat the Fowl Pox Virus	Viljoen
BOL/5/016	Diagnosis and Molecular Characterization of the Foot-and-Mouth Disease Virus	Crowther
BYE/9/006	Rehabilitation of the Chernobyl-Affected Territories	Crowther
CMR/5/011	Nuclear Techniques for Improving Local Ruminant Productivity	Boettcher
CMR/5/012	Diagnosis and Surveillance of Major Animal Diseases Using Molecular Biology Techniques	Crowther
COL/5/020	Use of Protein Banks for Improving Pork Production	Vercoe
CPR/5/014	Increasing the Productivity of Crop/Livestock Production System	Vercoe
ELS/5/010	Improving Nutrition Practices and Reproductive Efficiency in Cattle	Vercoe
ERI/5/003	Monitoring and Control of Transboundary Animal Diseases	Viljoen
ETH/5/012	Integrating Sterile Insect Techniques for Tsetse Eradication	Feldmann Viljoen
HON/5/002	Improvement in the Nutritional and Sanitary Conditions of Cattle to Enhance their Productivity through Nuclear Methods	Vercoe
INS/5/029	Supplementary Feeding and Reproduction Management of Cattle	Vercoe Boettcher
INS/5/032	Improving Beef and Dairy Cattle Production in Yogyakarta	Vercoe Boettcher
INT/5/148	Establishing Quality Systems in Veterinary Testing Laboratories	Viljoen Crowther
IRA/5/012	Preparation of ELISA Kits for Diagnosis of Foot-and-Mouth Disease	Crowther
IVC/5/028	Surveillance and control of African Swine Fever	Diallo Unger
KEN/5/025	Development of Diagnostic Tests and Vaccines for Livestock Diseases	Unger
MAG/05/12	Increasing Self-sufficiency in Domestic Meat and Milk Production	Vercoe
MAL/5/025	Food Safety Monitoring Programme for Livestock Products	Cannavan
MLI/5/019	Improving Pneumopathies Diagnosis in Ruminants Using PCR	Viljoen
MON/5/012	Monitoring of Residues in Livestock Products and Surveillance of Animal Diseases	Cannavan Crowther
MON/5/013	Diagnosis and Surveillance of Transboundary Animal Diseases and Production of Diagnostic Reagents	Crowther Viljoen
MYA/0/006	Human Resource Development and Nuclear Technology Support	Crowther
MYA/5/011	Development of Supplementary Feeding Strategies Based on Local Feed Sources	Vercoe
MYA/5/012	Diagnosis and Control of Swine Vesicular Disease and Swine Brucellosis	Crowther
MYA/5/013	Integrated Approach for Enhancing Cattle Productivity	Vercoe

NAM/5/007	Control of Animal diseases in Northern Namibia	Viljoen
NER/5/011	Upgrading Laboratory Services for Diagnosis of Animal Diseases	Diallo Unger
NIC/5/007	Determining Drug Residues in Bovine Meat Exports	Cannavan Byron
NIR/5/032	Control and Eradication of African Swine Fever	Crowther
PAN/5/014	Improving Cattle Production and Quality Control for Monitoring of Animal Diseases	Crowther Viljoen
PER/5/027	Use of Nuclear Techniques to Improve Alpacas Productive and Reproductive Methods	Boettcher
RAF/0/013	ICT-Based Training to Strengthen LDC Capacity	Crowther Boettcher
RAF/5/046	Increasing and Improving Milk and Meat Production (AFRA III-2)	Boettcher
RAF/5/053	Assistance to OAU/IBAR PACE Programme for the Control and Eradication of Major Diseases Affecting Livestock	Viljoen Lelenta
RAF/5/054	Improvement of Livestock Productivity through an Integrated Application of Technologies (AFRA III-4)	Boettcher
RAF/5/055	Support to African Union's Regional Programmes for Control and Eradication of Major Epizootics	Viljoen
RAS/5/035	Improving Animal Productivity and Reproductive Efficiency (RCA)	Vercoe Boettcher
RAS/5/041	Production of Foot-and-Mouth Disease Antigen and Antibody ELISA Reagent Kit (RCA)	Crowther
RAS/5/044	Integrated Approach for Improving Livestock Production Utilizing Indigenous Resources and Conserving the Environment (RCA)	Garcia Boettcher
RER/5/012	Regional Control of Brucellosis in Sheep and Goats (core 2003–2007)	Crowther
SAF/7/002	Development of Veterinary Vaccines and Strengthening Drug Residue Laboratory Capabilities	Crowther Viljoen
SIL/5/006	Improving the Productivity of N'dama Cattle	Boettcher Vercoe
SUD/5/028	Epidemiology and Control of Snail-borne Diseases in Irrigated Areas	Crowther
SUD/5/029	The Characterization and Quality Assured Production of an Attenuated Theileria Annulata vaccine	Crowther
TUN/5/021	Fodder Shrubs as Feed Resources to Improve Livestock Productivity	Vercoe
URT/5/021	Livestock Development in Zanzibar After Tsetse Eradication	Boettcher Vercoe Viljoen
YEM/5/004	Improving the Diagnosis of Animal Diseases	Crowther
YEM/5/006	Quality Management for Upgrading Animal Disease Control	Crowther Viljoen
ZAI/5/014	Upgrading Laboratory Services for Diagnosis of Animal Diseases	Crowther
ZIM/5/010	Improvement of Veterinary Diagnostic Laboratory Services	Unger

Publications

In Press

Improving Animal Productivity by Supplementary Feeding of Multinutrient Blocks, Controlling Internal Parasites, and Enhancing Utilization of Alternate Feed Resources

IAEA-TECDOC-1495

ISBN 92-0-104506-9

This publication is a comprehensive overview of the practical aspects of developing and using urea-molasses multinutrient blocks in different parts of the world. Livestock farming is important for provision of animal protein for human consumption, and as a source of income for many poor farmers in developing countries. With the increase in human population and economic growth of many Asian countries, the demand for livestock products will increase considerably in the coming years. However, the main constraint to livestock development in these countries is the scarcity and fluctuation of the quality and quantity of the year-around animal feed supply. Increased population and industrialization are making the arable land scarce and in addition a large area of arable land is being degraded due to human activities. For sustainable development of the livestock sector it is essential to secure sufficient supply of balanced feeds from resources, which do not compete with human food. The conventional feeds such as soya bean, groundnut, rapeseed meals etc., are either not available or are available at a very high cost. Therefore, there is an urgent need to efficiently utilize locally available feed resources such as tree and shrub leaves, agroindustrial by-products and other lesser known and new plants adapted to harsh conditions and capable of growing in poor, marginal and degraded soils. Another important limiting factor for enhancing animal productivity in the tropical countries is heavy internal parasitic load in livestock. Results from the regional IAEA TC project entitled Improving Animal Productivity and Reproductive Efficiency RAS/5/035 are presented in this publication. It is hoped that this publication will be of great practical value to extension workers, students and researchers, and to those thinking of using such feed supplementation technology or of starting commercial production.

FAO/IAEA Manual on Measurement of Methane from Ruminants

This manual will be published by Springer

This manual stems from a training workshop on Methodologies for Determination of Methane from Ruminants that was held in Zurich in 2005 under the Coordinated Research Project (CRP D3.10.24.) on Development and Use of Molecular Techniques for Predicting and Enhancing Livestock Productivity. The main objective of this CRP is to reduce methane (a greenhouse gas) emission from livestock and divert the energy being lost in methane production to increasing livestock production and simultaneously, reducing environmental pollutants. The key aspect of this work is having the ability to measure methane emissions from livestock so that the effect of any changes that are made to reduce methane emissions can be measured quantitatively. However, the methods for measuring methane from animals *in vivo* are complex and the capacity to measure methane from whole animals requires some specialized equipment, careful planning and experience. If the specialized equipment can be purchased, then for most of the techniques used to measure methane *in vivo*, the remaining equipment can be constructed quite simply and cheaply. The objective of producing this manual is to provide researchers starting work in this field with all the necessary information they need to decide on the best method to use in their environment and then to establish that technique in their organization. This manual explains in detail the following 6 methods that are used to measure methane from ruminants:

- SF6 tracer technique
- Respiration chambers
- Tunnel System for methane determination using an infra-red detector and GC
- Chamber/box system for methane determination using a GC
- Indirect method for methane determination by infusion of labelled short chain fatty acids
- Direct method for methane emission by infusing labelled methane

The manual provides both theoretical and very practical details, including diagrams, equations and photographs, to enable a research team to set up and measure methane emissions from ruminants. It is also a source of references to key publications in this field.

In Preparation

Improving the reproductive management of smallholder dairy cattle and the effectiveness of artificial insemination services in Africa using an integrated approach

Results from the IAEA Technical Cooperation AFRA Project on 'Increasing and improving milk and meat

production', implemented with technical support of the Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture.

Formulation of Guidelines for Manure Management in Asian Livestock Production Systems for Achieving Agricultural Sustainability

A publication about developing guidelines for efficient manure management in Asian livestock production systems is being prepared based on an Expert Meeting that was held under the IAEA/RCA Regional Technical Cooperation Project (RAS/5/044). The specific objectives of the nutrition component of the project are to improve animal productivity and decrease discharges of selected greenhouse gases (methane and carbon dioxide), and selected nutrients (nitrogen and phosphorus) into the environment. This publication is focussed on the management of nutrient waste component of the project.

Livestock manures and other agricultural waste products represent a valuable resource, which if used appropriately with a minimization of losses can replace significant amounts of fertilizer in areas with intensive livestock production. On the other hand the large volumes of animal manure are not only a source of valuable plant nutrients but also a threat to aquifers and surface waters. As livestock production intensifies there are serious concerns that poor management and use of manure could jeopardize the sustainability of the production system because of environmental damage and disease transfer, and reduce productivity levels to well below their potential. In this publication information about state-of-the-art manure management practices, current practices in Asia, amounts of manure produced and barriers to effective manure management is combined to provide guidelines for the development of a sustainable, environmental friendly and sanitary livestock production in Asia. It highlights the pressing need for holistic research into strategies and technology for management and treatment of manure, residues and wastes, which can ensure a sustainable use of nutrients and reduce environmental impacts, including odour and ammonia emissions, greenhouse gas emissions and the spread of diseases

Managing Prenatal Development to Enhance Livestock Productivity

The need for a book dealing with managing prenatal development to improve livestock productivity was identified during a Consultants meeting on Research Needs for Improvement of Livestock Productivity in Developing Countries Through Manipulation of Nutrition in utero, held in October 2005.

There is a growing demand worldwide for livestock products and the role of developing countries in meeting this demand will increase. Within this, the current production systems will come under increasing pres-

sure because of the access to feed resources and other environmental challenges. The reproductive female will be under the most pressure in the future because she will be expected to reproduce consistently, and at the very least, annually. The female will also face nutritional and other environmental challenges in meeting the developmental needs of the embryo and foetus throughout gestation and in the preweaning period. Therefore, the foetus is exposed to various challenges that are mostly, but not exclusively, of a nutritional nature. The question is whether these challenges impact on foetal development and subsequent health, growth, reproductive and lactational characteristics of the offspring.

The objectives for writing this book are to provide a quantitative assessment of the role of, and current state of understanding of the mechanistic basis to, environmental plasticity in producing healthy and productive livestock. The book will contain review papers covering all the key livestock species as well as chapters covering relevant information on non-livestock species.

Publication on The Use of Non-structural Protein of Foot-and-Mouth Disease Virus (FMDV) to Differentiate between Vaccinated and Infected Animals.

The results of the Coordinated Research Project on the Use of Non-structural Protein of Foot-and-Mouth Disease Virus to Differentiate between Vaccinated and Infected Animals are being prepared and will be published shortly.

Publication on Developing, Validating and Standardizing Methodologies for the Use of PCR and PCR-ELISA in the Diagnosis and Monitoring of Control and Eradication Programmes for Trypanosomosis

The publication has been completed and will be submitted for publication.

Publications in Scientific Journals and Conference Proceedings

Tests used for diagnosis and surveillance of livestock diseases- aspects of kit validation; producer and end-user responsibilities

J. R. Crowther , H. Unger ,G. J. Viljoen

OIE Scientific and Technical Review, December 2006.

The Joint FAO/IAEA Programme of the IAEA in Vienna, Austria, has a long experience in helping to develop and validate assays and has provided strong support in developing OIE norms. This paper will focus on ELISA and PCR as the major technologies exploited in diagnosis and surveillance. Problems involving the terminology and factors in kit production, supply and

validation are examined, in particular emphasizing the importance of robustness and ruggedness of tests. The responsibilities for achieving quality controlled data to solve diagnostic and surveillance of producers, distributors, users and national and international organizations are discussed. The roles of internal quality control (internal proficiency testing) and external quality assurance (external proficiency testing) as well as aids to solving problems with kits are examined.

Publications by Project Counterparts

Detection of *T.b. rhodesiense* Trypanosomes in Humans and Domestic Animals in South East Uganda by Amplification of Serum Resistance-Associated Gene;

J.C.K. Enyaru, et al.;

Ann. N.Y. Acad. Sci. 1081:311–319 (2006)

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 John Crowther at j.crowther@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>

Information on New FAO titles:

To be regularly informed on FAO new titles, subscribe to FAO-Bookinfo, the free electronic newsletter from the FAO Sales and Marketing Group. Please send an email to mailserv@mailserv.fao.org leave the subject blank and then put in the first line of the message the following: Subscribe FAO-Bookinfo-L.

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>
- A training package to help artificial insemination (AI) technicians to improve the performance of AI and field services provided to farmers is now accessible from the AP&H Section website (http://www-naweb.iaea.org/nafa/aph/public/d3_pbl_1_10.html). It was produced under an IAEA Technical Cooperation Project – RAF/0/013 – ICT – Based Training to Strengthen LDC Capacity with the collaboration of the Animal Production & Health Section of the Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture. This package is also available as a CD ROM for users who have no access to internet connection.



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