



# Animal Production and Health Newsletter

JOINT FAO/IAEA DIVISION OF NUCLEAR TECHNIQUES IN FOOD AND AGRICULTURE  
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Dear Colleague,

As 1996 begins an opportunity is provided to look back and review our activities during 1995 and perhaps more importantly look forward and consider what lies ahead for the coming year and beyond. Without question 1995 was an historic year for the Division with the appointment of a new Director, Jim Dargie, and the subsequent changes in direction and tempo that this has gradually brought about.

Perhaps of greatest significance for the Division in 1995 was the development of a five- year strategy plan for each of its six Sections linked to the preparation of a far more transparent programme of work and budget for the 1997/98 biennium. The technical staff of the Animal Production and Health Section and the Animal Production Unit (APU) at the Agency's Laboratories worked together to focus with some clarity on our own collective programme. With the participation of all technical staff from both groups (AP&HS/APU) we spent a considerable amount of time evaluating the product of our efforts from the past ten years, and attempted to learn from our successes and failures about how we might better devise and implement new programmes that will best serve our counterparts in the less industrialized countries.

It would not be appropriate for us to include the full strategy statement in this Newsletter but I am sure you will get the flavour of its direction and objectives when reading the various activities detailed later. In essence though, we feel that much of the technology transfer needed for the effective use of radioimmunoassay and ELISA has now been completed and that we now need to provide support for the application of these technologies in a more focused manner to solve specific problems. We believe that cost-benefit analyses will provide data that point to the considerable social and economic advantage of solving specific problems affecting animal agriculture. These advantages should convince national and international authorities that it is essential to sustain the use of nuclear and related technologies to improve animal productivity and resolve animal health problems!

In supporting studies aimed at improving animal production we have concentrated on the identification of problems related to current national artificial insemination programmes and the development of effective systems for utilizing locally available feed resources to provide diets that will meet the needs for enhanced ruminant production. In the disease diagnosis programme we have concentrated only on the major diseases affecting livestock with the primary emphasis on support for monitoring and surveillance of these disease in on-going control and eradication programmes.

You will have grasped already that the new strategy does not represent a radical departure from our prior approach but rather is more of a shift in emphasis from the diagnostic technology to the disease and/or production problem that needs to be addressed. In developing our strategic plan however we did consider new areas for support. In particular, we felt that many of the breakthroughs in nucleic acid technologies were of tremendous relevance to specific problem areas in developing countries. These concepts were enforced by the requests received from many of you for support in these areas. Accordingly, in this issue of the NEWSLETTER is an announcement that we intend to develop a new FAO/IAEA Coordinated Research Programme concentrating on the use of the polymerase chain reaction for confirming the diagnosis of specific diseases such as rinderpest and

contagious bovine pleuropneumonia. More radically we have spent some time this year developing a possible programme in aquaculture which will concentrate on using micro-satellite DNA technology to develop an effective system for progeny testing on small-holder fresh-water farms.

Underpinning all these activities, and in many ways our most important emphasis, has been the development of an external quality assurance programme both for FAO/AEA ELISA kits and for RIA progesterone measurement. In the longer term we see this activity in the disease programme as part of the international move towards assay standardization and assurance that the assays are accurate and reproducible. This will result in wider acceptance of laboratory results that will be needed to facilitate international livestock trade under the new liberalized trade agreements such as GATT. During the next 2 - 3 years we hope that many of the national veterinary laboratories we assist will be able to gain our "recognition" status and thus play a vital role in international livestock trade programmes.

Despite the fact that we have spent a considerable amount of our time and energy on the development of this new plan, we have attempted to improve our current programmes by providing both comprehensive and appropriate support. We have provided technical backstopping to some 70 Technical Cooperation projects and over 65 Research Contracts and organized 5 Research Coordination meetings, 7 Training Courses and 3 Consultants Meetings. We have developed three new computer software programs and published a number of international articles on livestock production and health issues.

Staff changes within the Section have fortunately been fairly limited since the last Newsletter although we are pleased to welcome Dr. Amarjit S. Nanda to the Section as the technical officer responsible for Asia. Dr. Nanda replaces Oswin Perera and comes with an impressive pedigree of basic and on-farm research into the reproductive performance of a variety of indigenous breeds of cattle.

Finally, and we remain aware of this at all times, our primary function is to help you improve livestock productivity in your country. I hope that we continue to meet your needs and may I, on behalf of all of those involved in the animal production and health programme, wish you all the very best for the coming year.

With best wishes,

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**(B) FORTHCOMING EVENTS**

**(1) FAO/IAEA Regional Training Workshop on "Use of Immunoassay and Related Techniques for Studies on Animal Production in Africa", Radioisotopes Application Division, Egyptian Atomic Energy Commission, Cairo, Egypt, 13 - 30 April 1996**

This training workshop is being organized under the framework of the African Regional Co-operative Agreement for Research, Development and Training related to Nuclear Science and technology (AFRA). The objective of the course is to provide some basic and advanced information on nutrition and reproduction and their interactions in ruminant livestock, including practical training in radioimmunoassay for measuring the reproductive hormone progesterone and in feed evaluation techniques. The course is aimed at young scientists presently involved in national projects aimed at improving the productivity of ruminant livestock species. It is not aimed at those in managerial or administrative positions. Participants will be trained in the appropriate use and quality control of nuclear and other techniques to monitor livestock nutritional and reproductive indices. Emphasis will also be placed on the analysis and interpretation of research data. The course is open to 15 participants from AFRA Member States participating in AFRA Project VIII - Radioisotopes in animal reproduction and nutrition. The course will be conducted in English. The closing date for applicants was 15 January 1996 and all those that have applied for the course will be shortly notified if their applications was successful or not.

**(2) FAO/IAEA Coordinated Research Programme on the Seromonitoring and Surveillance of Rinderpest in Africa - Phase III**

From 1987 to 1993, this programme was funded by the Swedish International Development Authorities (SIDA). In early 1993, a proposal was presented to the European Union and it was anticipated that after 1993 the continuation of the programme would be funded through the EU. This would involve the establishment of a central epidemiology unit at PARC headquarters in Nairobi and the continued support for national diagnostic laboratories to conduct sero-monitoring and disease surveillance operated by the Animal Production and Health Section of the Joint FAO/IAEA Division. This has not yet materialised and the primary support for the sero-monitoring activities has been the support available under IAEA's programme of Technical Cooperation and the FAO "Empres" Programme.

We are now, however, confident that the programme can start in mid-1996. The funding was agreed by the EDF committee and the project documents were sent to Brussels for signature. Research contract proposals have been received from 21 countries and any PARC countries wishing to participate should apply immediately.

**(3) FAO/IAEA Interregional Training Course on the Use of Molecular Techniques (PCR, DNA probes) for the Diagnosis and Monitoring of the Major Livestock Diseases, IAEA Laboratory, Seibersdorf, 21 October - 15 November 1996**

**Participation:** The training course is open to 16 participants from developing Member States of FAO and IAEA.

**Background of the course:**

In developing countries diseases still sharply reduce livestock production. In the past 9 years, the main activity of the Animal Production and Health Section of the Joint FAO/IAEA Division was the development and transfer to developing countries of internationally accepted, validated and standardised enzyme-linked immunosorbent assay (ELISA) kits for diagnosing diseases, the monitoring of disease control programmes and for studying the epidemiology of diseases. New techniques like the polymerase chain reaction (PCR) and DNA probes are now increasingly used to complement the established diagnostic techniques for infectious diseases. The Animal Production and Health programme will increasingly support such molecular-based technologies.

**Purpose of the course:**

To introduce the concepts of the use of PCR and DNA probes in animal disease diagnosis and in the monitoring of disease eradication programmes. To provide practical training in these techniques. To provide training in the set up of a diagnostic PCR laboratory in a developing country.

**Participants' qualifications:**

Applicants should be veterinarians or senior technicians and intimately involved in the diagnostic work at the bench level. They should be experienced in laboratory techniques and have a working knowledge in serology and in the diagnosis of animal diseases.

**Nature of the course:**

The course will be of a practical nature supplemented through theoretical lectures. The lectures will deal with the basics of molecular techniques. Lectures will concentrate in particular on the routine use of diagnostic PCR and DNA probes in animal disease diagnosis. Information will be provided on how to set up a diagnostic PCR laboratory. The lectures will be followed by practical exercises on these techniques. The epidemiological lectures will concentrate on the use of these techniques in the monitoring of disease control programmes, their diagnostic performance and their limitations.

**Application procedure:**

Nominations should be submitted in duplicate on the standard application forms for training courses. Completed forms should be endorsed by and returned through the official, established channels (e.g. the Ministry of Foreign affairs, the National Atomic Energy Authority, or the office of the United Nations Development Programme). They must be received by 19 August 1996. Nominations received after this date or applications sent directly by individuals or private institutions cannot be considered. Completed and endorsed application forms may be submitted by facsimile.

It is suggested that advance information of the nominations is submitted by telex/fax with the following information: name, age, academic background, present position and full working address (incl. telex, telephone and facsimile numbers), to enable the IAEA to make a preliminary evaluation of candidates.

**Language certificate:**

In the case of countries in which English is not an official or working language, nominations must be accompanied by a separate certificate of the candidate's proficiency in English. This certificate must be issued by a language school, cultural institution or an embassy of a country in which English is spoken.

**Administrative and financial arrangements:**

Nominating governments will be informed in due course of the names of selected candidates and at that time, full details will be given of procedures to be followed with regard to administrative and financial arrangements. The IAEA will pay the full cost of the participants' air travel from their home countries to Vienna and return. During their attendance at the course, participants will receive from the IAEA a stipend sufficient to cover the cost of their accommodation, food and incidental expenses. The organisers of the course do not accept liability for the payment of any costs or compensation that may arise from damage to or loss of personal property, or from illness, injury, disability or death of a participant while he/she is attending the course, and it is clearly understood that each government, in nominating candidates, undertakes responsibility for such coverage. Governments would be well advised to take out insurance against these risks.

**(4) FAO/IAEA Regional Training Course on the Use of Immunoassay Technologies for the Diagnosis and Control of Foot-and-Mouth Disease in the East Asia and Pacific region, Animal Health Research Centre, Ho Chi Minh City, Vietnam, 4 - 22 March 1996**

**Participation:** The training course will be open to 15 participants from developing FAO and IAEA Member States in the East Asia and Pacific region.

**Background of the course:**

In Asia, foot-and-mouth disease is one of the most important diseases affecting livestock, and one major constraint to effective control at both the national and regional level is a lack of diagnostic capability within the countries of the region. In the past eight years, the main activity of the Animal Production and Health Section of the Joint FAO/IAEA Division has been the development and transfer to developing countries of internationally accepted, validated and standardized enzyme-linked immunosorbent assay (ELISA) kits for diagnosing diseases, the monitoring of disease control programmes, and for studying the

epidemiology of diseases. Such kits now include foot-and-mouth disease, and these are available for both the detection of the causative virus and the detection of an animal's response to natural infection or vaccination.

**Purpose of the course:**

The purpose of the course is to train scientists from the region in ELISA technology and in the use of the FAO/IAEA ELISA kits for the diagnosis and monitoring of control programmes against foot-and-mouth disease. Special emphasis will be given to quality control and trouble shooting. Furthermore, the course will provide training in data management in animal disease diagnosis using the ELISA and in the principles of sero-epidemiology in the monitoring of disease control programmes; and will demonstrate and explain the use of PCR in foot-and-mouth disease diagnosis.

**Participants' qualifications:**

Applicants should be experienced in laboratory techniques and have a working knowledge of foot-and-mouth disease serology. Applicants should be veterinarians or senior technicians and intimately involved in the diagnostic level at the bench level.

**Nature of the course:**

The course will be of a practical nature with emphasis on the use of the FAO/IAEA ELISA kits and epidemiological principles. Practicals in the use of the FAO/IAEA software will familiarize the participants with the use of computer software programs in animal disease diagnosis. Lectures will be given on immunology with particular reference to immunoassays (indirect, competitive and antigen capture ELISA). The epidemiological lectures will concentrate on the use of ELISAs in the monitoring of disease control programmes, the diagnostic performance of these test kits, and their limitations. The lectures will be complemented by practicals. PCR technologies for the diagnosis and study of foot-and-mouth disease will be introduced and demonstrated.

The closing date for applicants was 31 December 1995 and all those that have applied for the course will be shortly notified whether their application was successful or not.

**(C) PAST EVENTS**

- (1) First FAO/IAEA Research Coordination Meeting of Coordinated Research Programme on "The Use of ELISA for Epidemiology and Control of Foot-and-Mouth Disease and Bovine Brucellosis in Latin America", Rio de Janeiro, Brazil, 25 - 29 September 1995**

The meeting was held at PANAFTOSA, Rio de Janeiro, Brazil, and was attended by 10 Research Contract holders (5 Foot-and-Mouth Disease, 5 Brucellosis) from 8 Latin American countries, 5 Agreement holders, 3 Consultants and 3 Observers.



The Research Contract and Agreement holders presented progress reports on their respective research. Since this CRP is dealing with Brucellosis and FMD, the last two days involved presentations of results within a common group, whilst the discussion and elaboration of individual work plans were undertaken separately.

The brucellosis group has now tested around half of the samples needed for full validation of the competitive ELISA for separating vaccinated from naturally infected animals. It is concluded that this ELISA will detect antibodies to brucellosis caused by infection and not by vaccination in sera but that further validation is required. Results were discussed emphasizing statistical methods of assay validation using ROC (receiver operator characteristics) analysis. A common workplan for the final year of this CRP was agreed.

The programme originally aimed to validate a liquid phase blocking antibody FMD ELISA for detection of antibody to FMDV. This test would also detect antibody resulting from vaccination. It would be used to identify antibody-negative animals in geographical areas where the vaccine is not used and infection is absent. Therefore, it is considered to be a useful test for certification of FMDV status of animals. Due to a recent substantial change in the international trade regulations (OIE has approved a new category in the International Zoo-sanitary Code called "FMD-Free practising vaccination"), the need for a test which can discriminate between antibodies due to infection vs vaccination has become apparent but as of yet no internationally validated test exists. Having accepted this, it was still considered highly appropriate to complete validation of the PANAF-TOSA/Pirbright/FAO/IAEA liquid phase blocking antibody ELISA as the assay is essential in determining vaccination success and in countries that have ceased vaccination. Results obtained so far were discussed and a common workplan for the coming and final year of this CRP was agreed upon.

It was concluded that work under this CRP is progressing well and it is recommended that all Research Contracts be renewed for a further year. Some difference of opinion exists on the value of the current PANAF-TOSA/Pirbright/FAO/IAEA liquid phase blocking antibody ELISA, which does not discriminate between vaccinated and naturally infected animals. Given the lack of any viable alternative at present, it is recommended to complete the agreed validation of the original assay (with some agreed modifications). The final RCM of this CRP should take place at the IAEA in Vienna in early 1997.

(2) **FAO/IAEA Interregional Training Course on "Improving the Productivity of Ruminant Livestock through 'On-farm' Assessment of Nutrition - Reproduction Interactions Using Nuclear and Related Techniques". Seibersdorf, Austria, 25 September - 3 November 1995**

A total of 177 applications were received for participation in this course. This reflects the increasing interest in an integrated multidisciplinary approach to solving constraints limiting the productivity of livestock in developing countries. It is foreseen that future FAO/IAEA activities on improving animal production will adopt this multidisciplinary approach and focus on nutrition-reproduction interactions.

It was a difficult task to select the course participants among this large number of applicants. Eventually, 21 participants from 21 countries were chosen. They were: Mr. M. Musanti (Argentina), Ms. I. Petrova (Bulgaria), Mr. Zhang Degang (China), Ms. M.T. Graviria-Scioville (Colombia), Ms. G. Fukuda Suzuki (Honduras), M. J. Bestari (Indonesia), Ms. R. Muinga (Kenya), Mr. R.L. Rasoloarison (Madagascar), Mr. M.S. Kumwenda (Malawi), Mr. Z. A. Mohd Jelani (Malaysia), Mr. P. Toolsee (Mauritius), Mr. C. Lamothe Zavaleta (Mexico), Mr. D.R. Khanal (Nepal), Mr. S.M.M. Siddiqui (Pakistan), Mr. C. Gomez (Peru), Mr. P. Nowakowski (Poland), Ms. K. Ahmed (Sudan), Mr. A. Swaid (Syria), Mr. S.L. Solomona (Western Samoa), Mr. A. M. Al-Mulsi (Yemen) and Mr. J. Mwenya (Zambia).

The course lasted for six weeks and covered monitoring of reproductive performance "on-farm", evaluation of feeds and nutritional status of livestock "on-farm", nutrition-reproduction interactions, project design and computerized analysis of animal production data. Practicals were held on blood and milk sample collection and processing, radioimmunoassay (RIA) for the measurement of progesterone using FAO/IAEA RIA kits, the measurement of blood metabolites and minerals and the computerized analysis of animal production data. At the end of the course, each participant presented a research proposal that reflected the subjects covered during the course. Field trips were organized to the experimental dairy and sheep farms of the Veterinary University of Vienna, the Department of Nutrition of the Veterinary University of Vienna, The Federal A.I. Institute in Wels, an Oilseedmill in Aschach and dairy farms in Styria, Salzburg and Upper Austria. Lectures and practicals were given by FAO/IAEA staff and outside lecturers to provide a high level of expertise in individual subjects. The outside lectures were from both developed and developing countries.

The course was certainly successful, as proven by the positive evaluations, enthusiasm and hard work of the participants and the lecturers and the high calibre of the research proposals presented. We would particularly like to thank Dr. R. Tagle (Uruguay), Dr. O. Perera (Sri Lanka), Dr. B. McBride (Canada), Dr. F. Sundstøl (Norway), Dr. E. Bamberg (Austria), Dr. F. Wittwer (Chile), Dr. J. Leibetseder (Austria), Dr. K. Buchtela (Austria), Dr. M. Bryant (UK), Dr. R. Wetteman (USA), Dr. E. Pérez Guitérrez (Costa Rica), Dr. F. Oberlehner (Austria) and Dr. Fischerleitner (Austria) for their contribution as lecturers during the course. We would like to express our gratitude to all those involved in the course for their enthusiastic inputs and collaboration.

**(3) FAO/IAEA Regional Training Course on the Use of Immunoassay and Molecular Methods for Animal Disease Diagnosis and Control, Rabat, Morocco, 9 October - 3 November 1995**

The training course was held at the Laboratoire Officielle de Contrôle des Médicaments et Sérums Vétérinaires, Rabat, Morocco. It was attended by 22 participants from 17 French speaking African countries. They were: Mr. L. Gilbert Aplogan (Benin), Mr. L. M. Millogo (Burkina Faso), Mr. Jean-Paul Yomo (Cameroon), Mr. G. Kondolas-Oualybangah (Central African Republic), Mr. D. Bikinkita (Congo), Mr. M. Sadia and Mr. J. Kassi Kassi (Côte d'Ivoire), Mr. Z. Belew (Ethiopia), Mr. I. Pita Bah (Guinea), Mr. H. Andriamboavonjy Ralaivoavy (Madagascar), Mr. M. Diallo (Mali), Ms. H. Uteene (Mauritius), Ms. B. Mata (Mozambique), Mr. B. Yaou (Niger), Mr. M. Mbacké Seye (Senegal), Ms. R. Ben Osman (Tunisia) and Mr. N. A. Egala (Zaire). Local participants were: Mr. M. Abbadi, Mr. F. Oumellouk, Mr. A. Mansouri, Mr. M. El Maadoudi and Mr. B. Harif. The majority of the

lecturers also came from the region. They were: Dr. E. Couacy-Hymann (Côte d'Ivoire), Dr. K. Tounkara (Mali), Mrs. M. Diop (Senegal) and Dr. D.McKay (United Kingdom).

The purpose of the course was to train scientists in the use of the FAO/IAEA ELISA kits for the diagnosis of brucellosis, foot-and-mouth disease and contagious bovine pleuropneumonia. Formal lectures were given on basic immunology, enzyme immunoassays and on aspects on the use of computers for data collection from an ELISA reader.

The number of different ELISAs demonstrated emphasized the flexibility of the technique but also familiarized the participants with problems that can arise. During the final week, lectures and practicals on DNA probes were given to introduce molecular techniques. During the last days, sensitivity and specificity of tests were determined and sample sizes and sampling frames were discussed to put the diagnostic techniques learned earlier into a practical context. The local arrangements for the course were excellent and we would like to thank the staff of the laboratory for their assistance.

(4) **First Research Coordination Meeting of the FAO/IAEA Coordinated Research Programme on "Improvement of Ruminant Livestock Productivity in Developing Countries through the Use of Progesterone RIA to Increase Efficiency and Quality of Artificial Insemination Services", Vienna, 13 - 17 November 1995**

The meeting was held at the Vienna International Centre and was attended by 14 Research Contract holders, 4 Agreement holders, 1 Technical Contract holder, 1 consultant and 2 observers. The Contract holders came from 7 Asian and 7 Latin American countries whilst the advisers were from a wider geographic distribution. During the meeting, the participants presented the tentative work plans and, in a few cases, preliminary results. Group discussions were held on the structure, layout and possible additions to the FAO/IAEA developed computer program AIDA (Artificial Insemination Database Application). A full afternoon was dedicated to practical work on the different ways of entering data and displaying the results in the computer application. Contract holders were assigned to specific Agreement holders for more detailed discussions of their one-year survey protocol. The Second RCM will take place in early 1997, probably in Southern Australia.

It was understood that these studies on the artificial insemination service can be undertaken in many ways; however, the best way to interchange information and compare results between countries is to work to standard protocols. Thus an FAO/IAEA work plan was thoroughly discussed, slightly amended and finally agreed. The Research Contact holders are committed to the protocols set out in the AIDA program for the one-year survey. This includes methods of sample collection, preservation and analysis, and the use of AIDA field forms for data collection. The uniform working protocol will highlight the major constraints and allow the implementation of corrective measures during the second phase of the programme, after the second RCM.

The main conclusions and associated recommendations arising from the meeting were as follows:

**Conclusions:**

- (a) There was an initial disparity between tentative research work plans of the Research Contract holders and the objectives and suggested methodology of the CRP.
- (b) There were inconsistencies in the interpretation of the methods of sample collection, preservation, and analysis; in the understanding of certain fertility indices used in the programme; and in the definition of quality of semen used.
- (c) The value of the AIDA program was not uniformly appreciated and this was reflected in the inability of Contract holders to use this tool to implement their data processing.
- (d) Valuable information relevant to the purpose of the survey which was not part of AIDA was identified.
- (e) There is a danger that research other than the survey may divert Contract holders from original objectives during the first phase of the programme.
- (f) The meeting allowed valuable discussions resulting in agreement on a uniform work plan and on the use of AIDA for data entering and preliminary analysis.

**Recommendations:**

- (a) The protocols set in the AIDA program should be rigorously followed. This includes the methods of sample collection, preservation and analysis, and the use of AIDA field forms for data collection.
- (b) The Research Contract holders have to be committed to the AIDA survey and its complimentary study in oestrus detection in post-partum cows as a first priority in their research activities.
- (c) The AIDA program should be modified to include the parameters identified at the meeting and the upgraded version distributed as soon as possible.
- (d) Whenever possible, resources should be provided to evaluate the quality of semen.
- (e) Inseminators should be monitored according to the procedures recommended in the AIDA protocol without advising them of possible errors.
- (f) The field survey should be concluded well in advance of the next scheduled RCM to allow proper analysis of the data.
- (g) It is essential that frequent communication occurs between Research Contract holders, Agreement holders and the FAO/IAEA scientific secretary to ensure the success of the programme.

(5) **FAO/IAEA/ILRI Workshop on the Diagnosis of Tick-borne Diseases Using Immunoassay Methods, International Livestock Research Institute, (ILRI), Nairobi, Kenya, 6 November - 1 December 1995**

Tick-borne diseases (TBD) are a major constraint to livestock production globally. In East and Southern Africa East Coast Fever (ECF) causes considerable losses not only due to animal mortality and reduced productivity but also due to the costs of control programmes which rely largely on the prevention of tick infestation by application of acaricides. In the past, the use of ELISAs in the diagnosis of TBD was impeded through the low specificity of the ELISA but through the use of recombinant antigens developed at ILRI, the sensitivity and specificity of the ELISA has been considerably increased. The ELISA now offers many advantages over conventional diagnostic techniques in the monitoring of TBD control programmes.

This workshop provided the participants with ELISA training in the diagnosis of TBD using ILRI developed reagents and it marked the start of a new programme of support on TBD. The ELISA kits for TBD - at present under validation - will become available in the near future in the form of FAO/IAEA/ILRI ELISA kits.

The workshop was attended by 11 participants, eight participants from Africa being supported through IAEA and 3 participants supported through ILRI. The majority of the lecturers came from ILRI. The course concentrated on the ILRI developed ELISAs for the detection of antibodies to *Theileria parva*, *T. mutans*, *Anaplasma marginale*, and *Babesia bigemina*. It included also lectures and practicals on the use of the FAO/IAEA ELISA software and its use in animal disease diagnosis. Epidemiological lectures were given on the use of ELISAs in the monitoring of TBD control programmes, the diagnostic performance of these test kits and their limitations.

The local arrangements both for the course itself but also for the accommodation were excellent and we would like to thank the staff of ILRI for their enthusiastic support throughout the course.

(6) **FAO/IAEA Consultants Meeting on "The Application of Molecular Techniques in Animal Disease Diagnosis in Developing Countries", Vienna International Centre, 20-23 November 1995**

The polymerase chain reaction (PCR) technique has become one of the most widely used techniques of molecular biology, and for good reason; it is a relatively rapid and accurate means of producing microgram amounts of DNA from minute quantities of source material (in extreme cases from a single cell or organism) and can be used on samples which have deteriorated beyond levels suitable for other research or diagnostic techniques. Application of the PCR technique for animal disease diagnosis in developing countries has been attempted by a number of individuals for a variety of purposes with inconsistent results. The technique has the potential to augment on-going and future surveillance, control, and eradication programmes, and the demand for this technology from FAO and IAEA Member States has been high. In order to explore the application of PCR and related molecular techniques to animal disease diagnosis within the context of present and future FAO/IAEA programmes, a Consultants Meeting was held to achieve the following objectives:

- Review the applications of PCR in animal disease diagnosis.
- Determine the applicability of PCR in developing countries.
- Determine the general requirements for establishing and sustaining PCR in developing countries.
- Identify the animal diseases appropriate for application of diagnostic PCR in developing countries.
- Determine the research and development needs required for transfer of specific PCR diagnostic technology to developing countries in terms of training requirements, reagents supply, and/or kit production.
- Make recommendations with regard to the implementation of an FAO/IAEA Coordinated Research Programme in this area.

The consultancy team consisted of a group of scientists with a variety of animal disease and molecular biological backgrounds in developing as well as developed countries. They were Dr. Tom Barrett (United Kingdom), Dr. Adama Diallo (France), Dr. Steven Edwards (United Kingdom) and Dr. Karl-Erik Johansson (Sweden).

The conclusions and recommendations of the Consultants were:

- (a) There are specific applications of the polymerase chain reaction (PCR) and related molecular techniques in animal disease diagnosis which can be defined and for which there are needs in developing countries.
- (b) Development and transfer of this technology should be accomplished on a selective basis taking the following into account:
  - (1) Will the technology be useful to specific recipients?
  - (2) What is the comparative cost/benefit in relation to other diagnostic techniques?
  - (3) What is the probability of success and subsequent sustainability of the technology?
- (c) Facilities and equipment for diagnostic PCR are expensive and must be dedicated. Sites for transfer of this technology in developing countries must be evaluated, and the potential recipients' contribution determined, before transfer occurs.
- (d) Rinderpest, Peste des Petits Ruminants and Contagious Bovine Pleuropneumonia are three diseases of importance to developing countries for which PCR diagnostic techniques have been developed to a level suitable for standardization and transfer. Trypanosomiasis, tick-borne diseases, foot-and-mouth disease, and important avian and swine diseases are areas which require further research, but should be kept in mind for extensions of this effort.
- (e) Diagnostic kits should be developed, but rigorous training and external quality assurance programmes will be the principal determinants of success in this effort.
- (f) Cost/benefit analyses should be used to help recipient countries understand the importance of sustaining the transferred technology after FAO/IAEA support is terminated.

(g) An FAO/IAEA Coordinated Research Programme (CRP) should be established to initiate the standardization and transfer of this technology, and its subsequent use for confirming the diagnosis of rinderpest and contagious bovine pleuropneumonia.

(h) Rigorous general and disease-specific training in molecular biology was identified by the Consultants as the most important component affecting the success of this technology transfer. Another important factor identified was the ability of a participating laboratory to provide all of the facilities (dedicated areas, constant electrical supply, etc.) and most of the basic equipment necessary to the use of the PCR technique for disease diagnosis. Therefore, it was recommended that the establishment of an FAO/IAEA Coordinated Research Programme and subsequent Technical Cooperation Projects in this area be linked logistically with evaluation of current facilities and equipment, and with intensive training of the staff members at the participating laboratories. It was considered pointless to assist a laboratory in achieving the physical capability of performing diagnostic PCR without having several permanent employees of that laboratory who are thoroughly trained in the general field of molecular biology and the disease-specific diagnostic technique.

(i) Initially, it is envisaged that standardization of diagnostic kits would occur as a combined effort between the IAEA Laboratories at Seibersdorf, the appropriate reference and training centers, and the participants in the FAO/IAEA Coordinated Research Programme. Provision of the necessary equipment and diagnostic kits to the developing country diagnostic laboratories would occur only after the disease-specific molecular diagnostic training of participating laboratory staff is well underway or has been completed.

Subject to available funding, it is proposed to initiate a new FAO/IEAE Coordinated Research Programme during 1996 based on the recommendations given by the consultants. The guidelines for this Programme will be announced in the next Newsletter.

#### **(D) STATUS OF EXISTING COORDINATED RESEARCH PROGRAMMES**

##### **(1) Development of Feed Supplementation Strategies for Improving the Productivity of Dairy Cattle on Smallholder Farms in Africa**

The Programme has 14 Research Contracts and 5 Research Agreements and no further awards can be considered. The Second RCM will take place at the Institut Agronomique et Vétérinaire Hassan II, Rabat, Morocco, from 1 - 5 April 1996.

##### **(2) Development, Standardization and Validation of Nuclear-based Technologies for Measuring Microbial Protein Supply in Ruminant Livestock for Improving Productivity**

Based on the FAO/IAEA Consultants Meeting held at the Vienna International Centre, Austria, from 22-24 May 1995, a new FAO/IAEA Coordinated Research Programme on the above topic is being initiated as from 1996. It is anticipated that 8-10 Research Contracts and Research Agreements will be awarded in the coming months. The first RCM of the programme is being planned for July/August 1996 at a venue still to be decided.

**(3) Use of Immunoassay Methods for Improved Diagnosis of Trypanosomiasis and Monitoring of Tsetse and Trypanosomiasis Control Programmes in Africa**

This programme has 15 Research Contracts and 4 Research Agreements and no further awards can be considered. The third RCM is being planned for September 1996 in Dakar, Senegal.

**(4) Development of Supplementation Strategies for Milk producing Animals in Tropical and Sub-tropical Environments through the use of Nuclear and Related Techniques**

The programme has 16 Research Contracts and 6 Research Agreements and the final RCM is planned for early 1997.

**(5) Improving the Diagnosis and Control of Foot-and-Mouth Disease in South East Asia Using ELISA-based Technologies**

This programme has 9 Research Contracts and 3 Agreements. A Technical Contract has been awarded to the World Reference Laboratory, Pirbright, UK, for the supply of FMD reagents. The 2nd RCM is planned to be held in the region in early 1997.

**(6) The Use of ELISA for Epidemiology and Control of Foot-and-Mouth Disease and Bovine Brucellosis in Latin America**

This programme has 5 Research Contracts dealing with Foot-and-Mouth Disease and 5 dealing with brucellosis. There are 4 Research Agreement holders in the programme. The final RCM will take place at the VIC, Vienna, in early 1997.

**(7) Improvement of Ruminant Livestock Productivity in Developing Countries Through the Use of Progesterone RIA to Increase the Efficiency and Quality of Artificial Insemination Services**

This programme has 12 Research Contracts. The award of two further Research Contracts, three Research Agreements and a Technical Contract is pending. The Second RCM will be held in early 1997.

**(E) DEVELOPMENTS AT THE ANIMAL PRODUCTION UNIT, SEIBERSDORF**

**1. General**

In the last NEWSLETTER, you were informed that Peter Wright, who was Unit Head for the last 5 years, returned to Canada in July to the new National Centre for Diagnostic Virology in Winnipeg, Manitoba. During the interim period, a search for a new Unit Head has been ongoing. Dr. R. Jacobson of New York State, USA has been serving as Acting Head of the Unit while on sabbatic leave from Cornell University and his 6-month term will end on February 15, 1996. Dr. Jacobson writes that in addition to the usual administrative work, it has been challenging and enjoyable to work with officers and staff of the Unit and Section



toward forging a team effort in meeting the needs of our counterparts. I hope that a few new ideas will emerge that will eventually make a difference... for you.

We have been involved in some new initiatives. As a result of a consultant's meeting held in Vienna in November, the Unit/Section has solidified a new thrust into nucleic acid technologies that will emphasize training of our counterparts. We've been aided considerably in this new endeavor by the addition of Dr. Mark Robinson to the Unit who comes from the United States Agriculture Research Service Laboratory in Pullman, Washington. He has been outfitting a Biotechnology Laboratory in the APU. He, along with Roland Geiger from the Section, were instrumental in organizing the Consultant's Meeting and Mark has finalized a report which is summarized in Section C of the NEWSLETTER. We are grateful that Mark is on the scene to supply needed expertise, ideas, and perspective.

We have been rethinking the ways in which the Unit and Section may provide more effective backstopping for technical problems and strategies that should help developing countries continue their efforts toward improved animal productivity after TCPs and RCMs have run their course. Sustaining the initiatives that have been made over the last decade through the FAO/IAEA programmes is a major concern and challenge. We think that with the advent of international trade agreements, laboratory testing proficiency will become a major issue for developing countries. When the demand for diagnostic services increases, the support for them will also increase. What may be perceived as a problem in trying to meet these new needs should be seen as an opportunity. We are, therefore, actively working on new ideas that will help you to get ready for the challenge; these somewhat conceptual and futuristic thrusts have been both interesting and challenging. Keep an eye out for more on these subjects in later issues of the NEWSLETTER...

We are moving into a Quality Assurance Programme for the Seibersdorf Unit that will include all of the elements of "Good Laboratory Practice" (GLP). We are developing Standard Operating Procedures for every component of our operation and plan to eventually offer these "SOPs" to those of you who are interested in meeting international standards that will allow your laboratories to become licensed. We also are continuing in our effort to emphasize and expand the External Quality Assurance Program for our counterparts under the direction of Dr. Barbara van der Eerden. The EQAP recognizes laboratories for proficiency in conducting tests. Details of this programme are given later in this section of the NEWSLETTER.

With a leading role of Mario Garcia from the Section, who has developed a data base for all research and technical cooperation projects supported by the Section/Unit programme, we are creating new segments for that data base that will assist us in keeping contemporaneous records of all requests for kits and supplies, and will also help us to track all shipments to counterparts. It should improve our efficiency and accuracy in dealing with exactly what you requested.

We are continuing to fulfill our role as an OIE Collaborating Centre for ELISA and Molecular Techniques in Animal Disease Diagnosis. Dr. van der Eerden represented the laboratory at the biannual meeting of the OIE Standards Commission in Paris, in September. The OIE is recognized by the World Trade Organization as an international standard-setting body that provides guidelines and standards for health regulations applicable in the

international trade of animals. Our advisory role will become more important as we provide recommendations for standards and guidelines. Accordingly, we are producing a chapter for the next edition of OIE's "Manual of Standards for Diagnostic Tests and Vaccines". The chapter, entitled "Guidelines for Validation of Diagnostic Assays for Infectious Diseases", will attempt to identify the problems with validating assays and provide suggestions for the various approaches that may be relevant to your situation.

The next part of the NEWSLETTER discusses two examples of work on-going at the Unit in cooperation with Technical Officers from the Section and with our external advisors. Although we are currently in a transition phase in the Unit and so are at a low ebb in personnel numbers, we are working toward enhancing our applied research efforts. In addition to the two examples given below, we have been identifying short-term and long-term projects that will clarify and enhance the diagnostic capacity of test kits and services we provide. An example of a much needed study is whether there is a standard way that pipette tips can be cleaned for reuse that will not increase the variability in the assay. Another study needs to be done to determine whether a highly controlled cleaning procedure can be developed to allow reuse of microtitre plates. This kind of practical, applied research is directed at cutting costs so as to enhance sustainability of the assays you have been using under CRPs and TCPs.

Although I will not be here to contribute to the next issue of the NEWSLETTER, I look forward to watching the development of (and possibly contributing from a distance to) the future FAO/IAEA programmes. It is an important time for the Unit and its team effort with the Section. The selection of the new Unit Head, which should occur in the near future, will be pivotal in bringing creative and aggressive leadership to the Unit. We need to have your feedback on how we can better provide services to your programme. Let us know what you are thinking in this regard. We will be very pleased to receive your ideas.

## **2. Applied Research and Service Functions**

### **2.1 *Trypanosomiasis Antigen Capture ELISA***

An *in vitro* production system for routine bulk production of monoclonal antibodies is being established at the APU for use in the trypanosomiasis antigen capture ELISA. Various minor changes to the assay were initiated in order to increase the specificity and sensitivity of the test. The following assay adjustments have been made:

- (1) the use of high binding capacity plates;
- (2) constant shaking for 15 minutes at 37°C;
- (3) dilution of control and test serum samples;
- (4) addition to the serum diluent buffer of 0.5% normal mouse serum as a liquid phase blocking agent;
- (5) addition to the conjugate diluent buffer of 0.1% bovine serum albumin as a liquid phase blocking agent;
- (6) use of tetramethylbenzidine chromogen and hydroxide peroxide;
- (7) use of non-corroixive 1M phosphoric acid solution as a stopping solution;
- (8) incorporation of standardized internal quality control samples such as a strong positive, moderate positive, and negative antigen control as well as a conjugate control;

- (9) expression of results as percent positivity of the strong positive antigen control, and
- (10) use of a specialized computer software program developed for the ELISA kit.

The revised version of the Ag-ELISA has been delivered as a ready-made kit to the Research Contract holders in the various countries in Africa. The changes made to the test to improve sensitivity and specificity were presented at the British Society for Parasitology Seminar on Trypanosomiasis and Leishmaniasis held from 3-6 September 1995 in Glasgow, United Kingdom. Studies will be continued to improve the sensitivity of the test without jeopardizing the excellent specificity.

#### 2.2. *Addressing a Specificity Problem in the Indirect Brucella Assay - Example of on-going applied research*

It is our responsibility to work toward resolution of unexpected problems that arise from time to time in FAO/IAEA kits. We were informed by the counterpart of a Technical Cooperation Project that they were unable to achieve an appropriate level of agreement between the ELISA and Complement fixation-confirmed Rose Bengal test results. This is a highly unusual finding that was confirmed at our laboratory. We are in consultation with Dr. Klaus Nielsen about the problem and hope to find an explanation. These are the kind of inputs from you that help us to assure the validation of the assays provided by the FAO/IAEA. Local conditions, including different infectious agents that are unique to your indigenous population of cattle, may cause such cross reactivities in ELISA that would not be expected. You should always be on the lookout for such unusual findings and report them to us so we can seek a resolution.

#### 2.3. *Changes in the RIA kit for Progesterone*

Recently, a new shipment of progesterone RIA kits went to counterparts. The manufacturer of our kits, DPC, had changed some of the kit components which required that the assay be recalibrated at the laboratory in a combined effort with Section and Unit personnel. The new kit has antibody and tracer that are different from previous kits, the tracer is colourless, and overnight incubation causes higher maximum binding (i.e. up to 70%). Adequate incubation time for the new kit is thus 3-4 hours at room temperature. As a consequence of this, all antibody-coated tubes you may have been accumulating from previous kit shipments are no longer useful because when used in conjunction with the new tracer, they will render wrong results. All old antibody-coated tubes should thus be discarded. An updated manual is being prepared to include the new assay characteristics. A copy of it will be included in the February shipment of kits.

#### 2.4. *International Field Trials for the Brucellosis Competitive ELISA and the FMD ELISA*

The APU has been involved with advisors from Canada, Italy, the UK and PANAFOSA in Brasil in assisting counterparts in their work to validate these new kit formats. A report of the recent RCM in Rio de Janeiro, in which progress on these projects was reviewed, is provided elsewhere in the NEWSLETTER. Please refer to the report for an update.

### **3. Quality Assurance Programmes**

A lot of work for nothing or a little work for a lot of return?

If you buy a car that promptly quits running, you have reason to be upset with the car manufacturer. If your physician makes a misdiagnosis, resulting in an unnecessary treatment, you may be much more upset. If you are a farmer and a state laboratory uses ELISA technology to misdiagnose your herd resulting in depopulation of the herd (and depletion of your bank account), you may be furious. In every case when errors are made, whether in manufacturing or in diagnostics, the client suffers. To avoid such errors, quality control is being implemented at all levels in society to give assurance to clients that the product you purchased or service you received is first-rate.

#### **3.1. Why my laboratory?**

But you say, "I run a good laboratory, I follow the exact procedure in the FAO/IAEA ELISA manual, my internal quality control is adequate (the assay controls run within the upper and lower control limits), and the reagents are stored properly, so this doesn't apply to me." Are you sure? Any laboratory in industrialized countries that is working on products for human use must comply with quality assurance principles or their work is not accepted. The reason is that there are many instances where small and even imperceptible changes in how things are done in the lab can have a considerable affect on a assay. The only way to demonstrate that such problems do not occur is to have a certified laboratory. In the current atmosphere of international trade agreements involving animals and animal products, you will eventually be held accountable for the results you provide so it is in your best interest to enroll in a system that will help you to avoid such a problem. It is a scientific and practical imperative that has become reality for all progressive laboratories world-wide.

#### **3.2. How can it help my laboratory?**

Involvement in a Quality Assurance Programme has many positive implications for a laboratory. It gives the director of that laboratory assurance that she/he is providing the best possible diagnostic service. It tells national officials that laboratory is using objective, external criteria for evaluating its proficiency and that it can compete with the best in the world. It also assures interested international investors and traders that the laboratory is providing results that are recognized by relevant authorities.

International trade in livestock and livestock products is increasing and the animal health regulations are becoming stricter. They are being set by your Government, or your neighbour's Government, and by the World Trade Organization (WTO), the Office International des Epizootics (OIE), the European Union, and the Americas, etc. These regulations are making it mandatory that internationally accepted Quality Standards be established for all the activities concerning livestock and livestock products. Suppose your country is trying to control or eradicate a disease. The moment your country declares itself free of a disease, or it falls under some other classification in International Animal Health Code, you can be assured that the reliability of your testing laboratory will be called into question. That is exactly why the FAO/IAEA initiated an External Quality Assurance Program (EQAP); to help you meet the standards that are going to be required of your laboratory and to provide documentation that states that your laboratory results are valid.

### **3.3. What is involved in QA programmes?**

A Quality Assurance Program for your laboratory includes not only proficiency testing such as we are sponsoring through the EQAP (see also the Newsletter of July '95), but also includes many other elements that help laboratories assure that their work is done by international standards. The guidelines for a total quality assurance program for laboratories have been developed by the International Organization for Standardization (ISO). This is a worldwide federation of organizations that deal with standards, representing 111 Member States. The mission of the ISO is to promote the development of standardization and related activities in the world in order to improve and facilitate the international exchange of goods and services. They have published several documents that define all elements of standardization and good laboratory practice. One guide (ISO Guide 25) outlines "General requirements for the competence of calibration and testing Laboratories." The basic message in this guide is that all activities within a laboratory need to be documented. We will send you a copy of this guide early in 1996.

### **3.4. Are any Research Contract or Technical Cooperation Project holders involved in Quality Assurance Programmes?**

The answer is yes. Under these programmes, participants receive a panel of unknown samples which is tested and the results, which are kept confidential, are evaluated in Vienna and then shared with the participant. Scientists using the FAO/IAEA RIA kits have been involved in proficiency testing for several years.

Since 1991, the rinderpest network has operated an successful External Quality Assurance Programme in Africa.

In October 1995, an expanded External Quality Assurance Program for the rinderpest competitive ELISA was initiated in 20 African laboratories. An External Quality Control (EQC) serum test panel was sent to participating laboratories together with a questionnaire. Also, we requested the participants to return the internal quality control (IQC) data from the computer that acquired the data from previous runs of the rinderpest ELISA test kit (the data acquired by the computer is transferred by the participant to a floppy disc that was provided).

An identical EQAP for 34 laboratories, using the Agency's Brucella indirect ELISA test kit in Africa, South America and Asia, was started in November, 1995.

### **3.5. What will be done with the data?**

The data are totally confidential and any reports that compile data from many laboratories will **not** identify the participating laboratory! As soon as all results from the EQC serum test panel, the questionnaire and the IQC data are returned to the Agency for a given EQAP round, Dr. van der Eerden will prepare a interim report and will distribute it to all participants. After the completion of two succesful rounds of the EQAP, the individual laboratories will receive recognition for their competence in performing the ELISA.

### **3.6. What next?**

The second dispatch for the EQAP for rinderpest is scheduled in March / April. The WAREC countries will also participate. An interim report of the results of the first dispatch of October 95 will be sent to you. The second dispatch of the EQAP for Brucella is scheduled in May 1996. As soon as the results of the first dispatch are processed and analysed you will receive a interim report. A third EQAP for the Foot and Mouth Disease ELISA (antibody and antigen detection) will start in June/July '1996. The laboratories in receipt of these kits will receive information before the actual dispatch. The EQAP for the trypanosomosis ELISA will start later in 1996.

### **3.7. Maybe you are thinking: "I'm not much interested in this extra work"!**

Please come to a full stop and think about the consequences (and maybe reread the first part of this article). If you don't send in your results, not only are you affected but everyone in the programme is affected! We cannot help you or your colleagues if you don't send in the questionnaires, your results from the test panel, and your internal quality control data. We need your data for comparisons with the data from all other labs; without that, we cannot assess the efficiency of any laboratory. So we urge you to send us your results in a timely manner. In case you can foresee you will be unable to return the results on time, could you please inform us. If you don't think this is important, it might be advisable to reread the first part of this article. It really is important to you. So, we are ready to work with you and assist you. Your responses are thus critical to the success of the programme.

### **3.8. Comments or suggestions?**

If you have any comments, suggestions or questions, do let us know. The EQAP is open for constant improvement. Also if you have relevant information on Quality Assurance that is of importance to your colleagues, feel free to send it to us for further distribution.

**Thank you!**

To all of you who are responding to the EQAP, thank you very much! It really is just a little work with a considerable return on your investment.

## **(F) PUBLICATIONS**

### **Printed**

"Improving Animal Productivity by Nuclear Techniques", Garcia, M., Jayasuriya, M.C.N., Perera, B.M.A.O., IAEA Yearbook 1995.

"Annual Rinderpest Sero-monitoring Results throughout Africa (Phase II) 1994".

### **In Press**

"Development of Feed Supplementation Strategies for Improving Ruminant Productivity on Smallholder Farms in Latin America through the Use of Radioimmunoassay Techniques", IAEA-TECDOC.

## **In Preparation**

"Immunoassay Methods for the Diagnosis and Epidemiology of Animal Diseases in Latin America". Proceedings of the Final RCM of the SIDA-funded CRP.

## **(G) ANIMAL PRODUCTION & HEALTH SOFTWARE PROGRAMS**

### **1. SID Version 3.0**

Due to unforeseen difficulties in the programming within EPI-Info 6.0, we decided to reconsider alternative computerised database management options. Following a great deal of discussion within the Animal Production and Health Section and several experts in the field of epidemiology, (a database package which can be used for the various diseases), it was decided to use Access® for Microsoft Windows®.

Access® is designed such that one has unparalleled access to data, combined with the ease-of-use made possible with Windows. Unlike EPI-Info Access® is not an epidemiological package as such. Some of the features available in EPI-Info, e.g. sample size calculator are not available in Access®. The great advantage of Access®, however, over EPI-Info is that the interface can be made very user friendly. To make full use of all the features available one does need to be computer literate. As in SID 1 and 2 the program produces standard reports with the relevant data compiled.

By the end of 1995, the first version of the sero-monitoring database will be sent out to a few laboratories within the PARC programme for evaluation. With the recommendations and comments accrued, a final version of SID 3.0 will be made available by the beginning of next year. Following this a comparable database will be developed for the Disease and Serological Surveillance as part of the Global Rinderpest Eradication Campaign. Midway through next year programs should become available for other diseases such as FMD, Brucellosis, etc. It is foreseen that all these databases will be linked to a serumbank database and that results for the different diseases can be compared.

Please contact Dr. Wicher Holland, Animal Production and Health Section, IAEA [Fax: 43 (1) 20607, E-mail: holland@ripo1.iaea.or.at, Phone 43 (1) 2060 26056] for further information.

### **2. Database for Artificial Insemination Services - AIDA**

The database has been primarily designed for project counterparts of the FAO/IAEA Coordinated Research Programme on "Improvement of Ruminant Livestock Productivity in Developing Countries through the Use of Progesterone RIA to Increase Efficiency and Quality of Artificial Insemination Services" to store data and provide reports related to the Artificial Insemination Service given by a particular AI Centre, AI or farmer's cooperative.

Data of farms, cows, semen and AI technicians are stored and partially analyzed to serve as a tool to evaluate the quality of the AI Service and to identify constraints that may be hampering the fertility. Also, it serves to correlate progesterone concentrations in milk/blood samples with fertility and to facilitate data interpretation. The software includes a useful User's manual to facilitate the installation procedure and to guide users on data entering, editing and printing reports.

The database can easily be used by anyone who conducts or is in charge of an AI Service in cattle, buffalo and to some extent in sheep. It is not suitable for other livestock species because of different reproductive patterns, especially in the ovarian activity and oestrus behaviour. AIDA was extensively reviewed during the First Research Coordination Meeting of the above CRP and it was found highly valuable for entering field and laboratory data, for providing immediate tabulated results and trends and for keeping uniformity among all users allowing them to compare results in a more reliable manner. Users also recommended the inclusion of few parameters like farmer activities, methods of semen thawing, quality of semen batch, weight and body condition at calving.

The Animal Production and Health Section has received an unexpected high number of requests for this software. Most of the requestors were provided with free copies. Future requests will be shipped after release of the updated version by end of December 1995.

Please contact Dr. Mario García, Animal Production and Health Section, IAEA [Fax: 43 (1) 20607, E-mail: garciam@ripol.iaea.or.at, Phone 43 (1) 2060 26048] for further information.

### **3. New ELISA programme, EDI Vers. 2.11**

The initial version of the programme was distributed to all counterparts and there was a general consensus that the new version is easier to use than the older ones. However, it turned out that there was a major error in the calculation module of the competitive ELISA and also a few minor "bugs" in other modules. This was corrected and the older version, EDI 2.1 should now have been replaced by everybody with the new version of the programme, EDI 2.11. If you do not have the new version, please contact us.

Please remember that the program has to be configured for your ELISA reader and if there is already a functioning version of EDI installed on the computer, it is recommended to write the configuration of the old program down before you install the new one. Once installed the parameters must be changed accordingly.

In some laboratories there was a problem with the handshake of the reader. **In the new program the handshake is disabled. If there are problems with the interfacing of the reader and the computer this has also to be changed on the reader and the handshake has to be switched off.** Please refer to the ELISA reader manual for how to change the handshake.

The error in the calculation of the PI values was only found after the program was used on a routine basis and we depend on your comments to identify these errors. Please therefore let us know if there are any more errors you found in the program. If you have any other comments or suggestions concerning the program, please do not hesitate to let us know!



At present, the program works only with the Multiskan MCC and the Multiskan Plus Mk II (equivalent to the Immunoskan Plus) but shortly it will also become available for use with the SLT and Dynatech ELISA readers.

**(H) XXIV<sup>th</sup> ANNUAL MEETING OF ESNA 12 - 16 September 1994, Varna, Bulgaria**

**Report of Working Group 2 Advanced Methods in Animal Sciences by Chairman, Prof. M. Cristaldi - Univ. of Rome "La Sapienza", Italy - & Prof. B. Todorov - VISVM of Fac. Vet. Med., Stara Zagora, Bulgaria**

The working group held six sessions in which 30 papers including 5 posters were presented by scientists from 12 countries.

***Session 1, ELISA Technology***

There were three oral communications: two from Macedonia and Serbia (group of Drs. Bosnakowski and Hristovski), one from Bulgaria (group of Prof. Todorov). The authors' contributions and the resulting discussion demonstrated that ELISA methods, when correctly applied, showed positive antibody responses in more than 70% of tested cattle. Also stressed was the importance of the comparison between the results from blood and milk samples and the epidemiological approach to the disease detection.

***Session 2, Tracer techniques***

There were four communications: one from Poland (group of Prof. Bobek), two from Bulgaria (group of Dr. Hristov) and one from Prof. Binnerts (The Netherlands), moreover, one poster was presented from a Turkish group. The first contribution from Sechman *et al.* demonstrated the relevance of estradiol in the shell gland development of domestic hen and in their blood flow. Prof. Binnerts discussed the importance of trace elements in toxicology and their chemical and physiological characteristics (Cu/Mo, Cu/Zn, Fe/Rn). Hristov *et al.* illustrated the role of rumen protozoa and bacteria in the digestive function and propose <sup>35</sup>S as tracer. Sel *et al.* from Turkey demonstrated concentrations of cloramphenicol in wild and breed trout.

***Session 3, Reproduction and Radioimmunoassay (RIA) Technology***

Four communications were presented: two from Hungary (group of Drs. Balogh and Thuroczy), two and one poster from Bulgaria (group of Prof. Georgiev & co-workers) and one poster from Turkey (group of Kilicoglu *et al.*). The Hungarian colleagues proposed a radiometric method (Tc-99m) for the detection of sperm motility and a RIA assay for diagnosis of hyperadrenocorticism in dogs. Drs. G. Penchev and S. Georgieva from Bulgaria showed the histostructural changes induced by total-body gamma irradiation in the pig testis and in the sexual hormones, respectively. The poster of the Turkish colleagues showed the use of ultrasonography in the gynecology of domestic cats. The poster presented by Drs. Petko Georgiev and Jordan Nicolov showed the pathological consequences

of experimental rumen acidosis in sheep; in the subsequent session the same co-authors presented a communication on the biochemical and hormonal aspects of the same problem in ruminants.

#### ***Session 4, RIA Technology***

Eight oral communications and one poster belonged to this session: the first from Cyprus, two (Drs. I. P. Georgiev and T. Georgieva) and two (Drs. Kutsarov and Iliev) from Bulgaria, one above-mentioned communication of Drs. Nikolov and Georgiev, one from Poland (group of prof. Bobek), one poster-oral communication from Bulgaria (group of Prof. Todorov) and the poster presented by the Turkish colleagues.

Koumas and Papachristoforou demonstrated the effects of birth season on the puberty in female Chios sheep and in Damascus goats. Georgiev and Georgieva investigated the relationships between biological parameters and milk yield in dairy cows. The Bulgarian group of Kutsarov investigated the hormone increase in the chronic immobilization conditions of ewes by the RIA technology. The group of Bobek and Sechman showed no differences in the resistance to heat stress between not-treated and ascorbic acid treated chickens. Dragoev *et al.* demonstrated that combined effects of pesticide "AGRIA 1050" and gamma irradiation cause multiple alterations in the amino acid contents of lamb meat. Cinar and Sulu studied the effect of monensin on the hormonal values of Holstein calves.

#### ***Session 5, Toxicology***

Four communications and one poster from Bulgarian researchers were presented: the group of Drs. Pavlov and Popov studied the influence of nitrates on the toxicological indices in ruminants. Dr. Angelov studied the radiomimetic effects of sulfur mustard on chickens (the work was presented by Mrs. Andonova since the authors were unable to participate in the meeting). In the poster and in the oral presentation of Dr. Vasiliev's group the effects of chronic heavy metal intoxication on the hormonal values in sheep were presented.

#### ***Session 6, Physiology, Pathophysiology and Microbiology***

The session had six communications and three posters: Dr. Murgali from Turkey presented the use of lignolytic fungi to improve the nutritional value of wheat straw; Ivanov *et al.* from Bulgaria studied the behavioural and nervous reactions of ewes during milking machine use. Yarkov *et al.* studied the negative influence of a pyrethroid insecticide on chicken phagocytosis; Aminkov and Pascalev studied the cardiopulmonary effects of fentanyl and lidocaine mixture used for epidural blockade in dogs; Hubenov presented in two posters studies on the pathogenesis of haemotransfusion shock in cattle observing the acid-base and haemocoagulation changes; the poster of Turkish and Japanese colleagues presented an electrophoretic study of a native cattle in Turkey. The group of Angelov studied experimental treatments of rumen acidosis (in this session also Mrs. Andonova read two communications).

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