Research Coordination Meeting on “The Use of Irradiated Vaccines in the Control of Infectious Transboundary Diseases of Livestock”.

The First Research Coordination Meeting of a Coordinated Research Project (CRP) on “The Use of Irradiated Vaccines in the Control of Infectious Transboundary Diseases of Livestock” was held from 11-15 October 2010 in the Vienna International Centre, Vienna, Austria. It was attended by nine Research Contract Holders and three Research Agreement Holders.

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

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

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

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

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

**Conclusions:**

- The Animal Production and Health Laboratory, Seibersdorf will play a lead role in developing standard operating procedures for attenuation of *T. evansi*. This will involve *in vitro* culture of trypanosomes and determination of their metabolic activity following different doses of irradiation.

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

In Sri Lanka, the objectives in the next year will be to determine the dose of irradiation that arrests the development of *Haemonchus* larvae in the abomasum wall. This will include assessing the potency of irradiated larvae in inducing an immune response in goats and determine resistance to challenge. Among the parameters monitored will be egg counts, haematology, pepsinogen levels and antibody responses to excretory/secretory (ES) antigens.

There are few reports of irradiated vaccines for *F. hepatica*, so considerable developmental work will be required in Ethiopia where studies are being initiated. This will entail identifying the optimum attenuating irradiation dose for the larval metacercariae, determining the number of irradiated metacercariae required for oral vaccination and the effect of different vaccination schedules in inducing protective immune responses. An essential component of the research is the production of infective metacercariae using laboratory reared snails. Parameters measured will include antibody responses, cytokine level and post mortem findings of fluke development.

A similar programme of work is envisaged for *F. gigantica* studies in Sudan. Basic facilities for production of metacercariae will be established, irradiation dosage determined and efficacy of vaccination assessed by measuring antibody levels, cellular immune responses together with liver enzyme changes and associated pathology.

The available vaccines for *I. multifiliis* that have been prepared by chemical treatment or freeze-thawing are not effective in protecting fish. It is planned to prepare trophont and theront culture facilities to enable generation of material for irradiation. The metabolic activity of the parasites before and after irradiation will be monitored by Q RT-PCR in order to identify the most effective dosage. Fish will be vaccinated with irradiation attenuated parasites and the immune response in serum and at the skin surface compared with that induced by a formalin-killed vaccine. Fish will be challenged to determine efficacy of protection.

Complementary studies on *Th. annulata* will be done in Turkey and China. In Turkey, the vaccine source will be schizont-infected cultures, while in China piroplasm-infected red cells. In both instances preliminary trials will determine the most effective dose of irradiation to inhibit the development of the parasites prior to their use in vaccination trial in animals.

The participants endeavour to keep in contact with the Technical Officer and the Research Agreement Holders to inform on the progress of their work, and should submit a report to the IAEA at the end of the first year.