



## ***BPRC's contribution to the progress of medical science: research with non-human primates***

Medical research still requires the use of animals, since many of these studies can only be performed using non-human primates, mainly because they are so closely related to humans. Certain non-human primate species are the only ones that can be infected by the same pathogens that naturally infect humans. In addition, certain novel medicines and treatments are highly specific, and are only effective in humans and their closest relative, the non-human . For this very reason, non-human primates are used for testing in order to guarantee the safety and efficacy of such vaccines/medicines/treatment methods before they can be administered to humans. To this end, Dutch law stipulates that non-human primates may be used as experimental animals only if no suitable alternatives are available.

The Biomedical Primate Research Centre (BPRC) is a specialised scientific research institute with long-standing expertise in research on diseases that threaten public health, including infectious diseases such as malaria, AIDS, hepatitis, and tuberculosis, as well as auto-immune diseases like rheumatoid arthritis and multiple sclerosis. The BPRC has also been involved in research aimed at improving organ transplantation. BPRC's research is focussed on:

- a) Gaining insight into processes that cause diseases, thus enabling the development of increasingly effective and/or safe medicines;
- b) Determining the efficacy and safety of new medicines and treatment methods.

The BPRC also studies the development of alternatives for the use of animals, in accordance with the guiding principles referred to as the 'three Rs': Replacement, Reduction, and Refinement. In this way, it may become possible that animals will eventually no longer be needed for future research.

What are BPRC's research achievements so far?

## *Transplantation*

- Specific genetic characteristics of humans play a major role in whether or not a transplanted organ will be rejected. Research in non-human primates has increased the success rate of selecting the most suitable match for an individual patient.
- The discovery that a blood transfusion prior to an operation has a positive effect on the survival of a transplanted organ was made in non-human primates. This is now common in human clinical practice.
- The first medicines used in humans to prevent the rejection of transplanted organs (immunosuppressants) were tested extensively in non-human primates. This also applies to new methods of treatment to prevent rejection: i.e. new combinations of drugs.
- Drugs are important in enhancing successful transplantation. A number of medicines currently used to stimulate the production of red blood cells after bone marrow transplantation were tested initially in non-human primates.
- When organs are transplanted into humans, following a standard procedure creates a better chance of the new organ's acceptance. Research in non-human identified the methods that would not only be safe but would also work in practice.
- Severe rejection may occur after a bone marrow transplantation, and the severity is influenced by the bacteria (micro-flora) inhabiting the patient's colon. The chance of a transplant rejection can be reduced by modifying this micro-flora. This was initially studied in non-human primates, and is now practised successfully in many hospitals.
- Studies in skin and kidney transplantations in non-human primates have resulted in new treatment methods that reduce rejection.

## *Chronic diseases*

- Studies in non-human primates have resulted in a considerable increase in scientific information regarding how arthritis develops. These studies have clarified which factors mostly influence sensitivity for arthritis.
- Many questions still remain regarding disease progression in multiple sclerosis and its treatment. Research within the BPRC has led to new information concerning the mechanisms that may be involved in the course of multiple sclerosis. This knowledge is used to develop and evaluate new methods of treatment.
- Research in non-human primates has resulted in safe and effective treatments for autoimmune diseases such as multiple sclerosis and arthritis.
- Research at the BPRC has contributed significantly to the early recognition of possible severe side effects of new medications for chronic diseases such as multiple sclerosis and arthritis.
- A significantly refined model for multiple sclerosis in marmoset monkeys has been developed. Using this model, a possible explanation has been found for the previously poorly understood association between Epstein Barr virus infection and multiple sclerosis.
- The marmoset monkey model for multiple sclerosis provides new insights into the onset and mechanism of the disease, and offers opportunities for new treatments.
- The BPRC non-human primate models for multiple sclerosis have been used for the preclinical safety and efficacy testing of potentially therapeutic antibodies.
- A marmoset model for chronic arthritis has been developed.

## ***Chronic diseases (cont.)***

- Several new medicines for arthritis have been evaluated in the rhesus monkey model. Further evaluation is currently ongoing in the clinic.
- Antibodies against the cartilage protein collagen in the joints of arthritis patients and the arthritis primate models are highly comparable.
- A novel model for chronic Parkinson's disease has been developed in marmoset monkeys. In this model, disease symptoms can be suppressed through neuro-feedback.
- A novel model for Alzheimer's disease has been developed in marmoset monkeys. This model is being used to evaluate the safety and efficacy of potential medicines.

## *Infectious diseases*

- The first genetically engineered Hepatitis B vaccine was tested in non-human primates for its efficacy. This vaccine is now administered worldwide, and protects people from this infectious disease.
- A specific hepatitis virus (hepatitis delta), which can cause liver cancer when co-infection with hepatitis B virus occurs, was discovered and characterised at the BPRC.
- Non-human primates were used to determine how the number of HIV-virus particles in the bloodstream relates to the chance of actually developing AIDS. This information is now used to monitor infection and adjust treatment in human patients. Each year, approximately 2 million people die of AIDS worldwide.
- HIV vaccines are first tested in non-human primates, and the results of these studies are used to select the vaccine preparations that will proceed to human trials.
- Protection in humans, induced by HIV vaccines, is influenced by the genetics of the recipient; this observation has been confirmed in rhesus macaques.
- The effects of HIV vaccination in combination with antiviral treatment have been studied in non-human primates at the BPRC. This is currently being applied in clinical practice.
- Chimpanzees who are exposed to human immunodeficiency disease are infected but do not develop the acquired immunodeficiency syndrome. Studies of the mechanisms that are involved in preventing immunodeficiency in the chimpanzee has led to a better understanding of AIDS development in humans.

## ***Infectious diseases (cont.)***

- A promising peptide vaccine has shown partial protection from HIV infection and AIDS development in infected rhesus monkeys. This concept is currently being followed up.
- A tuberculosis model that better assesses protective vaccine effects has been developed in non-human primates.
- BPRC's non-human primate model for tuberculosis contributes to the selection of possible vaccine candidates in a preclinical setting.
- Studies in non-human primates have given important information regarding mechanisms involved in different life-cycle stages of the malaria parasite. This information can be used to identify new targets for drug development. Worldwide, around 0.7 million people die annually from malaria.
- Research at the BPRC has made an important contribution to the development, production, and testing of the first Dutch vaccine against malaria tropica, the most severe form of malaria.
- BPRC has contributed to the development of new anti-malarial drugs, which are currently being evaluated in the clinic.
- BPRC has contributed to the discovery of a new drug target in malaria parasites.
- A malaria vaccine formulation showed major side effects when it was administered to rhesus macaques, and as a result it was subsequently withdrawn from planned human trials.
- BPRC has developed a new culture system for a primate malaria that uniquely allows research into the biology and possible treatment of dormant malaria parasites.

## ***Infectious diseases (cont.)***

- The BPRC has developed a genetically modified monkey malaria parasite. The availability of this parasite will make it easier to assess the safety and efficacy of new malaria vaccines and drugs.
- The BPRC provides a global service to screen non-human primates for virus infections. New primate viruses have been discovered in samples sent for screening from zoos or rehabilitation centres.
- BPRC has implemented primate models for influenza infection, which are being used to evaluate vaccines and drugs.
- By comparing different primate species, the most suitable influenza infection model has been developed.
- BPRC has developed West Nile virus infection models in non-human primates, which are being used to evaluate antiviral drugs and vaccines. A new vaccine that provided protection against infection has been evaluated successfully.

## **Reducing animal testing: the 3 Rs and animal welfare (cont.)**

- The BPRC has identified the genetic background of all its animals. As a result, animals can be better selected for particular research projects, overall reducing the number of animals required for experiments.
- The BPRC has developed significantly fewer invasive methods to determine the disease process of multiple sclerosis in marmoset monkeys and arthritis in rhesus monkeys.
- For multiple sclerosis, the BPRC has succeeded in developing a much milder methodology to obtain a still relevant disease model.
- In tuberculosis, the BPRC has discovered a relationship between clinical chemistry parameters measured in blood samples and the chances of surviving this disease. Hence, certain studies can be shortened, consequently reducing stress levels in the monkeys.
- *In vitro* research has demonstrated that inflammation parameters can differ dramatically between different tissues.
- Reduction of animal use in BPRC's research on new antimalarial drugs has been nominated by the Dutch animal protection organisation for the 'leef in het lab' award 2014.
- The BPRC has developed a laboratory method to test and develop adjuvants (agents that enhance the effectiveness of vaccines). In 2008 this study was nominated for 'Audience Award for Alternatives' (Publieksprijs Alternatieve Dierproeven).

## ***Reducing animal testing: the 3 Rs and animal welfare (cont.)***

- Cell cultures have been developed to study diseases of the central nervous system, such as multiple sclerosis. These methods are now being used, whenever possible, although in the final analysis animals still have to be used. Nevertheless, the number of animals needed for this research has been reduced.
- New methods have been developed to breed and house monkeys in a research institute. Living conditions of the animals resemble those of their natural habitat, thus improving animal welfare.
- BPRC has employed personnel that work on 'Positive Reinforcement Training' of the animals to reduce and prohibit stress. This refinement has a positive influence on animal welfare as well as study outcomes.
- A large programme for food- and non-food enrichment is operative and in continuous development.
- BPRC continues to play an important role in the training of animal caretakers and other specialists in the area of animal training and enrichment.
- BPRC organises national and international meetings to promote and improve animal welfare in primate centres/research institutes.
- BPRC has developed a manual offering standard food enrichment recipes for macaques and marmosets. This manual is made freely available, and over 300 copies have been distributed worldwide.
- Due to the modern facilities, all animals at the BPRC are housed socially.

## ***Reducing animal testing: the 3 Rs and animal welfare (cont.)***

- The BPRC is continuously refining procedures. A programme has been implemented to further improve veterinary procedures and animal housing.
- For the optimal social housing of animals, BPRC has employed specialists that focus on animal breeding, selection of the most suitable animals for an experiment, and introduction/training of animals in the experimental facilities.
- Animal breeding is a continuous task for the veterinarians and the colony manager. Contraception is applied to actively regulate the number of offspring, but with care taken to interfere as little as possible with the complex social structure of the breeding colonies.
- The BPRC has developed less- and non-invasive technologies to identify specific markers. As an example, we are able to measure long-term stress by examining the hair of the animals. Results show that the modern housing and careful approach have significantly reduced their stress levels.
- The BPRC has a routine lab for fast and effective measurements of animal health. This allows for a quick and adequate response when needed.
- Tissues and blood samples from non-human primates are stored in documented databases. This is being used by many international researchers and fits very well in the '3R' concept.
- The BPRC regularly organises symposia and scientific meetings on Alternatives for primate use and Refinement of research methodologies.

