NC3Rs

National Centre for the Replacement, Refinement
and Reduction of Animals in Research

REALISING THE POTENTIAL

Annual Report 2008

CONTENTS

The 3Rs

Replacement refers to methods which avoid or replace the use of animals defined as 'protected' under the Animals (Scientific Procedures) Act 1986.

Reduction refers to methods which minimise animal use and enable researchers to obtain comparable levels of information from fewer animals or to obtain more information from the same number of animals, thereby reducing future use of animals.

Refinement refers to improvements to husbandry and procedures which minimise pain, suffering, distress or lasting harm and/or improve animal welfare.

Picture credits:

Pages 3, 28	Arif Gardner
Pages 4, 5, 8-13, 26	Darrin Jenkins
Page 14	Teri Pengilley
Page 22	AstraZeneca (Pictured: State-of-the-art UK facilities, providing stimulating environments in which dogs can socialise)

FOREWORD

I was honoured this year (effective from July 2008) to be appointed Chair of the NC3Rs Board. A detailed account of the work and achievements of the Centre during 2008 is to be found within these pages, together with a look forward to plans for 2009 and beyond. In this foreword I want to reflect briefly on the opportunity we have now, through the NC3Rs, to bring real and lasting benefits to science and animal welfare without hindering or compromising the quality or productivity of research.

We live in fascinating times; the investment in biological and health sciences research, in tandem with remarkable advances in the technology available for probing and analysing physiological and pathological processes, continues to pay huge dividends. Increased understanding of biological systems in health and disease continues apace, and is matched by an appetite to ensure that scientific advances are translated quickly and effectively into provision of improved healthcare and other important benefits.

In addition, we have a responsibility to ensure that when those same scientific advances provide opportunities to deliver improvements in animal welfare (through the 3Rs), these are seized and exploited fully.

We must recognise that animals will remain, for the foreseeable future, an important part of life science research. However, it is important that that recognition is matched by acknowledgement of our collective responsibility to make sure that wherever possible we bring to bear our best science and technology to improve animal welfare through application of the 3Rs.

It is here, in the alignment of the best that scientific research has to offer with continued improvements in animal welfare that the NC3Rs has had, and will continue to have, a pivotal role to play. I have been impressed how the Centre, and the team of gifted and enthusiastic scientists led by Dr Vicky Robinson, have legitimised research focused on delivery of the 3Rs. Through the medium of the Centre many among the UK scientific community have for the first time considered whether the fruits of their research could provide realistic opportunities to replace, reduce and refine the use of animals in research and testing.

It is in realising those opportunities that the NC3Rs plays such an important role in our national scientific landscape, and I feel privileged to be associated with that work.

Professor Ian Kimber, Chairman

PROMOTING RESEARCH

A major aim of the NC3Rs is to promote high quality research in the 3Rs as a central component of advancing the life sciences — increasing knowledge, improving human and animal health and medicine, and contributing to national competitiveness. The Centre does this by funding research, recognising and rewarding excellence, and working with the UK's major research funders.

"The NC3Rs is now a mainstream funding source attracting high quality proposals, many from top researchers in the UK who clearly want to address the 3Rs."

Professor Nancy Rothwell DBE, Deputy President and Deputy Vice-Chancellor, University of Manchester

Funding 3Rs research

The Centre currently has two routes for investing in 3Rs research, a main funding scheme that awards grants of up to three years and a Small Awards Scheme. Both schemes are now generating valuable research outputs across the biosciences.

The NC3Rs is the UK's largest funder of 3Rs research, and investment in the main funding scheme has increased year-on-year since the Centre was opened, from £500,000 in 2004 to £2.6 million in 2008, with a total of £8 million invested to date. New increased funding awarded by the Government, via the Medical Research Council (MRC) and the Biotechnology and Biological Sciences Research Council (BBSRC), as part of the last comprehensive spending review will ensure this continues.

Grant applications are judged using the MRC scoring criteria so that only the highest quality research is awarded. Ten grants were awarded in 2008 (see Research grants for 2008). This is equivalent to a 20% success rate and is therefore comparable with the UK's major bioscience funding bodies. In order to continue to drive the 3Rs research agenda, two priority areas for investment were highlighted in 2008 in addition to 'response mode' funding. These were '*3Rs in fish*', under which three awards were made; and '*Refinement in rodent husbandry, care and procedures*' for which two projects were funded.

By positioning 3Rs research within the mainstream of the life sciences, the NC3Rs has begun to change the scientific environment in the UK. Advances in science, with measurable impacts on the 3Rs, are being published by researchers funded by the NC3Rs and this has led to a shift in practices in other research groups in the UK and elsewhere. Examples can be found in the sections on the 3Rs in cardiovascular research (page 8) and wound healing research (page 12).

The Small Awards Scheme, run in partnership with the Laboratory Animal Science Association (LASA), aims to support relatively small scale endeavours to acquire information or skills relating to the 3Rs such as research, training and exchange visits. In 2008, eleven awards were made including *'Validating non-invasive collection methods in African bat populations'* and *'Chick embryo replacement of mouse models to study the regulation of gene expression'* (see **www.nc3rs.org.uk/fundedsmallawards** for the complete list).

Research grants for 2008

The following ten research projects were funded by the NC3Rs in 2008.

Professor Sue Barnett and Dr Mathis Riehle, University of Glasgow (£294,404) The development of an *in vitro* model of CNS injury to identify factors which promote repair

Professor Andrew Cossins and Professor Ernst Wit, University of Liverpool (£512,584) Development of a mechanistically informative genome-wide, *in vitro* chemicals screening technology

Dr Atticus Hainsworth, St George's London (£43,288) Carotid artery endothelial growth: a novel *in vitro* assay

Dr Ioanna Katsiadaki, Cefas (£398,640) Validating a sexual development test using the 3-spined stickleback for addressing the 3Rs in fish toxicity testing

Professor Robert Newbold and Professor Michael Donovan, Brunel University (£299,052) Development and validation of mechanisms-based *in vitro* transformation assays for carcinogen screening

Dr Keith Redhead, Intervet UK Ltd (£26,988) Replacement *in vitro* assays for the quantification of clostridial vaccine antigens

Dr Paul Simons, Professor Philip Hawkins and Professor Mark Pepys, University College London (£302,128) Inducible SAA transgenic mice: a refined model of human amyloidosis

Professor Phil Stephens, Professor David Kipling and Professor David Thomas, Cardiff University (£243,624) Establishment and validation of a stable, cell-based diabetic wound bioassay

Dr Siouxsie Wiles and Professor Shiranee Sriskandan, Imperial College London (£270,784) Reduction and refinement of murine models of bacterial infection

Dr Jun Zou and Professor Christopher Secombes, University of Aberdeen (£254,548) Development of leucocyte cell lines for immunological research in teleost fish

Full abstracts are at www.nc3rs.org.uk/fundedresearch

"This year's prize-winning research is an excellent example of the 3Rs being applied to an exciting area of science."

Professor Patrick Vallance, Senior Vice President of Drug Discovery, GlaxoSmithKline

Rewarding excellent research

To raise the profile of excellent research that has had positive 3Rs and scientific impacts, the Centre has an annual 3Rs Prize of £10k, sponsored by GlaxoSmithKline. The prize is awarded to a piece of research published in the last two years and this year's winner is Mr Thomas Johnson, University of Cambridge, who has developed a system for testing new treatments for sight-threatening conditions such as glaucoma and macular degeneration[1].

New treatments involving the transplantation of stem cells into the eye may prove beneficial for patients by protecting the vulnerable nerve cells within the retina, and may even cure blindness. Improving the outcome of injecting stem cells into the eye is hindered by a lack of model systems which not only replicate the challenges of transplanting cells into a living eye, but also enable experimental manipulation. Currently, researchers inject stem cells into the eyes of living animals, but such experiments are time-consuming, potentially stressful to the animals, and require the use of large numbers of animals to achieve statistically significant data.

To overcome these limitations, Mr Johnson, working under the supervision of Dr Keith Martin at the Centre for Brain Repair in Cambridge, obtained retinal tissue from euthanased rats and optimised culture techniques to maintain the tissue for up to 17 days. His winning publication demonstrates that the cultured tissue remains healthy, maintains its layered architecture, and continues to generate a variety of proteins that are characteristic of the retina. Most importantly, the cultured retina responds to cell transplantation in a similar way to the eyes of living animals. The culture system also allows well-controlled experimental manipulation and avoids the problem of graft rejection.

Using the new method, an eight-fold reduction in the number of animals used is possible because eight retinal cultures can be obtained from a single rat. The use of animals has also been refined because the animals are humanely euthanased instead of undergoing interocular injections and the associated welfare costs. Using this technique, future research will be able to improve the efficacy of stem cell therapy while utilising far fewer animals than would otherwise be necessary.

Because of the high quality of entries for the 3Rs Prize, the selection panel also awarded two Highly Commended prizes. The recipients were Mr Charalambos Tymvios, Imperial College London, for a publication on the NC3Rs-funded research on refining a model of pulmonary embolism (see page 9) and Dr Jenny Morton, University of Cambridge, for a publication describing a refinement of the tests to measure cognitive deficits in mice used for neurodegenerative disease research[2].

Working with the major research funders

To embed the 3Rs into all publicly funded research involving animals, the NC3Rs also works with the other major science funders in the UK. Over the last year, the NC3Rs has continued to fulfil and develop its roles as a source of expertise on the 3Rs and a catalyst for joint activities between the UK's funders of research using animals. This work is facilitated by renewed funding from the Wellcome Trust for a scientific post and for the first time has included the involvement of the Department for Environment, Food and Rural Affairs (Defra) and the Natural Environment Research Council (NERC). The development of guidelines, peer review, and joint sponsorship of workshops were among the activities undertaken in 2008.

The most far-reaching activity has been the joint publication of 3Rs guidance for researchers and associated veterinary and animal care staff using vertebrates (live animals or animal products) in research funded by the Research Councils (MRC, BBSRC and NERC), Defra, NC3Rs, and Wellcome Trust.

Bringing together, harmonising and expanding existing material from the different funders in one document, the guidelines exceed the legal minima and provide an easy and authoritative reference for researchers. They include links to advice on designing experiments to minimise the number of animals used, and also the most appropriate ways to house, transport, handle and restrain animals to minimise distress.

1: Johnson TV & Martin KR (2008) Development and characterization of an adult retinal explant organotypic tissue culture system as an ***in vitro*** intraocular stem cell transplantation model. ***Investigative Ophthalmology & Visual Science*** **49** (8), 3503-12

2: Morton AJ, Skilings E, Bussey T & Saksida LM (2006) Measuring cognitive deficits in disabled mice using an automated interactive touchscreen system. ***Nature Methods*** **3** (10), 767

"Working with the NC3Rs has helped strengthen the MRC's commitment to ensuring that the 3Rs are fully embedded in the research we fund."

Sir Leszek Borysiewicz, Chief Executive, MRC

Researchers applying for funding from any of the funding bodies must show that they comply with the principles set out in the guidance. This is the first time such principles have been tied to funding. To facilitate compliance, referees of grant applications, and the panels and committees that make decisions on funding, are asked to assess whether the proposed research and practices meet the principles in the guidance and whether full consideration has been given to the 3Rs. Where issues are identified, these must be adequately addressed by the applicant or the research will not be supported.

The NC3Rs continues to review all grant applications involving non-human primates, dogs, cats or equines that are submitted to BBSRC, MRC, Wellcome Trust and, where appropriate, other members of the Association of Medical Research Charities (AMRC), and advises on 3Rs issues. A total of 142 applications have been reviewed to date.

Working closely with other funding bodies, the NC3Rs aligns the best science with the 3Rs, through joint research calls and workshops. In 2008, this included a joint call for proposals with BBSRC on the use of invertebrate models, the tissue engineering initiative with BBSRC (see page 17) and a workshop to highlight scientific and technological advances which could refine the use of chronic implants in animal research, held jointly with the Wellcome Trust. Some animal studies involve implantation of recording, stimulation or restraint devices that need to stay in place for months or years. These chronic implants can have a negative impact on animal welfare because of the potential for pain and discomfort from the implant surgery and anaesthesia or problems afterwards, such as infection, inflammation or rejection.

The workshop brought together scientists and veterinarians who work with implants, and experts from specialities such as orthopaedic and reconstructive surgery, dentistry, and biocompatible materials, for information exchange and to identify opportunities for collaboration directed at improving animal welfare and delivering more efficient science.

INNOVATIVE APPROACHES IN CARDIOVASCULAR RESEARCH

Diseases of the heart and circulatory system are the main cause of death in the UK, accounting for one in three (35%) deaths each year. There is extensive investment in research to understand these diseases and develop effective new treatments. However, many of the current experimental models have limitations for investigating human disease and there is an acute need for improved approaches. Funding from the NC3Rs, awarded in 2006 to two research groups at Imperial College London, is now helping to deliver progress.

Funding of 3Rs research in pulmonary embolism has led to improved scientific results which will inform new treatments.

An enhanced animal model for pulmonary embolism research

Dr Michael Emerson has refined an animal model of pulmonary embolism which could help in understanding how the disease develops and testing potential new drug treatments. A paper describing the new model, which has led to an improved scientific approach and less suffering for the animals, was published in early 2008[1].

Pulmonary embolism is a potentially fatal condition where the pulmonary artery, the major vessel that supplies blood to the lung, becomes blocked by a blood clot that has formed in a vein elsewhere in the body and travelled to the lungs via the bloodstream. Symptoms can include difficulty breathing, chest pains when drawing breath, and heart failure, collapse or sudden death in the most severe cases.

The animal model most commonly used involves injecting conscious mice with a blood clotting agent to induce a massive pulmonary embolism which leads to paralysis and often death.

The effectiveness of drugs is assessed by their ability to prevent paralysis or death and the tissues of the mice are also analysed to better understand the causes of the disease. There are clear animal welfare concerns in inducing a fatal and painful condition in mice and the procedure is classified under the Animal (Scientific Procedures) Act 1986 (ASPA) as causing 'substantial' suffering. Additionally, the model has limitations in that it only reflects the most extreme stage of the disease. The early stages, which are most amenable to treatment, are not represented.

Dr Emerson, and his colleague Mr Charalambos Tymvios, were funded by the NC3Rs to develop a refinement to the model which uses non-fatal levels of clotting agent in anaesthetised mice. By introducing radioactively labelled platelets into the bloodstream of the mice, the gradual formation of clots in the cardiovascular system can be monitored over time in unconscious animals.

Dr Michael Emerson

Results obtained using the refined animal model are significantly improved over those derived with the original model, both in terms of minimising animal use and increasing the amount of scientific data generated. The procedure has been refined from 'substantial' to 'unclassified' and animal use has been reduced from 200 to 30 mice in a typical experiment because multiple readings can be carried out in the same animal and fewer are needed as controls. The data generated is also more representative of the early stages of the disease which makes it more valuable in the search for treatments. Dr Emerson has also been inspired to look for non-animal alternatives to his research, including the use of human platelets, and has a Small Award to progress this work.

1: Tymvios C, Jone S, Moore C, Pitchford SC, Page CP & Emerson M (2008) Real-time measurement of non-lethal platelet thromboembolic responses in the anaesthetized mouse. *Thrombosis and Haemostasis* **99** (2), 435-440

Professor Sian Harding

Developing stem cell replacements for use in heart disease research

Large numbers of animals are currently used in heart disease research as there are no suitable alternatives available. There is considerable interest in the use of heart muscle cells, known as cardiomyocytes, derived from stem cells, to investigate heart disease and to potentially treat patients with damaged hearts.

Professor Sian Harding, Dr Nadire Ali, and Dr Jamie Wright, funded by the NC3Rs, are applying their expertise in this area to use these cells as a replacement for animals in heart disease research.

Cardiomyocytes can be created in the laboratory by inducing human embryonic stem cells to develop into heart muscle cells. Their origin means that they are more relevant for the investigation of human disease. The added advantage of using stem cells as a source of cardiomyocytes, rather than adult cells dissected from animal or human tissue, is that they can be kept alive in the laboratory for up to five months rather than only a few days. Before the cardiomyocytes derived from stem cells can be used as a model for the human heart, it is necessary to determine whether their functional properties and responses to drugs are comparable with those of adult cardiomyocytes from human hearts.

An important characteristic of cardiomyocytes is that they beat in the same way as an intact heart. Professor Harding and her colleagues have compared the response of stem cell-derived cardiomyocytes and adult cells to drugs that affect the speed at which they beat [1]. They found that the stem cell responses were similar to adult cells, confirming their potential as a model system.

Using the stem cell-derived cardiomyocytes has the potential to replace approximately 180 animals per year in Professor Harding's laboratory alone and, based on the scientific literature, in approximately 1500 studies worldwide, each using around 10-50 animals per year. The long term potential for reduction of animal use is even greater in pharmaceutical testing, where these cells may provide valuable information on new medicines to treat heart disease whilst avoiding the use of animals.

The stem cell-derived cardiomyocytes are now being systematically compared with models currently in use, through partnerships with pharmaceutical companies. The public-private collaboration '*Stem cells for safer medicines*' is co-ordinating and funding these efforts.

1: Brito-Martins M, Harding SE & Ali NN (2008) ß1- and ß2AR responses in cardiomyocytes derived from human embryonic stem cells: comparison with failing and non-failing adult human heart. *British Journal of Pharmacology* **153**, 751-759

Using the stem cell-derived cardiomyocytes has the potential to replace approximately 180 animals per year in Professor Harding's laboratory alone, and in 1500 studies worldwide, each using around 10-50 animals per year.

Dr Jamie Wright

REPLACING ANIMAL USE IN THE STUDY OF WOUND HEALING

A wide range of animal species and models are used to study wound healing but they have variable relevance to human disease, can be technically demanding, and are associated with animal suffering. Research funded by the NC3Rs at the Universities of Lancaster and Cardiff is helping to provide new solutions that replace the use of animals in this area.

Dr Bojun Zhao

New vision for eye research

As a result of NC3Rs funding, Dr Nigel Fullwood, Dr Frank Martin and Dr Bojun Zhao, University of Lancaster, are pioneering a new technique with the potential to have a significant impact on ophthalmology research worldwide. Using cow eyes obtained as a by-product of the meat industry, they have developed a fully functional model of the front part of the eye, the cornea, which provides a more efficient way of investigating eye injury and disease, while also reducing and replacing the use of animals.

Maintaining the complex architecture of the cornea in the laboratory is a considerable challenge and most research on eye disease and injury is still conducted in live animals. However, the researchers have now shown that this model acts similarly to a live cornea so that, for example, if the model cornea is damaged it heals normally.

One of the unique factors of the model is that the outside of the cornea is exposed to air, analogous to a normal eye, and therefore more accurately replicates the natural environment. In addition, a major advantage of the model is that the cornea is easier to manipulate and more parameters can be monitored than in the eye of a live animal.

Dr Fullwood and colleagues have investigated the use of the model to study alkaline burns to the cornea from domestic or industrial chemical accidents[1]; The treatment for alkaline burns is difficult, and the prognosis poor, and this system may lead to the development of better treatments without the use of large numbers of animals. Dr Fullwood has already demonstrated that the model provides a suitable system to investigate gene therapy treatments for corneal wound healing and inflammation[2].

With further development the model cornea could also be utilised in routine toxicity testing. During the development of new chemicals and drugs, many compounds undergo tests on live animals to establish their potential to cause injury to the eye. In combination with other non-animal alternatives, Dr Fullwood's current work to validate and automate his system could lead to a reduction in animal use for this purpose.

Publications describing the model have generated significant interest from other researchers in both the UK and worldwide. A number of groups both in Europe and Asia have already adopted this model for use in corneal research.

A novel approach to identify new wound healing medicines

Professors Phil Stephens and David Thomas, and their colleagues at Cardiff University, received grants from the NC3Rs in 2006 and 2008 to develop a cellular tool intended to improve discovery of therapies to treat chronic diabetic wounds and significantly reduce the use of animals in the discovery process.

Impaired wound healing, such as diabetic foot ulcers, occurs in 3-5% of the population over 65 and dealing with diabetic foot problems costs the UK health service over £38 million per year. The animals used to study wound healing do not adequately reproduce the dysfunctional wound healing seen in patients and, as a result, there is a need for improved models.

Professor Stephens' team have isolated normal and diseased skin cells from diabetic patients and compared them in order to identify genes that are expressed differently in the diseased cells. They now have a source of cells that represent the disease and are using them to develop a high-throughput system for screening potential medicines by looking at the expression of reporter genes.

Dr Nigel Fullwood

It is anticipated that when the funding ends in three years time, the team will be close to commercialising the research and developing a high-throughput screening system. Such a reproducible, diabetic wound model system will provide a unique resource for the wound healing community to advance scientific knowledge and improve drug discovery while simultaneously reducing the number of animals required to achieve this. Such an approach could be translated to other disease areas such as rheumatoid arthritis or osteoarthritis.

1: Zhao B, Ma A, Martin FL & Fullwood NJ (2009) An investigation into corneal alkali burns using an organ culture model. *Cornea*, In press

2: Zhao B, Allinson SL, Ma A, Bentley AJ, Martin FL & Fullwood NJ (2008) Targeted cornea limbal stem/progenitor cell transfection in an organ culture model. *Investigative Ophthalmology and Visual Science* **49**, 3395-3401

ACCELERATING INNOVATION IN THE 3RS

Providing an engaging and intellectually stimulating environment for scientists to advance the 3Rs as an integral part of their research is essential to the Centre's objectives. The NC3Rs organises a range of symposia and workshops to solve the latest challenges in research involving animals, including exploring opportunities for using new technologies, horizon scanning, raising the profile of the 3Rs, and promoting collaborations between diverse disciplines and sectors.

"The NC3Rs should be congratulated on their achievements in inspiring scientists to exploit their research to benefit the 3Rs."

Professor Clive Page, Chair, Biosciences Federation Animal Sciences Group

By attracting mainstream researchers from a range of disciplines and sectors, events organised by the NC3Rs provide unique forums for discussion and interaction on the 3Rs. In 2008, the NC3Rs organised (or supported) nine events, with a total of more than 700 attendees from over 200 organisations including representatives from universities, industry, funding bodies, regulatory agencies and Government.

In order to maximise impact and align with the UK's life sciences research priorities, the NC3Rs works closely with scientific partners. For example, in 2008, the Centre organised workshops on tissue engineering with BBSRC and the refinement of chronic implants with the Wellcome Trust. The workshops are part of integrated programmes of activities led by the NC3Rs which will impact on the future direction of research. The collaborations and ideas instigated are fostered, progressed, and shared through peer-reviewed publications, and expert working groups.

Examples of how effectively this approach has worked in practice can be found in the sections on tissue engineering (see page 17) and nausea and emesis (see page 18).

In addition to topic-specific workshops, the NC3Rs also hosts symposia to increase knowledge and raise awareness on the 3Rs. In 2008, as part of its established programme of symposia, the NC3Rs held its second 3Rs symposium with the Biosciences Federation Animal Sciences Group which represents all of the UK's leading learned societies. In September, the Centre hosted its annual symposium specifically for animal technicians, sponsored by AstraZeneca, and recognised as part of the professional development of animal technicians by the Institute of Animal Technology.

In November, the fourth annual meeting on the welfare of non-human primates was held, focusing on the topical issue of breeding and supply. With speakers from Israel, Mauritius, and the USA, the meeting attracted over 90 delegates from the scientific and regulatory communities.

EVENTS IN 2008

Stakeholder Meeting
16 January, London
Annual event providing a review of progress and initiatives.

The 3Rs: from fundamentals to application – joint with Biosciences Federation
19 March, London
Symposium to highlight scientific and technical advances which have implications for the 3Rs.

Engineering tissue alternatives to animals – joint with BBSRC
30 April, London
Meeting to showcase the potential of tissue engineering as a means to replace animals.

Toxicokinetics and the 3Rs
29 May, London
Workshop to discuss the role of toxicokinetic information in improving chemical risk assessment and advancing the 3Rs in regulatory toxicity testing.

Minimising non-human primate use in monoclonal antibody development
26 June, London
Meeting to explore opportunities for minimising non-human primate use in chronic studies, including reproductive toxicity.

Animal Technicians' Symposium
17 September, London
Annual meeting focusing on animal welfare and refinement issues.

Refinement of the use of chronic implants in animal research – joint with Wellcome Trust
1 October, London
Workshop to highlight scientific and technological advances which refine the use of chronic implants.

3Rs and wildlife – run by the Central Science Laboratory
2 October, York
Symposium funded under the Small Awards Scheme to promote implementation of the 3Rs in wildlife studies.

Primate Welfare Meeting
25 November, London
Annual event bringing together individuals with a common interest in the welfare of laboratory-housed non-human primates.

REALISING THE 3RS POTENTIAL IN BASIC RESEARCH

Discovering opportunities for the 3Rs in basic research is inherently difficult because of the nature and variety of the research questions and the lack of scientific and technological solutions. The NC3Rs has embraced this challenge by stimulating scientists to consider how the 3Rs can be exploited in their research in two areas — the use of tissue engineering and the study of multi-system reflexes. This has facilitated identification of novel applications, development of new approaches, and translation of research across disciplines and sectors.

"The knowledgeable and interactive approach of the NC3Rs has energised the field of tissue engineering. Translating this enthusiasm into useful replacement methods will take time and patience and continued funding from a wide range of industry and government agencies." *Professor Kevin Shakesheff, Director, Centre for Biomolecular Sciences, University of Nottingham*

In many fields of research, there is growing acknowledgment that alternative approaches have an increasingly important role in overcoming current challenges with some animal models. The Centre has created an environment for successful discovery, translation, and uptake of alternative approaches by: engaging leading scientists, encouraging the application of their expertise to replacing animal use, and supporting ideas that emerge from NC3Rs workshops and collaborations.

The NC3Rs works in rapidly advancing and difficult research areas to promote new opportunities for minimising animal use. Frequently, these approaches and technologies can be relevant to other fields or applied research, opening up the potential for further reduction of animal use.

Fulfilling the replacement potential of a rapidly advancing research area

Engineering human tissues to treat disease by replacing dead, diseased or non-functioning tissue in the body has huge potential to provide new therapies for a wide range of medical needs. The success of this relies on accurately recreating the characteristics of the live tissue in the laboratory. For instance, interactions between different cell types and between cells and their extracellular matrix must mimic the environment in the body. There are a wide range of tissue engineered products currently available, including cartilage, tendons, skin and corneas.

Tissue engineering has the potential to boost scientific research by providing new experimental tools with which to address research questions in diverse fields. Because tissue engineering aims to reproduce what happens *in vivo*, it also has the potential to replace animal use. Until recently, the clinical focus of the field has meant that opportunities for replacing animal use, for example in biological research or in toxicological risk assessment, have not been fully recognised. There are pioneering examples of skin models which have been validated and accepted as alternatives to tests involving animals for skin corrosion and irritation, but there is much greater scope for exploiting the technology.

To increase the profile of tissue engineering as a tool to replace the use of animals, the NC3Rs has been working with BBSRC, a major funder of tissue engineering in the UK. This partnership has allowed increased investment in 3Rs applications of the technology through joint strategic funding. In 2008, a jointly-sponsored workshop brought together over 100 scientists from leading groups working on a range of tissue engineering products to share progress on areas where there is the greatest potential to replace animal studies. This meeting has resulted in new collaborations between scientists from a wide range of disciplines. A review of the workshop, which includes the examples that were presented, such as asthma, diabetes and toxicity testing of new drugs, will be published in 2009.

Building on the success of the first workshop, the NC3Rs and BBSRC are hosting a two-day symposium and networking event in 2009. The focus of the symposium is to encourage uptake and commercialisation of tissue engineering and to showcase areas where tissue engineering could replace animal models in basic research and safety assessment.

"Working with the NC3Rs challenged me to consider the possibility of using non-sentient models to identify emetic liability rather than using mammals such as the ferret. This has led directly to a collaboration to investigate the behavioural response of social amoeba to a range of emetic agents."

Professor Paul Andrews, Dean of Postgraduate Studies, St George's, University of London

Tackling replacement in a complex system

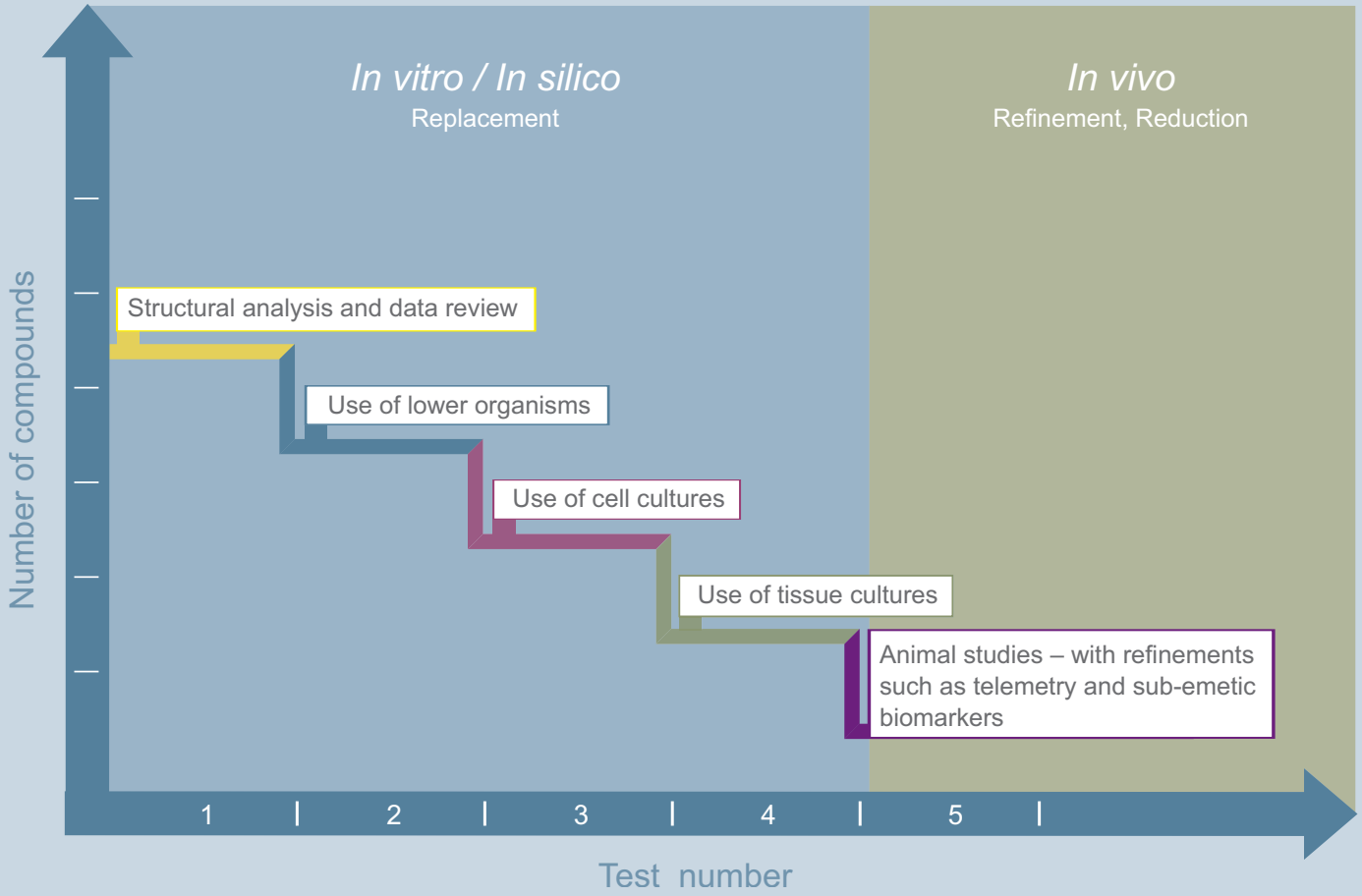
Identifying opportunities to reduce and replace the use of animals to study the body's complex responses to stimuli is exceptionally difficult. Studying multi-system reflexes mediated through neural pathways, where a number of tissues or organs are involved, has conventionally been considered unfeasible without using living animals. The NC3Rs is leading an ambitious project to review animal use and opportunities for replacement in multi-system reflexes, using nausea and emesis (vomiting) research as a model case that will set a precedent for other fields.

Nausea and emesis are amongst the most common symptoms encountered in medicine as either symptoms of disease or side effects of treatments. Studying nausea and emesis is important to understand the mechanisms behind the symptoms, to develop new treatments, and to reduce the number of drugs in development that have nausea and emesis as side effects. Emetic liability is a significant problem with more than 50% of marketed drugs having nausea as a side effect and 33% having nausea and emesis as side effects. This can reduce patient compliance. In addition, nausea and emesis have contributed to some important drugs being stopped in development.

Many animal species are used to study nausea and emesis, including ferrets, shrews, mice and dogs. However, there is considerable variability in the responses of the animal models to drugs known to cause emesis in man, making the choice of species for preclinical investigations difficult. Some of the most commonly used animals, such as rodents, do not have a vomiting reflex and understanding and assessing the subjective human sensation of nausea in animals is exceptionally difficult. Additionally, these studies can cause animal suffering due to the effects of inducing emesis such as reduced food intake, weight loss and dehydration.

The NC3Rs project was initiated by a workshop attended by experts from academia, industry and regulatory bodies to explore opportunities for alternative approaches to overcome the challenges of the current animal models, and minimise animal use. Recommendations from the workshop have been incorporated into a hypothetical screen for emetic liability in which undesirable compounds would be screened out using *in vitro* and *in silico* methods, thus reducing the eventual use of animals. For example, nematodes and social amoeba have already been shown to respond to some substances that cause emesis in humans. The challenge is to understand whether these responses can be used to predict nausea and emesis in humans, as this could ultimately be exploited as a screen for emetic liability.
A review of the workshop is in press in the *British Journal of Pharmacology*. Further work is currently underway to explore the full potential of these solutions to reduce animal use and improve knowledge of nausea and emesis to provide faster and enhanced development of medicines.

A hypothetical screen for emetic liability



A stepwise approach to establishing emetic liability to ensure that only the most promising drugs are tested in animals (Modified from Holmes A, Rudd J, Tattersall D, Aziz Q & Andrews P (2009) Opportunities for the replacement of animals in the study of nausea and vomiting. *British Journal of Pharmacology – in press*).

STIMULATING CHANGE IN SAFETY AND TOXICITY TESTING

Animals are used in the pharmaceutical and chemical industries to test the effectiveness, safety and toxicity of drugs and chemicals prior to exposure in man and to the environment. There are opportunities to minimise this use by assessing the scientific rationale of the tests and the utility of the data obtained. The NC3Rs works with experts from companies, universities and regulatory bodies in the UK, elsewhere in Europe and the USA to apply new ideas and technologies to the use of animals in industry. The Centre's integrated approach has led to changes in practice and regulatory guidance and a demonstrable reduction in the use of animals in specific areas.

"Many people used to see work on alternatives to animals for toxicity testing of chemicals as an activity separated from mainstream research. The NC3Rs is facilitating the start of a real change to this perception — the 3Rs are now a legitimate part of the overall science agenda in the UK."

Dr Phil Botham, Head of Human Safety, Syngenta

Working in partnership with the chemical industry

Recent changes to European legislation regulating the safety of chemicals have significant implications for the use of animals. Under the Seventh Amendment to the Cosmetics Directive, the majority of animal tests on cosmetic ingredients are to be banned from 2009, with the ban extending to all tests on ingredients by 2013. Also, the 2007 REACH (Registration, Evaluation, Authorisation and restriction of CHemicals) regulation which covers the majority of chemicals may require evaluation of 30,000 existing chemicals, potentially requiring the use of large numbers of animals.

These regulatory changes have generated an urgent need for alternative approaches, both to meet the demands of the Cosmetics Directive and avoid an increase in animal testing for the purposes of REACH. While much progress has been made in development of non-animal alternatives for some short-term tests such as skin corrosion and eye irritation, replacement of animals for other purposes, such as detection of the wide range of potential adverse effects after longer-term exposure to a chemical, remains a significant scientific challenge. A number of organisations around the world have established initiatives in this area, including the NC3Rs.

The Centre's work with the chemical industry (i.e. industrial chemicals, plant protection products, cosmetics and consumer products) is progressed through its Regulatory Toxicology Forum. The Forum, which was set up in 2007, brings together practising toxicologists and regulators to discuss the 3Rs at both strategic and operational levels, to support the UK's position as a leading player in progressing the 3Rs in the regulatory arena in Europe. This work has received substantial support from the chemical industry, and funding for a scientific post at the NC3Rs to take forward initiatives proposed by the Forum has been provided by The Dow Chemical Company, SC Johnson, Syngenta and Unilever.

Given the large number of activities in this field internationally, the Forum has identified areas where the NC3Rs can provide added value over and above existing initiatives to improve the scientific basis of chemical risk assessment and implementation of the 3Rs.

One important theme identified by the Forum has been the potential for greater use of information on exposure to a chemical, to support the 3Rs in risk assessment. Risk assessment involves consideration of both the potential adverse effects of a chemical (i.e. the 'hazard') and the amount an individual is likely to be exposed to, and to date the focus of 3Rs initiatives has primarily been on the hazard side of this equation. The NC3Rs has therefore concentrated much of its efforts on exposure-related projects.

REACH and other legislation allow specific toxicity tests, usually conducted in animals, to be waived if it can be shown that exposure to humans or the environment is unlikely to be significant. However, there is only limited information on how justification for exposure-based waiving can be made in practice. Using inhalation toxicity as a test case, the NC3Rs has commissioned a scoping study to assess the amount and usefulness of available information that could inform exposure-based waiving opportunities. The findings of this study are now being developed for publication as guidance on the relevant options and information resources that can be helpful in building a robust case for the regulators which would allow the use of animals to be avoided.

Another project is focusing on assessment of internal exposure to a chemical (i.e. toxicokinetics) and its use in chemicals testing. Currently, the pharmaceutical industry uses toxicokinetic data to inform and reduce their animal studies but its use is less common in the chemical industry. Toxicokinetic information can be used in the selection of dose levels for animal tests, which helps to avoid the use of excessively toxic doses, resulting in less animal suffering. Improved dose selection can also help to avoid additional animal studies being triggered to understand the effects seen at these high doses.

To stimulate cross-sector discussion on whether this approach could be applied further to safety assessment in the chemical industry, and to consider 3Rs approaches to obtaining toxicokinetic data, the NC3Rs organised an international workshop on *'Toxicokinetics and the 3Rs'* in May.

Around 70 individuals from national and international pharmaceutical and chemical industries and regulatory authorities attended the event. To communicate the key points that emerged at the workshop and engage a wider audience, a review paper is currently in preparation to be submitted for publication in 2009.

Gathering evidence to support drug development

Further progress has been achieved with the NC3Rs partnership with the pharmaceutical industry, supported by renewed funding from the Association of the British Pharmaceutical Industry (ABPI) in 2008. This project has continued to expand and the NC3Rs is now working with scientists from 43 pharmaceutical and biotechnology companies and regulatory bodies from the UK, elsewhere in Europe and the USA to deliver a comprehensive programme of work in diverse areas of pharmaceutical discovery and development. Five expert working groups have been taking an evidence-based approach to explore and validate new opportunities to minimise the use of a variety of species including non-human primates (herein referred to as 'primates'), dogs and rodents.

During 2008, experts involved in the NC3Rs/ABPI initiative to minimise the use of primates have continued to share, generate and analyse data which supports the replacement and reduction of primate use in drug discovery and development in two areas; pharmacokinetic analysis for drug candidate selection and the safety testing of monoclonal antibodies. This has involved a range of approaches, including commissioning research and organising a workshop. The next steps for 2009 will be to publish and disseminate the results of this work – facilitating wider uptake in the scientific and regulatory communities.

Pharmacokinetic analysis for drug candidate selection

Understanding what happens to a drug when it is administered, known as pharmacokinetics, is an important aspect of the drug discovery process to identify drug candidates with an appropriate profile in humans. Many drugs are cleared from the body too quickly to have an effect and are therefore unsuitable for development. Analysis of pharmacokinetic data can prevent further investigation of such drugs and avoid unnecessary subsequent animal use.

Drug clearance is mediated by a number of mechanisms including elimination by the kidneys and enzyme-based metabolism. Opinion is divided across industry on the most appropriate method to predict human pharmacokinetics and currently a range of species, including primates, dogs and rodents, and *in vitro* approaches are used. In recent years, there has been a growing body of

literature supporting the use of the primate to provide this information. However, both published and unpublished data show that, for compounds that are cleared from the body by metabolism, alternative techniques using human cells can provide predictions with comparable accuracy to *in vivo* models, thus having the potential to replace the use of animals.

This year, the NC3Rs has been working with the pharmaceutical industry to devise and implement a research strategy which will provide data to support and increase the use of the *in vitro* methods. To increase confidence in the robustness and reproducibility of the methods, the Centre has commissioned research to generate *in vitro* metabolism data on a standard set of over 50 compounds which have previously demonstrated the utility of the primate. This new data set has been compared to equivalent *in vitro* data provided by Pfizer for the same compounds and

" The success of the NC3Rs can be seen by the number of company experts and regulators from around the world that become actively engaged in their programmes. The partnership with ABPI has been successful, because the organisation has listened to the challenges companies face and designed scientific programmes that both support the development of new medicines as well as driving the 3Rs."

Dr Richard Barker, Director General, ABPI

initial analysis indicates that the ***in vitro*** methods can provide enhanced information compared to that obtained from primates. The findings of the commissioned research have been presented at national and international conferences and are being prepared for publication in 2009.

Monoclonal antibodies

Over the last decade, monoclonal antibodies (mAbs) have been developed as therapies to treat a variety of diseases from cancer to rheumatoid arthritis. The advantages of mAbs over chemical entities, such as greater specificity for the target protein and slower elimination from the body, have been exploited in areas of unmet medical need to provide significant advances in treatments. This success, combined with the ability of mAbs to target proteins deemed intractable by chemical means, has led to significant investment in the development of biotherapeutics such as mAbs by the pharmaceutical and biotechnology industries.

Monoclonal antibodies are highly specific for both their target and species, and this has an impact on the choice of species used in preclinical testing to predict safety in man. Primates are often the only relevant species for testing and with the rapidly increasing number of mAbs in the pipeline this means that the use of these animals is rising. To address this, the NC3Rs has been working with the industry for the last two years to develop alternative, scientifically robust, approaches to minimise the use of primates.

The project was initiated by the NC3Rs in 2006 with a workshop to investigate how mAbs could be safely developed without the use of primates. A hypothetical mAb development pathway produced at the workshop was published in 2007 [1] and the Centre has subsequently been focusing on collecting and analysing data to validate the pathway. Working with experts from 15 international companies and regulatory bodies, the NC3Rs has acted as an independent broker for data sharing across the industry. Data has been shared on over 100 mAbs including information on study design and the use of rodents rather than primates. Based on these data, the working group has focused on minimising primate use in chronic toxicity studies, including reproductive toxicity.

In order to share the working group's findings and proposals with a wider scientific audience the NC3Rs held an international workshop in June 2008. Discussions on reproductive toxicity studies concentrated on whether they are always necessary, whether rodents could be used instead of primates, and whether study designs which reduce primate use provide sufficient safety information. Opportunities for minimising primate use in other chronic toxicity studies were also discussed, including whether all currently used dosage levels are necessary and questioning the need for studies of longer than six months.

"Opportunities exist for industry to think creatively about science-based biotherapeutic development. The NC3Rs has challenged some traditional approaches to safety assessment and opened up avenues of alternatives to primate use. This dialogue has resulted in thoughtful reduction of primate use and must continue in order to have an impact globally."

Dr Laura Andrews, Vice President of Pharmacology and Toxicology, Genzyme

1: Chapman K, Pullen N, Graham M, Ragan I (2007) Preclinical safety testing of monoclonal antibodies: the significance of species relevance. *Nature Reviews Drugs Discovery* **6** (2), 120-126

An end to acute toxicity testing in drug development

A collaboration between 18 European pharmaceutical companies and contract research organisations, coordinated by the NC3Rs, to review the value of acute toxicity data has continued to develop in 2008, including influencing the revision of the International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) M3 guidelines, gathering data to challenge the use of acute toxicity studies to provide information on overdose in man, and refining the use of maximum tolerated dose (MTD) studies. The success of this collaboration has been highlighted by the launch of an equivalent project on acute toxicity testing of chemicals by the European Partnership for Alternative Approaches to Animal Testing.

Acute toxicity studies have historically been carried out in animals in order to satisfy regulatory guidelines, prior to new medicines being administered in man. Single dose acute toxicity is the only test in pharmaceutical development where death of the animal is an endpoint. This initiative has reviewed the utility of single dose acute toxicity tests in informing drug development and has resulted in a 70% reduction in animal use for acute toxicity testing (equating to approximately 15,000 animals per year) in the companies involved in the collaboration.

Data sharing between the companies has shown that single dose acute toxicity testing is of limited value in assessing safety in humans and the information needed can be obtained from other less harmful tests which are already carried out as part of the drug development process. This finding was published in early in 2008[1], attracting attention from the general and specialist media in the UK and elsewhere.

Even without a change to the regulatory requirements this initiative has led to a measurable reduction in animal use. However, for this impact to be realised worldwide regulatory change is required. In 2008, draft revisions to the ICH M3 guidelines were published. Importantly these incorporated the recommendations of the NC3Rs/industry partnership and referenced its publication. Since ICH brings together all of the regulatory bodies worldwide, the revision which is expected to be adopted in 2009, should result in an end to single dose toxicity testing for pharmaceuticals to support first time use in man. The European Medicines Agency (EMEA) draft position paper on acute toxicity studies has also incorporated the recommendations made by the working group.

Avoiding the use of single dose toxicity testing by using data already generated in the drug development process, without the need for lethality, represents a significant step in reducing animal use. Data on acute toxicity can be extrapolated from studies in animals which determine the MTD of a drug. These studies are less harmful to the animals used, nevertheless, there is scope to further refine animal welfare. To explore this, the NC3Rs/industry partnership has been sharing data on the clinical endpoints used in different therapeutic areas and the identification of early and objective indicators for MTD studies, in order to develop better humane endpoints.

1: Robinson S ***et al*** (2008) A European pharmaceutical company initiative challenging the regulatory requirement for acute toxicity studies in pharmaceutical drug development. ***Regulatory Toxicology and Pharmacology* 50** (3), 345-352

"Changing the regulatory guidelines for acute toxicity testing seemed unachievable at the outset. Having the NC3Rs as a neutral and scientific forum for data sharing, hosting discussion, and widely disseminating the work was a key factor in our success."

Dr Sally Robinson, Principal Toxicologist, Global Safety Assessment, AstraZeneca

ACUTE TOXICITY PROJECT — KEY STEPS SINCE 2003

March 2003
Presentation to ABPI on a proposal to establish a cross-industry working group and to call for members

June 2003
First meeting (6 member companies)

Presentation to EFPIA to call for additional members

April 2004
Initial questionnaire on study design shared

June 2004
Outcome of initial questionnaire presented at 9th FELASA Symposium

January 2005
Seventh meeting (15 member companies)

Agreed to share specific compound data

August 2005
Presentation of findings from compound data sharing at 5th World Congress of Alternatives and Animal Use in the Life Sciences

September 2005
Presentation to EFPIA to gather support for findings

December 2005
Presentation of findings from compound data sharing to EMEA Safety Working Party and discussion of possibility of raising as a topic for ICH M3 revision

March – June 2006
Data collected from 6 additional US companies

Presentation to PhRMA to solicit support for findings

October 2006
Leaflet detailing the work disseminated for use at conferences and for contract research organisations to use with companies not involved in the working group

November 2006
Workshop held with regulators from UK, Europe, Japan and the USA

March 2007
Thirteenth meeting (18 member companies)

Presentation at 10th FELASA Symposium

Discussions with those involved in revision of ICH M3

January 2008
Press release highlighting publication of paper in *Regulatory Toxicology and Pharmacology*

April 2008
Response submitted to ICH M3 to support regulatory recommendations

June 2008
ICH M3 at step 2 and EMEA draft concept paper on acute toxicity both including citations to publication

December 2008
Questionnaire to European poison centres to collect information on the value of acute toxicity studies for overdose

VIEWS ON THE 3RS

The NC3Rs has conducted a survey to provide information on how scientists who use animals understand and implement the 3Rs. This is the first survey of scientists to focus on the 3Rs and the results provide information for benchmarking progress and the development of the Centre's strategy. The results of the survey, carried out by People, Science and Policy, were collated in 2008 and highlight some interesting themes.

The use of animals in scientific procedures in the UK is regulated by the Animal (Scientific Procedures) Act 1986 (ASPA). In England, Wales and Scotland, the Act is administered by the Home Office via a three-level system of licences which cover the person carrying out the regulated procedures (the personal licence), the programme of work (the project licence) and the establishment at which the procedures are undertaken (the certificate of designation).

The 3Rs are not referred to as such in the ASPA [1], but they are implemented through requirements that a project licence cannot be granted unless the work *'...cannot be achieved satisfactorily by any other reasonably practicable method not entailing the use of protected animals'* and the procedures *'...use the minimum number of animals, involve animals with the lowest degree of neurophysiological sensitivity, cause the least pain, suffering, distress or lasting harm, and are most likely to produce satisfactory results'*.

Therefore, under the regulatory framework, a project licence cannot be granted unless the 3Rs have been considered and are implemented. Personal licence holders are responsible for the welfare of the animals on which they have performed regulated procedures, and the application of the 3Rs is integral to ensuring this responsibility is put into practice.

Development and uptake of the 3Rs is also supported at the institutional level by the Ethical Review Process (ERP) which was introduced by the Home Office in 1999. Scientists with project and/or personal licences (and those who support and advise them) are a main audience for the NC3Rs and understanding their level of awareness and use of the 3Rs, and perceived barriers to implementation, provides a basis for informing the Centre's strategy and a baseline for measuring attitudes over time.

The opinion survey was conducted between July and October 2007 using an online questionnaire targeted at project and personal licence holders. The questionnaire was developed following in-depth interviews with licence holders and was overseen by an expert steering group. It was distributed to all designated establishments via the Home Office's Certificate Holder's circular. Although this was identified as the most expedient distribution method, circulation to licensees was dependent on subsequent dissemination by the Certificate Holder. Responses were received from 1,955 licensees which is a response rate of approximately 14% of all possible respondents. The data gathered was weighted to reflect the overall distribution of licensees using information provided by the Home Office. A majority of those who responded (76%) were scientists (most of the remaining 24% were animal care staff); 37% of the scientists were project licence holders and 94% were personal licence holders. 74% of the scientists worked in academia and 15% in industry (most of the remaining 11% worked for Government bodies).

The survey highlights a number of trends among scientists who hold project and/or personal licences which are summarised below.

- Generally there is a good understanding of the definitions of the 3Rs. However, there is confusion over the definition of refinement with 52% of scientists incorrectly defining it as improving experiments to yield better data [2].
- The modular training courses required to attain a project and personal licence often provide the first introduction to the 3Rs for scientists (57%).
- More than four out of ten of the scientists (43%) consider all three 'R's to be equally relevant to their work, with the majority indicating that implementation of the 3Rs would not be detrimental to the quality of their results (82%).
- The majority of scientists (73%) do not think the use of animals can ever be completely replaced, with 77% of those involved in designing experiments indicating that nothing would allow them to address their research objectives without the use of animals.
- Almost all scientists consider the 3Rs when designing and carrying out experiments (95%), however, this figure declines significantly to 26% when writing up findings for publication.
- Less than half the scientists find the ERP helpful in replacing (31%), reducing (42%) or refining (46%) animal use.
- Most scientists identify data sharing or collaboration between research groups (77%) or companies (60%) as factors which would allow fewer animals to be used.
- Lack of scientific or technological innovation is seen as the main obstacle to implementing the 3Rs by 33% of scientists, with only 6% identifying insufficient funding as a factor.
- Approximately, one-third of scientists have developed techniques that reduce (34%) or refine (30%) the use of animals. This falls to 12% for replacement techniques.
- Only 6% of scientists have applied for 3Rs research funding. The main reasons for not doing so are not having an opportunity to do so (48%), lack of relevance to research interests (31%) and unfamiliarity with funding sources (25%).

Further information can be found at **www.nc3rs.org.uk/opinionsurvey**

1: They are, however, referred to in the Guidance on the Operation of the ASPA

2: Respondents were given the correct definitions of the 3Rs before continuing with the survey

LOOKING AHEAD TO 2009

The upcoming year marks an important milestone, the anniversary of the publication of Russell and Burch's 'The principles of humane experimental technique' which first described replacement, reduction and refinement. In the intervening years since the publication in 1959, the 3Rs have become widely adopted as a legal and ethical framework for the use of animals in research and testing in the UK and elsewhere. In more recent years, polls of public opinion in the UK have repeatedly demonstrated that support for the use of animals is conditional on the application of the 3Rs, and the latest independent research, commissioned by the Government, shows that there is increasing support for investment in alternatives to the use of animals. It is against this back-drop of regulation, ethics and public opinion that the NC3Rs was launched and, indeed, 2009 marks another important date — the Centre's fifth anniversary.

After 50 years of the 3Rs we have an opportunity for reflection and celebration, and we are very pleased to be joining forces with the Fund for the Replacement of Animals in Medical Experiments to sponsor a meeting in Nottingham to do so with many of those who have played leading roles in championing the 3Rs. However, our main focus will be to use the anniversary to look forwards and to stimulate new interest and fresh ideas. In 2009, we will be launching a new annual studentship scheme using our new Government funding, to embed the ethos of the 3Rs in tomorrow's research leaders from the start of their careers. With funding available for five places each year we are confident that we will quickly build up an impressive portfolio of talent and research. We are also delighted to be working with the New Scientist magazine on an essay competition to stimulate original and imaginative ideas from young scientists on how the 3Rs might be advanced in the next 10-20 years. The magazine's worldwide readership of over 800,000 provides a significant new pool of potential ideas and entrants for the prize, which will include the publication of the winning essay in the New Scientist.

Since 2004, we have been committed to ensuring that the NC3Rs occupies a unique position in the 3Rs field. Our goal has been to put the 3Rs at the heart of the life sciences — driving and exploiting the use of new technologies to maximise opportunities to not only replace, reduce and refine animal use, but to support science and innovation in the UK. We have started to move the 3Rs agenda away from the rhetoric and towards the delivery of benefits, and it is this approach that has gained support from the scientific community and provided credibility to the 3Rs as a desirable research goal and output. This year we have seen real impact of the work we have been leading and funding. However, as the Centre approaches its fifth year it is important to evaluate our strategy and approach, and to do this there will be a review of the NC3Rs in 2009. This will provide us with an opportunity to re-examine what we have achieved and plan for the future, as we develop our business case for the next Government spending review.

Dr Vicky Robinson, Chief Executive

FINANCIAL SUMMARY

This annual report describes the NC3Rs activities for the calendar year 2008. The NC3Rs accounting period, however, runs from 1 April to 31 March each year. The information provided below covers the period 1 April 2007 to 31 March 2008.

FOR THE YEAR ENDED 31 MARCH 2008

The total income for this financial period was £2,891,904; 24% higher than the period April 2006 to March 2007. This largely reflects additional funding from the MRC and BBSRC. Funding from industry also increased.

The annual budget is agreed by the NC3Rs Board. Total expenditure increased from £1,540,917 in 2006/07 to £2,171,709 in 2007/08. This can primarily be accounted for by increased grant expenditure and programme costs.

Board costs include travel for members to meetings and associated honorariums. In the period April 2007 to March 2008, Board costs were £25,388; 162% higher than in the previous financial year. This was a result of recruitment costs (e.g. advertising) for the new NC3Rs Board chairman.

This year categorisation of expenditure for programme and operating costs has been revised to ensure greater transparency. Programme costs now cover initiatives led by the NC3Rs Office. This includes costs for workshops, symposia, working groups and the salaries of staff that lead and support these initiatives. In the period April 2007 to March 2008, expenditure on programme costs was £715,862; 22% higher than in the previous financial year. This reflects the cost of ongoing and new initiatives. Operating costs now include staff salaries for core administrative duties, staff travel and training, recruitment, stationery and publishing costs. In the period April 2007 to March 2008, expenditure on operating costs was £150,036; 18% higher than in the previous financial year.

Grant expenditure was £1,280,423 in the period April 2007 to March 2008; 57% higher than in the previous financial year. This reflects the ongoing expenditure of grants awarded in 2004, 2005, 2006 and 2007. Note that grants are awarded for a period of up to three years. Funding committed but not yet spent on grants is carried forward.

An independent accountant oversees the management of the NC3Rs finances. For logistical reasons the NC3Rs uses the MRC accounting systems and is subject to its auditing procedures. The NC3Rs is grateful to the MRC for generously providing office space and infrastructure support including IT, payroll and personnel services.

Income

	2007/08 (£)	2006/07 (£)
Government	2,630,000	2,079,000
Charity	62,904	98,065
Industry	199,000	148,850
Total	**2,891,904**	**2,325,915**

Expenditure

	2007/08 (£)	2006/07 (£)
Board costs	25,388	9,701
Programme costs	715,862	588,024
Operating costs	150,036	127,539
Grant costs (includes Small Awards)	1,280,423	815,653
Total	**2,171,709**	**1,540,917**

Research funding expenditure

Financial year	Total awarded (£)	Total spent spent (£)
2004/05	523,148	118,379
2005/06	988,425	268,990
2006/07	1,467,222	815,653
2007/08	2,467,711	1,280,423

BOARD, PANEL MEMBERS, AND STAFF

NC3Rs Board

Professor Ian Kimber (Chair)
University of Manchester

Dr Vicky Robinson
NC3Rs

Dr Julia Fentem (Deputy Chair)
Unilever

Professor Paul Flecknell
Newcastle University

Dr Lesley Heppell
BBSRC

Professor Jane Hurst
University of Liverpool

Dr Maggy Jennings
RSPCA

Dr James Kirkwood
UFAW

Dr Tony Peatfield
MRC

Professor Nancy Rothwell, DBE
University of Manchester

NC3Rs 3Rs prize selection panel for 2008

Professor Ian Kimber (Chair)
University of Manchester

Professor Michael Festing
University of Leicester

Professor Paul Flecknell
Newcastle University

Dr Stella Hurtley
Science

Dr Declan Mulkeen
MRC

Professor Clive Page
King's College London

Dr Ian Ragan
CIR Consulting Limited

Professor Patrick Vallance
GSK

Sir Mark Walport
The Wellcome Trust

Professor Paul Wiles
Home Office

NC3Rs grant assessment panel for 2008

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University of Manchester

Professor Innes Cuthill
University of Bristol

Professor Andrew Derrington
University of Kent

Professor Paul Flecknell
Newcastle University

Dr Jeffrey Fry
University of Nottingham

Professor Jane Hurst (Deputy Chair)
University of Liverpool

Professor Ian Kimber
University of Manchester

Professor Sheila MacNeil
University of Sheffield

Mr Terry Priest
University of Manchester

Dr Alan Teale
Retired (formerly University of Stirling)

Professor Dominic Wells
Imperial College London

Dr Carl Westmoreland
Unilever

NC3Rs staff

Dr Vicky Robinson
Chief Executive

Programme Managers

Dr Anthony Holmes	Replacement
Miss Carol Kilkenny	Reduction
Dr Mark Prescott	Refinement
Dr Kathryn Chapman	3Rs and the pharmaceutical industry
Dr Alison Cook (until July 2008)	3Rs and the chemical industry
Dr Stuart Creton (from August 2008)	3Rs and the chemical industry
Dr Harriet Warburton	Research Funding
Mr Tim Watson	Communications
Ms Ashley Scott	Operations Manager

ACKNOWLEDGMENTS

The NC3Rs would like to thank the following organisations for their generous financial support:

- **MRC**
- **BBSRC**
- **Home Office**
- **Wellcome Trust**
- **ABPI**
- **GlaxoSmithKline**
- **AstraZeneca**
- **Unilever**
- **The Dow Chemical Company**
- **SC Johnson**
- **Syngenta**
- **LASA**

The NC3Rs would also like to thank Harlan for their provision of animals for NC3Rs grants.

The NC3Rs would like to thank the following Board Members whose term ended in 2008:

Dr Bryan Howard
LASA

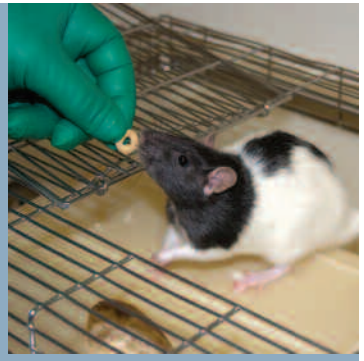
Dr Jon Richmond
Home Office

Professor Malcolm Rowland
University of Manchester

Dr David Smith
AstraZeneca

GLOSSARY

ABPI	Association of the British Pharmaceutical Industry
AMRC	Association of Medical Research Charities
ASPA	Animal (Scientific Procedures) Act 1986
BBSRC	Biotechnology and Biological Sciences Research Council
Defra	Department for Environment, Food and Rural Affairs
EFPIA	European Federation of Pharmaceutical Industries and Associations
EMEA	European Medicines Agency
ERP	Ethical Review Process
FELASA	Federation of European Laboratory Animal Science Associations
ICH M3	International Committee on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use – Topic M3
LASA	Laboratory Animal Science Association
MRC	Medical Research Council
MTD	Maximum Tolerated Dose
NERC	Natural Environment Research Council
PhRMA	Pharmaceutical Research and Manufacturers of America
REACH	Registration, Evaluation, Authorisation and restriction of CHemicals



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