Chapter 8

The use of animals for research in the pharmaceutical industry

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Introduction

8.1 The pharmaceutical industry conducts or supports approximately one third of the animal research that is undertaken in the UK. Some of this is basic research that seeks to examine normal biological processes and the nature of disease (see also Chapters 5 and 6). However, most has more specific, applied objectives and concerns the development of new medicines or vaccines, improved diagnosis or better methods of toxicity testing. Since the process of producing medicines has changed significantly over time, we begin with a brief overview of developments from the late 19th century to the present. We then describe the way medicines are currently produced in terms of eight stages. These are: discovery and selection of compounds that could be effective medicines (stages 1 and 2), characterisation of promising candidate medicines (stages 3 and 4), selecting candidate medicines and ensuring their safety (stage 5), clinical studies on humans (stages 6 to 8), and also research carried out to support the medicine once it has been marketed. For each stage we describe the way in which animals are used in the process, and give some examples of specific experiments. As in the case of research described in the previous chapters, welfare implications for the animals involved in pharmaceutical research are as diverse as the types of research and must be considered on a case by case basis. In this chapter we focus on the use of animals on the development of medicines for use in humans. We also consider briefly vaccines[1] and veterinary medicines.

The development of the pharmaceutical industry

8.2 The modern pharmaceutical industry has its origins in the chemical industry of the late 19th century and the first half of the 20th century. A first peak of research activity concerned the development of treatments for war injuries and infectious diseases arising from mass migrations during and after the First World War. During the Second World War and subsequently, a much more systematic approach to the discovery of new medicines led to a significant increase in both medical discovery[2] and industrial activity.[3]

8.3 Early pharmaceutical research drew on existing animal models that were used in experimental physiology, extending established scientific traditions of using animals in research. New potential medicines were not directed at a specific target such as a cell receptor, as they are today. Rather, the effect of medicines was measured in relation to the general physiological response of an animal, such as changes in blood pressure. This method of screening for potentially beneficial effects of medicines used large numbers of animals, and was inefficient and cumbersome. As pharmaceutical research expanded in the 1950s and 1960s, the use of animals expanded in parallel. In the 1980s, novel techniques, improved facilities, computer technology and new materials became available and were integrated into the research and development process. The use of alternatives to solely animal-based research and development, such as cultured cells, also expanded.

1 See World Health Organization (2003) *State of the Art of New Vaccines: Research and development* (Geneva: WHO), available at: http://www.who.int/vaccine_research/documents/en/stateofart_excler.pdf. Accessed on: 29 Apr 2005.

2 HistoryWorld *Combined Medical Timeline (Wellcome Trust)*, available at: http://www.historyworld.net/timelines/timeline.asp?from=existing&D=1925&selection=&tid=yocb&title=Combined%20Medical%20Timeline&back=existing.asp. Accessed on: 26 Apr 2005.

3 Corley TAB (1999/2000) *The British Pharmaceutical Industry Since 1851*, available at: http://www.rdg.ac.uk/Econ/Econ/workingpapers/emdp404.pdf. Accessed on: 26 Apr 2005.

8.4 From the late 1980s these developments continued to transform pharmaceutical research and development. Information technology became more efficient, allowing the integration of rapidly expanding amounts of data generated by advances in basic biological knowledge. This information was integrated with data from new technologies such as high-throughput chemistry and biology, genomics, pharmacogenetics, advanced diagnostic imaging and the application of bioinformatics. Since the 1980s, the continued expansion of pharmaceutical research in the UK has also been accompanied by the increasing use of a wide range of modern methods, which we describe below (see Figure 8.1).[4] The use of these methods was one factor that contributed to the decrease in animals involved in commercial research during the same period, from 60 percent (or 2.1 million) of the total number of procedures in 1987, to 36 percent (or 1 million) of the total in 2003.[5]

Capture the potential of the human genome industrialisation of drug discovery

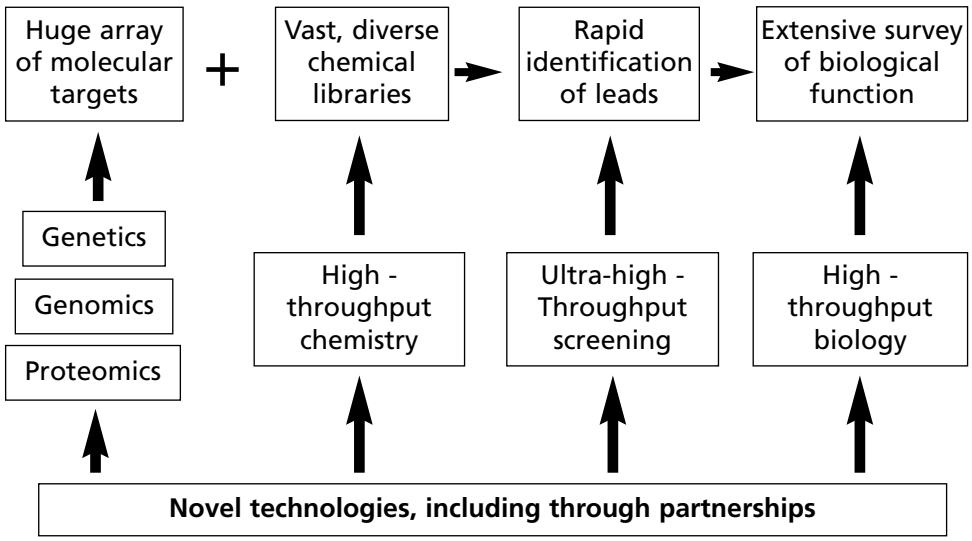

Figure 8.1: The industrialisation of drug discovery to capture the potential of knowledge of the human genome

Source: GlaxoSmithKline

Use of animals in current pharmaceutical research and development

8.5 The discovery and development of new medicines[6] entails a very complex range of different methodologies (Figures 8.1 and 8.2). The process, undertaken primarily by the pharmaceutical industry, takes an average of 10–15 years.[7] In this section, we describe it in terms of eight stages,

4 The Association of the British Pharmaceutical Industry (ABPI) *The Development of Medicines*, available at: http://www.abpi.org.uk//publications/briefings/Dev_Medicines.pdf. Accessed on: 26 Apr 2005.

5 This figure includes the use of animals by companies outside of the pharmaceutical sector, for example in toxicity testing and ecotoxocity testing of products that might have an impact on the environment. See Home Office (2004) *Statistics of Scientific Procedures on Living Animals Great Britain 2003* (London: HMSO), p22. In financial terms, the use of animals is only a small part of the total required to produce a licensed new medicine. Estimates of animal cost do not usually exceed five percent. See 71st Stephen Paget Memorial Lecture, available at: http://www.rds-online.org.uk/pages/news.asp?i_ToolbarID=6&i_PageID=176. Accessed on: 26 Apr 2005; AstraZeneca (2003) *Take a Walk Along the Path to a New Medicine*, available at: http://www.astrazeneca.com/sites/7/imagebank/typeArticleparam502178/seeking_new_medicines_v15.html. Accessed on: 26 Apr 2005.

6 We use the term discovery to refer to research that aims to find novel connections between diseases, molecular targets and well-characterised therapeutic interventions. We use the term development to refer to laboratory and animal studies designed to test the mechanisms, safety and efficacy of an intervention prior to its applications to humans (pre-clinical development) and to trials involving human participants to determine further the safety and efficacy of potential drug candidates (clinical development).

7 Network Science (2004) *The Process of Drug Development*, available at: http://www.netsci.org/scgi-bin/Courseware/projector.pl?Course_num=course1&Filename=top.html. Accessed on: 26 Apr 2005.

beginning with target identification and ending with the launch of the new product (see Table 8.1).[8] Data from animal research are crucially important to researchers in the pharmaceutical industry when deciding whether a potential medicine will be effective and safe for use in humans.[9]

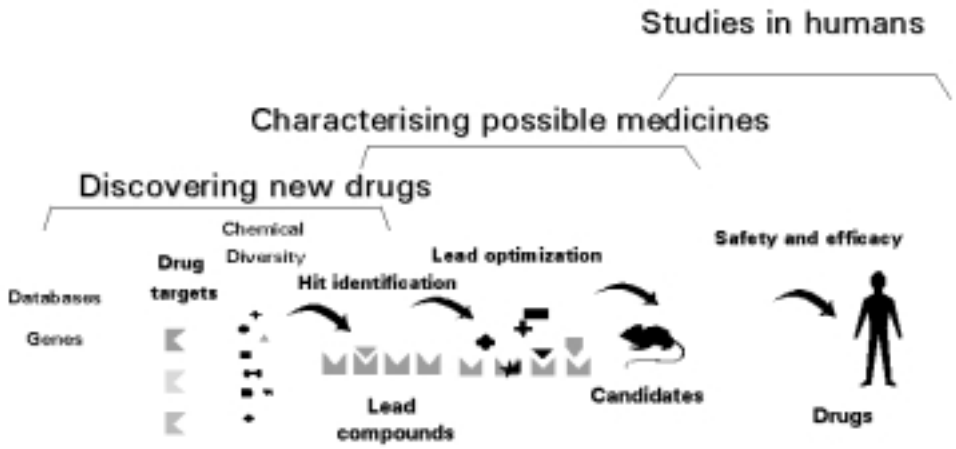

Figure 8.2: Overview of the activities involved in modern drug discovery and development

Source: GlaxoSmithKline

Table 8.1: Overview of the process of discovery and development of medicines

Objective	Stage no.	Description	Average number of compounds entering each stage	Average use of animals
Discovery and selection of potential new medicines	1	Target identification	–	5–15%
	2	Identification of possible medicines	1,000,000	
The characterisation of promising candidate medicines	3	Lead identification	1,000	60–80%
	4	Lead optimisation	200	
Ensuring the safety of selected candidates	5	Selecting candidate medicines	17	10–20%
Clinical studies on humans	6	Concept testing	12	Generally none
	7	Development for launch	9	Generally none
	8	Launch phase	2.2	Generally none

8 See AstraZeneca (2003) *Take a Walk Along the Path to a New Medicine*, available at: http://www.astrazeneca.com/sites/7/imagebank/typeArticleparam502178/seeking_new_medicines_v15.html. Accessed on: 26 Apr 2005.

9 Samuels G (2003) *Medicines: Tried And Tested - In Animals?*, available at: http://www.abpi.org.uk/publications/publication_details/mttur/mttur_ani.asp. Accessed on: 26 Apr 2005.

Stages 1 and 2: discovery and selection of compounds that could be effective medicines

8.6 Early stages of the discovery process can be divided into two stages. Stage 1 involves target identification (seeking, for example, to identify receptors for active molecules), and stage 2 relates to the identification of possible medicines. Both stages make use of advances in genetic and basic biological research, and of new, automated technologies including:[10]

- **high-throughput chemistry**: systematic exploration of the diversity of chemical structures to increase the number of possible candidates; the aim is to produce a shortlist of novel molecules that have the potential to be safe and effective medicines;
- **ultra**[11]**-high-throughput screening**: automated analysis of a very large number of novel molecules in cell-based *in vitro* assays, which are analysed by automated systems using advanced robotics;
- **high-throughput biology**: technologies such as automated administration of medicines and automated blood collection via catheters into blood vessels, which then allow a more rapid and detailed analysis of the full range of effects in whole animals.

The very large amounts of data generated from these new methods are then integrated and analysed further by means of statistical and computational methods.

Stage 1: target identification

8.7 The search for new medicines begins by focusing on areas that are of potential interest to pharmaceutical companies. These include medicines that can be used to address unmet medical needs (for example, Alzheimer's disease), interventions against diseases that affect a great number of people, such as malaria or HIV/AIDS, medicines that are sometimes referred to as 'lifestyle drugs', such as Viagra or Propecia,[12] and improvements to existing medicines.[13] Pharmaceutical companies also sometimes seek to develop new medicines even if the medical need is already met because there appears to be access to a profitable share of the market.

8.8 Effective medicines maximise their effect on a specific biological pathway and minimise effects on all other pathways. The identification of useful *targets*, such as disease-associated genes or proteins that function as receptors for active molecules of new medicines, is therefore crucial. Information from the sequencing of the human and animal genomes is also important for the identification of disease mechanisms and for understanding how a person's genes can affect both disease processes and their responses to medicines.[14]

Stage 2: identification of possible medicines

8.9 In the next stage, compounds that might interact with the selected targets are submitted for high-throughput screening (or HTS), which is the automated testing of tens or even hundreds of thousands of compounds in a systematic way using cell based in vitro assays. Compounds or 'hits' that are judged to be the most interesting are then examined further. At the start of the process there is an average of one million compounds; at the end, numbers have decreased to about 1,000.

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10 AstraZeneca (2003) *Enabling technologies*, available at: http://www.astrazeneca.com/article/11177. GlaxoSmithKline (2003) *New Automated Approach will Transform Discovery Research at GSK*, available at: http://science.gsk.com/news/features/030613-tres.htm; GlaxoSmithKline (2003) *GSK Progresses Its Plan to Automate Discovery Research Processes*, available at: http://science.gsk.com/news/features/031021-hlw.htm. Accessed on: 26 Apr 2005.

11 The prefix 'ultra' refers to the very high throughput enabled by miniaturisation and automation.

12 Viagra was developed to treat impotence. Propecia is intended to help patients who suffer from baldness.

13 See paragraphs 3.13, 14.40, 14.58 and 15.83 for a brief discussion on similar medicines, sometimes known as 'me-too' drugs.

14 See Nuffield Council on Bioethics (2003) *Pharmacogenetics: ethical issues* (London: NCOB).

Use of animals

8.10 The molecules that are studied in stages 1 and 2 are screened against animals, animal tissues and cloned human receptors. The numbers of animals involved are small, probably less than ten percent of the total number used in pharmaceutical research.[15] Animal tissues are used for some *in vitro* tests, but cloned human receptors are preferred as these are more selective. GM mice are most commonly used to assess the importance of a drug target by examining the effects of deleting genes responsible for the synthesis of proteins such as receptors or other potential drug targets. The way in which the welfare of these animals is affected depends on the precise nature of the genetic modification that has been applied. Phenotypic effects may range from a lack of detectable changes to stunted growth and developmental abnormalities, and early death (paragraphs 4.57–4.58). Assessments need to be on a case by case basis as it is difficult to make generalisations.

Vaccines

8.11 Advances in genomic research have had a significant impact on the use of animals in the vaccine discovery process, often reducing the number involved or, leading to the replacement of animals such as primates with genetically modified mice.[16] Bacterial and viral genomes have been sequenced and potential vaccine targets are tested in high-throughput screening. The main difference in comparison to drug development is that the potential medical product under test is usually not an inorganic chemical molecule, but a biological product such as a fragment of a virus. Mapping of the human genome has also allowed the discovery of biological products that may eventually protect from, or even treat, diseases such as cancer.[17]

Stages 3 and 4: the characterisation of promising candidate medicines

8.12 In stages 3 and 4 the pharmacological properties of potential medicines are characterised more fully. These techniques combine use of non-animal approaches such as computer studies and analysis, chemistry and cell culture, with animal-based techniques such as advanced surgery, behavioural analysis, imaging such as MRI, and tissue and body fluid analysis (see paragraphs 4.53–4.56). New technologies such as telemetry now allow much more information to be obtained from each animal. For example, data from multiple measurements of physiological parameters such as heart rate or levels of neurotransmitters can be combined. With regard to welfare, post-operative pain can be controlled by pain relieving medicines, but sometimes they may interfere with experiments on pain and may not be given (see Box 8.3). The choice of pain relieving medicine can therefore be critical. Occasionally, distress can also be caused by devices used in telemetry (see paragraph 4.56).[18]

Stage 3: identification of 'leads'

8.13 Potential drug compounds ('hits') that have been identified by means of high-throughput screening are further examined in this stage, commonly using more complex cell cultures or assays based on animal or human tissue. The number of compounds entering this phase is usually in the hundreds. Through 'hit-to-lead chemistry', these hits are converted into a

15 The statistics collected by the Home Office do not include these data and companies vary in how they implement the various stages, making this figure difficult to estimate.

16 See The Associate Parliamentary Group for Animal Welfare (2005) *The Use of Animals in Vaccine Testing for Humans*, p21, available at: http://apgaw.org/userimages/Vaccinetesting.pdf. Accessed on: 26 Apr 2005; see also paragraph 6.35.

17 For example, see Berthet FX, Coche T and Vinals C (2001) Applied genome research in the field of human vaccines *J Biotechnol* 85: 213–26.

18 Morton DB, Hawkins P, Bevan R *et al*. (2003) Seventh report of the BVAAWF/FRAME/RSPCA/UFAW Joint Working Group on Refinement: Refinements in telemetry procedures *Lab Anim* **37**: 261–99.

significantly lower number of compounds known as 'leads'. Lead compounds are chemicals that influence the target in a way that indicates that they have high potential to be developed into effective treatments.

Stage 4: lead optimisation

8.14 Lead compounds are further refined by synthetic chemical modification, leading to the identification of a subset of the compounds that fulfil the requirements for clinical usefulness.[19] Animal and non-animal techniques are used to test for attributes such as absorption, duration of action and delivery to the target. The results determine whether the lead compounds have the potential for subsequent testing in human trials, and therefore the qualities to become candidates for medicines.

Use of animals

8.15 Most of the animals used by the pharmaceutical industry are involved in stages 3 and 4, comprising up to 80% percent of the total. Some techniques, such as methods for administering a medicine and measuring the level in blood, are generic for all types of research and testing (see paragraphs 4.31–4.59), but specific animal models of disease are used in particular areas of research. For example, one model may be used to identify targets for compounds to treat acute tissue damage after a stroke, whereas another may seek to find targets relevant to long-term recovery from a stroke (see Box 8.2). As we have said, an animal need not share all properties of humans to be an effective model. It is sufficient for the model to be similar in relevant aspects of the disease being studied (see paragraph 4.10).

8.16 The involvement of GM animals, usually mice, during stages 3 and 4, is becoming increasingly common. They are generally used either to determine if a gene is important as a target (target validation) or, once its importance is known, as a much more specific animal model of a disease.[20] Some tests of bioavailability (the degree or rate at which a medicine or other substance is absorbed or becomes available at the intended site in the body after administration), drug disposition and pharmacogenetic models[21] may also be used in a more limited way at this stage.

19 Physico-chemical, pharmacokinetic and toxicological properties are important criteria in assessing potential clinical usefulness.

20 Wellcome Trust (2003) *Transgenic mice*, available at: http://www.wellcome.ac.uk/en/genome/technologies/hg17b012.html Accessed on: 26 Apr 2005.

21 See MacGregor JT (2003) The future of regulatory toxicology: impact of the biotechnology revolution *Toxicol Sci* **75**: 236–48.

Box 8.1: The characterisation of promising candidate medicines (stages 3 and 4): example of animal research undertaken during of the development process of a new medicine

Jin, Q, Nie H, McCleland BW *et al*. (2004) Discovery of potent and orally bioavailable N,N'-diarylurea antagonists for the CXCR2 chemokine receptor *Bioorg Med Chem Lett* 14:4375-8.*

The aim of this research was to test the ability of a series of compounds to bind to the CXCR2 chemokine receptor (thus blocking its function). CXCR chemokines are signalling molecules that play an important role in transporting neutrophils (a type of white blood cell) to sites of inflammation in disease processes involved in arthritis, asthma and reperfusion injury (where the body's attempt to restore blood flow to an injury causes damage by oxidation).

A non-animal *in vitro* assay was used to identify compounds which may bind to the CXCR receptor. Six compounds were identified and their affinity for the CXCR2 receptor, as well as their effect in a living body, was investigated. The degree of binding to the CXCR2 receptor was then assessed in cell lines originally derived from the kidneys of Chinese hamsters.

In a further test, the compounds were injected into groups of three rats. This was first done intravenously and then, in a later experiment, injected into the peritoneal cavity. This experimental format is designed to both reduce the number of animals used and experimental variation. At various intervals after administration of the compounds, blood samples were taken from the lateral tail vein of the rats. Further *in vitro* studies using components of rat and human liver cells were carried out to investigate the way that the liver metabolises these compounds. These cells were obtained from euthanised rats and from human tissue which had been removed during surgery. The research yielded a new class of CXCR2 compounds that are potent and effective in binding and blocking CXCR2 receptor function.

* This is an example of animal research that has been carried out in the UK and published in a peer-reviewed journal. Details relate to this specific example and should not be taken to represent a 'typical' animal experiment. It is important to note that individually published experiments usually form one part of a continuing area of research, and the significance of the results may therefore be difficult to interpret.

Box 8.2: The characterisation of promising candidate medicines (stages 3 and 4): example of animal research undertaken during the development process of a new medicine

Irving EA, Vinson M, Rosin C *et al*. (2005) Identification of neuroprotective properties of anti-MAG antibody: a novel approach for the treatment of stroke? *J Cereb Blood Flow Metab* **25**: 98–107.*

It had been previously hypothesised that a protein called myelin-associated glycoprotein (MAG) was a contributing factor to the lack of regeneration of the CNS after injury, such as stroke. This research project demonstrated that the antibody specific to this protein, anti-MAG, possessed the ability to neutralise the inhibitory effect of MAG on neurons following an induced stroke and, in addition, protected certain CNS cells from cell death *in vitro*. Rats given the antibody improved in their motor function ability after the stroke compared with control animals, measured by their ability to walk along a cylindrical beam. The authors concluded that the data indicated potential for the use of the antibody as a therapeutic agent for the treatment of stroke.

Under anaesthesia, small tubes were inserted into the brains of rats to enable the induction of a stroke. Two weeks later the rats were anaesthetised and a stroke was induced by causing a transient blockage of an artery in the brain for 90 minutes. Rats that displayed circling stereotypic behaviour one hour following the surgical procedure were judged to be suitable models and therefore only these rats were included in the study. During the following week, the rats were administered with the test antibody at 1, 24 and 72 hours after the stroke either into the brain or intravenously. They were then euthanised.

* This is an example of animal research that has been carried out in the UK and published in a peer-reviewed journal. Details relate to this specific example and should not be taken to represent a 'typical' animal experiment. It is important to note that individually published experiments usually form one part of a continuing area of research, and the significance of the results may therefore be difficult to interpret.

8.17 Information about research carried out during stages 3 and 4 is often provided through oral communications and posters at scientific meetings, and is later reported in scientific publications.[22] Many thousands of such posters and publications are published annually by industry. More recently, the Home Office has begun to make available abstracts of licensed research (see Box 13.4), which are likely to include many types of experiment undertaken to identify and optimise pharmaceutical leads. We consider issues relating to publication of research in more detail in Chapter 15 (see paragraph 15.35).

22 See PubMed, a service of the US National Library of Medicine, which includes over 15 million citations for biomedical articles dating back to the 1950s, available at: http://www.ncbi.nlm.nih.gov/pubmed. Accessed on: 26 Apr 2005.

Vaccines and veterinary medicines

8.18 Characterisation of vaccines and other biological products during stages 3 and 4 has a number of special features. First, the product may require modification so that it can be administered and remain effective as it is absorbed and transported around the body. Secondly, vaccines commonly contain an adjuvant (e.g. aluminium hydroxide) which is used to increase the effectiveness of the immune response. Both vaccine modification and testing of adjuvants involve the use of animals. The product is often administered to animals and their immune responses are measured by sampling blood and tissue.[23] For example, vaccines against tetanus are tested for potency in mice or guinea pigs. Animals are given the tetanus vaccine (which should confer protection) and later receive what would be expected to be a lethal or paralytic dose of tetanus toxin. If the vaccine has the required potency, the toxin will cause no adverse effects for the animals (see also Box 8.5).[24] In the past, many more tests were required during which the animals showed symptoms of the disease, which could be severe and even lead to death. This methodology has been replaced in many cases by earlier, more humane, experimental endpoints (see paragraph 5.22), such as changes in weight, body temperature or behaviour.[25] In addition, blood and tissue markers of infection are increasingly used.[26]

8.19 The development of new veterinary medicines often involves studies that use the same species for which the medicine is intended. Usually, animals with specific diseases are used as models, although animals spontaneously affected by the disease or condition are also used in field studies.[27] The effect on the animals is specific to the area of research, and may depend also on the state of their health. For example, in the case of the severe respiratory disease pasteurellosis, which affects cattle, 450 calves were used in a programme to develop a vaccine and a significant proportion suffered from the disease.[28] The vaccine that was developed has now been used successfully to bring the disease under control.[29] In field trials potential suffering is usually avoided by comparing the new vaccine to existing treatments (if available) rather than using placebos as a comparison.

Stage 5: selecting candidate medicines and ensuring their safety

8.20 The aim of stage 5 is to decide whether promising compounds could be tested in trials involving human volunteers. Questions that need to be addressed include:

- Do particular compounds meet the quality threshold to be a successful medicine?
- Would the medicine be safe and effective for humans?
- How best could the medicine be administered?

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23 Leenaars PPAM, Hendriksen CFM, de Leeuw WA *et al*. (1999) The production of polyclonal antibodies in laboratory animals ECVAM Workshop Report 35 *ATLA* **27**: 79–102.

24 See Weisser K and Hechler U (1997) *Animal Welfare Aspects in the Quality Control of Immunobiologicals: A critical evaluation of animal tests in pharmacopoeial monographs* (Nottingham: FRAME, ECVAM and the Paul Ehrlich Institut).

25 See Hendriksen CFM and Morton DB (Editors) (1999) *Humane Endpoints in Animal Experiments for Biomedical Research* Proceedings of the International Conference, 22–25 Nov 1998, Zeist, The Netherlands (London: Royal Society of Medicine), available at: http://www.lal.org.uk/endpoints1.html. Accessed on: 26 Apr 2005.

26 Griffin JF (2002) A strategic approach to vaccine development: animal models, monitoring vaccine efficacy, formulation and delivery *Adv Drug Deliv Rev* **54**: 851–61.

27 This is regulated under the authority of Animal Test Certificates (ATC). See Veterinary Medicines Directorate (2004) *Animal Test Certificates*, available at: http://www.vmd.gov.uk/lu/amelia/amelia13n.pdf. Accessed on: 26 Apr 2005.

28 This would have included the animals used as the positive controls (to prove the bacteria could cause the disease) and unprotected animals that had been administered trial vaccines that proved ineffective.

29 National Office of Animal Health (2002) *Vaccination of farm animals*, available at: http://www.noah.co.uk/issues/briefingdoc/22-vaccfarmanimals.htm. Accessed on: 3 May 2005.

- How much of the medicine will be active in the body?
- Is it possible to produce enough of the active compound at an acceptable cost?

8.21 Once a candidate drug has been selected, toxicity studies are then conducted on animals, completing the pre-clinical phase of the development process.[30] The increased knowledge gained in the earlier stages of the modern drug-discovery process means that potential medicines are now better characterised by the time that the toxicity studies begin. Extrapolations are made from animal and non-animal data to predict safety and the initial dose of medicines to be used in humans. The use of toxicity databases, toxicogenomics, proteomics and high-throughput screening (see paragraph 8.6) play an important role in providing additional information and helping to reduce the use of traditional toxicity studies. Together with data from non-animal studies, pre-clinical results of these tests are submitted to regulatory authorities in the application for permission to conduct clinical studies in human volunteers. The final outcome of this stage is a candidate drug that meets the safety criteria set by regulatory bodies and has the potential to be developed into a successful and commercially viable product.

Use of animals

8.22 At this, and subsequent stages, toxicity tests on animals are undertaken to meet the requirements of regulators that a potential medicine demonstrates an acceptable balance of safety and efficacy (see paragraphs 9.6–9.21). The custom and practice of regulatory agencies has been to rely on data from animal research when making these judgements, although increasingly more data from validated non-animal methods are generated and accepted. Toxicity studies account for between five and 20 percent of animal use by the pharmaceutical industries. In 2003, pharmacological safety and efficacy evaluation constituted ten percent of the total number of animal procedures in Great Britain.[31] Animal tests at this stage are much more uniform compared to the experiments carried out in drug discovery and they need to be conducted in a format that is accepted worldwide. Some of the most important tests, and associated welfare implications, are described in Chapter 9, in which we discuss toxicity testing in more detail. (The scope of Refinements,[32] and the application of the Three Rs in toxicity testing more generally, are considered in Chapters 11 and 12).

Vaccines and veterinary medicines

8.23 Before administering a novel vaccine to human volunteers researchers need to ensure that the candidate vaccines will not infect trial participants with the disease (as might be possible with live vaccines) or lead to an inappropriate immune response, such as producing antibodies that have adverse effects. It also needs to be ascertained whether the agent, or additives such as adjuvants, are likely to cause direct irritation at the site of application. The

30 The pre-clinical phases (discovery, selection and characterisation of promising candidate medicines and toxicity studies) take on average four to five years to perform, and cost on average £200 million. See Network Science (2004) *The Process of Drug Development*, available at: http://www.netsci.org/scgi-bin/Courseware/projector.pl?Course_num=course1&Filename=top.html. Accessed on: 26 Apr 2005.

31 Eighty-three percent of toxicological procedures were performed to comply with legislative or regulatory requirements. See Home Office (2004) *Statistics of Scientific Procedures on Living Animals Great Britain 2003* (London: HMSO).

32 Refinements can be implemented through the use of more sensitive markers of toxicity (for example, blood tests or remote monitoring) and more humane endpoints. See Organisation for Economic Co-operation and Development (2000) *Guidance Document on the Recognition, Assessment, and Use of Clinical Signs as Humane Endpoints for Experimental Animals Used in Safety Evaluation*, available at: http://www.olis.oecd.org/olis/2000doc.nsf/4f7adc214b91a685c12569fa005d0ee7/c125692700623b74c12569bb005aa3d5/$FILE/00087372.PDF. Accessed on: 26 Apr 2005.

common types of test required are single- and repeated-dose toxicity assessments and testing to determine any local irritation (see paragraphs 9–9-9.18).[33] Specific tests are also required to determine how effective the vaccine is in protecting animals against challenge with the pathogen. In order to assess efficacy, the test vaccine is administered to animals and the disease is subsequently induced. If the vaccine does not protect the animals they may experience pain or suffering related to the disease, although humane endpoints are usually chosen. Animals are euthanised when these are reached.

8.24 Veterinary medicines are generally evaluated for safety using the species in which they will eventually be used (see paragraph 8.19).

Stages 6–8: clinical studies on humans

8.25 Potential new medicines are first tested on small groups of healthy human volunteers, and then on progressively larger groups of patients.[34] These tests are organised into four consecutive trials, Phases I–IV (see Figure 8.3). Experimental medicine has enlarged the application of existing clinical tests that are used in these studies. They include advanced blood and tissue diagnostics, and imaging techniques, such as MRI or PET scanning (see paragraph 5.12 and Box 11.1).

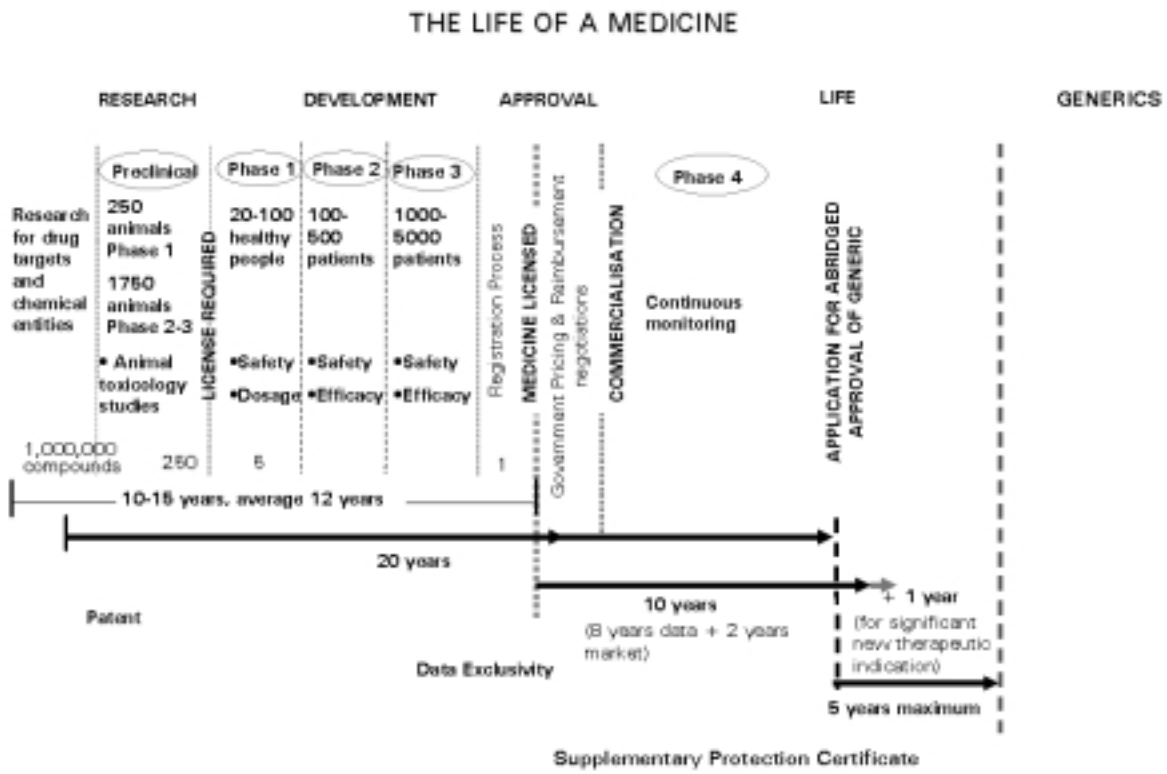

Figure 8.3: The path to modern medicine – phases of pre-clinical and clinical development

Source: GlaxoSmithKline

33 The European Agency for the Evaluation of Medicinal Products (1997) *Note for Guidance on Preclinical Pharmacological and Toxicological Testing of Vaccines*, available at: http://www.emea.eu.int/pdfs/human/swp/046595en.pdf. Accessed on: 26 Apr 2005.

34 See ABPI (2003) *Clinical Trials - Developing New Medicines*, available at: http://www.abpi.org.uk/publications/briefings/clinical_brief.pdf. Accessed on: 26 Apr 2005.

Stage 6: concept testing

8.26 Typically, no more than a few candidate medicines for any given disease enter this stage. In Phase I of clinical trials, they are first tested in a limited number of healthy volunteers (see Figure 8.3). The purpose is to determine how well the active ingredient is actually tolerated in humans and whether it has the desired effect, to obtain information about suitable dosage and to determine whether it has characteristics that would allow it to be developed into a medicine.

Use of animals

8.27 During the subsequent Phases II–IV, additional animal studies that aim to ensure the safety of the particular medicine and its application are undertaken. For example, if a medicine is intended to be given to women of childbearing age, reproductive toxicology would be assessed in animals prior to Phase II studies (see paragraphs 9.22–9.23 and Box 8.4).

Stage 7: development for launch

8.28 If testing in healthy volunteers (Phase I) and a limited number of patients (Phase II) is successful then large-scale trials involving human volunteers are carried out (Phase III). Phase III trials involve between 1,000 and 5,000 patients, and provide the basis for the final decision as to whether to continue or abandon the project. The size and scale of Phase II and III studies make this stage the most expensive part of drug development (see Figure 8.3).[35]

Use of animals

8.29 During clinical studies on humans, Phases I–III, a comprehensive set of safety tests in animals continues to be carried out. The project team that is developing the medicine liaises with the internal ethics committee and regulatory authorities, to define the tests that are required to ensure safety (see paragraphs 9.4–9.25).

Vaccines and veterinary medicines

8.30 The clinical development of vaccines may require further safety tests in animals, which are broadly similar to those required for human medicines.[36] The exact nature of these tests depends on the results of clinical trials.[37] The data required for a marketing authorisation for a veterinary medicine concern proof of efficacy and bioavailability of a product.[38] The scale and scope of the data provided are generally less comprehensive than for human medicines, although in some cases specific emphasis is given to certain areas. For example, in the case of food-producing animals, evidence is required on the potential for residues of new medicines to accumulate in food.[39] Bioavailability studies are similar to those undertaken for human medicines (see paragraph 9.24), although more-invasive muscle tissue samples may be taken in order to test for residues.

35 The average length of clinical phases (concept testing and development for launch) is eight to twelve years and the average cost of clinical phases is £350 million, see The Association of the British Pharmaceutical Industry *The Development of Medicines*, available at: http://www.abpi.org.uk//publications/briefings/Dev_Medicines.pdf. Accessed on: 26 Apr 2005.

36 The tests described in Chapter 9 may also apply to vaccines.

37 The European Agency for the Evaluation of Medicinal Products (1997) *Note for Guidance on Preclinical Pharmacological and Toxicological Testing of Vaccines*, available at: http://www.emea.eu.int/pdfs/human/swp/046595en.pdf. Accessed on: 26 Apr 2005.

38 Veterinary Medicines Directorate (2002) *Application Form for a Marketing Authorisation*, available at: http://www.vmd.gov.uk/lu/forms/appform1a.pdf. Accessed on: 2 May 2005.

39 See Veterinary Residues Committee *Fact Sheet*, available at: http://www.vet-residues-committee.gov.uk. Accessed on: 2 May 2005.

Stage 8: launch phase

8.31 At this stage, the data from all of the pre-clinical and clinical studies are collated and sent to the regulatory agencies (see paragraphs 9.4 and 13.49–51). The average time for regulatory approval is 1.5 years.

Use of animals

8.32 There is usually no animal use at this stage.

Support for the marketed medicine

8.33 Once a medicine is approved by the regulatory agencies, Phase IV clinical trials monitor long-term effects in large numbers of patients and evaluate economic aspects of the medicine. Extensive programmes to capture information on disease epidemiology and the outcomes of using the medicine may be established. This information gathering may also include sampling, for example to obtain pharmacogenetic data, to inform the very first stages of drug discovery. New indications and new formulations are also closely examined. Medicines originally intended for treatment of one disease are sometimes found to have beneficial effects for others (see Boxes 8.3 and 8.4).

Box 8.3: Testing approved drugs for a novel use: example of animal research undertaken after a medicine is on the market

Fox A, Gentry C, Patel S, Kesingland A and Bevan S (2003) Comparative activity of the anti-convulsants oxcarbazepine, carbamazepine, lamotrigine and gabapentin in a model of neuropathic pain in the rat and guinea pig *Pain* **105**: 355–62.*

The aim of this research was to find out whether drugs that are currently used to treat epilepsy could also be effective as pain killers for persistent neuropathic pain. This form of pain is produced by the nervous system itself, 'phantom' limb pain in amputees being one extreme example. Clinical management of neuropathic pain is very difficult as it responds poorly to opiates and non-steroidal anti-inflammatory medicines. It is treated primarily with anti-epileptic medicines and anti-depressants, although both are associated with significant use-limiting adverse effects. The researchers concluded from their experiments on guinea pigs and rats that some of the anti-epileptic medicines administered were able to relieve neuropathic pain, although the effects differed between the two species. These new medicines were not accompanied by the use-limiting side effects exhibited by current treatments for neuropathic conditions.

In some animal models for neuropathic pain, the spinal or facial nerves of the animal have been fused, leading to the development of a long-lasting pain response. In this example, guinea pigs and rats had the sciatic nerve in one leg surgically exposed, and one third to one half of its thickness was tied with a suture under anaesthetic. The aim of this intervention was to reproduce the exacerbated response that sufferers from neuropathic pain experience in response to a normally mild stimulus. The animals were allowed to recover for approximately two weeks after surgery. Post-operative painkillers were not used since the development of pain was the object of the study. Following recovery, the researchers assessed the pain response by applying increasing pressure to the paws of the animals. The threshold at which the animal flinched was measured for both the injured and the uninjured hind paw after which greater pressure was not applied. The medicine under test was then administered and the same procedure was carried out for up to six hours thereafter, and repeated for up to six days.

A further experiment was carried out on rats to measure the pain response to a stimulus that would not usually cause pain. Thin filaments were applied to both hind paws, starting with a low force. This was repeated five times at intervals of one or two seconds and the response noted. The researchers waited for at least five minutes between using successively stiffer filaments. The filament force that produced a withdrawal of the paw was denoted as the threshold for the stimulus, after which greater pressure was not applied. Thresholds were determined prior to and up to six hours following drug administration. All animals showed an increased sensitivity to pain following the surgical procedure.

* This is an example of animal research that has been carried out in the UK and published in a peer-reviewed journal. Details relate to this specific example and should not be taken to represent a 'typical' animal experiment. It is important to note that individually published experiments usually form one part of a continuing area of research, and the significance of the results may therefore be difficult to interpret.

Box 8.4: Use of thalidomide

Thalidomide is a notorious example of the failure of methodology in pharmaceutical research.* The example is frequently used to argue that the results of animal studies cannot be applied to humans. Thalidomide was licensed as a sedative after safety tests performed on animals were approved by the regulatory authorities. Between 1957 and 1961 it was prescribed to pregnant women as a treatment for morning sickness and other symptoms. In December 1961 *The Lancet* published a letter by Dr W.G. McBride, an Australian obstetrician, stating that he had observed frequent limb deformities in babies of women who had taken thalidomide during pregnancy. It later emerged that more than 10,000 children around the world had been affected.†

Research published five months after Dr McBride's letter confirmed that thalidomide given to pregnant rabbits resulted in the birth of litters with similar limb deformities to those in humans. Subsequent research showed that offspring of mice, rats, hamsters, macaques, marmosets, baboons and rhesus monkeys suffered comparable effects. Although the licensing of thalidomide involved animal research, tests on pregnant animals were not undertaken as this was not a legal requirement. Partly in response to the thalidomide tragedy, the UK passed the Medicines Act of 1968, which regulates the testing and supply of medicines in the UK (see paragraph 13.49).‡

Strict measures have been put in place in many countries to prevent the use of thalidomide by pregnant women. At the same time, the drug has been found to be an effective treatment for other conditions. Celgene Corporation has begun developing the medicine for a range of potential indications, including AIDS-related, dermatological and cancer-related conditions.∫

In 1998 the US Food and Drug Administration granted marketing clearance to Celgene's Thalomid for the treatment of erythema nodosum leprosum (ENL), a severe and debilitating condition associated with leprosy (Hansen's disease). The Authority also imposed unprecedented restrictions on the distribution of the medicine. These included restriction on those who were permitted to prescribe thalidomide, a requirement for a negative pregnancy test result within 24 hours of starting therapy and weekly testing during the first month of use. Women were also required to use two reliable forms of contraception simultaneously while taking the drug.**

* See RDS Thalidomide, available at: http://www.rds-online.org.uk/pages/page.asp?i_ToolbarID=5&i_PageID=1070 . Accessed on: 2 May 2005.

† Powell RJ (1996) New roles for thalidomide *BMJ* **313:** 377–8.

‡ ABPI Law - Approval of medicines, available at: http://www.abpi.org.uk/amric/basic5.asp. Accessed on: 2 May 2005.

∫ See Celgene Thalomid, available at: http://www.celgene.com/Products.aspx?s=1. Accessed on: 21 April 2005; see also Pollard M (1996) Thalidomide promotes metastasis of prostate adenocarcinoma cells (palii) in L-W rats. Cancer Lett 101: 21-4

** Food and Drug Administration (1998) FDA approves thalidomide for Hansen's disease side effect, imposes unprecedented restrictions on distribution, available at: http://www.fda.gov/bbs/topics/answers/ans00887.html. Accessed on: 2 May 2005.

Use of animals

8.34 Limited animal use may be required for new indications, new formulations or in studies of possible adverse effects in patients. However, there is much reliance on archived animal and human testing data.

Vaccines

8.35 An exception to limited use of animals at this stage occurs in vaccine testing. Immunisation is a very cost-effective public health intervention and billions of doses of vaccine are administered each year for the prevention of a range of diseases.[40] Relatively large numbers of animals are used for toxicity testing of batches of these vaccines. This is because the exact composition and properties of many biological products are very difficult to control and may alter after production.[41] Continuous safety and efficacy testing of production batches of vaccines is therefore carried out.[42]

8.36 Depending on the type of test, there may be serious welfare implications. For example, if death is the required endpoint, or if it is the most convenient stage for reliable

40 See World Health Organization (2003) *Vaccines, Immunization and Biologicals – Statistics and Graphics*, available at: http://www.who.int/vaccines-surveillance/StatsAndGraphs.htm. Accessed on: 26 Apr 2005.

41 For example, the reactivation by mutation of inactivated viruses needs to be monitored and assessed.

42 The recent report by the Associate Parliamentary Group for Animal Welfare on vaccine testing describes why such quality control is required, the animals that are used, the pain and distress that they experience and the current use, and prospects for, Replacement, Reduction and Refinement. See The Associate Parliamentary Group for Animal Welfare (2005) *The Use of Animals in Vaccine Testing for Humans*, available at: http://apgaw.org/userimages/Vaccinetesting.pdf. Accessed on: 26 Apr 2005.

observation, then it may be used, subject to regulatory approval. While the terminal stages of a lethal endpoint may not involve much, if any, suffering as the animal may be comatose, the suffering that may have taken place beforehand can be substantial and may have involved symptoms such as inappetence (lack of appetite), malaise, convulsions or paralysis (see also Box 8.5).

Box 8.5: Examples of animal suffering in the context of quality control of vaccines for human use*

Tetanus potency test

Batches of tetanus vaccine are tested for potency in mice or guinea pigs. The standard method involves testing a new vaccine against a reference vaccine at three different concentrations. It has been estimated that 66–108 animals are usually used for each test. The animals are administered with the vaccine under the skin and four weeks later with a single dose of tetanus toxin. This dose could be lethal or paralytic. Control animals (that receive no vaccine) and those animals that are unprotected because the test vaccine they receive is unsuitable or is at an ineffective concentration suffer paralysis and death.

Diptheria (absorbed) potency test

Guinea pigs are immunised with test samples of diphtheria vaccine, and are subsequently infected with diphtheria bacteria four weeks later. In the EU, both lethal and non-lethal amounts of the bacterial toxin are permitted for this test and the endpoints are death or skin inflammation respectively. At least three dilutions each of the test vaccine and a reference vaccine are used, together with one untreated control group. A minimum of 70 animals is used to test each vaccine batch and both methods cause severe pain and distress for those animals that are unprotected (see above). There is no agreement on whether the lethal or non-lethal methods cause greater suffering.

* See The Associate Parliamentary Group for Animal Welfare (2005) The Use of Animals in Vaccine Testing for Humans, available at: http://apgaw.org/userimages/Vaccinetesting.pdf Accessed on: 26 Apr 2005.

The validity of animal models used in pharmaceutical research

8.37 We have described why and how animals are used in pharmaceutical research and have illustrated with several examples the range of welfare implications that they may experience. Many people who are concerned about animal suffering are critical of the permissibility of animal research on ethical grounds. However, there are critics who also object to the use of animals in pharmaceutical research on scientific grounds. They question the transferability and predictability of data obtained from animals, and its reliability for the accurate assessment of the safety of new therapeutic interventions, as shown by the following respondents to the Consultation:

> '…animal experimentation is positively harmful to human health… [It] does not provide information that is relevant to human medicine because the data cannot be transferred to humans with any degree of reliability. In fact, studies of the predictability of animal experiments consistently show them to be worse than random guesswork… Adverse drug reactions are the fourth leading cause of death in the Western world, killing over 100,000 individuals every year in the US alone. Clearly, the animal tests are failing to protect people.'
> *Animal Aid*

> '…claims that animal experiments have instilled a misplaced sense of the relative danger of a drug are supported by the incidences of false negatives and false positives known to be attached to such tests.'
> *Cris Iles-Wright*

8.38 We have shown above that producing a new medicine is a lengthy and complex process, and that decisions on the compounds that should proceed to the next stage are taken using a wide range of information. Tests on animals play a vital role, but they are not the only source of information that is used to determine safety and efficacy (see Figure 8.3). Some critics of animal research and testing tend to attribute any problems with the final product solely to the use of animal testing. We consider the general question of whether or not

animals are useful models for humans in medical research in paragraphs 10.27–10.32). Systematic limitations faced by any modelling approach are addressed in paragraphs 10.33–10.36), and the findings of scientific reviews on the critical evaluation of research involving animals are discussed in paragraphs 10.37-10.43).

8.39 We observe that claims that animal research is failing to protect people from adverse drug reactions (ADRs) need to be treated with some caution. ADRs[43] have a number of causes. Many of these are avoidable, for example where they arise from prescription errors, where people have been given or have taken the wrong medicine, or from interactions between different medicines taken simultaneously. In 2004, researchers conducting the largest prospective analysis in the UK of ADRs as a cause of admission to hospital found that more than 70% were avoidable and could have been predicted by taking into account pharmacological properties of the medicines involved.[44] While ADRs may be the direct result of administration of one specific medicine, the question remains whether this is proof of the failure of the animal model (or any other model) involved in the development process, or a methodological problem. As we have said, phases I–II of human clinical trials in the development of a medicine include up to 5,000 patients to monitor efficacy and safety. If severe ADRs occur during these trials, the development of the medicine is not usually taken further. However, ADRs may occur at very low statistical frequencies, for example 1 in 10,000, and hence may not be revealed at this stage (see paragraphs 10.33 and 10.1). In making inferences about the occurrence of ADRs, and the role that animal research plays, it is therefore unhelpful to generalise. ADRs can occur for a number of reasons and could, in principle, also be caused by a medicine that, hypothetically, had been developed without the use of animals.

8.40 Some also argue that the withdrawal of medicines from the market is indicative of the fact that animal research does not help to prevent ineffective or harmful medicines being used by humans.[45] In the UK, the Medicines and Healthcare products Regulatory Agency (MHRA) monitors whether medicines on the market meet the appropriate standards of safety, quality and effectiveness. When there is sufficient evidence to suggest that the risk of taking a medicine outweighs its benefit to patients, the Committee on Safety of Medicines (CSM) and MHRA take appropriate regulatory action to protect the health of patients, and may initiate steps to withdraw medicines from use. Between 1995 and 2005, 18 medicines were withdrawn from the UK market by companies or by the Licensing Authority on grounds of safety (see Box 8.6). A study conducted in 1994 on medicines withdrawn between 1961 and 1992 concluded that in the UK, 49 were taken off the

43 Edwards and Aronson define an ADR as 'an appreciably harmful or unpleasant reaction, resulting from an intervention related to the use of a medicinal product, which predicts hazard from future administration and warrants prevention or specific treatment, or alteration of the dosage regimen, or withdrawal of the product.' See Edwards IR and Aronson JK (2000) Adverse drug reactions: definitions, diagnosis, and management *Lancet* **356**: 1255–9.

44 The researchers, using Edwards and Aronson's definition of ADRs (see previous footnote), sought to ascertain the burden of ADRs though a prospective analysis of hospital admissions to two large general hospitals in the UK. Every patient aged over 16 years who was admitted to these hospitals (18,820 patients) over a six month period was assessed to determine if the admission had been caused by an ADR. It was found that 1,225 admissions were related to ADRs (equalling 6.5%, which is consistent with an estimate of 5% based on pooled data from several studies worldwide). Three types of avoidability were assessed: definitely avoidable (7-10%: the ADR was due to treatment inconsistent with present day knowledge of good medical practice), possibly avoidable (60-66%: the ADR could have been avoided by an effort exceeding the obligatory demands of present day knowledge of good medical practice) and unavoidable (25-30%: the could not have been avoided by any reasonable means). See Pirmohamed M, James S, Meakin S *et al*. (2004) Adverse drug reactions as cause of admission to hospital: prospective analysis of 18 820 patients **BMJl 329:** 15–9. See also Waller P and Rawlins **P A User's Guide to the Safety of Medicines,** available at: http://www.dsru.org/pat_guide_1.html. Accessed on: 2 May 2005; Kohn LT, Corrigan JM and Donaldson MS (Editors) (2000) *To Err is Human: Building A Safer Health System*, available at: http://www.iom.edu/report.asp?id=5575. Accessed on: 26 Apr 2005.

45 See BUAV Don't we need animal experiments to make sure drugs are safe for humans?, in *Frequently asked questions about vivisection*, available at: http://www.buav.org/faqs.html. Accessed on: 2 May 2005.

market (see Box 8.7). These withdrawals were mainly due to inadequate evidence of efficacy in widespread clinical use, loss of therapeutic interest or poor market performance. To what extent the withdrawal of medicines can be attributed exclusively, or in part, to the use of animals in research would need to be assessed in individual cases (see paragraphs 10.27–10.43).[46]

Box 8.6: Medicines withdrawn in the UK for safety reasons 1995–2005

Name of medicine (brand name)	Year action taken	Primary safety concerns
Naftidrofuryl oxalate injection (Praxilene)	1995	Cardiotoxicity
Pemoline (Volital)	1997	Liver toxicity
Troglitazone (Romazin)	1997	Liver toxicity
Fenfluramine (Ponderax)	1997	Heart valve disease
Dexfenfluramine (Adifax)	1997	Heart valve disease
Sertindole (Serdolect)*	1998	Disorders of heart rhythm
Tolcapone (Tasmar)†	1998	Liver toxicity
Mibefradil (Posicor)	1998	Drug interactions
Trovafloxacin (Trovan)‡	1999	Liver toxicity
Grepafloxacin (Raxar)	1999	Disorders of heart rhythm
Pulmonary surfactant (Alec)	2000	Increased mortality
Cisapride (Prepulsid)	2000	Disorders of heart rhythm
Droperidol (Droleptan)	2001	Disorders of heart rhythm
Cerivastatin (Lipobay)	2001	Muscle toxicity
Levacetylmethadol (Orlaam)	2001	Cardiac arrhythmias
Kava kava	2003	Liver toxicity
Rofecoxib (Vioxx)	2004	Myocardial infarction/stroke
Valdecoxib (Bextra)	2005	Serious skin reactions

* Sertindole has since been reintroduced under very restricted conditions.

‡ Tasmar, Trovan and Orlaam were licensed through the centralised procedure with the European Commission as the Licensing Authority.

‡ Trovafloxacin was never marketed in the UK.

Source: MHRA

46 See also Chapter 6, footnote 40.

Box 8.7: Medicines withdrawn from the market*

Between 1961 and 1992 a total of 131 medicines were withdrawn from France (63), Germany (58), UK (49) and USA (41) (note that some were withdrawn from more than one country). Only ten were withdrawn in all four countries. In the UK the 49 withdrawn medicines can be separated into four groups, as follows:

1) Medicines withdrawn after long-term use, which were marketed before detailed animal or clinical tests, or in use despite known toxicity, and later replaced by a medicine for the same indication with less toxicity

Product	Year of launch	Year withdrawn
Aspirin (paediatric form)	1899	1986
Aminopyrine	1900	1975
Clioquinol	1930 (1900)	1981
Dipyrone	1930	1977
Oxyphenisatine	1955	1978
Oxyphenbutazone	1962	1984
Phenacetin	1900	1980
Phenformin	1959	1982

2) Medicines withdrawn because of toxicity (generally carcinogenicity) revealed by animal tests that were continued after launch

Product	Year withdrawn
Alclofenac	1979
Chlormadinone	1970
Danthron	1987
Fenclofenac	1984
Indoprofen	1983
Megestrol	1970
Methapyrilene	1979
Polidexide	1975

3) Medicines withdrawn for reasons unrelated to standard investigation of toxicity (i.e. particular type of toxic effect apparent after launch)

Product	Reason for withdrawal
Alphaxalone	Allergy to excipient
Cromoglycate (eyedrops)	New formulation (untested)
Desensitising vaccines	Allergy
Doxylamine	Alleged teratogenicity
Factor VIII	Risk of AIDS transmission
Growth hormone (natural)	Possibility of CJD transmission
Guanethidine (eyedrops)	New formulation (untested)
Indomethacin-R	New formulation (untested)
Mebanazine	Toxic interaction with diet or other drugs
Nialamide	Toxic interaction with diet or other drugs
Phenoxypropazine	Toxic interaction with diet or other drugs
Thalidomide	Teratogenicity not tested for
Zomepirac	Allergy

Continued

4) Medicines withdrawn due to unexpected toxicity

Product	Year of withdrawal	Reason
Benoxaprofen	1982	Toxicity in the aged
Benziodarone	1964	Hepatotoxicity
Domperidone injection	1986	Cardiovascular effects
Feprazone	1984	Multiple
Ibufenac	1968	Hepatotoxicity
Methandrostenolone	1982	Endocrine effects
Metipranolol	1990	Ophthalmological
Mumps vaccine	1992	Neuropsychiatric
Nomifensine	1986	Haematological
Practolol	1975	Rare idiosyncrasy
Prenylamine	1989	Cardiovascular
Propanidid	1983	Allergic type
Sulphamethoxypyridazine	1986	Haematological
Suprofen	1987	Nephrotoxicity
Temafloxacin	1992	Multiple
Terodiline	1991	Cardiovascular
Thenalidine	1961	Haematological
Triazolam	1991	Neuropsychiatric
Tryptophan	1990	Multiple
Zimeldine	1983	Neuropsychiatric

* Spriet-Pourra C and Auriche M (1994) *Drug Withdrawal From Sale*, 2nd Edition (Richmond: PJB Publications).

8.41 Lastly, animal research is also undertaken by the pharmaceutical industry to refine the predictive capacity of data obtained from animal and human studies. For example, researchers seek to identify how results from different species can be best integrated in order to develop better predictions of how the medicine will be distributed, absorbed and excreted in the human body (see Boxes 8.8 and 9.4).

Box 8.8: Example of animal research undertaken to improve the predictability of pharmacokinetic data

Aviles P, Pateman A, San Roman R *et al*. (2001) Animal pharmacokinetics and interspecies scaling of sordarin derivatives following intravenous administration *Anitmicrob Agents Chemother* **45**: 2787–92.*

The aim of this research was to compare the pharmacokinetics of a group of synthetic antifungal agents against reference compounds in different animal species and to assess whether human pharmacokinetics could be reliably predicted from this information.

The antifungal agents that were used belong to a new group of synthetic chemicals called sordarin derivatives which have been shown to prevent the growth of fungal pathogens. Opportunistic fungal pathogens remain a common cause of death in immunocompromised patients, such as those with HIV/AIDS or those receiving chemotherapy or immunosuppressive therapy.

The study used *Cynomolgus* monkeys, rats, mice and rabbits. Gunn rats, which have impaired liver function, were used to assess the processing of sordarin derivatives when they pass through the liver. A representative sordarin derivative was administered intravenously to animals. In mice, this was achieved by puncture of the tail vein. The compound was administered through tubes inserted into the jugular veins of rats and into marginal ear veins of rabbits. In monkeys, administration was performed via the cephalic vein. Each compound was administered once. In mice, blood samples were taken by cardiac puncture using a needle at eight intervals after administration. Three mice were euthanised by cervical dislocation at each sampling point. Samples of rat blood were taken from the end of the tail. Rabbits were sampled using a tube placed in the central artery of the ear. Samples of monkey blood were obtained from the posterior of the animals by direct venepuncture using a needle at ten intervals after administration.

Blood samples were allowed to clot and then centrifuged to separate the serum (the clear yellowish

Continued

fluid obtained upon separating whole blood into its solid and liquid components). Data obtained from the serum were used to construct concentration-time curves to evaluate the effectiveness of the substance on the growth of fungal protein. Comparisons between the species used were made and assessed against data from previous studies. The researchers concluded that integrating the pharmacokinetics data from the different animals used would result in better predictions for the way the sordarin derivatives are metabolised in humans and therefore contribute to the study design of initial clinical trials in humans.

* This is an example of animal research that has been published in a peer-reviewed journal. Details relate to this specific example and should not be taken to represent a 'typical' animal experiment. It is important to note that individually published experiments usually form one part of a continuing area of research, and the significance of the results may therefore be difficult to interpret.

Summary

8.42 Pharmaceutical research and development has been transformed over the past 50 years because of the availability of advanced information and diagnostic technologies, and an increased understanding of genetics. At present a wide range of advanced methods that do not involve animals is used together with animal research. Although there has been a substantial decline in the total use of animals, pharmaceutical research remains responsible for a significant proportion of the animal experiments conducted in the UK each year. A very wide range of basic and applied medical and veterinary research projects is supported or conducted by pharmaceutical companies as part of the search for new medicines and vaccines for use in humans and animals. We described eight different stages in the development process. The majority of animals (60-80%) are used in the characterisation of promising candidate medicines; less (5-15%) are used in the preceding discovery and selection process. GM mice are most commonly used in the early stages of development of new medicines to assess the importance of a drug target, although they are also used increasingly in later stages (target validation) or as animal models of a disease (see Chapter 7).

8.43 The welfare implications for animals involved in research are as varied as the research itself. Non-experimental factors, such as housing, husbandry and the training of those handling the animals, especially in relation to the implementation of Refinements, all influence welfare. Some techniques, such as methods for administering a medicine and measuring the level in blood, are generic for all types of research. In the case of specific animal models of disease, welfare implications depend on the symptoms of the disease. A special case is the production of vaccines. Since the exact quality of biological products is often very difficult to control, tests to assess potency and toxicity are carried out on each batch, which may lead to symptoms ranging from lack of appetite to paralysis for animals such as monkeys, mice and guinea pigs.

8.44 The use of animals in pharmaceutical research and development in the future is difficult to predict. The following are among the many possible outcomes:[47]

- the use of animals may continue to fall as the use of advanced methods increases;
- the use of animals may remain static, but advanced imaging, sensing and biomarkers will allow extraction of even more information in an increasingly refined way; or
- the use of animals may rise because the increasing volume of information from the early stages of drug discovery presents the possibility of more and more new medicines.

47 See ABPI (2001) *Statistics, Animal Research and Development of Medicines*, available at: http://www.abpi.org.uk/publications/briefings/40301-ABPI-Brief-Statistics.pdf Accessed on: 2 May 2005. Various sources suggest that the use of animals is falling when compared with research and development activity undertaken by the largest pharmaceutical companies (see Samuels G (2003) *Medicines: Tried And Tested - In Animals?*, available at: http://www.abpi.org.uk/publications/publication_details/mttur/mttur_ani.asp; GlaxoSmithKline (2002) *The Impact of Medicines: Corporate and Social Responsibility*, available at: http://www.gsk.com/financial/reps02/CSR02/GSKcsr-10.htm#ref), or that the numbers of animals used may vary from year to year (AstraZeneca (2003) *Animal research*, available at: http://www.astrazeneca.com/Article/11174.aspx). All accessed on: 2 May 2005.