THE WISDOM OF RUSSELL AND BURCH

6. Replacement

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The total replacement of animal procedures should be our common goal, not least because animal models cannot tell us what we need to know about human conditions and responses

In *The Principles of Humane Experimental Technique*,[1] Russell and Burch defined a *Replacement technique* as "any scientific method employing non-sentient material which may in the history of experimentation replace methods which use conscious living vertebrates". They recognised that, while this definition can be applied readily to plants and micro-organisms, "a more difficult question arises when we consider free-living metazoan invertebrates".

They decided to exclude invertebrates "from consideration as objects of humanitarian concern", describing their uses as *comparative substitution* and arguing that "to shed obsessional tears over the fate of these organisms would bring the whole concept of humanity into contempt".

This may seem rather harsh in today's animal welfare circles, but it is in line with many current practices involving vertebrates as well as invertebrates, including the use of pesticides, the trawling of fish from the sea, and many agricultural procedures.

Russell and Burch distinguished between *absolute replacement*, in which vertebrate animals are not required at all, and *relative replacement*, where animal are still required, but "are exposed, probably or certainly, to no distress at all".

In the *absolute replacement* group, they included the use of metazoan endoparasites, higher plants, micro-organisms, and non-living physical and chemical systems. In view of the stance they had taken, they did not mention invertebrates, but nematodes (e.g. *Caenorhabditis elegans*) and insects (e.g. *Drosophila melanogaster*) would be regarded as absolute replacements today.

In the *relative replacement* group, they included "non-recovery experiments on living and intact, but completely anaesthetised animals", "and "experiments where animals are still required, but only to furnish preparations after being painlessly killed".

The last-named category included "work on the isolated cells, tissues, or organs of vertebrates". They saw tissue culture as a bridge between *relative* and *absolute replacement*, and considered mammalian tissue cultures to be "one of the most important replacement techniques, and indeed one of the most important developments in biology". Most of us would have no hesitation in agreeing with that.

However, *replacement* can also be categorised in other ways. For example, a distinction can be made between *partial replacement*, where animals are subjected to regulated procedures as part of the programme of work, and *total replacement*, where animals are not subjected to regulated procedures at all. *Partial replacement* might involve the exposure of an animal to a toxic chemical, after which it is killed and its cells or tissues are subjected to further tests *in vitro* (i.e. *ex vivo*). By contrast, where all the experimental procedures are applied to cells or tissues *in vitro*, this would be classed as *total replacement*.

Nevertheless, in my opinion, one of the most important distinctions is between *direct replacement* and *indirect replacement*. In the former case, a replacement technique is used to give results which are directly comparable with those which would be obtained by the animal procedure that is being replaced.

A classical example of *direct replacement* is a test for irritancy, based on the application of chemicals to isolated rabbit eyes, instead of to the eyes of intact rabbits. The problem with this approach, where the aim is to identify chemicals likely to cause irritancy in human eyes, is that the uncertainty of the *ex vivo/in vivo* (i.e. isolated rabbit eye to *in situ* rabbit eye) equivalence must now be added to the uncertainty of the *in vivo/in vivo* (rabbit eye to human eye) equivalence. In addition, the isolated rabbit eye may be even more unlike the human eye than the *in situ* rabbit eye, so the data it provides will be even more difficult to interpret and apply.

Seeking genuine *indirect replacement* procedures is much more intelligent, if it involves defining the information it was hoped to get from the animal procedure, then obtaining it from much more advanced experimental techniques. For example, the metabo-

lism of drugs can be studied by using human hepatocytes *in vitro*, instead of administering the drugs to rats or dogs, which have different complements of drug metabolising enzymes. *Direct replacement* offers the possibility of direct relevance, where the object of interest can be studied, rather than an inadequate and imperfect model.

To return to the wisdom of Russell and Burch, it is interesting to note that it is in their chapter on *Replacement* that they discuss the difference between fidelity and discrimination, and the importance of the high-fidelity fallacy.[2] The danger is in assuming that, since vertebrates — and especially the higher vertebrates, such as Old World monkeys — are *generally similar* to humans, data from experiments applied to them will be *specifically relevant* to humans. Russell and Burch believed that "progress in replacement has been restricted by [this] plausible, but untenable, assumption". Common sense says that they were right, but the presumption of high-fidelity remains the justification for much research and testing with animal models today.[2]

Russell and I followed the same zoology degree course at Oxford, in what was then called the Department of Zoology and Comparative Anatomy. We studied every order of animals from the Protozoa (e.g. *Amoeba*) to the Hominoidea (great apes and humans), and the emphasis was on how animals had evolved from common ancestors via adaptive radiation, in ways appropriate to their particular environments and lifestyles, how originally-common features could evolve to be the solutions to different problems, and how the same kinds of problems could be solved in different ways. It would never have occurred to us that one type of animal could precisely and satisfactorily model another one.

While it may be true that our current thinking about *replacement* is somewhat different from that of Russell and Burch in 1959, to me, at least, their discussion on fidelity and discrimination is of timeless value, especially when coupled with the encouraging words of their view that "*Replacement* is always a satisfactory answer; but *reduction* and *refinement* should, wherever possible, be used in combination".

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References

[1]Russell, W.M.S. & Burch, R.L. (1959). *The Principles of Humane Experimental Technique*, xiv + 238pp. London, UK: Methuen.
[2]Balls, M. (2013). The Wisdom of Russell and Burch. 3. Fidelity and discrimination. *ATLA* **41**, P12–P13.

The Principles of Humane Experimental Technique is now out of print, but the full text can be found at http://altweb.jhsph.edu/pubs/books/humane_exp/het-toc. An abridged version, *The Three Rs and the Humanity Criterion*, by Michael Balls (2009), can be obtained from FRAME.