Methods in clinical pharmacology – a tale of two worlds

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Experimental studies of drug effects in man in 2008 are not to be undertaken lightly. Older heads of departments may still nostalgically remember the days when a study started about one week after the three page protocol was written. The current generation knows that 80 page protocols, 300 page Investigational Medical Product Dossiers, ethics committees, competent authorities, and GCP and GMP loom on the rocky path to the first measurement in the first subject to be recruited. No wonder these clinical scientists look upon the world of epidemiological investigation with envy. A meta-analysis, or other form of systematic review, requires little more than an internet connection to the electronic library and your own creativity and data analysis capability, although the personal effort can be of course just as large as for an experimental study. Yet the cost per produced publication is a fraction of that of a prospective intervention study involving human subjects. The same goes for the owners of large registries of observational data. So, there are two different worlds in which a clinical pharmacologist can operate. One world contains existing data that need to be integrated or (re)analysed, while in the other the data are carefully collected with enormous effort and at a huge cost.

This issue of the *Journal* includes a number of important reviews and observational studies from the world of existing data, starting with Edward Calabrese, who outlines the importance of hormesis. Hormetic dose response curves, which indicate opposite actions at different concentrations, have been shown in various biological systems, from isolated cells to intact animals and humans. This is perhaps not surprising and is in line with Paracelsus's observation that everything becomes toxic at high concentrations]. The mechanism of the hormetic dose response is not always known. The observation that male rats have enhanced penile erection after yohimbine at low plasma concentrations but seem to lose interest at high concentrations suggests the inevitable hypothesis that the high dose may have given them a headache. Whatever the mechanism, biphasic dose response relations will make drug development a lot harder and may be the reason for

some development failures, as the probability of hormesis is seldom taken into account [1].

The huge database of Australian veterans and their families ($n = 273\,228$) provided an opportunity for Clare Ringland and colleagues to study potentially dangerous combinations of treatments that may cause 5HT toxicity. Of course, the database does not allow definite conclusions, as it is not known from the data if the drugs were really taken together, or indeed taken at all, and, if so, if they were actually taken concomitantly, but the authors make a credible case that physicians do not always take this into account. Unfortunately, we will not know if serotonin syndromes actually occurred in this population, as those data were not available [2]. This is a good example of how such registries can yield data that cannot be obtained experimentally, as an ethics committee would not allow obvious medication errors to be introduced randomly.

When researchers from Israel, Germany, and Italy came together to investigate possible effects of fluoxetine and paroxetine in pregnancy using data from teratological information databases in their countries, they found an association between the use of SSRIs in the first trimester and cardiovascular anomalies. Of course, such data do not allow definitive conclusions; after all the women were also depressed and smoked more, and causality cannot be assumed in the absence of a randomized prospective study after randomization. This is again the best one can do to investigate this difficult area, which is fraught with potential biases and confounders. The data do not change clinical practice, as one hopes that these medicines are not used in pregnancy anyway, unless the harms (to both mother and child) and benefits are carefully evaluated [3]. Another huge database of more than 10,000 Canadians was investigated by Sylvie Perreault in Quebec where she detected that better adherence to statins (or at least the possession of the tablets as the taking of the tablets remained of course unobserved) gave a better outcome [4].

So, where in this issue of the *Journal* are the carefully performed, well controlled, randomized experiments that

are also part of the work of the experimental clinical pharmacologist? Yuka Tochihara *et al.* save the day, with a study of the effect of folic acid on endothelial function [5]. They made sure that folic acid concentrations went up during hemodialysis in one group and down in the other, and this affected the pulse wave in the radial artery, measured with a SphygmoCor® device, about which more later. Unfortunately, there was no placebo control, nor placebo run-in and the authors do say that more needs to be done, which leaves us in expectation of more experimental work in this interesting patient category with chronic endothelial dysfunction. There are also contributions on two of the oldest medicines known to man, opium and ethanol (alcohol), demonstrating that important findings are not limited to the new molecules. One is from Andrew Somogyi and colleagues who did extremely important experiments to find the optimal dosing regimen for tincture of opium in addicts and Remco Zoethout experiments with ethanol showing that it is possible to keep ethanol concentrations in the plasma stable for a long time. This may not come as a surprise to some of our readers but we do not advise to try his method at home!

The preponderance of database studies and evaluations of registries and meta-analyses analysis (if not in this issue, a number of papers certainly in the *Journal* number of patients observed) contrasts with the November issue of the *Journal* 30 years ago (1978), when the published work was largely experimental or methodological. Peto [6, 7] has always maintained that systematic reviews and randomized evidence with adequate power are complementary; a meta-analysis on its own is no more than a stimulus to get randomized data. Meta-analysis predicted the effects of aspirin in myocardial infarction almost perfectly, but failed badly when mis-predicting an effect of magnesium sulphate in myocardial infarction [8]. So, randomized experiments with important clinical outcomes, as well as the more physiologically oriented experiments that have always been part of the clinical pharmacologist's toolkit, remain essential. The equipment used for obtaining experimental data is of course an essential part of this toolkit.

Development of the experimental toolkit

We publish as supplementary material to this issue of the *Journal* two historical films. One was made at the National Institute of Medical Research (NIMR) in the laboratory of Henry Dale [9] and William Paton in 1948 and the other at the Royal College of Surgeons' laboratory in the 1960s. Both films are extremely amusing, and it is fascinating to watch John Vane performing data manipulation or Gustav Born supervising an experiment smoking a pipe. Try to spot as many famous British pharmacologists as you can. The Pythonesque quality of the films is also not to be

missed, but there is another reason why we think this is important material.

These films represent life in the laboratory in those days in quite a bit of detail. What comes through in both films is the intense relationship researchers had with their equipment, which was entirely analogue and mechanical and generally home-built. The recording kymographs were connected to isolated organs or tissues by strings, pulleys, and levers, and any malfunction (which presumably occurred frequently) could immediately be seen and corrected. This is in stark contrast to modern equipment, which is digital and connected to computers with proprietary software, and of course has a CE mark, so that when it breaks down it has to be sent somewhere foreign. Perhaps of more concern is that the equipment is not at all transparent in its workings to the researcher, who takes it straight out of the box. Potentially, it can produce results that are meaningless. The pulse wave analysis work in this issue was done with a SphygmoCor® device [5]. This can be bought as a neat package, with dedicated software, and it records and analyses pulse waves using a very stable and reliable system, which anyone can use with only a little training. However, it is very difficult to find out what goes on inside. We believe there is a point here for the practicing and future translational scientist (otherwise known as the experimental clinical pharmacologist). We should ask ourselves if we know enough about our toolkit. And assuming that one never knows enough, we launch in this issue of the *Journal* a new series on methods in clinical pharmacology.

BOX 1

Potential topics for methods articles. These articles will be marked with the logo to indicate that they are methodology papers

- Measurement of endothelial function
- Pharmacodynamics of the proteome
- Metabolomic analysis by NMR and MS
- Modern assay techniques for drugs
- Immunomonitoring and evaluation of the function of the components of the immune system
- Pulmonary function testing and pulmonary challenge models
- Gene chip and micro-array analysis
- Neurophysiological and cognitive measurements
- Assessment of brain function by fMRI/PET
- Polysomnography
- Measurement of renal function-Inflammatory ex vivo and in vivo models in man
- Dermatological measurement techniques
- Measurement of gut motility
- Endocrinological challenge tests
- Studying drug-drug interactions
- Data mining and advanced biostatistical techniques
- Pharmacogenetic measurement techniques
- -In vivo studies of receptors
- -Assessing bioavailability
- -Randomised trial design
- -Pharmacoepidemiological study design
- Pharmacoeconomical methods
- -Measurement of adherence

To do so we remind you of the past, with a commentary on the first pharmacological recordings with the kymograph, a venerable piece of equipment on which many drug effects were recorded for the first time. The paper includes some original recordings [10]. Pharmacologists do not have to operate kymographs any more, and unlike the pharmacologists in the NIMR they do not have to do their experiments covered in soot (which was used to get the black background on the revolving drum of the kymograph and was apparently produced by burning benzene). However, they still need to know the technical aspects of many techniques. When you look at the blood pressure curves obtained by the inventor of the kymograph, Carl Ludwig, with an entirely mechanical piece of equipment, you realize that this is exactly the same as the Sphygmo-Cor® recordings of pulse waves, only simpler and more transparent for the non-engineer.

There is a need for papers that give a good technical overview of the physiological or biochemical background of a certain technique or group of techniques, explaining the working of modern equipment and its major applications, together with instructive examples. In the box we list potential subjects that could be covered, but the subjects do not have to be limited to this list. We shall guard against papers that cover an area that is too broad ('Measurement of the CNS effects of drugs') or too narrow ('Recording the first heart sound by phonocardiography') but otherwise we leave the content to experts in any types of methods useful for experimental clinical pharmacology. Such papers should have a high 'how-to' nature, and there are ample opportunities for multimedia support; colour graphics in the publication of software and video as supplementary material are all possible. The editorial board will ask known experts for contributions, but will also look with interest and an open mind and accept suggestions for contributions on this topic. Do contact the editorial office first to make sure there is no duplication.

We intend to publish these methodological papers under one heading, and eventual bundling of the contributions is expected to make a very useful reference for anyone wanting to enter the experimental world. The historical material may be amusing, because it appears to reflect a lost world, but we need experiments and experimental techniques more than ever. There are still many exciting findings yet to be made, and these will in time find their way into systematic reviews. We hope to keep publishing messages from both worlds.

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Supporting information

Additional Supporting Information may be found in the online version of this article:

Video Clip S1. A Career in Pharmacology. The video clip is in Quicktime.

Video Clip S2. Pharmacology at the National Institute of Medical Research 1948. The video clip is in Quicktime.

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