REFERENCES

Horder, Dodds, E. C. & Moran, T. (1954). *Bread.* London: Constable.
Jackson, S. H., Doherty, A. & Malone, V. (1943). *Cereal Chem.* **20**, 551.
Jones, C. R. & Moran, T. (1946). *Cereal Chem.* **23**, 248.
Kent-Jones, D. W. & Amos, A. J. (1947). *Modern Cereal Chemistry*, 4th ed. Liverpool: The Northern Publishing Co. Ltd.
Kent-Jones, D. W., Amos, A. J., Martin, W., Scott, R. A. & Elias, D. G. (1956). *Chem. & Ind.* p. 1490.
Moran, T. & Drummond, J. (1945). *Lancet*, **248**, 698.

Problems and pleasures of human experiments

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Introduction

No one, I believe, has so far made experiments on his fellow men to find out how to feed his tame rats, yet many people have worked on rats with the firm belief that their results could be applied directly to the nutrition of their fellow men. Investigations on animals have been of inestimable value in nutritional research, but Claude Bernard (1865) saw the dangers of generalizing too widely from the results obtained from them: 'Les expériences pratiquées sur le chien ou sur la grenouille ne pouvaient, dans l'application, être concluantes que pour le chien et pour la grenouille, mais jamais pour l'homme'. And we have always felt that if one's chief concern was man the crucial experiments should, if possible, be made on him.

Whatever the nature of the investigation, whether it concerns bread or anything else, experimental work on man presents problems and gives pleasures which are to be found in no other kind of work. Strangely enough, man is the only mammal for which a vivisection licence is not required. This makes things easier in some ways, more difficult in others. It makes things easier because no records have to be kept for the Home Office, and there is no necessity to get a new certificate signed by the President of the Royal Society or one of the Royal Colleges every time the experimental lay-out is changed. It makes things more difficult because the responsibility of the investigator is very much greater. He is not protected by the Home Office and, although if he is working with fellow scientists he can explain to them the nature of the experiment and get their consent, if he is working with children, for example, the whole responsibility rests with him. The parents or guardians must, of course, agree to the investigation being made, and be told in general terms the nature of the experiment, but they are unlikely to understand it fully, and they have to trust the investigator. It goes without saying that no experiment must ever be made on a human being that could within reason be expected to do him any permanent harm. This limits the nature of the experiments, and one hesitates, for example, to deprive a child for any length of time of any food or dietary constituent that is known to be beneficial to him. It also limits the criteria by which the effects of a food or a diet may be judged, so that these are restricted mainly to body measurements, clinical examinations (including special ones such as radiological and dental examinations) and analysis of products of the body that can easily be obtained. These are generally blood, urine and faeces. Man has the advantage of being a comparatively large experimental animal, and even children generally provide plenty of material on which the necessary tests may be made.

The design of the experiment

Nutritional experiments on man can be divided into two main groups. Firstly there are detailed studies on a small number of individuals. These may include many kinds of clinical examinations and tests, but they usually involve in addition the quantitative collection and subsequent analysis of the excreta and duplicate portions of the food. This is the laborious and difficult part of the work, and it is what limits the number of subjects which can be investigated at one time. Secondly, there are feeding experiments on larger numbers, in which the effects of the diet are judged simply by changes in weight, height and other body measurements, the results of clinical examinations, or changes in the concentration of some constituent in the blood or in isolated specimens of urine.

As in all research, before beginning to design the nutritional experiment the first thing to do is to define the problem, and then to consider the experimental approach that is most likely to solve it. It is no use setting out vaguely, for example, to discover whether brown bread is a better or worse food than white bread. The problem must be much more exactly defined. Are we interested, as the 19th century investigators were, in the digestibility of the two sorts of bread? Are we interested in the two breads as purveyors of vitamins? Are we concerned with their proteins, or with their effect on mineral metabolism? These are some of the ways in which we ourselves have had to deal with bread from time to time, and each of them demands a different experimental approach.

Having defined the problem, we must next consider what kind of experiment is likely to give a clear-cut answer and how best we can demonstrate any differences that there may be between the breads. Should we use adults or children as our subjects? Should they be previously well-nourished or undernourished in respect of the constituent in which we are interested? How many subjects do we need? Should half of them eat white bread and half eat brown bread all the time, or should half begin on white and half on brown and then cross over in the middle? Individuals vary very much in their ability to absorb and utilize the constituents of their diets, and if we are doing metabolic work on small numbers, a crossed-over experiment is better, so that each person acts as his own control, but if we are working with large numbers the other approach may be preferable. Then we must decide what proportion of the diet should be made up of bread and of what the rest of the diet should consist. How much control do we need to have over the rest of the diet? Do we need a preliminary period before the main experiment begins to allow the subjects time to become adjusted to the experimental diet, or to become depleted of some dietary constituent? For how long should we plan to continue our experiment? If we are doing metabolic work, weighing all the food and collecting all the urine and faeces, there are obvious practical difficulties about asking people to do this for very long periods at a time. If we are making observations which are less demanding on the subjects' lives, periodic weighings and clinical examinations for example, then we can go on much longer. But man has a long life compared with most laboratory animals, and any experimental period is likely to be a small proportion of it. The investigator has the same life span as his subjects, so he clearly cannot carry his experiment through many generations. Critics are apt to say afterwards that the experiment was not continued for long enough, particularly if it does not give the result they expected, but in general the policy should be to continue for at least the shortest time which we think will give a scientifically valid result. In designing experiments, whether on animals or man, it is wise to consider beforehand whether the results are likely to lend themselves to statistical treatment and, if so, to consult a statistician at the outset about the design of the experiment. Then we have to decide by what criteria we are going to judge the effects of the two kinds of bread. For what constituents are we going to analyse the blood? What particular points are we looking for in our clinical examination? How do we assess quantitatively the results of our clinical examination? These are just a few of the questions that have to be considered in planning nutritional experiments on man, and the answers to all of them depend on the particular problem that we set out to solve. The proper design of the experiment is all-important, and no amount of statistics can put right an investigation that was badly projected. The practical side is important too, and ideals may have to be surrendered for practical reasons, but it is well worth while planning the perfect experiment in the first instance, and then seeing how nearly the perfect design can be put into practice.

The practical working out of the design

I am going to illustrate this part of my paper by referring to some of the problems and pleasures we have had in making the two types of nutritional investigation to which I have referred. Most of our experiments with bread involving balance studies were made during the early years of the war, when we were primarily interested in the effects of the phytic acid in brown flour on calcium absorption. We have never had any great difficulty about finding subjects for our investigations, but there is no doubt that in wartime or in times of food shortage it is easier to persuade people to act as subjects than it is in times of peace and plenty. In the United States it is customary to pay the experimental subjects, but in this country work has almost always been done on volunteers. Krebs & Mellanby (1942) used conscientious objectors; we have always worked with colleagues, and we think it is very desirable that the investigator should act as an experimental subject, and do nothing to others that he is not prepared to do to himself. Any sort of nutritional experiment is bound to curtail one's social activities, and our metabolic experiments lasted for nearly a year, with only one weekend free every 4 weeks. In these balance studies all our food had to be weighed, and a similar portion of food taken for analysis. All urine and faeces had to be collected, which meant that a bottle had to travel with us everywhere we went. We had to rely on our subjects to remember to collect all their urine and faeces—we could not put them into a metabolism cage as we would an animal! As a matter of fact it is not hard to remember after a day or two, and it soon becomes such a habit to collect all the urine that one has quite a guilty feeling in not doing so when the experiment is over.

In our investigations we had four men and four women in the experimental party and we studied the calcium absorption from various kinds of bread—white bread, brown bread, white bread and brown bread with added calcium carbonate, the two breads with added calcium phosphate, white bread with added sodium phytate and brown bread after the phytate had been removed (McCance & Widdowson, 1942*a*,*b*). In each experiment we compared two kinds of bread and we used a crossed-over design. We worked in weekly periods, and each experiment lasted for 3 weeks, with a 3-day preliminary period when the kind of bread was changed. We had all our meals at the laboratory, and after every meal one-fifth as much as each person had eaten of each food was weighed out and put into his bowl for analysis. The food was collected in the same bowl for a week. Those were the days before Waring Blendors were so common, and at the end of the week the food in the bowl was mixed by hand in a large mortar. The storage and transport of urine and faeces was one of our problems. All our subjects slept at home so they needed vessels at home as well as at the laboratory, and these had to be collected up by car every week. In those pre-Blendor days, the faeces like the food, were mixed by hand, with a thick glass rod flattened at the end.

These were some of our problems, but the work brought us great pleasures too. At the end of our experimental study of rationing during the first months of the war, for example (McCance & Widdowson, 1946), we all went to the Lake District in the depths of winter to test our physical fitness on our experimental diet.

Our feeding experiments in Germany (Widdowson & McCance, 1954) also had their problems. We worked in orphanages where none of the staff or children spoke a word of English. At one home the couple in charge believed that it was morally wrong for children to have as much to eat as they wanted, and we had to overcome this. Food was very short at the time, and we held large stocks of flour which had to be protected from thieves. At the beginning of the investigation we took great care to make our groups of children as similar as possible in every way. Each group contained the same number of boys and girls, of similar ages, and of similar height, weight and clinical gradings. Then came an epidemic of scarlet fever in one home, and several children had to go to the fever hospital and were lost to the experiment, thus upsetting the balance of the groups. The parents of others came out of prison, and demanded to have their children home, which upset the groups still further. Fresh children were admitted to the orphanages, and we had to decide whether to include them in the experiment or not.

The diets were very simple and the feeding arrangements at the orphanages ran smoothly. A dietitian was in charge at each orphanage. The five bread groups at the larger home had their meals in different rooms, and the five breads all looked different, so there was no danger of getting them mixed up. The children developed such a sense of loyalty to their own particular bread that when a party of them was taken to the zoo, each child instinctively picked out a piece of his or her own colour from the parcel of scraps that was taken to feed the animals.

At the beginning of the experiment we had serious qualms about allowing children to eat a diet containing so much white bread, and we were prepared to terminate the experiment at any moment, should any signs of B-vitamin deficiencies appear. As it turned out that was never necessary. The medical officers of health in the two towns in Germany where we were working supported us from the beginning. I think they regarded us as a relief organization, and were only too thankful when we suggested providing the children with extra food. They supported us still more strongly when they saw how all the children were improving as a result of our bread, and for us also, of course, it was a great pleasure to see how much good children could derive from bread—whatever its colour.

The interpretation of the results

Probably the first nutritional experiment on man was made about 600 B.C. by Daniel, a young man 'in whom was no blemish, but well favoured, and skilful in all wisdom, and cunning in knowledge, and understanding science . . . . . .

Then said Daniel . . . . . . Prove thy servants, I beseech thee, ten days; and let them give us pulse to eat, and water to drink.

Then let our countenances be looked upon before thee, and the countenance of the children that eat of the portion of the king's meat: and as thou seest, deal with thy servants.

So he consented to them in this matter, and proved them ten days.

And at the end of ten days their countenances appeared fairer and fatter in flesh than all the children which did eat the portion of the king's meat.' (*Daniel* 1, 4, 11–15).

It seems that the countenances of all ten experimental subjects appeared 'fairer and fatter in flesh' than those of any of the subjects in the control group 'which did eat the portion of the king's meat' and, when the result is as clear-cut as this, statistical analysis of the results is quite unnecessary. How should one interpret the results of Daniel's experiment? Should one conclude that pulse and water is a better diet than the king's meat for all persons in all circumstances? Certainly not. Children might not have grown so well on it. If the experiment had continued for more than 10 days, deficiencies of the pulse and water diet might have shown up. All one can conclude from Daniel's experiment is that over a period of 10 days a diet of pulse and water produced fairer, fatter faces in healthy young men than a diet of king's meat. Daniel clearly expected this result, and fortunately for him he obtained it, but this is not always so, and an honest investigator must always keep an open mind. If he expects one result and gets another, he must be prepared to think again. Often an unexpected result is more interesting than the expected one, of which we had a good example while we were working in Germany (Widdowson, 1951). For 6 months we followed the heights and weights of about 100 children in two orphanages, A and B. These children were living on the German rations, which were not enough for them. We then gave extra brown bread to the children in orphanage A but not to the others, and we continued to weigh and measure the children for another 6 months. During the first half year the children in home A gained three times as much weight as those in home B, although their rations were the same. The children in home A then received the extra bread, but from the time they began to eat it they grew more slowly, and the children in the other home B, who got no extra food, began to gain weight at a much faster rate. We knew that the children in home A ate the extra bread, for we weighed all their food, and their calorie intakes were 20% higher during the second 6 months than the first, and yet they gained less weight. Are we to conclude that extra brown bread stops children growing, or was there some other factor at work which was outweighing any beneficial effects of the extra calories we were providing?

At the beginning of the investigation home B was presided over by Fräulein Schwarz. Just at the time when we began to give the extra bread to the children in home A, the authorities transferred Fräulein Schwarz from home B to home A. Fräulein Schwarz was a very harsh, unsympathetic woman, and children and staff lived in constant fear of her reprimands and criticisms, which were often quite unreasonable. She chose mealtimes to scold the children publicly and she would single out individual children for special ridicule. By the time she had finished the food would be cold, all the children would be in a state of agitation and several would often be in tears. Anyone who is embarking on human experiments will do well to remember 'Better is a dinner of herbs where love is, than a stalled ox and hatred therewith' (*Proverbs*, 15, 17).

REFERENCES

Bernard, M. C. (1865). *Introduction a l'Étude de la Médicine Expérimentale.* Paris: J. B. Baillière et fils.
Krebs, H. A. & Mellanby, K. (1942). *Lancet*, **242**, 319.
McCance, R. A. & Widdowson, E. M. (1942*a*). *J. Physiol.* **101**, 44.
McCance, R. A. & Widdowson, E. M. (1942*b*). *J. Physiol.* **101**, 304.
McCance, R. A. & Widdowson, E. M. (1946). *Spec. Rep. Ser. med. Res. Coun., Lond.*, no. 254.
Widdowson, E. M. (1951). *Lancet*, **260**, 1316.
Widdowson, E. M. & McCance, R. A. (1954). *Spec. Rep. Ser. med. Res. Coun., Lond.*, no. 287.

The report of the panel on flour

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The Panel on Composition and Nutritive Value of Flour was appointed in May 1955 by the Secretary of State for Scotland, the Minister of Agriculture, Fisheries and Food and the Minister of Health. The members of the Panel were nominated at the request of the Minister of Agriculture, Fisheries and Food by the President of the Royal Society (Lord Adrian, O.M.). The Chairman was Professor Sir Henry Cohen (now Lord Cohen of Birkenhead) and the other members were Dr A. C. Chibnall, Professor J. H. Gaddum, Professor R. A. Morton and Professor L. J. Witts (Great Britain. Parliament, 1956).