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A randomized, controlled, double-blind, cross-over, clinical trial of Q fever vaccine in selected Queensland abattoirs

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SUMMARY

A limited, randomized, blind, placebo-controlled trial of Q fever and influenza vaccines has been conducted in three Queensland abattoirs on a sequential analysis design. Ninety-eight subjects were given Q fever vaccine and 102 influenza vaccine. Q fever cases were observed in unvaccinated workers in all three abattoirs during the period of observation.

A total of seven Q fever cases in one group, one more than the number required to achieve statistical significance between the two vaccine groups, was reached after 15 months with the cases coming from two of the abattoirs. These Q fever cases were in the group which had been given influenza vaccine and none in that given Q fever vaccine.

Symptomless seroconversion rates of 24% were found in the remaining influenza virus vaccinees, and those without immunity were given Q fever vaccine.

INTRODUCTION

An accompanying paper [1] describes a large scale open trial of a Q fever vaccine (Q-vax, Commonwealth Serum Laboratories, Melbourne (CSL)) in four South Australian abattoirs during the period 1981–8. This established that in vaccinated subjects pretested for existing immunity, levels of reactogenicity were acceptable. One 30 μg dose of formalin inactivated Henzerling strain, Phase 1 *Coxiella burnetii* antigen appeared to confer complete protection against natural infection in the work place after 10–15 days had elapsed from vaccination and immunity appears to persist for at least 5 years as judged by epidemiological observations [1] and persistence of T lymphocyte sensitization to *C. burnetii* antigens [2].

Q fever was first described by Derrick [3] in the metropolitan abattoirs in Brisbane, Queensland, and since that time, the state has reported the highest incidence of Q fever of all Australian states.

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Because of this severe morbidity, after an initial report [4] of the South Australian trial, it seemed highly desirable to introduce vaccine prophylaxis into Queensland abattoirs.

Apart from this public health requirement, some vaccinologists and regulatory authorities consider that a randomized, blind, placebo-controlled trial is essential to establish the protective efficacy of any vaccine.

Although inactivated Q fever vaccines have been used to protect laboratory workers since the late 1940s and have given high rates of protection in several volunteer-challenge trials [5, 6], a blind placebo-controlled trial has not, as far as we are aware, been conducted anywhere in the world.

For the above reasons, it was decided to mount a small placebo-controlled trial in three Queensland abattoirs. Given the apparent protective efficacy in past open trials in laboratory workers, in volunteer-challenge trials and in South Australian abattoir workers, it would have been unethical to conduct a large scale randomized trial with a large placebo group exposed to acute Q fever and its significant sequelae. However, it did seem ethically acceptable to limit the trial to a comparison of two vaccines given under code, on a sequential analysis design [7], in the anticipation that if the Q fever vaccine was as protective as the open trial had suggested, then only a few Q fever cases in one vaccine group would be required to establish a significant difference between the two vaccine treatments.

This proposal was accepted by the University of Queensland Ethics Committee and a trial of Q fever vaccine compared with influenza virus vaccine was mounted with the results described below.

ORGANIZATION OF TRIAL AND METHODS

Abattoirs

Two country abattoirs, Kilcoy and Beenleigh, respectively some 120 and 50 km from Brisbane, and the Metropolitan Regional abattoirs, Colmslie, Brisbane, were chosen for the study. The country abattoirs offered the advantage that sick abattoir workers would be seen, not only by staff in the medical stations at the works, but also by a limited number of general practitioners, familiar with Q fever. The Metropolitan Regional abattoirs offered the advantage of a larger workforce and a well-staffed medical station which could monitor suspected cases of Q fever.

Subjects studied and vaccination protocol

Volunteers were sought from the workforce at each of the three abattoirs and the placebo-controlled nature of the trial was carefully explained to them. Those agreeing to participate gave comprehensively formulated and precise informed consent which very clearly covered the possibility that they might not get Q fever vaccine and therefore might not be protected. Some of the volunteers had taken part in past serological surveys as part of the Queensland Zoonotic Diseases Research Project and those who were known to be seropositive were excluded. The remainder were serotested for Q fever CF antibody according to standard protocol [1, 4, 8], and the negatives were allocated alternately to one or other vaccine groups. The next week they were skin tested [4] with a 1 in 300 dilution of the vaccine they were to receive. Skin tests were read 5–7 days later and positive

reactors [4] excluded. In order to prevent bias, the skin tests were read in an additionally blind manner. The reader was not informed of just which coded skin test vaccine dilution the volunteer had received until after a decision had been made on the skin test reaction. There was great attention paid to the standardization of the reading of the skin tests, with only two readers used throughout the trial and with consultative decisions made in all skin tests.

Those persons without Q fever antibody and a negative skin test were then injected subcutaneously with 0·5 ml of the coded vaccine of the group to which they had been allocated.

It would have been more convenient to skin test all the volunteers with the Q fever vaccine. That course of action was refrained from, in the protocol, in order to eliminate the possibility that even the small dose of skin test antigen might confer a degree of immunity to the members of the placebo group; i.e. those who were to receive the influenza vaccine.

Monitoring of vaccinees and other workers in the three abattoirs for Q fever cases; laboratory confirmation

Arrangements for the detection of cases differed slightly in the various centres. In Kilcoy, the two general practitioners in the town were the source of notification and verification. The town is circumscribed and the meat works are serviced by the same two doctors, via the abattoir Medical Centre, on a rotating and daily basis. In addition, the State Microbiological Laboratory, Brisbane, notified us of any suspect cases of Q fever with positive serological results, and the local private Pathology Services did likewise. In Beenleigh, the arrangements were similar to those in Kilcoy, with the difference that a single general practitioner serviced the Medical Centre at the works.

Detection of Q fever cases at the Metropolitan Regional abattoir was more difficult as workers lived in a number of different Brisbane suburbs. However, reports of positive serological results were received from the two Private Pathologists in the city, and from the State Microbiological Laboratory in addition to the notifications from general practitioners.

In addition to the above surveillance measures, sickness certificates brought by workers to the personnel branches of the three abattoirs were scrutinized for possible Q fever cases and, as Q fever is a compensatable disease, records of payments were also checked.

Laboratory confirmation of Q fever cases

Acute and convalescent phase sera were collected from all suspected Q fever cases and tested for complement fixing (CF) antibody to *C. burnetii* Phase 1 and 2 antigens at the State Microbiological Laboratory. As some Q fever patients may not develop CF antibody, or only develop it in late convalescence, sera were also tested in Adelaide by immunofluorescence (IF) on coxiella microdots with conjugates against IgM, IgG and IgA antibody [8]. The seven Q fever patients contributing to a definitive outcome in the trial all showed a fourfold or greater increase of CF and IF-IgM antibodies in serial specimens of sera.

Vaccines used

The vaccines used were CSL's Q fever vaccine (Q-vax) and Influenza A vaccine (Flu-vax). Both vaccines were presented in clear glass ampoules, which were identical, except that the first was labelled 15 and the second batch was labelled 01. It was impossible to detect any difference between the ampoules in the two batches; nor were there any detectable differences in the contents. A test of their visual appearance by staff and medical students in the Department of Social and Preventive Medicine, before the trial, showed that no observer was able to detect any difference. After the termination of the trial, the contents of ampoules, selected at random, were assayed for *C. burnetii* or influenza antigen in Adelaide, in order to verify that they were true to label and there were no discrepancies.

The sequential design and analysis

From the results of the South Australian vaccine trial it was possible to deduce that, whatever the true potency of the vaccine, it was not plausible that it would increase susceptibility to Q fever. Under these special circumstances, a one-sided test of protection versus no protection was legitimate, particularly if, as here, an initial non-reporting interval was assigned to eliminate cases of Q fever in persons vaccinated during the incubation period of a natural attack. To minimize delay in obtaining a result, one of Armitage's closed sequential designs, which are two-sided, was modified to meet the requirements of a one-tailed alternative hypothesis. The alternative hypothesis for this trial specifies an incidence parameter ratio of 9:1, that is, nine cases of Q fever expected among those given the placebo to one in those receiving Q fever vaccine (given equal numbers at risk); the null hypothesis specifies a parameter ratio of 1:1, which becomes 98:102 after allowing for the initial numbers in the two groups. Type I and Type II error probabilities were both set at 0·05; for these design parameters the maximum sample size is 21, but a result is possible after only six confirmed cases of Q fever in one or other of the comparison groups. This design is displayed graphically in Fig. 1.

RESULTS

Three hundred and seventy-three Q fever seronegative workers at the three abattoirs volunteered to take part in the trial. Twenty-three subjects gave a positive skin test to one or other of the two vaccines and were excluded from the trial. Finally, after some withdrawals, a total of 200 subjects remained for vaccination, 98 in one group and 102 in the other (Table 1). The numbers were not exactly balanced because although seronegative subjects were allocated alternately to each vaccine group, subsequent skin testing with one or other of the diluted vaccines gave more positive skin tests, and hence exclusion, in the Q fever vaccine than in the influenza vaccine group.

The two vaccinated groups at the three abattoirs were then followed for Q fever cases. Table 2 shows the distribution of verified Q fever cases in vaccinated and unvaccinated subjects in each abattoir. Figure 2 shows the cumulative total of Q fever cases in the influenza vaccine (placebo) group over time.

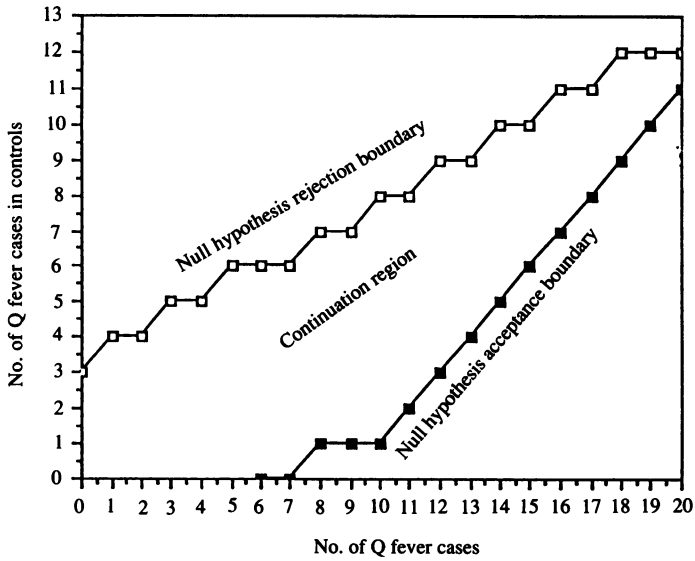

Fig. 1. Design for a closed, one-tailed sequential probability ratio test, $\alpha = \beta = 0{\cdot}05$; incidence ratio = 9·0.

Table 1. *Summary of numbers of volunteers at the three study abattoirs tested for Q fever antibody and skin tested with one or other of the coded vaccines, and if negative, then inoculated*

Abattoir	No. of volunteers screened	Skin test positive, either vaccine	No. inoculated and vaccine given: Q fever vaccine (15)	No. inoculated and vaccine given: Influenza vaccine (01)
Kilcoy	148	9	41	30
Beenleigh	103	5	21	28
Regional Metropolitan	122	9	36	44
Total	373	23	98	102

Table 2. *Number of vaccines given, under code, Q fever vaccine (15) or influenza vaccine (01), and followed for Q fever cases resulting from natural exposure in the three abattoirs. Data is also given on the number of Q fever cases in unvaccinated workers during the 15-month period of the trial*

Abattoir	Q fever vaccine (15): Vaccinees	Q fever vaccine (15): Cases	Influenza vaccine (01): Vaccinees	Influenza vaccine (01): Cases	Unvaccinated cases*
Kilcoy	41	0	30	5	8
Beenleigh	21	0	28	0	2
Regional Metropolitan	36	0	44	2	5
Total	98	0	102	7	15

* In the abattoir during the 15 months of the trial.

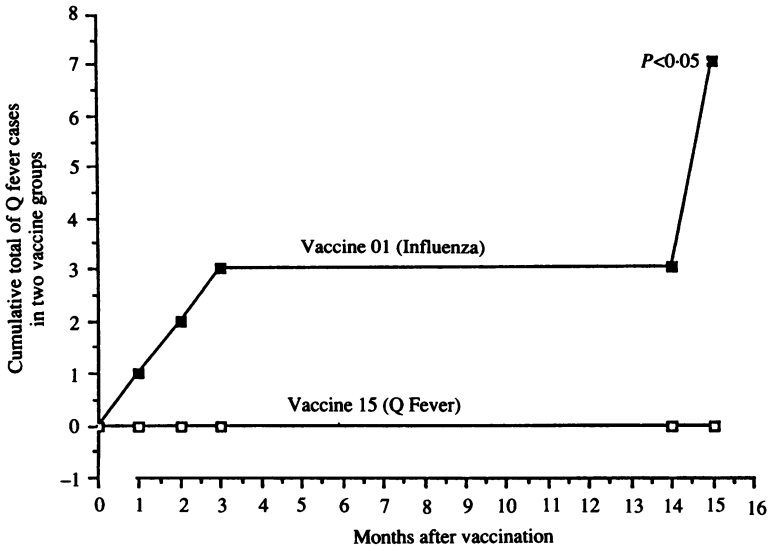

Fig. 2. Differences over time since vaccination in the cumulative totals of Q fever cases in two groups, one given Q fever vaccine (No. 15) and the other influenza vaccine (No. 01), see text.

The required sequential test boundary (six or more confirmed cases in one group, none in the other) was first passed 15 months after the commencement of vaccination. The trial was then terminated and the code for the vaccines broken; it was then found that all seven confirmed Q fever cases were in the group that had received influenza vaccine and that there were none in the group that had received Q fever vaccine.

When the rolls of the volunteers were checked, it was found that only three volunteers were lost to follow-up. All three were in the group that had been vaccinated with influenza vaccine.

All the remaining participants in the trial who had received influenza vaccine, but had not been ill, were then serotested and skin tested with Q fever antigen, and if negative in both tests, were offered (and given) Q fever vaccine. No further Q fever cases have occurred in the group after this one-way 'cross-over'. Comparisons of the results of the first and second Q fever serotesting in the group showed that 24% had seroconverted by complement fixation test, without illness, during the 15 months when they had been unprotected following the administration of influenza vaccine.

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