Q fever: hazard from sheep used in research

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The recent recognition that Q fever is endemic in Ontario and the known occupational risk of Q fever to research personnel working with sheep prompted a study to determine the prevalence of antibodies to the causative organism, *Coxiella burnetii*, in animals and staff at a Toronto animal research institute. Of 37 sheep 34 (92%) were found to be seropositive — that is, to have a titre of complement-fixing antibody to the phase II antigen of 1:8 or greater. Of 331 staff members tested, 18% were found to be seropositive, compared with 0.6% of a random sample of Toronto blood donors. The highest rate of seropositivity, 68%, was in the 28 animal attendants tested. Seropositivity was associated with working with sheep or fetal lamb tissue ($p < 0.0001$) and with visiting the animal facility ($p < 0.001$). Of the 59 seropositive staff members 63% had had no direct contact with sheep. There were 12 clinically apparent cases of Q fever, 2 of which required admission to hospital. Q fever remains a serious occupational hazard to staff working in research laboratories using sheep, even to those with indirect exposure to infected animals.

La confirmation récente du fait que la fièvre Q est endémique en Ontario et la reconnaissance du risque professionnel de fièvre Q auquel sont exposés les chercheurs scientifiques qui travaillent avec des moutons sont à l'origine de cette étude, qui avait pour but de déterminer la prévalence des anticorps contre l'agent pathogène, *Coxiella burnetii*, parmi les animaux et le personnel d'un institut de recherche chez l'animal de Toronto. De 37 moutons 34 (92%) se sont avérés séropositifs, c'est-à-dire, le titre d'anticorps fixant le complément contre l'antigène phase II était de 1:8 ou plus. De 331 membres du personnel 18% étaient séropositifs, comparativement à 0,6% d'un échantillon aléatoire de donneurs de sang de Toronto. Le plus fort taux de séropositivé, 68%, a été retrouvé chez les 28 animaliers. Une séropositivité était associée avec un travail nécessitant le contact avec des moutons ou avec du tissu embryonnaire d'agneau ($p < 0,0001$) et avec une visite à l'animalerie ($p < 0,001$). Des 59 membres du personnel séropositifs 63% n'avaient au aucun contact direct avec des moutons. On a compté 12 cas de fièvre Q cliniquement apparents, dont 2 ont nécessité l'hospitalisation. La fièvre Q demeure un risque professionnel sérieux pour le personnel de laboratoires de recherche qui utilisent des moutons; cela est le cas même pour les employés qui n'ont qu'un contact indirect avec des animaux infectés.

Q fever is a disease of those who work with or live in close association with livestock, particularly cattle, goats and sheep. The sheep has become a common animal model in medical research, and several outbreaks of Q fever have been reported in research centres using sheep.[1-6] We recently recognized that Q fever is endemic in Ontario[7] and reasoned that some research workers in this province may have acquired the disease through contact with animals. We therefore studied the prevalence of antibodies to the causative organism, *Coxiella burnetii*, in the animals and staff of a Toronto research institute that had been using a sheep model for perinatal research for the past 10 years. In this article we describe an outbreak of Q fever among the research personnel, which, despite considerable morbidity, was unrecognized before the study. As in other reported outbreaks,[3-5] the disease was a potential hazard not only to those directly involved with sheep research but also to those whose contact with the animals was brief and unintentional.

The setting

The Hospital for Sick Children is a 700-bed teaching hospital in Toronto with an active animal research program. The research institute owns about 220 sheep, primarily pregnant ewes. The animals were purchased from several suppliers in Ontario, but most came originally from western Canada; smaller numbers may have originated in the United States. The sheep were kept on a farm about 50 km northwest of Toronto and were transported when needed by truck to a holding area in the basement of the nurses' residence, which is situated across the street from the main hospital complex. They were then transported through a basement tunnel, in either open or closed carts, to the Gerrard wing of the hospital and taken up to the ninth floor in an elevator, which at other times is also used by hospital employees, patients and visitors. All animals were housed on the ninth floor, with as many as 20 sheep and several goats kept there at any one time, for an average stay of between 3 and 4 weeks.

Animal surgery was done on the eighth floor of the Gerrard wing (Fig. 1) and involved in-utero procedures resulting in either abortions or live births. The animals were taken to the eighth floor in an elevator that had rear doors opening to the surgical suites. Occasionally the sheep exited through the front doors and were led along the main corridor, thereby passing laboratories and offices not directly related to animal research. After surgery the animals were returned to the ninth floor and eventually to the farm.

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Air from the eighth and ninth floors is under positive pressure and is exhausted to the roof through charcoal filters. The incoming air duct is about 10 m from the exhaust duct. A xenon air-flow study done to investigate animal odours on the eighth floor had shown that air from the ninth floor leaked down to the eighth floor through the air ducts and elevator shafts.

Methods

Serologic testing was done on blood collected between July and October 1982. Samples were obtained from more than 90% of the sheep, animal attendants, technicians and researchers, as well as from other hospital staff, particularly those whose work might have taken them to the eighth or ninth floor of the Gerrard wing, and from other animals housed in the wing. Paired samples were not generally available for testing.

Serum was tested for complement-fixing antibody to phase II *C. burnetii* antigen by standard procedures[8] with the use of a commercially prepared antigen (Behring Institute). A titre of 1:8 or greater was considered positive.

A questionnaire was used to determine the extent and nature of the staff's contact with sheep or goats and whether any had had unexplained febrile illness of at least 48 hours' duration in the previous year. Such an illness was considered to be compatible with a diagnosis of Q fever.

Serum from 360 Toronto blood donors was also tested.

Chi-square tests with Yates's correction and *t*-tests were done for simple comparison of the data for the seropositive and seronegative groups.

Results

Of the 331 hospital staff who were tested, 59 (18%) were seropositive for *C. burnetii* antibody, with complement-fixing titres ranging from 1:8 to more than 1:2048. The highest rate of seropositivity (68%) was in those who were responsible for the total daily care of the animals in the research facility. However, as shown in Table I, antibodies were detected in other hospital employees, such as those working on the same floor as the animal operating rooms who had no direct contact with the animals. Many of these individuals were tested at their request or because their work brought them to the vicinity of the animal research unit.

In contrast to the findings in the hospital staff, of the 360 blood donors tested only 2 (0.6%) were seropositive for *C. burnetii* antibody.

Of the seropositive hospital staff 19 (32%) had high antibody titres (greater than 1:128), suggestive of recent infection.[9] Of these 19, 12 had a history of a recent febrile illness compatible with a diagnosis of Q fever, and seroconversion was documented in 2 (Table II). Two had been admitted to hospital; neither worked with animals, but the laboratories in which they worked were located on the eighth floor of the Gerrard wing, opposite the elevators used to transport the sheep. The clinical syndromes in the 12 usually consisted of a nonspecific, self-limited febrile illness. Hepatitis or atypical pneumonia had been diagnosed in four individuals by their own physicians.

The questionnaire was completed by 74% of those who were serologically tested — 52 (88%) of those found to be seropositive and 193 (71%) of those found to be seronegative for *C. burnetii* antibody. There was no significant difference in age or sex distribution, duration of hospital employment or contact with animals outside the hospital between the seropositive and seronegative groups (Table III). Seropositivity was, however, associated with visiting the animal facility at any time and with working with live animals or with sheep or fetal lamb tissue. The seropositive group also spent more time on the eighth or ninth floor of the Gerrard wing. Despite these associations, 37 (63%) of those who were seropositive had had no direct contact with sheep, although almost all recalled seeing sheep at some time in the hospital. Seropositive individuals were more likely to have had a recent febrile illness compatible with a diagnosis of Q fever than were those who were seronegative.

Of 37 pregnant ewes tested 34 (92%) were seropositive for *C. burnetii* antibody, having complement-fixing titres of 1:8 to 1:64. Of the four goats, four rhesus monkeys, four pigs, three dogs and one cat, rat, rabbit, chicken and mouse tested, only the goats and rhesus monkeys were also seropositive.

Discussion

In the past 2 years Q fever has again been recognized

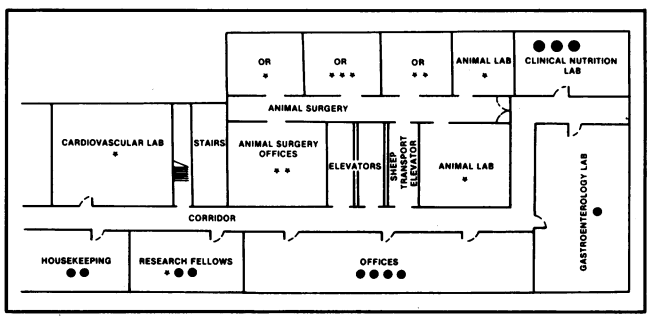

Fig. 1—Schematic plan of eighth floor of Gerrard wing, Hospital for Sick Children, Toronto. Symbols indicate location of staff members whose serum contained antibodies to phase II *Coxiella burnetii* antigen in a titre of 1:8 or greater: asterisks represent those working with sheep (n = 12); black dots represent those not working with sheep (n = 12). OR = operating room.

Table I—Distribution of staff of the Hospital for Sick Children, Toronto, who were seropositive for *Coxiella burnetii* antibodies

Staff	No. (and %) seropositive*/no. tested
Animal attendants	19/28 (68)
Surgical research staff	12/30 (40)
Others working on the eighth floor, Gerrard wing	12/38 (32)
Other	16/235 (7)
Total	59/331 (18)

*Having a titre of antibody to phase II *C. burnetii* antigen of 1:8 or greater by complement fixation.

as endemic in animal herds in Ontario. Increasing numbers of sporadic cases in humans are also being reported.[7] In the outbreak of Q fever in a Toronto research institute described in this article nearly one fifth of the 331 staff members tested were found to have complement-fixing antibody to phase II *C. burnetii* antigen. Individuals were often selected for serologic testing because they might have visited the animal facility; nevertheless, this rate of seropositivity is considerably higher than the 0.6% we found in 360 Toronto blood donors. As others have reported,[1,4,5] the prevalence of these antibodies was highest in those who had daily contact with animals, particularly sheep. However, there was also a considerable rate of seropositivity in those who visited the animal facility infrequently to perform experiments on other animals, and in those who worked in offices located along the eighth-floor corridor used to transport sheep. The eighth-floor hospital staff was presumably also exposed to aerosolized organisms carried in the air leaking from the ninth floor.

Q fever in humans is usually asymptomatic or mistaken for an influenza-like illness.[9] Occasionally a more severe illness, characterized by prolonged high fever, headache, and pneumonitis[10] or granulomatous hepatitis, occurs.[11] Physicians may fail to diagnose Q fever because of its nonspecific, protean manifestations or if there is no history of animal exposure. The 12 cases of Q fever at the Hospital for Sick Children were not recognized prior to the serologic survey, despite considerable morbidity, and were diagnosed retrospectively from the subjects' histories of a recent febrile illness and their antibody titres of 1:128 or greater. The "cryptic" nature of this outbreak is not without precedent.[4]

Rates of seropositivity for *C. burnetii* antibody at other Ontario research centres using sheep are of interest. At another University of Toronto research facility the rates were also high in 12 nongravid sheep (92%) and in 42 animal attendants (17%), but none of the 8 surgical researchers working with sheep were

Table III—Factors associated with Q fever among 244 staff members

Variable	No. (and %) of staff members or mean	
	Seropositive (n = 52)	Seronegative (n = 193)*
Male	25 (48)	78 (40)
Age (yr)		
Mean	35.1	35.1
Extremes	21; 60	16; 65
Duration of employment at the hospital (yr)	5.6	5.1
Ever went to eighth or ninth floor of Gerrard wing	46 (89)	117 (61)†
Daily time spent on those floors (h)	4.7	1.8†
Saw sheep in the hospital	48 (92)	116 (60)†
Worked with live animals	33 (64)	69 (36)†
Worked with sheep or fetal lamb tissue	23 (44)	17 (9)‡
Contact with sheep or goats outside of hospital	15 (29)	45 (23)
History of febrile illness in previous year	23 (44)	46 (24)§

*Some values or proportions in this group were significantly different from those in the seropositive group, at $p < \dagger 0.001$, ‡0.0001 and §0.01; for daily time spent on the two floors $t = 5.63$ with 243 degrees of freedom.

Table II—Clinical features of 12 staff members with antibody titres of 1:128 or greater and history of recent febrile illness compatible with a diagnosis of Q fever

Patient no.	Age (yr)/ sex	Aspects of illness			Highest RCFT*	Aspects of work	
		Month of onset	Duration (wk)	Nature		Done on floor where animals were kept	Involved animals
1	31/M	February	1	Fever, hepatitis	1 024	Yes	Yes
2	23/M	March	3	Fever, diarrhea, hepatitis	256	Yes	Yes
3†	32/M	March	2	Headache, diarrhea, myalgia	512	Yes	Yes
4‡	41/M	March	4	Fever, headache, cough, pneumonia	16 384	Yes	No
5	24/F	May	2	Fever, headache, cough	128	Yes	Yes
6	51/F	May	4/7	Fever, headache, cough, diarrhea	512	Yes	Yes
7	45/M	June	1	Fever, headache, cough, myalgia	256	Yes	Yes
8	33/F	June	1	Fever, headache, cough, myalgia	2 048	No	Yes
9	34/F	June	1	Fever, headache, cough, myalgia	256	Yes	Yes
10†	40/M	June	5/7	Fever, headache, cough, myalgia	512	No	Yes
11‡	47/M	June	5	Fever, headache, myalgia, phlebitis	1 024	Yes	No
12	28/M	July	4	Fever, headache, cough, hepatitis	512	Yes	No

*Reciprocal complement-fixing titre of antibodies to phase II *C. burnetii* antigen.
†Seroconversion documented.
‡Admitted to hospital.

seropositive (unpublished data). Sheep used at the University of Western Ontario, London, and at McMaster University, Hamilton, come from the same farm; however, the animals have lower rates of seropositivity, and no staff have been found to be seropositive (Dr. B. McLaughlin, laboratory services branch, Ontario Ministry of Health: personal communication, 1983). Previous studies have not shown a correlation between an individual animal's titre of antibodies to *C. burnetii* antigen and rickettsial shedding;[12] however, it may be that flocks (as opposed to individual animals) with low rates of seropositivity pose a smaller risk of disease transmission than do predominantly seropositive flocks.[13] More extensive studies correlating serologic and shedding data for animals and disease transmission to humans are required.

The causative agent of Q fever, *C. burnetii*, is hardy and highly infectious when in aerosol form after being shed from animal excreta or products of conception.[14] Gravid ewes shed particularly large numbers of organisms.[15] However, the following factors probably contributed to the spread of disease at the animal facility of the Hospital for Sick Children:

- The presence of infected gravid sheep.
- Unlimited access to the animal facility by hospital staff.
- Excessive transportation of sheep within the hospital.
- The lack of airtight segregation of sheep from other hospital areas.

The major factor in control of the outbreak was clearly the removal of the sheep from the hospital. Other control measures included the following:

- Disinfecting the animal facility, corridors and elevators.
- Reviewing the procedures for ventilation, transportation and waste disposal.
- Informing hospital personnel about the potential hazards of working with sheep and about the symptoms suggestive of Q fever.

These control measures are consistent with recently published recommendations for reducing the risk of Q fever in facilities where sheep are used for research.[16] No new cases of Q fever have occurred since the sheep were removed from the hospital.

Many institutions in North America are now doing research with sheep and are concerned about the potential for an outbreak of Q fever among research personnel. Our study confirms the risk of Q fever in animal research facilities, even to those with only indirect exposure to infected animals. Careful planning of such facilities is essential. The role of serologic monitoring, skin testing and vaccination requires further investigation, but the ideal prevention program probably includes skin testing and then vaccination for those whose results are negative.[17,18] The use of a vaccine in sheep and in susceptible research workers is presently being studied.

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