

A guide to Q fever and Q fever vaccination

Contents

Acknowledgments	3
Abbreviations	5
Foreword	6
Q fever: history of a major Australian discovery	8

Section 1

Q fever background

Impact of Q fever	12
Host range of Q fever	15
Mode of infection of human beings	18
Persons at risk	20

Section 2

Properties of *C burnetii* relating to pathogenesis and diagnosis

Properties of <i>C burnetii</i>	24
<i>C burnetii</i> : growth, survival, replication	26

Section 3

Q fever: Clinical presentation and course

Q fever infection: possible clinical outcomes	28
Acute Q fever	30
Chronic Q fever	36
Q fever endocarditis	40
Post Q fever fatigue syndrome (QFS)	44
Recrudescence granulomatous lesions	48

Section 4

Laboratory diagnosis of acute and chronic Q fever

Laboratory diagnosis of acute Q fever	50
Laboratory diagnosis of chronic Q fever	52

Section 5

Treatment

Treating acute and chronic Q fever	56
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Section 6

Q fever prevention

Q fever prevention	60
Pre-vaccination screening	62
Vaccination	69

Section 7

Vaccine adverse reactions

Common adverse reactions	74
Case studies: rare adverse events	76

Section 8

Frequently asked questions

FAQs	82
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Section 9a

Appendix 1

Properties of <i>C burnetii</i>	86
Infectivity and resistance	88
<i>C burnetii</i> : antigenic phase variation	89
<i>C burnetii</i> : growth, survival, replication	92

Section 9b

Appendix 2

General approaches to laboratory diagnosis	99
Techniques for detecting <i>C burnetii</i>	100
Serodiagnosis of Q fever	104
Laboratory diagnosis of acute Q fever	106

References

References	116
Q-VAX® Q Fever Vaccine & Q-VAX® Skin Test Product Information	122

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Acknowledgments

Contents

The contents of this booklet have been developed by Professor Barrie Marmion, AO.

Prof Marmion became involved in Q fever research with Dr Michael Stoker (Cambridge, UK) in an investigation (1949 to 1955) of Q fever in Britain. The Medical Research Council UK and the Public Health Laboratory Service sponsored the investigation as a sequel to the first isolation of *C burnetii* in the UK during an outbreak at a London hospital.

Prof Marmion's subsequent contributions with numerous co-workers include the definition of early and late stage Q fever endocarditis in the UK in the late 1950s and early 1960s, and trials of Q fever vaccine with Dr Richard Ormsbee (Rocky Mountain Laboratory, USA), CSL Ltd and many other collaborators in the 1980s, and in South Australia and Queensland in the early 1990s. The latter work also led to the definition in 1996 of the highly debilitating post Q fever fatigue syndrome via the Adelaide Q Fever Research Group and to continuing studies on the syndrome with Professor John Ayres' group in the UK who dealt with the 1989 Q fever outbreak in South Birmingham.

In 2003, Michael Stoker and Barrie Marmion were elected as honorary life members of the American Society for Rickettsiology.

CSL Biotherapies thanks Professor Marmion for his efforts and contribution, and in particular, for synthesising the extensive body of knowledge and research of this complex field in a manner relevant to the busy clinician.

Review

CSL Biotherapies thanks Associate Professor Denis W Spelman, Deputy Director, Infectious Diseases Unit, and Head, Microbiology, Alfred Hospital, Melbourne for reviewing the content and providing his valued feedback on the manuscript.

Disclosures: Professor Barrie Marmion and A/Prof Denis Spelman did not receive honoraria for this project and do not have financial interests in CSL Ltd.

Abbreviations

<i>C burnetii</i>	<i>Coxiella burnetii</i>
SCV	Small Cell Variant
LCV	Large Cell Variant
Ph 1	Phase 1
Ph 2	Phase 2
LPS	Lipopolysaccharide
CMI	Cell Mediated Immunity
ESR	Erythrocyte Sedimentation Rate
QFS	Q Fever Fatigue Syndrome
CFT	Complement Fixation Test
IFA	Immunofluorescence Assay
RIA	Radioimmunoassay
PCR	Polymerase Chain Reaction

Foreword



Q fever rarely attracts the public attention given to other infectious diseases, such as Legionella or meningococcal infections. Yet it is among the most costly and severe occupational zoonotic diseases in Australia, involving persons working directly with cattle, sheep and goats and others exposed indirectly to infection from domestic ruminants.

Acute primary Q fever may be highly incapacitating, but fortunately, is rarely fatal. Chronic sequelae,

such as Q fever endocarditis, granulomatous infection of bone, lung, or liver, or recrudescence of infection in the late stage of human pregnancy, illustrate the longer-term consequences of persistent infection with viable or replication-deficient *Coxiella burnetii*.

Since the last edition of this booklet, remarkable but preliminary advances have been made in understanding how this versatile pathogen *C burnetii* manipulates the intracellular environment of the host cell – the macrophage. Advances also have been made on the varying patterns of chronic persistence – as viable, recrudescence cells, or disabled, replication-defective cells, or sub-components that retain immunomodulatory antigens and lipopolysaccharide. Questions do remain and further research is required to provide answers for logical treatment of chronic sequelae.

A recent development is the cultivation of the Q fever organism in cell-free medium. While a remarkable scientific achievement, it is too soon to judge the implications of this for routine diagnosis and Q fever prevention.

Vaccine prophylaxis is the only feasible protection against airborne Q fever and its daunting acute and chronic outcomes. However, vaccine use may be complicated by cellular immune hypersensitivity to coxiella in subjects infected previously – a state probably maintained by persistence of organism or antigen. Vaccination of such persons may result in adverse reactions unless reactors are detected and excluded by skin and antibody tests before vaccination.

Pre-vaccination “screening” is generally very successful in avoiding undesirable adverse reactions. *However, it is emphasised that positive reactions in the screening test do not provide an absolute guarantee of predicting subsequent immunity on exposure. The primary purpose of pre-vaccination screening is to avoid adverse reactions to the vaccine.*

This and many other aspects of Q fever diagnosis and prevention are outlined in this updated booklet entitled “A guide to Q fever, and Q fever vaccination”. It is commended to those who have to deal with this complex and frequently underestimated disease.

Please note, the mini review in this booklet has the limited objective of assisting the physician in the recognition of acute and chronic Q fever, illuminating its laboratory diagnosis and explaining its vaccine prophylaxis. The booklet does not purport to be a review of the Q fever world literature. Unreferenced material can be attributed to unpublished experience of the Adelaide Q Fever Research Group (AQRG unpublished) and to common and established practice in the Institute of Medical and Veterinary Science (IMVS) Infectious Diseases Laboratories.

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Q fever: history of a major Australian discovery

Q fever is a severe, acute febrile illness of humans caused by *Coxiella burnetii*, a small facultative intracellular bacterium. The identification and isolation of *C. burnetii* was a joint achievement of Edward Derrick in Brisbane and Macfarlane Burnet in Melbourne.

In the mid 1930s, Derrick and colleagues first recognised the disease in abattoir workers and farmers in Queensland. Derrick documented the clinical disease and isolated the causative organism by inoculating blood and urine from Q fever patients into guinea pigs, which became febrile. The isolates were shown to be identical from various Q fever patients but unrelated to other fever-producing pathogens known at that time. Hesitant to give a definitive name, he called the disease 'Query Fever' – hence the name 'Q fever'.

Derrick sent infected guinea pig tissue samples to Macfarlane Burnet at the Walter and Eliza Hall Institute for Medical Research in Melbourne – who passaged the organism in mice. In smears and sections from liver and spleens of infected mice, Burnet found the cytoplasm of macrophages contained microcolonies of a minute bacterium, morphologically resembling rickettsiae.

Burnet found the cytoplasm of macrophages contained microcolonies of a minute bacterium, morphologically resembling rickettsiae

The plentiful growth of the organism in mouse spleen provided antigen suspensions, which facilitated development of simpler serological techniques for patient diagnosis and for serological surveys of native fauna and domestic ruminants.

It also became possible to compare strains of a similar organism isolated around the same time in the US at the Rocky Mountain Rickettsial Laboratory (from ticks from Nine Mile Creek, Montana USA). The Australian and American isolates were found to be essentially similar.

In the late 1930s, Herraald Cox at the Rocky Mountain Laboratory propagated the Q fever organism in chick embryo yolk sacs, which provided large numbers of bacteria for complement fixing antigen, agglutination tests, and for vaccine production.

Subsequently, the Q fever organism was found to differ from the classical rickettsia in cell structure, resistance properties, host cell tropisms, modes of replication, and antigenic relations. This led to its reclassification as a separate genus, *Coxiella burnetii* (in recognition of work by Cox and Burnet).

.....continued overleaf

Coxiella burnetii, is not related to the classical rickettsiae.....it is a separate genus

Q fever: history of a major Australian discovery

Recent studies (including the growth of *C burnetii* in cell-free media) have shown a close relationship to the Legionella group of organisms with which it shares many properties as a facultative intracellular pathogen.

At present, some 70 years on from Derrick's work, the coxiella genome has been sequenced. This and earlier genetic analyses confirm that *Coxiella burnetii* is not related to the classical rickettsiae such as typhus and spotted fever. Rather it is a separate genus and a near neighbour to Legionella group organisms.

Until recently, *C burnetii* was the only organism in the genus Coxiella, but recent work has discovered similar organisms in fresh water crayfish in Australia. While of phylogenetic interest, the new observations do not appear to be of importance for human health.

Section 1

Q fever background

- Disease impact
- Host range
- Transmission
- Persons at risk
- Mode of infection

Impact of Q fever

What is Q fever?

Q fever is a severe, acute febrile illness, which is a major problem in Australia and around the world. It is a zoonotic disease (i.e. spread from animals to humans) caused by the organism *Coxiella burnetii*.

Cattle, sheep and goats are the main reservoirs for infection in humans – although bandicoots, kangaroos and dogs also can be infected.¹⁻³ Several species of ticks are infected and parasitise bush animals.

Q fever occurs primarily in certain occupational groups – particularly workers from the livestock and meat industries. However, it is by no means restricted to these groups, as others in the general population may be infected (via airborne particulates or through fomites) through visits to high-risk areas or through proximity to infected animals or their contaminated products^{3,4} (see pages 15-21 for further information).

Prevalence of Q fever in Australia

Q fever is a notifiable disease in all Australian State and Territories – prevalence rates of notified cases of Q fever across Australia broadly reflect the intensity of local cattle and sheep husbandry and the associated processing industries. The highest per capita incidence typically occurs in northern New South Wales and southern Queensland (Table 1).

Table 1. Distribution of Q fever notifications in the year 2008⁵

	ACT	NSW	NT	QLD	SA	TAS	VIC	WA	Australia
Number of Q fever notifications	2	165	3	156	17	0	20	6	369

Q fever occurs primarily in certain occupational groups.... [but] is by no means restricted to these groups

Before the introduction of vaccination programmes, (first in the abattoirs in 1994 and later more widely in the rural community in 2001-2006) – there were around 500-800 Q fever cases annually across the country (Fig 1) and probably at least five times more unrecognised cases of clinical or subclinical infections.

(Q fever is prone to misdiagnosis as influenza or other respiratory infections due to the similarity of symptoms).

Q fever notifications have reduced substantially since the start of vaccination programs, with 369 notified cases in Australia in 2008 (Fig 1).

Q fever notifications have reduced substantially since the start of vaccination programs

Figure 1. Number of Q fever notifications in Australia 1991 to 2008⁶

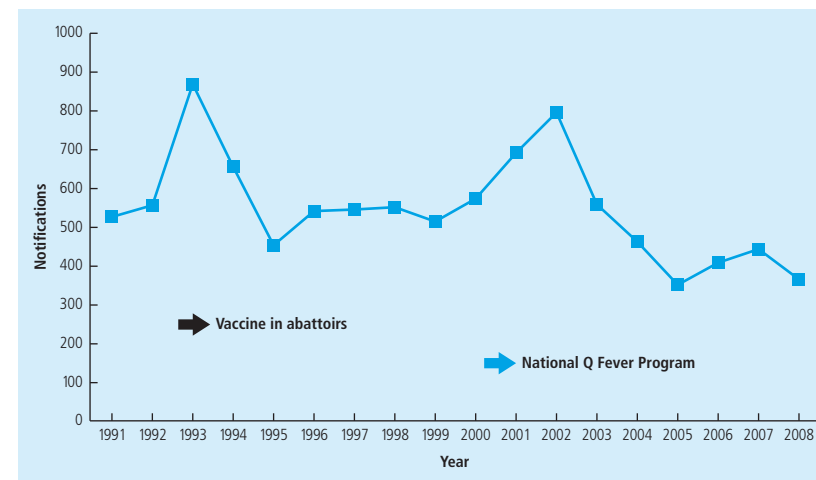


Figure based on notification data from Australian Department of Health and Ageing National Notifiable Diseases Surveillance System at http://www9.health.gov.au/cda/Source/Rpt_3.cfm

Impact of Q fever

Cost of the disease

Q fever-related costs to the community are substantial.

– A conservative estimate of the cost of Q fever WorkCover claims made to the Australian meat industry in 1993-1994 was around \$1 million annually with 1700 weeks of work lost each year.⁷

– Another study estimated medical costs alone as⁸

Around \$3,800 for each case of Q fever fatigue syndrome.

\$20,500 per patient for endocarditis with valve replacement surgery.

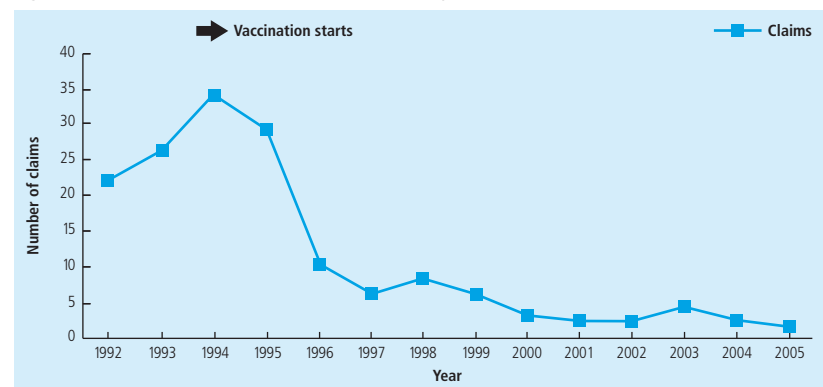
The figures do not include the added costs of compensation claims for these complications of Q fever.

Impact of vaccination

Vaccination programs have had a substantial impact on reducing the burden of Q fever⁹ and costs associated with the disease.¹⁰

Fig 2 illustrates the sharp decline in compensation claims for occupationally acquired Q fever in a large abattoir complex in Queensland after the implementation of a vaccination program in 1994.¹⁰ The reductions illustrated support vaccine efficacy data noted in formal clinical trials,^{11,12} and favourable predictions from cost-effectiveness analyses.⁸

Figure 2. Compensation claims for Q fever in a major Queensland abattoir complex¹⁰



Claims per annum for compensation for clinically and laboratory-proven, occupationally acquired Q fever. Reproduced from: Marmion, Barrie. Q fever: the long journey to control by vaccination. *MJA* 2007; 186 (4): 164-166. Copyright © 2007 The Medical Journal of Australia – reproduced with permission.

Host range of Q fever

Key points

- Worldwide, *C burnetii* infects many species of ticks, and wild and domesticated mammals, marsupials, macropods and birds.
- The epidemiology of human infection varies between countries due to differing reservoirs of infection as well as stock raising/processing practices.
- In Australia the principal sources of human infection are cattle and sheep.
- Feral goats also are a significant source. Kangaroo shooters and butchers may be infected.

Q fever: reservoirs of infection

- *C burnetii* has an exceptional host range and is found, for example, in ticks, native fauna, feral and domesticated mammals worldwide (with the possible exception of New Zealand)² – see Fig 3 overleaf.
- In Australia, coxiella is present in several species of ticks, bush animals such as bandicoots, rats, and in kangaroos and their ticks¹⁻³ – the latter represent a cycle of maintenance of the coxiella in the wild.
- In some species of tick, the transmission of infection occurs through the egg, and the cycle is complete. In other species of ticks, trans-ovarial transmission has not been shown and passage through an intermediate host (e.g. goats, domestic ruminants, etc) is necessary to maintain infection.^{2,3}
- Domestic ruminants – i.e. cattle, sheep and goats are the main reservoirs of human infection in Australia.
- Feral (i.e. introduced/escaped) mammals in the Australian bush such as goats, camels, horses, cats, dogs, foxes and rabbits are known from the world literature to be susceptible to Q fever infection² and hence, are potential reservoirs of infection.

Note: Some of the original research (by Derrick and others) relating to information in this section was published as far back as the late 1930s. For ease of referencing, information has been attributed predominantly to the following reviews, which contain details of the original references. (a) Stoker MP and Marmion BP. The spread of Q fever from animals to man: the natural history of a rickettsial disease. *Bull WHO*; 1955: 13: 781-806. (b) Lang GH. Coxiellosis (Q fever) in animals. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 23-48.

Host range of Q fever

- Apart from feral goats, which are well recognised as an important source of infection in Australian abattoir workers (and sometimes in persons visiting or living in the vicinity of abattoirs), our knowledge regarding infection in other feral animals and the significance for humans is limited.
- Q fever transmission to humans from tick bite or tick excreta has been recorded but is very rare.²
- Infected parturient mammals may shed particularly high numbers of *C burnetii* in the placenta, and in birth fluids,¹³⁻¹⁵ and may contaminate meconium and the surface of the neonate.
- The coxiella also may be shed in the milk of cattle, sheep or goats and unpasteurised milk and dairy products may be infective for humans.^{3,4}
- Additionally, *C burnetii* may be shed in urine during acute infection.^{3,15} For example, shedding in the urine of infected laboratory mice and guinea pigs may contaminate cage bedding – leading to aerosol infection of cage handlers and cross infection of other animals in the laboratory.

Domestic ruminants – i.e. cattle, sheep and goats.. are the main reservoirs of human infection in Australia

Importance of the host range of *C burnetii*

When investigating outbreaks of Q fever, it is important to consider all possible sources of infection, which can often be obscure (due the numerous potential reservoirs of the organism).

- For example, Q fever from exposure to infected parturient cats or dogs, and exposure to wild rabbits has been reported in Canada.¹⁶⁻¹⁸

Figure 3. Host range of Q fever

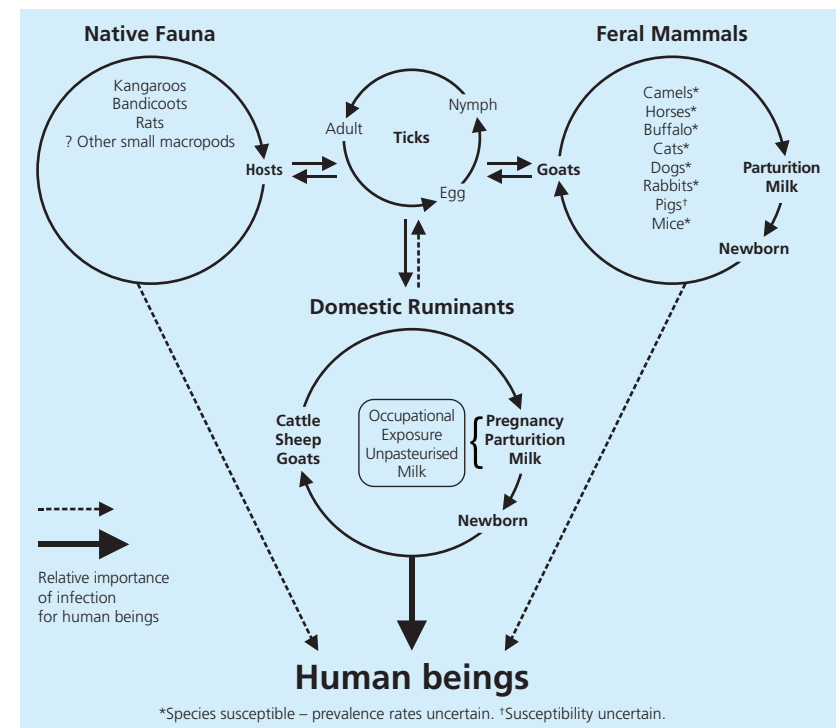


Figure courtesy of Prof Barrie Marmion – personal communication.

Mode of infection of human beings

Key points

- The predominant mode of infection of humans is via the respiratory tract, after inhalation of airborne dust or droplets containing the coxiella.
- Ingestion of infected, unpasteurised milk or dairy products causes infection (but not as consistently as infection by inhalation).
- Transcutaneous and intravenous infection is possible but rare.
- Sexual transmission has been reported but appears to be uncommon.

C burnetii transmission to human beings

- Human beings are infected with *C burnetii* by direct or indirect contact with infected animals, or products from these animals^{3,4} – see Fig 3 on previous page.
- Infected animals shed *C burnetii* in:^{3,15,19}
 - Urine
 - Milk
 - Faeces
 - Birth products (in particularly high numbers)
- The commonest route of infection is via the respiratory tract after inhalation of contaminated aerosols^{15,20} (fine droplets or dust) generated during parturition, or during the slaughtering process, and manipulation of the uterus, placenta and foetus from infected animals.
- Air samples in the vicinity of parturient animals are positive for *C burnetii* for up to two weeks after parturition.^{15,19}
- Soil and dust in animal holding areas may be contaminated with the organism from infected birthing animals, and the infected dust may be carried on work clothing, hair, straw, and other fomites, or on working dogs.^{3,19} *C burnetii* may then be liberated into the air in other environments at a distance – such as communal areas frequented by shepherds or meat processors or their homes. (This highlights the need to shower after work and launder working clothes).

Air samples in the vicinity of parturient animals are positive for *C burnetii*

- Human infection can occur via a number of routes and in many circumstances.
- After inhalation of the organism, the process of infection is thought to involve an intermediate step of multiplication in alveolar macrophages.²¹
- Consumption of unpasteurised infected milk or milk products (e.g. unripened cheese) may lead to infection⁴ but at a lower rate than through airborne exposure.
- Infection can occur through subcutaneous inoculation,¹⁵ but with a shorter incubation period than inhalation.
- Therefore, it is possible that infection might follow cuts with contaminated knives in the abattoir, or needle-stick injury when working with laboratory animals.

Human infection can occur by a number of routes

- Q fever has been transmitted by blood transfusion¹⁵ from donated blood collected in the late incubation period of primary infection.
- Transmission by tick bite involves possible contamination of the skin penetration area with infected tick excreta^{2,3} or perhaps inhalation of tick excreta during removal and disposal of the tick.
- Person to person infection during the course of acute primary Q fever via the respiratory tract is rare but has been reported.^{15,22}
- On the other hand, vertical transmission of the coxiella via the female genital tract clearly occurs perinatally, with infection of the foetus and possible carriage of *C burnetii* in the offspring.^{23,24}
- The male genital tract may be involved with infection of the testis and prostate and transmission of infection during intercourse.^{25,26} However, Q fever is not classified as a sexually transmitted disease.

Persons at risk

At-risk individuals

- Q fever is principally an occupational infection in Australia in certain groups, particularly workers in the livestock rearing and meat processing industries and in their dependent trades.
- However Q fever is by no means restricted to those working directly with cattle, sheep and goats on the farm, in the shearing team, or in the abattoir.
- Individuals in the community may be infected – e.g. by direct or indirect airborne infection from parturient animals, visits to stock saleyards, exposure to animal transporters, or residence near feed lots or abattoirs.
- Worldwide, there have been innumerable outbreaks in the general population and sources have not always been identified. Recent spectacular examples include:
 - (i) 147 cases in the general population in South Birmingham UK, arising from wind-borne infection from lambing sheep in pastures just south of the city limits.²⁷
 - (ii) 229 cases in visitors to an agricultural fair in Germany who visited a stall displaying newborn lambs from a single infected ewe.²⁸
 - (iii) Around 800 cases in a rural area of The Netherlands associated with intensive goat farming.²⁹ (The number of cases has reportedly increased to 1300).³⁰

Q fever is by no means restricted to those working directly with cattle, sheep and goats

- Outbreaks also have occurred in some unexpected or improbable settings, e.g.:
 - (i) Infection of a poker-playing group exposed to an infected cat giving birth to kittens in the same room.³¹
 - (ii) Infection of actors in a Nativity play involving a shepherd and his dog.³²
 - (iii) Outbreak in a school of Arts – possibly associated with unpacking a statue wrapped in straw.³³
 - (iv) Q fever outbreak in a cosmetics factory at which manufacture included powdered sheep placenta and foetal tissue.³⁴
- Table 2 provides examples of types of persons generally at risk of Q fever.

Table 2. Examples of persons at risk of Q fever

Abattoir workers
Farm workers
Veterinary personnel
Shepherds
Livestock transport workers
Shearers
Wool sorters
Pelt and hide tanners
Stockyard workers
Staff in veterinary microbiology labs
Maintenance engineers, electricians, plumbers, etc in risk environments
Dairy workers
Visitors to at-risk environments – e.g. research workers, teachers, school students, insurance agents, sales people, etc

Section 2

**Properties of *C burnetii*
relating to pathogenesis
and diagnosis**

Properties of *C burnetii*

Morphological variants of *C burnetii*

– *C burnetii* has two cell forms^{1,2}

LCV (large cell variant) – the intracellular replicating form of the organism.

SCV (small cell variant) – a highly resistant form for transmission to other cells and new hosts.

– LCV and SCV have different structures and antigens.

– Resistance to harsh conditions (see below) is likely a feature of the SCV.

(For detailed information and Figure on LCV and SCV – please see pages 86 and 87 of the Appendix 1)

Infectivity

C burnetii is a highly infective and efficient pathogen. Studies on the most virulent strains have estimated that one coxiella cell is sufficient to initiate infection of an animal.³

Resistance

C burnetii is able to resist physical and chemical stresses. The organism has the ability to survive in the dried state in harsh environmental conditions, e.g.³⁻⁵

– On wool at 15-20°C for 7-9 months (and almost twice as long at 4-6°C).

– On fresh meat in cold storage for > 1 month.

– On salt meat for 5 months.

– In dried cheese made from infected milk for 30-40 days.

– In skim milk at room temperature for > 40 months.

– In dry tick faeces for > 18 months.

C burnetii is able to survive harsh environmental conditions

C burnetii: antigenic phase variation

Along with morphological variation in the coxiella cell structure there also exists antigenic Phase variation in the cell wall antigens of the SCV.⁶

The sugar chains (O-chains) of lipopolysaccharide on the cell wall of the SCV exist in two forms:

– **Phase 1** – comprising a complete LPS chain,⁷ which is associated with virulence⁸ and immunomodulation of macrophage action.⁹⁻¹¹

– **Phase 2** – LPS with a truncated sugar chain.⁷ Phase 2 is avirulent in hosts with intact immune systems.⁸

Importance of phase variation

The phenomenon of phase variation has important clinical implications.

(a) Q fever vaccine.

Phase 1 LPS was identified as a key determinant of protective immunity. It is a major protective immunogen in Q fever vaccine and acts in concert with coxiella cell proteins that stimulate adaptive cellular immunity.^{9,12}

(b) Serodiagnosis of acute and chronic Q fever.

Differing patterns of antibody responses to phase 1 and 2 antigens develop during the acute and chronic Q fever.¹³ Antibody response patterns along with compatible clinical findings are used to diagnose different stages of Q fever disease.

Phase 1 is associated with virulence and immunomodulation of macrophage action

The information in this section is a short synopsis of the properties of *C burnetii* of diagnostic and/or pathogenetic significance. Please see Appendix 1 for detailed information on this subject.

***C burnetii*: growth, survival, replication**

Where does *C burnetii* grow?

- *C burnetii* is a facultative intracellular bacterium. Typically it grows/replicates inside its host cell. Recently, the successful growth of the organism in cell-free media has been reported.¹⁴
- ***In vitro***, in tissue culture systems, the organism will grow in many types of cells – e.g. yolk sac cells of chick embryo, and various mammalian cells in culture (but with varying difficulty in establishing infection).¹⁵
- ***In vivo***, in the whole animal, the principal host cell is the macrophage¹⁶ and other cells of the macrophage lineage (including monocytes, microglia and dendritic cells).

***C burnetii* pathogenesis**

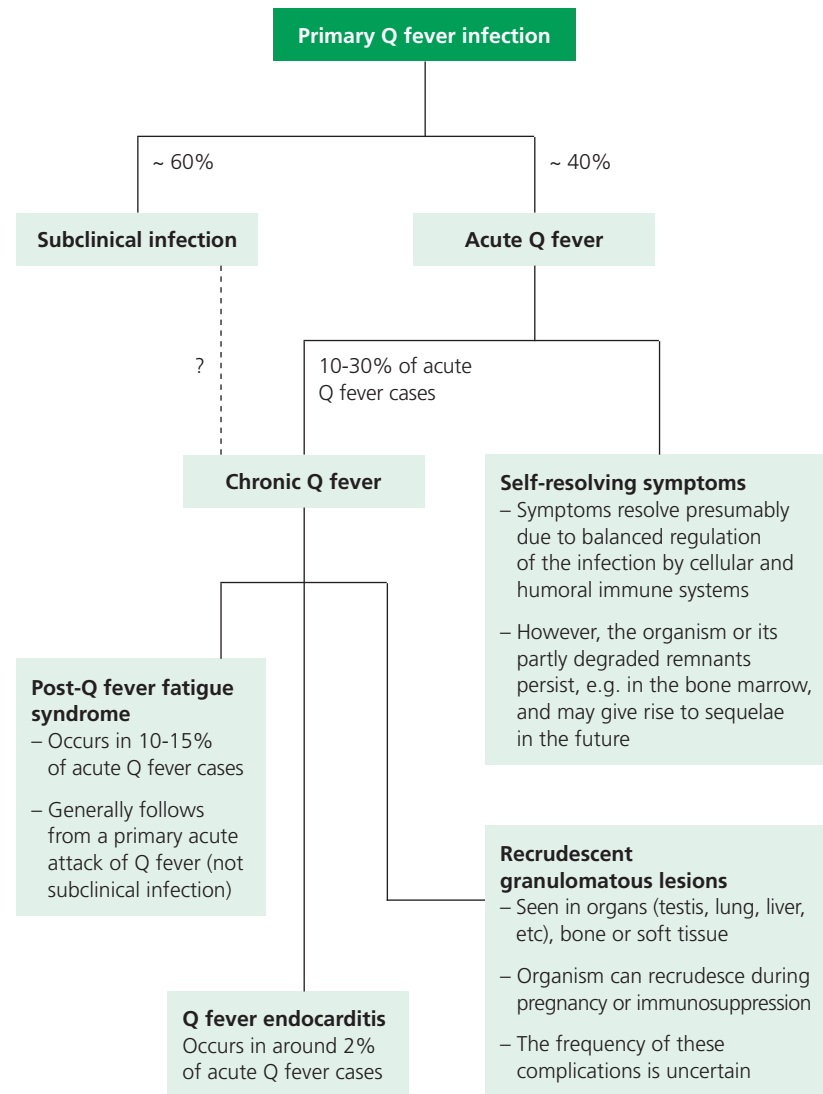
- After infection, *C burnetii* survives and multiplies in the macrophage phagosome.
- Its gene products modify interactions of the phagolysosome to utilise metabolic products from the macrophage autophagic pathway for replication.¹⁷
- The host's innate and adaptive immune responses appear to be inhibited during the long incubation period, but eventually effective humoral and CMI responses develop.
- Interferon-gamma plays a key role in the latter by restoring macrophage function to eliminate viable coxiellas.¹⁸
- Systemic and some other symptoms of acute Q fever are part of the CMI and acute phase cytokine responses.
- (See Appendix 1 for detailed information).

The principal host cell [for *C burnetii*] is the macrophage and cells of this lineage

Section 3

Q fever: Clinical presentation and course

Q fever infection: possible clinical outcomes



Subclinical infection

The commonest outcome after exposure to *C burnetii* appears to be subclinical infection (or a brief unremarkable illness), which only becomes apparent through future serological or skin testing.

This was illustrated in early clinical trials with Q fever vaccine in Australia, which found during the pre-screening process, that about half the abattoir population had antibody to *C burnetii* and another 4% had positive skin test but no detectable antibody¹ – indicating previous subclinical infection.

Acute Q fever

Although occurring less frequently than subclinical infection, Q fever may manifest as a severe acute febrile illness that commences abruptly and is extremely debilitating.

Key points – acute Q fever

- Primary acute Q fever is a severe acute febrile illness, which starts abruptly, with symptoms mimicking for example, those of influenza, *Mycoplasma pneumoniae* or *Legionella* spp infections.
- Hepatic involvement is common.
- Pneumonitis and neurological manifestations also may occur.
- Many of the severe systemic symptoms result from the action of potent immune mediators generated by the host's immune responses to infection.
- In the majority of cases, the acute illness resolves within 2-6 weeks.
- However, while afebrile, ~60% of patients may continue to experience low level post infection fatigue symptoms lasting 6-12 months, with eventual return to full health.

Acute Q fever

Symptoms and signs

Acute Q fever is an abrupt, severe, acute, typically self-limited febrile illness often described as an “influenza-like” illness.

- Indeed the systemic symptoms of Q fever (see Table 3 – i.e. abrupt onset, high fever, very severe headache, rigors, profuse sweats, muscle and joint pains, profound fatigue, photophobia, nausea and vomiting, etc are similar to those of influenza).
- However, the upper respiratory tract involvement and tracheobronchitis of influenza are not features of Q fever.
- None of the clinical features are diagnostically definitive. It is difficult to distinguish the systemic symptoms of Q fever infection from those of infection by *M pneumoniae* or *Legionella* spp.

Table 3. Characteristics at onset of acute primary Q fever^{2,4}

Fever, usually of abrupt onset and lasting for 5 to 50 or more days ²
Chills or rigors, lasting for 3 to 4 days ²
Profuse sweats ²
Very severe headache, which usually lasts while the patient is febrile ²
Myalgia and arthralgia ²
Profound fatigue and mental confusion or disorientation
Nausea and diarrhoea ²
Photophobia or blurred vision
Subclinical hepatitis, or rarely, frank jaundice
Weight loss during the acute phase of illness ⁴

The systemic symptoms of Q fever.....are similar to those of influenza

Aetiology of acute Q fever symptoms

The acute systemic symptoms and physical signs of Q fever and of other infections (e.g. *Legionella* spp, influenza, etc) are in part the result of the host's acute phase humoral, and particularly, cellular immune response to the infective agents.⁵

- The host's immune responses generate a wide range of immune mediators, such as cytokines, chemokines, and prostaglandins.^{6,7}
- These mediators have potent, and sometimes very severe effects on organ and metabolic functions.
- The presence of inflammatory markers such as C-reactive proteins and raised ESR⁴ (Erythrocyte Sedimentation Rate) also signal immune activation.

Clinical course of acute Q fever

- Acute Q fever develops after an incubation period of 15-25 days post-exposure depending on the dose^{8,9} (periods of up to 39 days have been reported).³
- In most cases the acute illness lasts 2-6 weeks.^{2,3}
- During this time there is often a striking loss of weight (6-12kg).^{4,9}
- Some patients may experience a biphasic febrile course² with the second episode responding to corticosteroid therapy rather than antibiotics.¹⁰
- Unusual complications in mid to late convalescence include peripheral thrombosis, bone marrow granuloma or necrosis, oesophagitis, inflammation of ovary or testis and prostate, pancreas, parotid, thyroid, etc.²

.....continued overleaf

In most cases the acute illness lasts 2-6 weeks...
There is often a striking loss of weight

Acute Q fever

- Despite rapid resolution of the acute illness, return to full health may be slow in many patients.
 - Some 64% have headache, joint and muscle pain, unusual fatigue, night sweats, mood changes and loss of libido during the next 6-12 months.¹¹
 - These individuals appear to have systemic symptoms of a post-infection fatigue syndrome, but with eventual return to full health.
 - Those that fail to recover after a year constitute the QFS (Q fever fatigue syndrome) group (see page 44).^{11,12}
- The majority (> 80%) of patients with acute primary Q fever are asymptomatic by 1 year from onset of illness.^{13,14}
- While acute Q fever is generally a self-limiting illness, death may occur in around 1% of untreated cases.¹⁵

Hepatitis

Hepatic involvement is a frequent presenting sign in Australian patients. Mostly there are mild-to-moderate biochemical abnormalities, including elevated alkaline phosphatase, amino transaminases and bilirubin levels.¹⁶ Some patients may present with mild hepatitis – others may be mildly jaundiced. Hepatosplenomegaly is common in Australian patients.^{3,16} Rarely, fatal liver necrosis occurs.¹⁶

Hepatic involvement is a frequent presenting sign in Australian patients

Pneumonitis

- In a small proportion of acute Q fever cases, small areas of pneumonitis occur after a few days of illness.³
- These sometimes develop into frank pneumonia with segmental consolidation on radiographs resembling bacterial pneumonia.¹³
- Pneumonitis is said to be less common in Australia than in Europe.³

Neurological manifestations²

- In addition to severe headaches, possible neurological manifestations include aseptic meningitis and/or encephalitis (0.2-1.3% of cases), photophobia, residual paraesthesia, and meningoencephalitis.
- Behavioural disturbances, cerebellar symptoms and signs, and cranial nerve neuritis also have been reported.

Other complications

- Pericarditis and myocarditis are each reported in around 1% of cases.¹⁷
- Primary infection in pregnant women (symptomatic or not), may be followed by abortion, premature delivery or low infant birth weight. The foetus may be infected during pregnancy. Additionally, Q fever may become chronic after delivery, resulting in recurrent miscarriages.¹⁷⁻¹⁹
- In women infected in the past, the organism may recrudesce during pregnancy.^{19,20}
- Patients with previous valvulopathy, pregnant women, or immunocompromised patients (e.g. with cancers, lymphomas, or HIV infection), are at increased risk of developing chronic Q fever.^{21,22}

Primary infection in pregnant women may be followed by abortion, premature delivery or low birth weight

Acute Q fever

A second episode of primary acute Q fever?

- After recovery from a primary Q fever attack, patients rarely, if ever, experience a second, *laboratory-validated* attack of *primary* acute Q fever.
- Nevertheless there are claims by abattoir workers who are frequently exposed, of a second or serial bouts of self-limited illness, symptomatically resembling Q fever, occurring a year or more after the initial infection.¹³
- Critical, in-depth laboratory study of such episodes is limited. However, when testing has been possible, the serological response patterns observed are those of a past infection or a secondary antibody response – not those of a primary infection.¹³

Comment

- The episodes may represent restimulation of heightened cell-mediated immune response upon re-exposure to airborne coxiella as antigen.¹⁰
- To increase our knowledge, ideally, future reports of these episodes should be investigated by examination of acute phase blood from these patients by PCR for *C burnetii* bacteremia, along with animal inoculation (among other investigations) – to confirm/exclude invasive infection.

After recovery from a primary Q fever attack, patients rarely, if ever, experience a second, laboratory-validated attack of primary acute Q fever

Acute Q fever: demographics²³

- In Australia, Q fever predominantly affects men, presumably reflecting occupational risks.
- However, the implementation of vaccination programs (first in abattoirs) appears to have shifted the demographics.
- A Department of Health and Ageing report on Q fever notifications and hospitalisations from 2003 to 2005 found that:²³
 - Rates were highest in adult males aged 15–39 and 40–64 years.
 - In the 1990s the rates were highest in males aged 15–39 years. Since then, highest rates have occurred in males aged 40–64 years.
 - The male to female ratio for both notifications and hospitalisations has generally been decreasing since the early 1990s, but remained relatively constant over 2003-2005 at 3.3 to 1 and 3.8 to 1, respectively.
- It is postulated that the wider use of Q fever vaccine in abattoirs may have led to the change in age distribution of male cases and an increasing proportion of women being infected – presumably indicating *C burnetii* exposure of at-risk groups outside the abattoirs.¹⁰

In Australia, Q fever predominantly affects men, presumably reflecting occupational risks.

Chronic Q fever

Key points

Chronic Q fever may take one of three forms

- (i) Q fever endocarditis: with anergic, poor cellular immune responses, cardiac valve vegetations containing viable coxiellas, and high levels of Q fever antibody.
- (ii) Continuing or recrudescent granulomatous infection: of bone, liver, testes or other organs. Recrudescent infection may also occur in the placenta and the foetus at the end of pregnancy.
- (iii) Post infection fatigue syndrome (QFS): with raised, impaired cellular immune responses, apparently no viable coxiellas, low or negligible antibody levels and clinical expression of a long lasting fatigue complex involving many body systems.

Predisposing factors for chronic Q fever

Certain conditions – e.g. pregnancy, immunosuppression, heart valve lesions and vascular abnormalities or prostheses, or specific immunogenetic backgrounds, predispose individuals to the development of chronic Q fever.^{17,18,22,24,25}

In these individuals, impaired/dysregulated immune responses to the organism or its antigens lead to the development of chronic sequelae. Additionally, long-term persistence of *C burnetii* or its antigens in the host and the host's response to these persisting antigens also may play a role.

Certain conditions – e.g. pregnancy, immunosuppression, heart valve lesions, etc.... predispose individuals to the development of chronic Q fever

Persistence of organism/antigens after acute Q fever

- *C. burnetii* has the ability to persist in the host after a primary infection.²⁶
- From infected humans (tissue samples from early stage endocarditis; products of conception at recrudescent infection at parturition; and from some granuloma in liver, bone or lung), viable coxiellas may be isolated and grown in eukaryotic cell culture, chick embryo yolk sac or laboratory animals.¹³
- In addition, in the absence of viable organisms, low-level persistence of coxiella genomic DNA and its antigenic cellular components has been seen in the bone marrow and/or peripheral blood of human beings up to 12 years after acute Q fever infection (even after symptom resolution and in the absence of long-term sequelae).^{26,27}
- A positive PCR detection of coxiella gene sequences does not necessarily equate to presence of viable organisms.
- At present we have little systematic insight into the regular site of persistence, if any, of viable coxiella that later recrudesce. The reproductive tract may be such a site and this requires further investigation.

Host responses

- The development of chronic Q fever is dependent in part on the host's immune response and the individual's ability to re-exert homeostasis over the immune response.
- Immunologic responses to coxiella infection span a wide spectrum influenced by predisposing conditions and/or the immunogenetic background of the individual as well as functions coded by the coxiella.

.....continued overleaf

Immunologic responses to coxiella infection [are in part] influenced by the immunogenetic background of the individual

Chronic Q fever

- At one end of the spectrum are raised CMI responses to *C burnetii* seen in Q fever fatigue syndrome, and at the opposite, is Q fever endocarditis with low level CMI response in the affected valves.²⁴
- Elimination of coxiellas as infective entities may vary in efficiency depending on the individual host's cellular immunogenetic repertoire.^{14,25}
- For example, monocytes from patients with Q fever endocarditis overproduce IL-10,²⁸ a mediator which is associated with impaired transendothelial migration of monocytes and lymphocytes, defective phagosome maturation, and reduced bactericidal activity of monocytes.²⁹⁻³¹

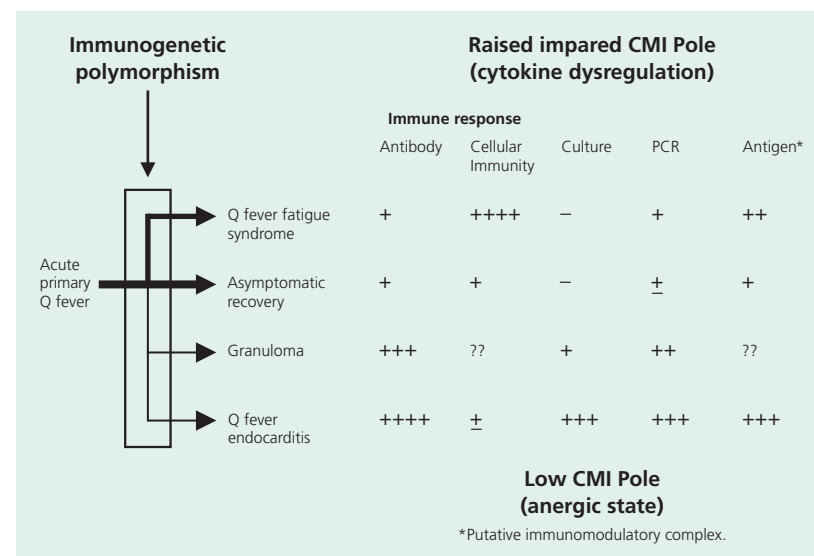
Consequence of persistence¹⁴

- In Q fever endocarditis and some granulomatous complications, a small number of living coxiellas survive. Infective activity continues at the organ level, and in granulomatous lesions, the organism may recrudesce at a later stage (e.g. during pregnancy).²⁴
- On the other hand, the persistence of coxiella as non-infective, immunomodulatory components produces no overt illness in most patients – due to re-exertion of immune homeostasis (i.e. balanced regulation of the activated CMI response to the persisting antigenic complexes).²⁴

Elimination of coxiellas as infective entities may vary in efficiency depending on the individual host's cellular immunogenetic repertoire

- In individuals that make a symptom-free recovery from the original acute infection, the only apparent evidence of antigenic stimulation from the persisting, coxiella cell components, is long-lasting or slowly-declining antibody levels, and heightened CMI response detected by positive intradermal skin tests during prevaccination screening.
- However, 10-15% of patients develop a long-lasting fatigue syndrome (QFS).¹⁴ In these individuals, the CMI is dysregulated and impaired.³²
- Finally, in Q fever endocarditis, persisting non-infective antigen complexes in the valve are associated with fibrosis and calcification and eventual mechanical dysfunction of valve in late stage endocarditis.¹³
- Fig 4 is a schematic representation of the postulated mechanisms in the generation of the various manifestations and sequelae after primary acute Q fever.

Figure 4. Polarity of immunological responses and clinical manifestations after primary acute Q fever¹⁰



??: CMI on *in vitro* cytokine release patterns not available. Tissue pathology patterns suggest intense CMI reaction. Figure supplied by Prof Barrie Marmion personal communication.

Q fever endocarditis

Endocarditis is the most serious manifestation of chronic Q fever. Observations of the Adelaide Q Fever Research Group suggest that in Australia, endocarditis follows about 2% of acute primary infections (although higher rates have been reported overseas).¹³

Background

- Typically, endocarditis develops or is detected within 1-2 years after the acute attack, but in some, it may not become apparent for 5-10 or more years.
- It is commonest in (but not restricted to) patients with pre-existing valvular heart disease due to underlying rheumatic fever or developmental abnormality.³³
- Prosthetic valves³⁴ and arterial grafts²² also may become infected.
- Such compromised persons should avoid working in at risk environments – and vaccination is highly advisable if exposure is unavoidable.
- The CMI response to *C burnetii* is impaired in Q fever endocarditis cases.³⁵
- Viable coxiellas may be cultured from blood, bone marrow or other tissues of patients with Q fever endocarditis.³⁶⁻³⁸

Clinical presentation

- Presenting symptoms typically are suggestive of cardiac involvement (heart failure or cardiac valve dysfunction).
- However, patients may also present with a less specific illness characterised by low grade fever (Table 4).²¹
- The diagnosis should be suspected when considering the possibility of subacute bacterial endocarditis but a series of blood cultures are negative.

The CMI response to *C burnetii* is impaired in Q fever endocarditis cases

Valve histopathology

- Valve vegetations in Q fever endocarditis are mostly small³⁸ compared with those due to other aetiologies of bacterial endocarditis and sometimes are in the stroma of the leaflet, or even on the mural endocardium.
- The vegetations may not be visible by conventional scanning techniques, e.g. ultrasound imaging, even via the trans-oesophageal route.¹³
- Histologically, Q fever endocarditis is characterized by significant fibrosis and calcifications, slight inflammation and vascularization, and small or absent vegetations.^{36,38}
- Large numbers of coxiella cells (live and dead) are present within macrophages and some are found extracellularly in areas of inflammation and necrosis.^{36,37}

Valve vegetations in Q fever endocarditis are mostly small...and may not be visible by conventional scanning techniques

Table 4. Clinical characteristics of Q fever endocarditis^{21,35}

Early presentation	Laboratory indices
Intermittent low grade fever	Raised ESR and CRP
Disabling malaise and fatigue	Increased IgM rheumatoid factor
Weight loss	Hyperglobulinaemia
Muscle aches and pains	Alterations of CD4/CD8 ratio
Chills and night sweats	Altered hepatic function tests
Changing patterns of cardiac murmurs	Increased creatinine indices
Signs of cardiac failure – dyspnoea; acute pulmonary oedema; angina; palpitations	Circulating immune complexes
Possible hepatosplenomegaly; purpuric rash; arterial embolism; microscopic haematuria	Anaemia
Clubbing of fingers	

Q fever endocarditis

Diagnosis

- Diagnosis most commonly involves measurement of high titres of antibody to *C burnetii* phase 1 and 2 antigens via immunofluorescence (IFA) or complement fixation (CF) assays – see ‘Laboratory diagnosis’ in Section 4 and Appendix 2.
- Very high levels of IgG and IgA antibodies to phase 1 and 2 antigens are found via IFA – with phase 1 IgG predominant. (IgG phase 1 titre $\geq 1:800$ is highly suggestive).^{35,39,40}
- Levels of IgM are low or negative^{39,40} (except in rare cases where the endocarditis has followed on from the acute primary attack).
- The above serological pattern, while “characteristic” of early stage Q fever endocarditis, *is not diagnostic on its own without compatible clinical findings*.
- In addition, the presence of *C burnetii* may be confirmed via isolation from peripheral blood samples and growth in cell culture or in laboratory animals, or by PCR assay of blood or bone marrow aspirates.¹³ (However, serology is most commonly used).

Phase 1 IgG is predominant in Q fever endocarditis.... with a titre of $\geq 1:800$ being highly suggestive

Late-stage Q fever endocarditis¹³

- During recent years, experience of the Adelaide Q Fever Research Group has also revealed a late stage ‘burnt out’ Q fever endocarditis in which valves removed for mechanical incompetence are distorted, fibrosed and calcified, but without obvious vegetations.
- Visually these are often assessed as due to “wear and tear” or “calcific aortic valve”.
- Features of the disease are shown in Table 5.

Table 5. Characteristics of late-stage ‘burnt-out’ Q fever endocarditis¹³

Histological examination (Giemsa) may reveal foci of particulate material – the residua of microcolonies once in macrophages
Examination of valve tissue shows: <ul style="list-style-type: none">– Substantial amounts of <i>C burnetii</i> DNA via PCR– Large amounts of coxiella antigen via immunofluorescence staining
Electron-microscopic examination may show degenerate LCV and SCV
Culture gives some positives, but samples are mostly negative in cell culture and AJ or SCID mice
Antibody titres are lower and depart from the “characteristic” serological profile of early stage endocarditis

Post Q fever fatigue syndrome (QFS)

In Australia, QFS is the most common chronic sequel to acute Q fever – affecting 10-15% of patients.¹¹

Clinical characteristics

- QFS usually follows on from an acute attack of Q fever¹¹ (but rarely, if ever, from subclinical infection).¹³
- The dominant symptom is an incapacitating fatigue out of all proportion to the degree of exertion that caused it (the exertion prior to illness would have been managed without difficulty).¹¹
- The debilitating fatigue may occur not during exertion, but later, even the next day, with the patient becoming bed-bound.¹¹
- *Importantly, the term ‘fatigue syndrome’ is misleading. QFS is characterised by a complex of symptoms that are not limited to fatigue* (see Table 6).
- For example: ethanol intolerance occurs in ~50% of QFS cases⁴¹ – patients often report ‘distressing hangovers’ after consumption of modest amounts of alcohol.¹⁰

The fatigue and disability of QFS last beyond a year and frequently more than 5 to 10 years

Table 6. Predominant symptoms of post Q fever fatigue syndrome (QFS)^{11,41}

Incapacitating fatigue	Pain or enlargement of lymph node groups
Nausea	Muscle tenderness on palpation
Constant headache	Arthralgia
Night sweats	Ethanol intolerance
Myalgia	Disturbed, unrefreshing sleep patterns
Intermittent fasciculation of muscle fibres	Irrational anger
	Loss of concentration and mental acuity

- A number of patients will experience some of the symptom complex listed in Table 6, i.e. fatigue and other features (without rigors or fever), for 6 months to a year after the initial attack of acute Q fever – followed by full recovery.^{11,13}
- However, QFS is an incapacitating, longer-lasting version of this symptom complex – the difference being that the fatigue and disability of QFS last beyond a year and frequently more than 5 to 10 years.^{11,12}

QFS: other features

- There is no overt organ involvement.
- Viable coxiellas are not isolated from QFS bone marrow or peripheral blood mononuclear cells (via animal inoculation or cell culture).²⁷
- Strong extracts of specimen are PCR positive for coxiella genomic DNA targets, but not when the specimen is diluted to an extent observed with a viable coxiella culture.¹⁴
- Coxiella antigens are present in samples examined in SCID mice.¹⁴
- Medium levels of antibody (IFA IgG Ph1: 10-80; Ph 2: 40-1280) usually are present.²⁷
- Cellular immune responses are heightened.³²
- Cytokine dysregulation appears to be a feature (high IL-6 levels, variable IFN-gamma, low levels of IL-10 and IL-2).³²

Cellular immune responses are heightened.....
cytokine dysregulation appears to be a feature

Post Q fever fatigue syndrome (QFS)

QFS diagnosis

PCR detection of DNA is not a stand-alone, definitive marker of QFS – (as low level coxiella DNA is detected by PCR in bone marrow of both QFS patients and those who have made an uncomplicated recovery).²⁷

The diagnosis may be considered given the presence of the stereotypic QFS symptoms outlined in Table 6 at a year or more following a laboratory-proven, clinically overt attack of acute Q fever and in the absence of any other infection or comorbidity that might be responsible for the fatigue syndrome.

(Fatigue syndromes may occur after infection with a number of other obligate or facultative intracellular bacterial infections and after some persistent virus infections, e.g. infectious mononucleosis or parvovirus B19).

Pathogenesis of QFS

At present there is no consensus on the pathogenetic process underlying QFS.

The following represents the current view of the Adelaide Q fever Research Group based on long-standing investigations.¹⁴

- Transient bacteremia generated during an acute primary Q fever infection is restricted by the host's adaptive cell-mediated and humoral immune response.
- A by product of the bacterial clearing process is persisting incompletely degraded (non-infective) coxiella cell components, which retain DNA sequences detectable by PCR and cell wall antigens (lipopolysaccharide).

- The residua persist in the host long after the initial infection, and it is hypothesised that these residua have immunomodulatory effects on cellular immune function and on macrophages and dendritic cells.
- In the majority (> 80%) of patients with acute Q fever there is symptom-free recovery with general homeostatic control of the immune stimulation caused by the persistent, coxiella cell debris.
- However, in a proportion (10-15%) of patients with immunogenetic susceptibility, the cell-mediated immune (CMI) response to the persisting antigen becomes dysregulated as there is a failure to exert homeostatic control.
- Cytokines and other immune mediators generated by the dysregulated CMI, give rise to QFS symptoms.³²

Cytokines and other immune mediators generated by the dysregulated CMI, give rise to QFS symptoms

Recrudescent granulomatous lesions

Background

- Recrudescent granulomatous lesions of liver, bone (around joints or in vertebrae), lung, testis and elsewhere in the genital tract – represent an additional chronic sequel to acute Q fever.⁴²⁻⁴⁵
- Recrudescence of the organism is known to occur during pregnancy, with infection in the placenta and of the foetus/newborn.
- Granulomatous complications do not always follow a clear, overt attack of acute primary Q fever. Presenting signs may include long-standing low-grade fever in some cases,⁴³ while others may present for the first time with symptoms of sudden-onset hepatitis, or pneumonia, osteomyelitis, or testicular swelling.^{13,42}

Features

- Granulomatous lesions may represent an intermediate stage between the anergy of endocarditis and the hypersensitive cellular immune responses of QFS (see Fig 4 on page 39).¹⁴
- Unlike endocarditis, there is marked lymphocytic infiltration of tissue, with collections of neutrophils, activated macrophages (multinucleate giant cells), and some cell necrosis.^{42,43}
- In granulomas there are variable numbers of coxiellas on microscopy – although PCR detection is positive.^{42,43}
- Information about attempts to culture coxiella from granulomas are scanty, but isolation has been successful from liver.³⁹
- The serological pattern for granuloma of the liver shows high levels of IgG antibodies to phase 1 and 2 antigens, some IgM antibody to the two phases (without substantial levels of IgA).³⁹ (NB: IgM rheumatoid factor must be removed from sera).

Recrudescence of the organism is known to occur during pregnancy, with infection in the placenta and of the foetus/newborn

Section 4

Laboratory diagnosis of acute and chronic Q fever

Laboratory diagnosis of acute Q fever

Acute Q fever: stages of illness

The illness falls into two main stages.

- (a) In the last few days of the incubation period (i.e. just before the onset of febrile illness) and in the first 8 to 12 days after onset, the Q fever organism may be present in the blood ('bacteraemia') and may also be found in the urine and in various tissues (see Appendix 2).
- (b) After day 10-12 from onset of illness, an antibody response develops, first in the IgM antibody class, and later in the IgG class – initially to the phase 2 antigen of the coxiella and later to the phase 1 antigen (Fig 6 in Appendix 2). IgM antibody levels peak in mid and late convalescence. After the febrile acute period, viable coxiella are rarely present.

Laboratory diagnosis: options

Based on the stages of illness, the options are:

- To detect the organism in the blood on day 1 to 10 and/or
- To show a change (increase in titre) in antibody response between a sample of serum taken up to day 10 (baseline, negative serum) and one or more samples taken on days 12-25 after onset of illness.

Practical issues

- Methods of direct detection of the coxiella are detailed in Appendix 2.
- In current practical terms, PCR detection of coxiella DNA sequences in the blood on days 1-10 is rapid² and is possible in some State reference laboratories.

After day 10-12 from the onset of illness, an antibody response develops

- Also in practical terms, the patient may not present to the doctor or the clinical diagnosis may not be considered until the end of the 'bacteraemic window' and the diagnosis then has to rest on demonstration of the four-fold or greater rise of antibody (between baseline and convalescent sera).
- If only a late or single convalescent stage serum sample is available, demonstration of raised antibody titre in the IgM antibody class provides an answer for clinical decision and treatment (see Appendix 2).
- The diagnosis is vitally dependent on establishing the date of ONSET of illness.
- Interpretation of serological or PCR results should be discussed with testing laboratory as described Appendix 2.
- Details of the relevant steps for the clinician to aid the laboratory diagnosis of acute Q fever are provided in Table 7.

Table 7. A clinician's guide to procedures for laboratory diagnosis of acute Q fever³

Specimens required	<ul style="list-style-type: none">– 5-10 mL of serum for antibody assays (clotted blood in gel separator 'vacutainer' tube).– 5-10 mL of unclotted blood (EDTA, not heparin or citrate) – for PCR and possible culture.
When to take specimens	<ul style="list-style-type: none">– Specimens should be collected as soon as the patient is seen (baseline specimen – may be negative), and collected again at 12-25 days after onset of illness.– If patient does not present (or Q fever is not suspected) until after day 10 – once again, collect specimen as soon as Q fever is suspected and another sample 5-7 days later.
Documentation	Should include best estimate of the date of onset of illness; data on chest X-ray; liver function tests; and inflammatory markers.
Notification	Laboratory-verified cases of Q fever are to be notified to the State Health Department.

Laboratory diagnosis of chronic Q fever

Chronic Q fever

Chronic sequelae to Q fever may take the form of:

- (a) Endocarditis on heart valves or the mural endocardium lining of the heart, or acute or chronic granuloma of the liver, lung, testes or bone among other sites. For clinical and pathological aspects – see previous section (Section 3).
- (b) A long-lasting post infection fatigue syndrome (QFS), which represents a different pathological response compared with the endocarditis-granuloma sequelae (see Fig 4 and Section 3).

The presence of organism in the blood may need to be supported by culture

Laboratory diagnosis: Q fever endocarditis or granuloma

- For endocarditis or granuloma, laboratory diagnosis depends on detecting a characteristic serological profile of high IgG and IgA antibody titres to phase 1 and phase 2 antigens (see Appendix II including Table 17).
- A compatible clinical picture also must be present (Section 3).
- *C burnetii* DNA may be detected by PCR assay in peripheral blood mononuclear cells or in biopsy specimens from granulomatous tissue.
- PCR assays also may be performed on fresh infected tissue, formalin fixed tissue and waxed embedded specimens (consult the laboratory).
- In diagnostically difficult cases the presence of organism in the blood may need to be supported by culture of the organism – consult with the reference laboratory.

Laboratory diagnosis – QFS

Diagnosis of QFS depends on a characteristic set of 8-10 symptoms (Table 6 in Section 3) following on a year or more after an overt acute attack of Q fever, and evidence of past and continuing antigenic stimulation from either viable coxiellas (rare) or persisting coxiella cell components (Appendix 2 and Table 17).

Table 8 provides a guide to optimising the laboratory diagnosis of chronic Q fever.

Table 8. A clinician's guide to procedures for laboratory diagnosis of chronic Q fever³

Specimens required	<ul style="list-style-type: none"> – 5-10 mL of serum for antibody assays (clotted blood in gel separator 'vacutainer' tube). – 5-10 mL of unclotted blood (EDTA, not heparin or citrate) – for animal inoculation/cell culture and PCR.
Other specimens	PCR assays also may be performed on biopsy specimens of fresh infected tissue, formalin fixed tissue, or waxed embedded specimens (consult the reference laboratory).
When to take specimens	Specimens should be collected as soon as the patient is seen (or as soon as chronic Q fever is suspected) and collected again about a fortnight later.
Documentation	<ul style="list-style-type: none"> – Look for documentary evidence of alleged acute Q fever episode and provide this to reference laboratory if required. – Current documentation should include best estimate of the date of onset of supposed chronic Q fever (endocarditis symptoms), or the date of presentation of granulomatous condition, or the length of time the patient has experienced QFS complex of symptoms. – Provide data on chest X-ray; and full blood workup including liver function tests and inflammatory markers; and results of ultrasound imaging (for endocarditis). – Note that visualisation of valve vegetations by ultrasound imaging may be difficult and transoesophageal imaging may be necessary.
Important considerations	<ul style="list-style-type: none"> – Clinicians should be aware that late-stage burnt-out Q fever endocarditis does not exhibit the characteristic serological pattern of early-stage Q fever endocarditis. – A diagnosis of chronic Q fever may not be made based on laboratory results alone. Diagnosis rests on the test results supported by compatible clinical findings (see Section 3).
Notification	Laboratory-verified cases of chronic Q fever are to be notified to the State Health Department.

Section 5

Treatment

Treating acute and chronic Q fever

Background

It is not the intention of this booklet to serve as an exhaustive reference on the treatment of Q fever. However, it is worth highlighting that the coxiella organism is sensitive to tetracycline spectrum of antibiotics and to ciprofloxacin, rifampicin, and to some extent, to cotrimoxazole (trimethoprim + sulfamethoxazole).^{1,2}

Most commonly, a tetracycline is used to treat acute Q fever.

Additionally, it is outside the scope of this booklet to cover in-depth, the treatment of Q fever in pregnant women or others in whom tetracycline is contraindicated or not tolerated. It is noted however, that long-term cotrimoxazole therapy may be used to treat Q fever during pregnancy.³

Treatment of acute Q fever^{4,5}

Adults	Children (> 8 years)	Other
The common treatment for acute Q Fever in an adult is Doxycycline orally 100mg 12-hourly for 14 days	For a child over 8 years of age the recommended regimen is Doxycycline 2.5mg/kg (up to 100 mg) orally 12-hourly for 14 days	For treatment of acute Q fever in younger children, or in pregnancy, specialist infectious diseases advice should be sought

Most commonly, a tetracycline is used to treat acute Q fever

Treatment of Q fever endocarditis⁶

- The treatment of Q fever endocarditis is more challenging – requiring a long course of combined therapy.
- Successful treatment has been achieved in some cases with tetracyclines or doxycycline, in combination with other antibiotics (e.g. cotrimoxazole, rifampicin, or a quinolone).
- However, the mortality and failure rates with the above regimens remains high – ranging from 54% for tetracycline or doxycycline alone to 33% for tetracycline associated with lincomycin/clindamycin, and 31% for cotrimoxazole associated with doxycycline.
- A study by Raoult and colleagues reported a shortened duration of therapy and reduction in the number of relapses with a combination of doxycycline and hydroxychloroquine sulfate (vs the established regimen of doxycycline and a quinolone).
Antibiotic treatment may need to be continued for several years.
For details, please refer to the original paper.

Antibiotic treatment [for Q fever endocarditis] may need to be continued for several years

Section 6

Q fever prevention

- Pre-vaccination screening
- Vaccination with Q-VAX[®]
Q Fever Vaccine

Q fever prevention

Prevention through vaccination

- The primary method of preventing Q fever is through vaccination.
- An effective vaccine against Q fever (Q-VAX® Q Fever Vaccine, CSL Biotherapies) has been available since 1989.^{1,2}
- In recognition of the importance of protecting individuals at risk of occupational exposure to *C burnetii*, large Q fever vaccination programs have been serially undertaken in Australia.³
 - 1991-1993: vaccination of workers in limited number of abattoirs
 - 1994-2000: increased vaccination coverage in large abattoirs in most States
 - 2001-2006 and subsequently, the Federally-funded National Q fever Management Program – extending vaccination to farmers, their families and employees in the livestock-rearing industry.
- Indeed a recent study has confirmed the effectiveness of the National Q fever vaccination program in substantially reducing the burden of Q fever (notifications and hospitalisations) in Australia.⁴

Who should be vaccinated?

- Q fever vaccine is recommended for those at risk of infection with *C burnetii* – **see Table 2 on page 21.**
- Ideally, vaccination should occur at least 2 to 3 weeks before the person starts working in an at-risk environment (as the risk of infection is highest in the first few years).
- Of particular note:
 - Women entering an at-risk occupation should be advised to seek Q fever vaccination before considering pregnancy – to avoid possible risks to the foetus (foetal infection/death).
 - Persons with cardiac abnormalities are at increased risk of Q fever endocarditis. If these persons cannot avoid working in high-risk environments, at least 3 weeks prior to commencing work, these subjects should undergo pre-vaccination screening and receive Q fever vaccine if indicated.

Who should not receive Q fever vaccine

Q fever vaccination is unsuitable in a number of instances. Those who should not be vaccinated are listed in Table 9.

Table 9. Persons who should not receive Q fever vaccine¹

Subjects with a history of Q fever
Anyone who has been vaccinated previously with Q fever vaccine
Persons with a history of likely exposure to the organism followed by an illness strongly suggestive of Q fever, or confirmed by laboratory tests. Every effort should be made to find serological evidence to verify the suggestion of past Q fever
Those with positive serology for Q fever antibody or a positive Q fever skin test
Anyone who is pregnant (note: the safety of Q fever vaccine in pregnancy has not been established – consequently, deferral of vaccination is recommended)
If there is known hypersensitivity to egg proteins or to any component of the vaccine

What happens if subjects with pre-existing immunity are vaccinated?

- Persons previously infected (either through acute illness or subclinical infection) exhibit 'sensitisation' to Q fever antigens – i.e. there is a heightened cell mediated immune response, which is perhaps maintained by low-level persistence of the organism or its antigen after infection.⁵
- Vaccinating these persons may, at times, lead to severe local inflammatory reactions and general systemic reactions.⁵ Such reactions are not inevitable – but they are unpredictable. When they do occur, they represent re-stimulation of the sensitised CMI response.
- Similarly, if a person who has received Q fever vaccine is revaccinated, the re-stimulation of vaccine-induced CMI may result in hypersensitivity reactions.⁵

Vaccinating persons [with pre-existing immunity] may lead to severe local inflammatory reactions and general systemic reactions

Pre-vaccination screening

General concepts

- The pre-vaccination screening test is used to exclude persons who are already sensitised to Q fever antigens and who may therefore experience serious hypersensitivity reactions if vaccinated.⁶
- Pre-vaccination screening involves first taking a detailed history to ensure the person being considered for vaccination has:
 - (i) a negative history of Q fever infection, and
 - (ii) has not been vaccinated with Q fever vaccine previously.
- Persons with a positive history of Q fever infection or those already vaccinated for Q fever do not require skin testing and serology as vaccination is contraindicated.
- Individuals with a negative history of previous Q fever infection or vaccination must undergo the following pre-vaccination tests:
 - (i) a skin test with intradermal injection of diluted Q-VAX® Skin Test and
 - (ii) a serological test for IgG antibody to *C burnetii*.
- The two assays measure different arms of the immune system.
 - The skin test detects the presence of acquired CMI and is the primary test in the pre-vaccination screen.
 - Serological testing detects antibody patterns indicative of past infection. It is an additional safeguard in case the skin test is performed incorrectly or gives an ambiguous reading.
- Detectable antibody may decline at a quicker rate than CMI and may be negative in a subject infected many years previously.⁷
- A substantially positive skin test is often (but not invariably) accompanied by antibody.

The skin test detects the presence of acquired CMI and is the primary test during pre-vaccination screening

Importance of detailed history

- A few subjects who have had laboratory-verified Q fever in the past, show no response to serological or skin testing.
- Despite the negative skin test and serology, these individuals may experience serious hypersensitivity reactions to Q fever vaccine.
- Therefore, *in all potential vaccinees, it is vital to obtain a detailed clinical history, and if possible, documentation of previous Q fever vaccination or laboratory results confirming Q fever disease.*
- Those who have worked in the livestock and meat industries for more than 10 years should be questioned particularly carefully.
- If there is any doubt about serological or skin test results, repeat the tests 2 to 3 weeks later (further explanation on indeterminate results provided on pages 67-68).
- Enquiries also should be made about aberrant reactions to other vaccines and history of hypersensitivity to drugs or environmental allergens, and autoimmune disease. In persons with such a history who are negative during prevaccination screening, careful consideration may be given to a trial dose of vaccine (see page 68) rather than proceeding directly with vaccination.⁸

Those who have worked in the livestock and meat industries for more than 10 years should be questioned particularly carefully

Pre-vaccination screening

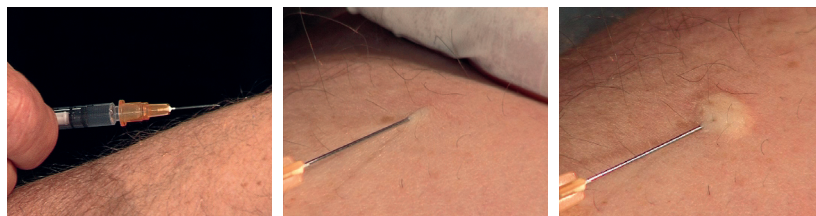
Skin test

- Only experienced personnel should perform and interpret skin tests.
- The correct process for skin testing as well as what can go wrong, are indicated in Table 10.
- In general, there is good correlation between a positive skin test and future immunity from infection – although this is not always the case.
- It is emphasised that **a positive skin test does not guarantee or prove that a person is immune to future exposure to *C burnetii*.**
- The main purpose of skin testing is to exclude individuals at risk of experiencing hypersensitivity reactions to vaccine.

Note: Q-VAX® Skin Test and Q-VAX® Q Fever Vaccine contain different amounts of purified, killed *C burnetii* – each has a specific purpose and different routes of administration. The two products **must not** be used interchangeably (i.e. the vaccine must never be used for skin testing or vice versa).

Only experienced personnel should perform and interpret skin tests

Prevaccination skin test: Intradermal injection technique



Place syringe at 10-15° angle to the skin. Bevel up.

Needle should be clearly visible through skin.

Inject slowly until formation of a bleb.

Table 10. Skin testing: correct and incorrect procedures^{1,8}

Correct procedure	<ul style="list-style-type: none"> – Skin testing is performed using Q-VAX® Skin Test. – Each 0.5 mL liquid vial is diluted in 14.5 mL of Sodium Chloride Injection (to a final volume of 15 mL). – The diluted Q-VAX® Skin Test should be freshly prepared, stored at 4°C and used within 6 hours. – If the injection site is not visibly clean, use methylated spirits to clean the site (do not use isopropyl alcohol). – A 0.1 mL dose of the diluted Q-VAX® Skin Test is injected intradermally into volar surface of the mid-forearm.* [See images on previous page]. – A positive reaction is any induration at the site of injection read 7 days after the test dose was given. – Any person with a positive reaction must not be vaccinated.
Errors	<ul style="list-style-type: none"> – Diluted skin test injected too deep – i.e. subcutaneously instead of intradermally. – Skin test performed in an inappropriate location (e.g. wrist). – Dilution errors in preparing the skin test. – Potency of diluted material reduced due to improper storage, or use after more than 6 hours post-dilution. – Test may be read too early – hence, an initial erythematous reaction at the injection site may be mistakenly read as a positive skin test. (It is important to wait 7 days after intradermal injection for a slow-developing induration). – Isopropyl alcohol used to clean injection site – (isopropyl alcohol may cause a skin thickening reaction, which may be misinterpreted as a positive result).

*Each 0.1 mL dose of diluted Q-VAX® Skin Test contains 16.7 ng of purified killed suspension of *C burnetii*.

Pre-vaccination screening

Pre-vaccination serology

- Serological testing during pre-vaccination screening measures IgG antibodies to *C burnetii* phase 2 antigen.
- A positive antibody test is indicated by:
 - CF antibody positive at 1 in 2.5 dilution
 - or
 - IFA positive at a dilution of 1 in 10 or more
 - or
 - Definitive positive absorbance value in EIA.
- IFA or EIA are the preferred tests – as CFT at 1 in 2.5 dilution is subject to false positive reactions from anti-complementary sera or low-level cross-reacting antigen from other bacteria (e.g. *Legionella* spp).⁹

Only persons with negative skin test and serology should be vaccinated

Pre-vaccination screening results: interpretation and action

- Table 11 depicts the recommended actions based on the results of pre-vaccination screening.
- Only persons with negative skin test and serology should be vaccinated.
- Note: a very small number of people who have had Q fever in the past may exhibit a negative response to serological and skin testing. Such persons may have severe reactions if vaccinated with Q-VAX® Q Fever Vaccine. For this reason, prior to vaccination, subjects should be carefully questioned regarding previous exposure to Q fever, the duration of such exposure as well as documented history of vaccination if available.
- Test results are indeterminate when skin test induration is negative or borderline (just palpable) *and* serology is equivocal in one or other serological tests (see following page for further information).

Pre-vaccination screening

Table 11. Interpretation and actions for serological and skin test results^{3,8}

Serology	Skin test	Interpretation/action
Positive antibody test	Positive Borderline Negative	Sensitised: do not vaccinate Sensitised: do not vaccinate Sensitised: do not vaccinate
Equivocal antibody test	Positive Borderline Negative	Sensitised: do not vaccinate Indeterminate: (see next page) Indeterminate: (see next page)
Negative antibody test	Positive Borderline Negative	Sensitised: do not vaccinate Indeterminate Non-immune: vaccinate

Notes:

- Positive skin test: induration present.
- Borderline skin test: induration just palpable (at limit of detectability).
- Negative skin test: no induration.
- Positive antibody test: CF or IFA positive according to laboratory's criteria or definite positive EIA absorbance value (according to manufacturer's instructions).
- Equivocal antibody test: CF or IFA results equivocal according to laboratory's criteria or equivocal EIA absorbance value (according to manufacturer's instructions).
- Negative antibody test: CF or IFA results negative according to laboratory's criteria or definite negative EIA absorbance value (according to manufacturer's instructions).

Indeterminate results^{3,8}

- Indeterminate results occur only in a small proportion of subjects.
- The finding may arise as a consequence of past infection with Q fever (where the markers of humoral and cell-mediated immunity have subsided), or may indicate the presence in the subject of immunity to antigens shared between *C burnetii* and other bacteria.

Pre-vaccination screening

Indeterminate results: possible courses of action

Typically, indeterminate findings have been dealt with as follows:^{3,8}

- (i) Repeat the skin test and interpret as per the guidelines for initial testing. Collect serum 2 to 3 weeks later to look for recall antibody response (i.e. a rapid rise in *C burnetii* IFA IgG antibodies to phase 1 and phase 2 antigens). A significant increase (defined as a 4-fold rise in titre of paired sera) indicates previous Q fever infection and vaccination is contraindicated.*

Or

- (ii) Inoculate the subject using **subcutaneous** injection of a 5 µg (0.1 mL) dose instead of the full 25 µg (0.5 mL) dose of the vaccine.* If there are no adverse effects 48 hours after the injection (i.e. severe local induration or severe systemic effects, perhaps accompanied by fever) – a further 0.4 mL (20 µg) dose of the vaccine is given within the next 2 to 3 weeks (i.e. before the development of CMI to the first dose).*

***Note:** The Q-VAX® Q Fever Vaccine PI does not list the above recommendations. For persons with indeterminate results, the Q-VAX® Q Fever Vaccine Approved Product Information states the following.

The risk-benefit decision of being vaccinated or not should be individually assessed and discussed with subject, in order to decide whether potential adverse events following vaccination outweigh the potential risk to that subject from Q fever infection and its associated complications.¹

Vaccination

Q-VAX® Q Fever Vaccine

- Q-VAX® Q Fever Vaccine contains a purified killed suspension of *C burnetii* (Henzerling strain) in the phase 1 antigenic state.¹
- The organisms are grown in the yolk sac of embryonated eggs, extracted, inactivated with formalin, and freed from excess egg proteins. Thiomersal 0.01% w/v is added as a preservative.¹
- Each 0.5 mL dose of vaccine contains 25 µg of formalin-inactivated *C burnetii*.¹
- The vaccination procedure is shown in Table 12.

Table 12. Vaccination procedure: Q-VAX® Q Fever Vaccine¹

Ensure the subject has a negative skin test and a negative antibody test result
Shake the Q-VAX® Vaccine pre-filled syringe gently before use
Inject 0.5 mL of solution subcutaneously . Do not administer vaccine intramuscularly
Revaccination must never be performed – due to the possibility of severe hypersensitivity reactions. The patient should be educated about the importance of this.

Immune response to vaccination

- Phase 1 whole-cell vaccines have been shown to be highly antigenic and protective against challenge.⁵
- In persons with negative pre-vaccination screening results – the serological response is chiefly IFA IgM antibody to *C burnetii* phase 1 antigen.^{7,10}

.....continued overleaf

Phase 1 whole-cell vaccines have been shown to be highly antigenic and protective against challenge

Vaccination

- While now established as an incorrect procedure, weakly seropositive individuals have been vaccinated in the past in clinical trials. In these individuals, the antibody response was mainly IgG to phase 1 and 2 antigens.^{7,10}
- The seroconversion rate is 80-82% between 3 and 19 months after vaccination dropping to 55-65% after 20 months.² However, 95% of vaccinees develop long-term cell-mediated immunity.^{2,11}
- (Note: lack of seroconversion after vaccination is not a reliable marker of lack of immunity).
- The duration of protective immunity following immunisation is unknown, but is believed to be in excess of 5 years.^{1,12}

Vaccine efficacy

- Strong protective efficacy of the vaccine has been demonstrated in open-label and placebo-controlled trials, and in post-licensing trials.^{1,2,12-14}
- A recent analysis of 7 trials estimated vaccine efficacy at 83-100%.¹⁵
- However, like all vaccines, 100% effectiveness cannot be guaranteed.
- Immunity to Q fever typically develops 15 days after vaccination.² During this time (and particularly during the first 10 days), exposure to the organism may lead to an overt infection.²
- Note: vaccine given during the incubation period of a natural attack of primary Q fever does not protect from development of disease.²

Immunity to Q fever typically develops
15 days after vaccination

- After extensive experience with the Q fever vaccine in clinical trials and through the National Q Fever Management Program, in recent years, very rare cases (< 1 person in 10,000 subjects vaccinated) of clinically diagnosed Q fever have been reported in vaccinated subjects.⁸ These occurred several months after vaccination (i.e. a timeframe within which immunity would have been expected to develop and not long enough after vaccination when immunity might theoretically have waned).
- Definitive investigation of these cases has been hindered by the unavailability of serial acute and convalescent phase sera and other specimens required for thorough analyses.
- However, about half of the presumptive failures appeared to be cases of acute primary Q fever⁸ based on the serological response pattern exhibited (rising titres of Q fever IgM antibodies, followed by IgG antibody – see Fig 6 in Appendix 2).
- Possible interpretations of the above results were:⁸
 - (i) idiosyncratic failure of the individuals to respond immunologically to the vaccine antigens;
 - (ii) the absence of watertight evidence that their “vaccination” was in fact with Q fever vaccine rather than with another vaccine – e.g. tetanus;
 - (iii) confusion on the part of the patient in identifying the pre-vaccination screening process as that of vaccination.
- There was some evidence suggesting the likelihood of (ii) above.
- In the remaining ‘vaccine failures’, IgG antibody was detected in early specimens with lower levels of IgM antibody.

.....continued overleaf

- Possible interpretations in remaining cases were:
 - (i) residual Q fever antibody from previous vaccination or sensitisation from subclinical infection coinciding with symptoms from another respiratory pathogen (e.g. *Legionella* spp, *Mycoplasma pneumoniae*).
 - (ii) a heightened restimulation of vaccine induced cellular immunity on exposure of the subject to living coxiella in the workplace – resulting in systemic Q-fever-like symptoms.

Investigating purported vaccine failures⁸

- The matter of vaccine failure is important and requires exploration of all possibilities.
- A suspect incident requires the following:
 - Adequate documentation of the vaccination.
 - Collection of serum and EDTA blood samples as soon after onset as possible as well as one taken at day 15-25 after onset of illness.
 - Determination of the serological response profile and inflammatory markers.
 - PCR and culture of the coxiella to detect invasion of the blood stream as found in primary Q fever.
 - In addition to the above, a wide screen is needed to exclude/confirm evidence of infection with other viral and bacterial respiratory pathogens.
- This complex investigation requires cooperation with a reference laboratory.
- In a vaccinated person, a clinical diagnosis of Q fever without in-depth laboratory evidence does not constitute vaccine failure.

In a vaccinated person, a clinical diagnosis of Q fever without in-depth laboratory evidence does not constitute vaccine failure

Section 7

Vaccine adverse reactions

- Common adverse reactions
- Case studies: rare adverse events

Common adverse reactions

Local and short-lived systemic reactions

The most common reactions after vaccination observed during clinical trials are minor local and systemic reactions, which are summarised in Table 13.¹ Reactions such as local tenderness and some erythema or oedema at the vaccination site shortly after inoculation, are similar to those observed with other bacterial vaccines.

Some people develop short-lived influenza-like symptoms.¹ The complex of systemic symptoms occurs in only a proportion of all potential vaccinees with negative pre-vaccination antibody and skin tests – and presumably represents a short-lived stimulation of the cellular immune system in immunologically unprimed individuals.

However, severe local or general systemic adverse reactions may develop in individuals already immune to Q fever if they receive Q-VAX®. Consequently, **diligent execution of the pre-vaccination screening protocol is fundamental to help minimise the risk of severe adverse reactions.**

Table 13. Common minor reactions to Q-VAX® Q Fever Vaccine¹

Reaction	Frequency of vaccine-induced reactions (%)
Local tenderness	48%
Local erythema	33%
Local oedema	0.6%
Fever (defined as > 38°C)	0.2%
Headache	9%
Other reactions (aching joints; swollen glands; flu-like symptoms; feeling faint; itching and induration at injection site)	15%
Later reactions (e.g. coincidental viral respiratory tract infections)	1.3%

Note: data relates to a clinical trial in South Australia in 464 vaccinated subjects.

Adverse reactions: post-marketing data

A range of adverse reactions has been reported during post-marketing use of the vaccine. These are shown in Table 14.²

Table 14. Post-marketing data: Q-VAX® Q Fever Vaccine adverse reactions²

Disorders of	Adverse reactions	Frequency of adverse reaction
Blood and lymphatic system	Lymphadenopathy	Very rare (<1/10,000)
Nervous system	Headache	Common (<1/10 and ≥ 1/100)
	Dizziness	Very rare (<1/10,000)
Gastrointestinal system	Nausea, vomiting and diarrhoea	Uncommon (<1/100 and ≥ 1/1000)
Skin and subcutaneous tissue	Delayed skin reaction (presenting up to 6 months after vaccination) at injection site (either vaccination and/or skin test site)	Common (<1/10 and ≥ 1/100)
	Hyperhidrosis	Uncommon (<1/100 and ≥ 1/1000)
Musculoskeletal system and connective tissue	Myalgia	Uncommon (<1/100 and ≥ 1/1000)
	Arthralgia	Very rare (<1/10,000)
General and administration site	Injection site inflammation (e.g. erythema, pain, warmth and swelling)	Very common (≥ 1/10)
	Injection site induration and/or oedema, pyrexia, malaise, fatigue	Uncommon (<1/100 and ≥ 1/1000)
	Injection site abscess formation, granuloma	Rare (<1/1000 and ≥ 1/10,000)
	Chills, chronic fatigue syndrome	Very rare (<1/10,000)

Rare adverse reactions – case studies

Other (rare adverse reactions)

In rare instances (i.e. less than 1 person in 1000 subjects vaccinated) severe local reactions such as abscess formation or delayed granulomatous induration (developing up to 6 months post-vaccination) have been reported.

Additionally, very rare cases (i.e. less than 1 person in 10,000 subjects vaccinated) of post-vaccination chronic fatigue syndrome have been reported. It is emphasised that these reactions are rare – but for interest, case studies are discussed in the following section.

1. Reactions in persons previously exposed to *C burnetii*³

Background

- A major obstacle to using the vaccine in the past was the failure to understand that prior immune sensitisation underlies the bulk of the more severe vaccine reactions.
- Reactions in those previously exposed to *C burnetii* have been greatly reduced by pre-vaccination screening for antibody and by intradermal skin testing.
- As mentioned previously, of the two, the important test is the skin test as an indicator of cellular immune hypersensitivity. The antibody test is performed as a safeguard for incorrect performance of the skin test.
- Importantly, antibody tests are not a substitute for skin tests – ~4% of abattoir workers infected in the distant past are antibody negative but skin test positive.¹

The important test is the skin test as an indicator of cellular immune hypersensitivity

Case study – Immune abscess

- A person who had worked in the meat processing industry for over 30 years underwent skin testing.
- The skin test was performed on skin in front of the wrist and was read as negative. (Note: the skin test was performed at an inappropriate site).
- The subject received Q fever vaccine.
- Within hours of vaccination, the vaccination site was acutely inflamed. Cultures testing for secondary bacterial infection were negative.
- The lesion became indurated and broke down, forming a sinus requiring surgical excision and drainage.
- The patient also suffered from general symptoms resembling QFS.
- The skin test site when read again a few days after vaccination, was positive.
- Exudate from the abscess site was tested for Q fever antibody and was strongly positive.

The skin test site when read again a few days after vaccination, was positive

Comment

- This appears to be an example of a person who was infected in the distant past whose skin test markers had declined to borderline levels and who had been tested on a thin area of skin instead of the forearm.
- The case highlights the need to enquire closely about any illness clinically resembling Q fever, particularly in those with a long history in high-risk environments.
- Importantly, as a safeguard, skin testing should always be accompanied by serology.
- Additionally, ambiguous skin tests should be repeated at a different site and be tested for recall antibody response (see page 68).

Rare adverse reactions – case studies

2. Reactions in persons with negative pre-vaccination skin tests³

Rare cases of covert/cryptic immunity due to past exposure may occur – i.e. prior exposure that is not detected during pre-vaccination screening. Some examples are given below.

Case studies – Protracted fatigue syndrome

- Two individuals who were negative during pre-vaccination screening, both developed pronounced fatigue symptoms (similar to QFS) at varying times after vaccination.
- One subject was connected to the meat processing industry while the other, a microbiologist, had worked in a veterinary laboratory.
- One case responded to extended low-dose corticosteroid therapy – and both recovered slowly over 2-3 years.

Comment

- It is presumed that these individuals were likely 'covertly immunised' prior to vaccination – through long-standing past exposure to the organism – with the markers of CMI and humoral immunity having declined to undetectable levels.
- In addition, the latter case may have been complicated by 'immune hypersensitivity/lability' as suggested by a past episode of cerebral malaria during time spent living in the Tropics.

Case study: Delayed granulomatous induration

- Finally, rare cases of delayed induration also have been reported – typified by the following case study.
- An individual living in a rural environment depicted negative results during pre-vaccination screening, and consequently, was vaccinated.

These individuals were likely 'covertly immunised' with the markers of....immunity having declined to undetectable levels

- There were no immediate adverse effects of note, but at a later time (between 1-8 months post-vaccination), the vaccination site became indurated and itchy, with desquamation of the skin – forming a mini-granuloma.
- Along with granuloma development at the vaccination site – the original skin test site also became positive.
- The induration gradually resolved.

Comment

- Once again, this may have been a case of covert/undetectable immunity in a subject previously exposed to *C burnetii*.
or
- Vaccination may have led to an initial suppression of CMI (in a manner similar to infection with live *C burnetii*) followed by delayed reaction as the likely result of 'burst through/re-emergence' of CMI – manifesting as an induration.

Overall pre-vaccination screening has essentially removed a major obstacle in the use of Q fever vaccine

Conclusions³

- Overall pre-vaccination screening has essentially removed a major obstacle in the use of Q fever vaccine, with resultant major public health benefits.
- However, with very large numbers of individuals vaccinated, a very small number react adversely even when negative in the pre-screen.
- It is important to enquire closely about past exposure to the organism, possible or laboratory-verified Q fever, adverse reactions to other vaccines, or autoimmune states, asthma, abnormal skin sensitivities, or any other pointers to unstable immune responses.

Section 8

Frequently asked questions

FAQs

1. How do you dilute the Q-VAX® Skin Test?

Each 0.5 mL of Q-VAX® skin test is diluted in 14.5 mL of Sodium Chloride Injection (to a final volume of 15 mL).¹

2. Can the diluted Q-VAX® Skin Test be used for more than one person within a 6-hour period?

Yes it may – provided storage and handling conditions are met (store the diluted Q-VAX® Skin Test at 4°C and use within 6 hours).¹

3. What are the recommendations when a patient has a borderline Q fever antibody test and a negative Q fever skin test?

This person's results are considered to be 'indeterminate'. See page 67 and 68 of Section 6 for possible courses of action.

4. Can the skin test be read after the 7 days specified in the PI?

The skin test must be read a minimum of 7 days after administration – although the skin test site may remain positive for 10 days or more. In the interest of avoiding confusion and maintaining consistency, it is useful to maintain a standardised timeframe of 7 days. However, if the skin test is not read at this time, it is worthwhile reading the test over the next few days, as a positive result will likely be maintained during this time.²

5. What is the time interval between administration of Q-VAX® Q Fever Vaccine, and protection?

Typically, immunity develops in approximately 15 days.³

It is useful to maintain a standardised timeframe of 7 days [for reading the skin test]

6. What is the minimum age for administering Q-VAX® Q Fever Vaccine to a patient?

No determination can be made about age. CSL Biotherapies has no data on the paediatric use of the vaccine.¹

7. Are doctors required to be registered/accredited to administer Q-VAX® Skin Test or Q-VAX® Q Fever Vaccine?

Doctors are not required to be registered/accredited. However, it is very important that anyone intending to administer Q fever vaccine has had appropriate training and is suitably experienced.

For information regarding training, contact the Department of Health in your State or Territory.

Section 9a

Appendix 1

Properties of *C burnetii*
relating to pathogenesis
and diagnosis

Properties of *C burnetii*

C burnetii has two cell forms: a large cell variant (LCV) and a small bacillary form called a small cell variant (SCV) – see Fig 5.

Morphological variants of *C burnetii*

Large cell variant (LCV)^{1,2}

- This has a thin cytoplasmic membrane and diffuse nuclear material.
- The LCV is the metabolically active, intracellular, replicating form of the coxiella with a structure that allows access of essential metabolites from the host cell.

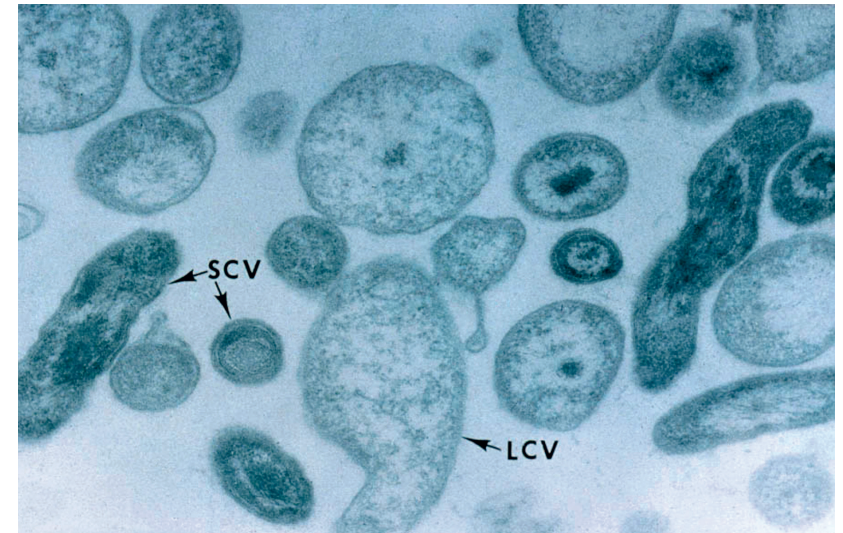
Small cell variant (SCV)^{1,2}

- SCV is rod shaped, has a thick cell wall with a highly cross-linked peptidoglycan layer and tightly condensed nuclear material.
- The SCV is metabolically inactive – it is considered to be the extracellular transit form with properties that ensure its survival in host tissues, substantial resistance to degradation in the external environment, and transmission to and entry into new hosts.

There are different antigens on the two morphological forms; LCV has a 29.5 KD protein whereas the SCV has other proteins and a lipopolysaccharide (LPS).¹

LCV is the metabolically active, intracellular, replicating form of the coxiella

Figure 5. Morphological variants of *C burnetii*



Electron micrograph of a preparation of cells in culture infected with *C burnetii* (supplied by Dr T McCaul to Prof B Marmion).

Spore formation?

- A morphological form with features resembling a spore is found in some LCV,² leading to a school of thought that the overall resistance of *C burnetii* to harsh conditions may be attributable to this 'spore'.
- However, there is little evidence to support the hypothesis as the spore has not been:³
 - Obtained in pure suspension free from LCV and SCV.
 - Shown to contain the complete genome of *C burnetii*.
 - Tested as a separate entity for infectivity, or thermal and environmental resistance.
- An alternative explanation is that this 'spore-like' form represents a stage of the assembly of a SCV.³
- It seems more likely that resistance to harsh conditions (see following pages) is a feature of the SCV.

Infectivity and resistance

Infectivity

C burnetii is a highly infective and efficient pathogen – with an extremely long biological half-life, which probably contributes to its infectivity.⁴ The infective dose probably varies between strains but it has been estimated through studies on the most virulent strains that one coxiella cell is sufficient to initiate infection of an animal.⁴

Resistance

C burnetii is able to resist a variety of physical and chemical stresses – resistance to elevated temperatures, dessication, osmotic shock, UV light, and chemical disinfectants, has been demonstrated.¹

As reported in Section 2, the organism has the ability to survive in the dried state in harsh environmental conditions, e.g.⁴⁻⁶

- On wool at 15-20°C for 7-10 months (and almost twice as long at 4-6°C).
- On fresh meat in cold storage for > 1 month.
- On salt meat for 5 months.
- In dried cheese made from infected milk for 30-40 days.
- In skim milk at room temperature for > 40 months.
- In dry tick faeces for > 18 months.

C burnetii is a highly infective and efficient pathogen

C burnetii: antigenic phase variation

Phase variation

Along with morphological variation in the coxiella cell structure there also exists so-called antigenic Phase variation in the cell wall antigens of the SCV.⁷ Phase 1 and 2 antigenic states differ in lipopolysaccharide characteristics.

Phase 1

- This is the virulent state in which the organism exists in human and animal infections.⁸
- In Phase 1, the lipopolysaccharide (LPS) of *C burnetii* is complex and has its full complement of sugar units and side chains (O-chains).
Similar to Phase 2 organisms, Phase 1 LPS contains KDO, D-mannose, D-glycero-D-manno-heptose, lipid A or a lipid A analog, and a complex mixture of fatty acids, including branched chain fatty acids.^{9,10}
However, unlike Phase 2 organisms, Phase 1 LPS also contains virenose, dihydrohydroxystreptose, and galactosaminuronyl-(1,6)-glucosamine.⁹⁻¹¹
- This complete LPS is of critical importance for virulence⁸ and macrophage modulation,¹²⁻¹⁴ and as a protective antigen in the vaccine.¹⁵

The complete LPS [of Phase 1 organisms] is of critical importance for virulence and macrophage modulation

***C burnetii*: antigenic phase variation**

Phase 2

- When an isolate in Phase 1 antigenic state is passaged in the yolk sac of chick embryo or cell culture,⁷ deletion mutants with truncated LPS side chains (O-chains)¹⁰ and without two unusual 3-carbon sugars, emerge and overgrow the Phase 1 coxiella cells.⁷
- This mixed population is designated overall as Phase 2.
- Pure lines of Phase 2 cells have been derived and shown to be avirulent for laboratory animals (except at very high doses).⁸
- Examination of a pure Phase 2 line of *C burnetii* shows gene deletion in the genomic sequences for carbohydrate synthesis.¹⁰
- Other explanations involving failure of gene expression also have been advocated.

Clinical importance of phase variation

From a clinical perspective, phase variation of *C burnetii* antigens holds particular significance for Q fever vaccination and serological diagnosis of disease.

Q fever vaccination¹⁵

- The Phase 1 antigen has been identified as a key determinant of protective immunity.
- This has helped to develop the modern version of Q fever vaccine containing a formalin-inactivated, highly purified suspension of Phase 1 *C burnetii*.

Phase 1 antigen has been identified as a key determinant of protective immunity

Serological diagnosis of clinical disease

- When *C burnetii* infects human beings, antibodies are produced against both antigenic phases of the organism.¹⁶
- In most cases, these antibodies appear (and increase in titre) in a characteristic order.¹⁶
- For example:
 - During the course of acute infection, antibody responses are initially directed mainly at Phase 2 antigen.¹⁷
 - Rising levels of IgM, followed by IgG and IgA antibodies to Phase 2 are detected by immunofluorescence at various times after the onset of illness – followed later by IgG antibody to Phase 1 antigen¹⁶ (see Appendix 2 and Fig 6 in the Appendix).
- Hence, serological tests can provide an indication of the stage of acute infection.
- Furthermore the pattern of antibody response (i.e. immunoglobulin class of antibody to particular antigenic phase) differs substantially between acute and chronic disease.
 - E.g. some antibodies, namely IgA antibody against Phase 1 antigen do not appear (or do so at very low levels) in acute Q fever – whereas they present at high titre in the early stages of Q fever endocarditis.¹⁷

When *C burnetii* infects human beings, antibodies are produced against both antigenic phases of the organism

***C burnetii*: growth, survival, replication**

Where does *C burnetii* grow?

C burnetii is a facultative intracellular bacterium. It grows/replicates inside its host cell (recently, growth in cell-free media has been reported).¹⁸

In vitro

In tissue culture systems, the organism will grow in many types of cells – e.g. yolk sac cells of chick embryo, and various mammalian cells in culture¹⁹ (but with varying difficulty in establishing infection).

In vivo

- *C burnetii* may be propagated in mice and guinea pigs and in embryonated eggs.¹⁹
- In the whole animal, the principal host cell is the macrophage²⁰ and other cells belonging to the macrophage lineage (including monocytes, microglia and dendritic cells).
- *C burnetii* is uniquely adapted to completing its lifecycle in the macrophage phagosome. Its genome codes for enzymes that operate optimally at the low pH of the phagosome.²¹
- Studies using *in vitro* and other model systems have begun to illuminate the complex mechanisms assisting the survival and replication of *C burnetii* in the host (see following pages).

C burnetii is uniquely adapted to completing its lifecycle in the macrophage phagosome

Survival – background

- *C burnetii* is a highly evolved and successful intracellular pathogen with enzymes uniquely adapted to operation within the acidic vacuole of the macrophage phagosome.²¹
- Survival and replication of *C burnetii* in the host is greatly dependent on the ability of virulent strains of *C burnetii* to subvert and avoid the microbicidal functions of macrophages.
- Current research using *in vitro* culture of macrophage cell lines and *ex vivo* growth of macrophages from patients has revealed the fascinating complexity of mechanisms involved. The extent to which the mechanisms are maintained/modified in an immunocompetent host remain to be elucidated.

Profound modifications of macrophage functions are coded by the coxiella

Survival in the acidic phagolysosome

- Macrophages (and specialised cells of this lineage) are the primary host cells for *C burnetii*.²⁰
- The coxiella completes its whole life cycle in the phagolysosome. Recent *in vitro* studies have revealed profound modifications of macrophage functions coded by the coxiella.
- The organism's enzyme systems operate optimally at the pH of the phagolysosome.²¹
- Additionally, the coxiella genome codes for the Type IV secretion system,^{22,23} which delivers effector proteins to the host cytosol.
- The effector proteins help to mediate functions including phagosome stalling and autophagic interactions, which nurture the environment favouring replication of the coxiella.²²
- By actively modulating the autophagic pathway and delaying lysosomal fusion, *C burnetii* likely creates an environment which supports intracellular differentiation and survival of the bacteria.²²

***C burnetii*: growth, survival, replication**

Modification of macrophage function

Mege and colleagues (Rickettsial Unit, Marseilles) have used *in vitro* and *ex vivo* model systems to identify numerous other mechanisms employed by *C burnetii* to downregulate early endogenous immune responses during LCV synthesis and replication.

The following additional facets of macrophage function modification by *C burnetii* have been identified.

- Phase 1 bacteria are internalised slowly/poorly by monocytes.²⁴
- Phase 1 LPS induces actin reorganisation, engagement of $\alpha\gamma\beta 3$ integrin, impairment of cross-talk between $\alpha\gamma\beta 3$ integrin and CR3 (complement receptor 3), and interferes with the spatial distribution of CR3 – resulting in reduced phagocytic activity of monocytes.^{13,25}
- Once internalised, *C burnetii* exist in the acidic vacuole at pH 4.5 – multiplying in small vacuoles, which fuse to form a phagolysosome.²⁴
- Phase 1 *C burnetii* escape intracellular killing in monocytes by inhibiting cathepsin fusion, which inhibits phagosome maturation.^{24,26}
- Additionally, the cytokine IL-10 plays a role by downregulating TNF- α production, thus enabling *C burnetii* survival and replication in monocytes. (Interestingly, monocytes of Q fever endocarditis patients overproduce IL-10).²⁷⁻²⁹
- Hence, the initial subversion of macrophage activation also facilitates the organism's survival and replication in host cells.

Numerous mechanisms [are] employed by *C burnetii* to downregulate early endogenous immune responses

Onset of clinical symptoms

- The incubation period (time from exposure to onset of symptoms) of Q fever may be up to 3 weeks (15-25 days).^{30,31} During this period the coxiella is able to multiply, initially in alveolar macrophages, and later in other parts of the body.
- Over time, the host's adaptive immune response emerges.
- Late in the incubation period large numbers of organisms are released into the peripheral blood, commencing a transient bacteremia²⁰ (from 2 days prior up to 12 days post symptom onset).
- The bacteremia facilitates spread of infection to other cells/organs in the body.²⁰

Immune response

- The onset of symptoms signals the emergence of the host's adaptive immune response – which includes antibody and cell-mediated immune (CMI) responses.
- The humoral and CMI responses work together to restrict and terminate the bacteremia (presently aided by antibiotic therapy).
- Antibodies are first detected in peripheral blood around day 7-10 after onset of illness.¹⁶
- Rising titres of antibodies to Phase 1 and Phase 2 antigens may be used to map the stage of acute illness (see Appendix 2 – Laboratory diagnosis).
- Antibodies facilitate the uptake of infectious microorganisms by macrophages.³²
- However, it is the cellular immune response that plays a critical role in controlling infection.^{20,32}

Around the time of symptom onset [begins a] transient bacteremia

***C burnetii*: growth, survival, replication**

Action of the immune response

- Presentation of *C burnetii* antigen-Ia complexes on the surface of phagocytic cells leads to T cell activation and the production of cytokines such as IL-1, IL-2, TNF- α and IFN-gamma.³²
- The soluble mediators promote T cell activation and proliferation and stimulate B cells to differentiate and secrete antibody.³²
- IFN-gamma is central to controlling infection – it restores the microbicidal properties of macrophages.²⁴
- IFN-gamma acts by:²⁴
 - Restoring cathepsin fusion, thus activating phagosome maturation and enabling intracellular killing of *C burnetii*.²⁶
 - Inducing vacuole alkalization – which prevents bacterial replication.
 - Killing *C burnetii* in infected macrophages and inducing apoptosis of *C burnetii*-infected macrophages, mediated in part by TNF.³³
- The plethora of immune mediators released as part of the acute phase CMI response give rise to the severe systemic (and other) symptoms of Q fever.

In a recent report, using mouse model systems (including IFN-gamma and TNF-alpha knockout mice) to study the immune response to *C burnetii*, Samuel and colleagues reported that T cells are critical for clearance of *C burnetii*, that IFN-gamma and TNF-alpha are essential for the early control of infection, and that B cells are important for the prevention of tissue damage.³⁴

IFN-gamma... restores the microbicidal properties of macrophages

Immune homeostasis

- Eventually, in the majority (>80%) of cases, immune homeostasis is reached. The acute symptoms subside and these patients recover slowly.
- However, antigenic cell components of the coxiella are not completely eradicated and may persist in varying forms in the bone marrow and other tissues of infected individuals.³⁵
- The persistence is evidenced by antibody levels that decline slowly (over months to years).³⁵
- The cellular immune response remains sensitised to *C burnetii* antigens – as evidenced by positive intradermal skin tests detected during prevaccination screening of persons infected in the past.
- Some individuals with predisposing conditions/immunogenetic background may develop chronic Q fever.^{36,37}

In the majority (> 80%) of cases, immune homeostasis is reached... [but] the CMI response remains sensitised

Section 9b

Appendix 2 Laboratory diagnosis of Q fever

General approaches to laboratory diagnosis

Acute Q fever

During the early, invasive stage of disease (and before antibody appears), laboratory confirmation of acute primary Q fever may be obtained by:

- Direct demonstration of the coxiella in blood, urine or tissue samples (Table 15 – following page).*
- Isolation in laboratory animals, or by its growth in cell culture.*
- PCR detection of *C burnetii* DNA prior to antibody detection – (note: PCR positivity doesn't necessarily indicate the presence of viable or replicating organisms).

***Note:** Due to the complexities and length of time associated with direct detection of the organism and isolation in laboratory animals/cell culture, these techniques are normally reserved for suspected chronic Q fever cases, particularly Q fever endocarditis.

Serology is the most commonly used diagnostic tool – i.e. observing the characteristic changing patterns of antibody responses in the IgM, IgG and IgA classes to Phase 1 and 2 antigens of *C burnetii* between symptom onset and early convalescence, which identifies a current infection, and distinguishes it from residual antibody from past exposure.

Chronic Q fever

Diagnosis of the various forms of chronic Q fever (i.e. Q fever endocarditis, granulomatous lesions, or QFS) requires a combination of direct detection of the coxiella and serological analysis mostly confined to specialist State or other laboratories. Consultation between clinician and the laboratory is strongly advised to facilitate the diagnostic process.

It is a mandatory requirement that isolation and propagation of *Coxiella burnetii* in animals, cell culture or chick embryos is done under level 3 bio-containment and by staff vaccinated with Q fever vaccine or known to have markers of immunity.

Techniques for detecting *C burnetii*

Direct detection of *C burnetii*

Table 15. Direct detection of *C burnetii* by microscopy

Direct detection technique	Properties
Staining of impression smears of infected tissues: – Gram stain – Modified Ziehl-Neelsen stain – Gimenez stain or Macchiavello stain or Flurochroming of coxiella genomic DNA by Hoechst 33258 stain	– SCV commonly seen – Gram variable – Weakly acid-fast – These methods are useful for staining infected tissue e.g. animal placentas, valve vegetations, infected yolk sac smears, but are insensitive for small numbers – Staining method not specific for coxiella
Detection by immunofluorescence or immunochemical histological staining	– Sensitive and specific methods – Particularly useful for detecting coxiella antigen in valve vegetations
Transmission electron microscopy	Detects SCV and LCV in tissues: For example, intra-cytoplasmic microcolonies or the remains of microcolonies once in the cytoplasm may be visualised in cardiac vegetations in Q fever endocarditis

Note: The information above reflects the experience of the Adelaide Q Fever Research Group.

C burnetii detection by PCR

C burnetii may be detected by PCR of target sequences in the genome. PCR techniques have facilitated rapid diagnosis of Q fever infection and sensitivity of detection. However, interpretation of the results requires care.

PCR in acute Q fever

- In acute Q fever, *C burnetii* can be detected by PCR in blood or serum during the bacteraemic period¹ – i.e. 10 days or so after onset of illness, before antibody has appeared or as it is just starting to be present) – see Fig 6 on page 107.
- The bacteraemia then declines – due to antibiotic treatment or perhaps because of the developing immune response.
The positive PCR reaction also may decline at this stage or may persist and overlap with the developing antibody response.
It is unclear whether this overlap indicates persistence of viable coxiellas or non-replicating but undegraded coxiellas retaining their DNA targets.
- It is emphasised that PCR positivity does not necessarily mean the presence of infective cells.

PCR in chronic Q fever

- For chronic Q fever, *C burnetii* DNA may be detected by PCR assay in peripheral blood mononuclear cells or in biopsy specimens from granulomatous tissue.
- PCR assays may be performed on fresh infected tissue, formalin fixed tissue, or waxed embedded specimens (consult the laboratory).
- For endocarditis and granulomas, PCR may be useful for detecting the presence of organism in the blood. However, in diagnostically difficult cases, PCR needs to be supported by culture of the organism (consult with the reference laboratory).

PCR positivity does not necessarily mean the presence of infective cells

Techniques for detecting *C burnetii*

PCR: Technical considerations

- As the technique is extremely sensitive, considerable attention must be given to avoiding cross contamination of samples by amplicon from previous test runs, or from positive specimens to others in the same test run.
- Various stages of the assay must be 'isolated' in rooms in different locations, with numerous negative controls (so called non-template controls) should be interspersed with samples. These have to be *negative* in each test run for the results to be taken as valid.

C burnetii isolation (cell culture/animal inoculation) as a diagnostic measure

Isolation of the coxiella may be performed in guinea pigs, hamsters and special lines of mice (AJI, SCID), in cultured eukaryotic cells (VERO, dog macrophage line DH82), or chick embryo yolk sac.

In general terms, isolation of *C burnetii* through cell culture or animal inoculation is not required for laboratory confirmation of acute Q fever. PCR detection of organisms in blood in the acute phase of illness, or more commonly, measurement of the serological response in early convalescence are rapid and cheap alternatives compared with culture.

There are a number of limitations associated with growth of the coxiella in laboratory animals or cell culture. These include the regulation requirement of Level 3 biocontainment laboratory and animal suites with vaccinated staff, special cages, dedicated autoclave equipment for sterilisation of cages, bedding, and reagents.

Isolation of *C burnetii*...is not required for [diagnosis] of acute Q fever

Additionally, detection of small numbers of coxiella by animal or cell culture inoculation may require holding the test substrate for weeks.

The limitations mean that the techniques for growing *C burnetii* are confined to reference or research laboratories. The laboratories should be consulted about the techniques, which typically would be used for unusual diagnostic problems, or the demonstration of viable organisms in the bloodstream of a patient with suspected endocarditis but who displays ambiguous evidence of valve involvement in medical imaging and low levels of PCR genomic targets on PCR assay.

It is too early to say whether the recent growth of *C burnetii* in cell-free media will greatly alter diagnostic approaches. If the isolates retain virulence, Level 3 biocontainment will still be required. (The relative sensitivity of animal eukaryotic cell culture and cell-free media for growth of *C burnetii* with specimens from suspected Q fever cases will need to be determined, etc).

The techniques for growing *C burnetii* are confined to reference or research laboratories

Serodiagnosis of Q fever

General concepts

- Serological testing is the most widely used diagnostic technique for Q fever (Table 16).
- The various assays differ in sensitivity and specificity as noted in Table 7.
- At present, in Australia, EIA is often used in clinical diagnostic laboratories.
- CFT, EIA and IFA with immunoglobulin class analysis are used in State Health laboratories.
- PCR may be used to complement serological testing.

Serological testing is the most widely used diagnostic technique... PCR may be used to complement serological testing

Table 16. Serological tests used to diagnose *C burnetii* infection

Test	Comments
Complement fixation (CFT)	<ul style="list-style-type: none">– Traditional test widely used to diagnose acute Q fever– Measures mostly IgG antibody and is less effective for detecting IgM, which forms first after onset of acute illness– Positive from mid-convalescence (see Fig 6)– Insensitive compared with IFA for detection of antibody after vaccination or from previous subclinical infection– Does not permit immunoglobulin class analysis of Q fever antibody responses– Can be unreliable due to presence of anticomplementary substances in the test serum
Immunofluorescence (IFA)	<ul style="list-style-type: none">– More sensitive than CFT– Detects early IgM antibody production– Can differentiate between and quantify antibodies from different immunoglobulin classes– Can be used to measure antibody response after vaccination
Enzyme immunoassay (EIA)	<ul style="list-style-type: none">– As sensitive as IFA– Can differentiate between antibodies from different immunological classes– Less flexible for measurement of changing antibody patterns as single dilution test formats are commonly used
Radioimmunoassay (RIA) Agglutination	<ul style="list-style-type: none">– Sensitive tests for special research purposes

Note: Assay handling and sensitivity can differ between laboratories. The information provided in the table reflects the experience of the Adelaide Q fever Research Group and Infectious Diseases Laboratories, IMVS.

Laboratory diagnosis of acute Q fever

Figure 6 is an idealised representation of the laboratory assays most relevant at various stages of a primary acute attack of Q fever.²

First 8 to 10 days after onset of illness

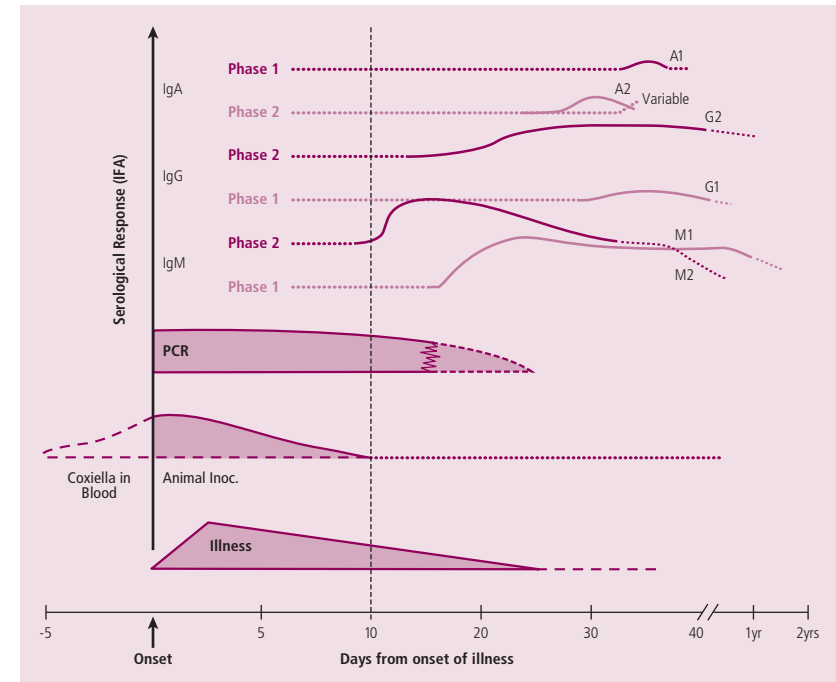
- Patients are bacteraemic but still antibody-negative.
- The coxiella may be detected most rapidly by PCR assay of the serum or plasma.
- Isolation of the coxiella in animals or cell culture requires Level 3 biocontainment facilities, and is slower and more expensive.
- Over 80% of specimens taken during this stage are PCR positive.¹

Day 10 to 12 from onset – up to day 20³

- IgM class antibody to phase 2 antigen (measured by IFA or EIA) appears first.
- This is followed later by IgM antibody to phase 1 antigen (See Fig 6).
- Later again, around day 15 to 20, IgG class antibody appears first to phase 2 antigen and later to phase 1 (see Fig 6).

IgM class antibody to phase 2 antigen (measured by IFA or EIA) appears first

Figure 6. Acute Q fever infection: bacteraemia and serological responses²



Note: This figure is an idealised representation based on the clinical and laboratory experience of the Adelaide Q Fever Research Group and Infectious Diseases Laboratories, IMVS. Results may vary based on the individual case.

Laboratory diagnosis of acute Q fever

Process⁴

- A baseline serum sample is taken at the patient's first visit (if the patient presents within the first 10 days of onset, this baseline sample will likely be seronegative or positive only at a very low level (Fig 6).
- A second sample is taken at 12-20 days after onset.
- The two sera are titrated in the same test run and detection of a 4-fold or greater rise in IgM antibody titre to coxiella Phase 2 antigen confirms the diagnosis of acute Q fever.
- The baseline sample is used for the PCR assay if the technique is accessible.
- If a positive antibody result is not obtained with the day 15-20 sample and a clinical suspicion of Q fever remains, it may be advisable to take an additional sample between 20-25 days after onset.

Detection of a 4-fold or greater rise in IgM antibody titre confirms the diagnosis of acute Q fever

Important notes⁴

- The interpretation of serological findings in a case of suspected Q fever is vitally dependent on the stage of illness when the serum sample is taken.
It is therefore essential to establish the exact date of ONSET – i.e. when the patient first became ill.
- *It is strongly recommended to consult with the reference laboratory doing the testing to discuss the date reported by the patient and the results obtained via serological testing.*
- The rule of a four-fold or greater increase in antibody titre remains the most stringent criterion for diagnosis of a current primary infection. Demonstration of a bacteraemia by PCR reinforces the validity of the diagnosis.

The real world⁴

- In everyday practice, patients may not present to the doctor (or Q fever may not have been suspected) until early to mid- convalescence (day 12-20 post-onset).
- At this stage of the illness, IgM antibody to Phase 2 and 1 antigens may already have reached a plateau.
- It is therefore advisable to take a blood sample as soon as a patient is seen and another sample 5-7 days later.
- Detection of late-rising IgG antibody to coxiella Phase 1 and 2 antigens may allow the diagnostic criteria of a 4-fold increase in antibody titre to be met (see Fig 6 on page 107).

It is essential to establish the exact date of ONSET of illness

Laboratory diagnosis of acute Q fever

EIA for serodiagnosis of acute Q fever⁴

- Typically, automated EIA is now used in clinical diagnostic laboratories for Q fever serodiagnosis.
- The test is highly sensitive and can measure antibody in IgM and IgG classes to Phase 1 and 2 antigens.
- Rather than providing titre values – the assay is most commonly formatted for testing a single serum dilution with antigen.
- The strength of the antigen-antibody reaction is quantified in terms of the intensity of a coloured end product.
- Results are most often reported as negative, low, medium, or high activity – in relation to a panel of positive or negative sera.
- Consequently, the 4-fold rise in antibody titre is more difficult to quantify in this routine assay format.
- If there is a change from low positive to high positive between acute and convalescent phase samples of serum, an increase of antibody can be inferred, although not in terms of titre.
 - The increase, along with the presence of an unchanging or increasing level of IgM antibody is secure enough grounds for a diagnosis of current Q fever for clinical treatment purposes.
 - However, it may be less persuasive for compensation purposes. In the latter instance, titration of the stored sera by IFA, and immunoglobulin class analysis is desirable.
- Once again, consultation with the reference laboratory regarding the interpretation of serological findings is strongly advised.

In early to mid convalescence...detection of late-rising IgG antibody...may allow the diagnostic criteria of a 4-fold increase to be met

EIA – single serum sample⁴

- In the real world of clinical practice, the possible sometimes diverges from the ideal, and a single serum sample is sometimes all that is available.
- Note: the practice of taking a single serum sample **is not** optimal.
- However, if circumstances arise when only a single serum sample is available, the evidence for a current infection may be argued from:
 - The strength of the positive reaction in EIA compared with the infrequency of that reaction value in a panel of sera from the unexposed population and the presence of antibody in the IgM class.
- The above deduction from a single sample along with compatible clinical observations, although not ideal – may provide reliable support for immediate therapy and other pressing clinical decisions.

Laboratory diagnosis of acute Q fever

Distinguishing past and current infection via serology

– After a primary acute Q fever infection, IgM antibody generally declines rapidly but is variable, and on occasion, may remain raised over several months.³

– However, IgG class antibody to *C burnetii* Phase 1 and 2 antigens may persist for many years after initial infection at unchanging titres ranging from 20 to 320 via IFA.^{2,5}

– Due to antibody persistence, at times, acute febrile illnesses caused by other agents (e.g. *Legionella* spp) are open to misidentification as Q fever in those frequently exposed to *C burnetii*. For example:

An abattoir worker presents with clinical symptoms due to non-Q fever agent

Serology shows presence of IgG antibodies to Phase 1 and 2 antigens due to past infection.

The symptoms are erroneously interpreted as current Q fever infection.

– Serological patterns suggesting past rather than a current acute infection include the following (also see Table 17):²

Low or medium IgG antibody titres that do not change between serial serum specimens.

The presence of antibody very early in the illness i.e. at a stage of the illness when it would not be present in an acute primary infection. (see Fig 6 on page 107).

IgG class antibody to *C burnetii* Phase 1 and 2 antigens may persist for many years after initial infection

Table 17. Idealised serological patterns at various stages of Q fever infection²

Clinical category	Time from onset of illness	Antibody titre								Antibody reading EIA	
		CFT		IFA							
				IgG		IgM		IgA			
		Ph 1	Ph 2	Ph 1	Ph 2	Ph 1	Ph 2	Ph 1	Ph 2	IgG	IgM
Acute Q fever	5 d	< 10	< 10	< 10	< 10	< 10	+ 10	< 10	< 10	Neg	Low
	12 d	< 10	20	< 10	80	20	1280	< 10	< 10	Low	High
	25 d	< 10	160	< 10	320	80	5120	< 10	< 10	High	High
	45 d	< 10	80	20	160	80	320	< 10	20-40	High	Low
Past Q fever infection (plus unrelated illness)*	6 d	< 10	20	< 10	320	< 10	< 10	< 10	< 10	Low	Low
	26 d	< 10	10	10	160	10	10	< 10	< 10	Mid	Mid
	40 d	10	20	< 10	320	< 10	< 10	< 10	< 10	Low	Low
Q fever endocarditis**	36-40 mths	1280	2560	5120	20,480	< 10	< 10	2560	640	High	Mid
	48 mths	1280	2560	5120	10,240	10	< 10	2560	640	High	Low
	60 mths	640	1280	5120	5120	10	< 10	2560	640	High	Mid
Granuloma*** of liver	10-40 d	?	320	1000	3000	1000	3000	< 10	< 10	?	?
QFS	6 mths	< 10	20	80	320	< 10	< 10	< 10	< 10		
	12 mths	< 10	20	80	320	< 10	< 10	< 10	< 10		

The titres given and the change in values are idealised examples. There is a wide variation between patients and some variation in sensitivity and formats of assays between laboratories.

– In acute Q fever it is the **increase** in titre that is the diagnostic feature of importance.

– In past infection, the values do not change significantly between serial samples.

– In chronic Q fever taking the form of endocarditis and/or granuloma, absolute titres are much higher in the IgG and IgA subclasses. Information about granuloma in bone, testes and liver is limited and original papers should be considered.

– In QFS, titres are essentially the same as those of a person infected in the past who has made an asymptomatic recovery.

IFA = titration of serum in microdots of coxiella by indirect immunofluorescence

EIA = enzyme immunoassay. Consult product leaflet of manufacturer.

*Period in days between serial samples during an illness other than Q fever, coinciding with antibody from past Q fever infection.

**Months after original acute Q fever when endocarditis presented as a clinical problem.

***Days from clinical presentation of granulomatous condition (as patients may not have experienced an overt attack of acute Q fever).

References

References

Recommended reading list:

Q Fever: The Biology of *Coxiella burnetii*. Williams JC & Thompson HA: eds. 1991. CRC Press Inc, Boca Raton, Florida, USA.

Q Fever. Volume 1. The Disease. Marrie TJ: ed. 1990. CRC Press Inc, Boca Raton, Florida, USA.

Section 1

1. Pope JH, et al. *Coxiella burnetii* in kangaroos and kangaroo ticks in western Queensland. *Aust J Exp Biol Med Sci* 1960; 38:17-27.
2. Lang GH. Coxiellosis (Q fever) in animals. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 23-48.
3. Stoker MG, Marmion BP. The spread of Q fever from animals to man; the natural history of a rickettsial disease. *Bull World Health Organ* 1955; 13: 781-806.
4. Marmion BP, Stoker MG. The epidemiology of Q fever in Great Britain; an analysis of the findings and some conclusions. *Br Med J* 1958; 2: 809-16.
5. Australian Government Department of Health and Ageing. National Notifiable Diseases Surveillance System. Number of notifications for the year 2008. Retrieved 22 May 2009 from <http://www9.health.gov.au/cda/Source/Rpt_2.cfm?RequestTimeout=500>
6. Australian Government Department of Health and Ageing. National Notifiable Diseases Surveillance System. Number of notifications by year, Australia, 1991 to 2008. Retrieved 22 May 2009 from <http://www9.health.gov.au/cda/Source/Rpt_3.cfm>
7. Garner MG, et al. *Aust N Z J Pub Health* 1997; 21: 722-30.
8. Kermode M, et al. An economic evaluation of increased uptake in Q fever vaccination among meat and agricultural industry workers following implementation of the National Q Fever Management Program. *Aust N Z J Public Health* 2003; 27: 390-8.
9. Gidding HF, et al. Australia's national Q fever vaccination program. *Vaccine* 2009; 27: 2037-41.
10. Marmion BP. Q fever: the long journey to control by vaccination. *MJA* 2007; 186: 164-6.
11. Shapiro RA, et al. A randomized, controlled, double-blind, cross-over, clinical trial of Q fever vaccine in selected Queensland abattoirs. *Epidemiol Infect* 1990; 104: 267-73.

12. Marmion BP, et al. Vaccine prophylaxis of abattoir-associated Q fever: eight years' experience in Australian abattoirs. *Epidemiol Infect* 1990; 104:275-87.
13. Syrucek L, et al. Isolation of *Coxiella burnetii* from human placentas. *J Hyg Epidemiol Microbiol Immunol* 1958; 2: 29-35.
14. Welsh HH, et al. Q fever in California. IV. Occurrence of *Coxiella burnetii* in the placenta of naturally infected sheep. *Public Health Rep* 1951; 66: 1473-7.
15. Marrie TJ. Epidemiology of Q fever. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 49-70.
16. Marrie TJ, et al. An outbreak of Q fever probably due to contact with a parturient cat. *Chest* 1988; 93: 98-103.
17. Buhariwalla F, et al. A dog-related outbreak of Q fever. *Clin Infect Dis* 1996; 23: 753-5.
18. Marrie TJ, et al. Q fever pneumonia associated with exposure to wild rabbits. *Lancet* 1986; 1: 427-9.
19. Welsh HH, et al. Air-borne transmission of Q fever: the role of parturition in the generation of infective aerosols. *Ann NY Acad Sci* 1957; 70: 528.
20. McKelvie P. Q fever in a Queensland meatworks. *Med J Aust* 1980; 1: 590-3.
21. Maurin M, Raoult D. Q fever. *Clin Microbiol Rev* 1999; 12: 518-53.
22. Mann JS, et al. Q fever: person to person transmission within a family. *Thorax* 1986; 41: 974-5.
23. Raoult D, Stein A. Q fever during pregnancy. A risk to women, fetuses and obstetricians. *N Engl J Med* 1994; 330: 371.
24. Raoult D, et al. Q fever during pregnancy: diagnosis, treatment, and follow-up. *Arch Intern Med* 2002; 162: 701-4.
25. Milazzo A, et al. Sexually transmitted Q fever. *Clin Infect Dis* 2001; 33: 399-402.
26. Kruszezwska D, et al. Possible sexual transmission of Q fever among humans. *Clin Infect Dis* 1996; 22: 1087-8.
27. Hawker JL, et al. A large outbreak of Q fever in the West Midlands: windborne spread into a metropolitan area? *Commun Dis Public Health* 1998; 1: 180-7.

28. Porten K, et al. A super-spreading ewe infects hundreds with Q fever at a farmers' market in Germany. *BMC Infect Dis* 2006; 6:147.
29. Delsing CE, Kullberg BJ. Q fever in the Netherlands: a concise overview and implications of the largest ongoing outbreak. *Neth J Med* 2008; 66: 365-7.
30. Professor Barrie Marmion personal communication.
31. Langley JM, et al. Poker players' pneumonia. An urban outbreak of Q fever following exposure to a parturient cat. *N Engl J Med* 1988; 319: 354-6.
32. Marmion BP, Stoker MG. The varying epidemiology of Q fever in the southeast region of Great Britain. II. In two rural areas. *J Hyg (Lond)* 1956; 54: 547-61.
33. Harvey MS, et al. An outbreak of Q fever in East Kent. *Lancet* 1951; 2: 1152-7.
34. Wade AJ, et al. Q fever outbreak at a cosmetics supply factory. *Clin Infect Dis* 2006; 42: e50-2.

Section 2

1. McCaul TF. The Developmental cycle of *Coxiella burnetii*. In: Q fever: The Biology. Williams JC, Thompson HA: eds. CRC Press 1991; p 223-58.
2. McCaul TF, Williams JC. Developmental cycle of *Coxiella burnetii*: structure and morphogenesis of vegetative and sporogenic differentiations. *J Bacteriol* 1981; 147: 1063-76.
3. Williams JC. Infectivity, Virulence, and pathogenicity of *Coxiella burnetii* for various hosts. In: Q fever: The Biology. Williams JC, Thompson HA: eds. CRC Press 1991; p 21-71.
4. Christie AB. In: Infectious diseases: epidemiology and clinical practice, 4th ed. Christie AB: eds. Edinburgh: Churchill Livingstone 1987; p 1098-1115.
5. Marrie TJ. Epidemiology of Q fever. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 49-70.
6. Stoker MG, Fiset P. Phase variation of the Nine Mile and other strains of *Rickettsia burnetii*. *Can J Microbiol* 1956; 2: 310-21.
7. Mayer H, et al. Chemical and immunochemical studies on lipopolysaccharides of *Coxiella burnetii* phase I and phase II. *Adv Exp Med Biol* 1988; 228: 577-91.
8. Moos A, Hackstadt T. Comparative virulence of intra- and interstrain lipopolysaccharide variants

of *Coxiella burnetii* in the guinea pig model. *Infect Immun* 1987; 55: 1144-50.

9. Izzo AA, Marmion BP. Variation in interferon-gamma responses to *Coxiella burnetii* antigens with lymphocytes from vaccinated or naturally infected subjects. *Clin Exp Immunol* 1993; 94: 507-15.
10. Capo C, et al. Subversion of monocyte functions by *Coxiella burnetii*: impairment of the cross-talk between alphavbeta3 integrin and CR3. *J Immunol* 1999; 163: 6078-85.
11. Dellacasagrande J, et al. alpha(v)beta(3) integrin and bacterial lipopolysaccharide are involved in *Coxiella burnetii*-stimulated production of tumor necrosis factor by human monocytes. *Infect Immun* 2000; 68: 5673-8.
12. Ormsbee RA, Marmion BP. Prevention of *Coxiella burnetii* infection; vaccine and guidelines for those at risk. In: 'Q Fever: The disease'. Marrie TJ: ed. CRC Press, Boca Raton, Florida. 1990: 225-48.
13. Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.
14. Omsland A, et al. Host cell-free growth of the Q fever bacterium *Coxiella burnetii*. *Proc Natl Acad Sci U S A*. 2009; 106(11): 4430-4.
15. Adelaide Q Fever Research Group unpublished. Prof Barrie Marmion personal communication.
16. Maurin M, Raoult D. Q fever. *Clin Microbiol Rev* 1999; 12: 518-53.
17. Voth DE, Heinzen RA. Lounging in a lysosome: the intracellular lifestyle of *Coxiella burnetii*. *Cell Microbiol* 2007; 9: 829-40.
18. Dellacasagrande J, et al. IFN-gamma-mediated control of *Coxiella burnetii* survival in monocytes: the role of cell apoptosis and TNF. *J Immunol* 1999; 162: 2259-65.

Section 3

1. Marmion BP, et al. Vaccine prophylaxis of abattoir-associated Q fever. *Lancet* 1984; 2: 1411-4.
2. Marrie TJ. Acute Q fever. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 125-160.
3. Spelman DW. Q fever: a study of 111 consecutive cases. *Med J Aust* 1982; 1: 547-8, 551, 553.

References

- Smith DL, et al. A large Q fever outbreak in the West Midlands: clinical aspects. *Respir Med* 1993; 87: 509-16.
- Vollmer-Conna U, et al. Production of pro-inflammatory cytokines correlates with the symptoms of acute sickness behaviour in humans. *Psychol Med* 2004; 34: 1289-97.
- Meghari S, et al. *Coxiella burnetii* stimulates production of RANTES and MCP-1 by mononuclear cells: modulation by adhesion to endothelial cells and its implication in Q fever. *Eur Cytokine Netw* 2006; 17: 253-9.
- Honstetter A, et al. Dysregulation of cytokines in acute Q fever: role of interleukin-10 and tumor necrosis factor in chronic evolution of Q fever. *J Infect Dis* 2003; 187: 956-62.
- Marmion BP, et al. Vaccine prophylaxis of abattoir-associated Q fever: eight years' experience in Australian abattoirs. *Epidemiol Infect* 1990; 104: 275-87.
- Ackland JR, et al. Vaccine prophylaxis of Q fever. A follow-up study of the efficacy of Q-Vax (CSL) 1985-1990. *Med J Aust* 1994; 160: 704-8.
- Professor B Marmion personal communication.
- Marmion BP, et al. Protracted debility and fatigue after acute Q fever. *Lancet* 1996; 347: 977-8.
- Ayres JG, et al. Protracted debility and fatigue after acute Q fever. *Lancet* 1996; 347: 978-9.
- Adelaide Q Fever Research Group unpublished. Prof Barrie Marmion personal communication.
- Marmion BP, et al. Q fever: Persistence of antigenic non-viable cell residues of *C burnetii* in the host. Implications for post Q fever infection fatigue syndrome and other chronic sequelae. Accepted *Q J Med* 2009.
- Bossi P, et al. Bichat guidelines for the clinical management of Q fever and bioterrorism-related Q fever. *Euro Surveill* 2004; 9: E19-20.
- Marrie TJ. Q fever hepatitis. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 171-177.
- Raoult D, et al. Natural history & pathophysiology of Q fever. *Lancet Infect Dis* 2005; 5: 219-26.
- Raoult D, et al. Q fever during pregnancy: diagnosis, treatment, and follow-up. *Arch Intern Med* 2002; 162: 701-4.
- Stein A, Raoult D. Q fever during pregnancy: a public health problem in southern France. *Clin Infect Dis* 1998; 27: 592-596.
- Syruck L, et al. Isolation of *Coxiella burnetii* from human placentas. *J Hyg Epidemiol Microbiol Immunol* 1958; 2: 29-35.
- Maurin M, Raoult D. Q fever. *Clin Microbiol Rev* 1999; 12: 518-53.
- Raoult D, et al. Q Fever 1985-1998. *Medicine (Baltimore)* 2000; 79: 109-23.
- Vaccine Preventable Diseases and Vaccination Coverage in Australia, 2003 to 2005. Q fever. *Communicable Diseases Intelligence* Volume 31 – Supplement – June 2007. Retrieved 20 May 2009 from <http://www.health.gov.au/internet/main/publishing.nsf/Content/cda-cdi31suppl.htm~cda-cdi31suppl-3.htm~cda-cdi31suppl-3l.htm>
- Helbig KJ, et al. Variation in immune response genes and chronic Q fever. Concepts: preliminary test with post-Q fever fatigue syndrome. *Genes Immun* 2003; 4: 82-5.
- Helbig KJ, et al. Immune response genes in the post-Q-fever fatigue syndrome, Q fever endocarditis and uncomplicated acute primary Q fever. *Q J Med* 2005; 98: 565-574.
- Harris RJ, et al. Long-term persistence of *Coxiella burnetii* in the host after primary Q fever. *Epidemiol Infect* 2000; 124: 543-9.
- Marmion BP, et al. Long-term persistence of *Coxiella burnetii* after acute primary Q fever. *Q J Med* 2005; 98: 7-20.
- Capo C, et al. Production of interleukin-10 and transforming growth factor beta by peripheral blood mononuclear cells in Q fever endocarditis. *Infect Immun* 1996; 64: 4143-7.
- Meghari S, et al. Deficient transendothelial migration of leukocytes in Q fever: the role played by interleukin-10. *J Infect Dis* 2006; 194: 365-9.
- Ghigo E, et al. Link between impaired maturation of phagosomes and defective *Coxiella burnetii* killing in patients with chronic Q fever. *J Infect Dis* 2004; 190: 1767-72.
- Ghigo E, et al. Interleukin-10 stimulates *Coxiella burnetii* replication in human monocytes through tumor necrosis factor down-modulation: role in microbicidal defect of Q fever. *Infect Immun* 2001; 69: 2345-52.
- Penttilä IA, et al. Cytokine dysregulation in the post-Q-fever fatigue syndrome. *QJM*. 1998; 91: 549-60.
- Marmion BP. Subacute rickettsial endocarditis: an unusual complication of Q fever. *J Hyg Epidemiol Microbiol Immunol* 1962; 6: 79-84.
- Fenollar F, et al. Risks factors and prevention of Q fever endocarditis. *Clin Infect Dis* 2001; 33: 312-6.
- Raoult D, et al. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 179-199.
- Andrews PS, Marmion BP. Chronic Q fever. Morbid anatomical and bacteriological findings in a patient with endocarditis. *Br Med J* 1959; 983-88.
- Brouqui P, et al. Immunohistologic demonstration of *Coxiella burnetii* in the valves of patients with Q fever endocarditis. *Am J Med* 1994; 97: 451-58.
- Lepidi H, et al. Cardiac valves in patients with Q fever endocarditis: microbiological, molecular, and histologic studies. *J Infect Dis* 2003; 187: 1097-106.
- Peacock MG, et al. Serological evaluation of Q fever in humans: enhanced phase I titers of immunoglobulins G and A are diagnostic for Q fever endocarditis. *Infect Immun* 1983; 41: 1089-98.
- Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.
- Ayres JG, et al. Post-infection fatigue syndrome following Q fever. *QJM* 1998; 91: 105-23.
- Nourse C, et al. Three cases of Q fever osteomyelitis in children and a review of the literature. *Clin Infect Dis* 2004; 39: e61-6.
- Tattevin P, et al. Granulomatous lymphadenitis as a manifestation of Q Fever. *Emerg Infect Dis* 2003; 9: 137-8.
- Janigan DT, Marrie TJ. An inflammatory pseudotumor of the lung in Q fever pneumonia. *N Engl J Med* 1983; 308: 86-8.
- Voigt JJ, et al. Liver and bone marrow granulomas in Q fever. *Gastroenterology* 1983; 84: 887-8.

Section 4

- Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.
- Turra M, et al. Diagnosis of acute Q fever by PCR on sera during a recent outbreak in rural south Australia. *Ann N Y Acad Sci* 2006; 1078: 566-9.
- Professor B Marmion. Personal communication.

Section 5

- Yeaman MR and Bacca OG. Antibiotic susceptibility of *Coxiella burnetii*. In: Q fever. The disease. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 213-23.
- Yeaman MR, Mitscher LA, Baca OG. In vitro susceptibility of *Coxiella burnetii* to antibiotics, including several quinolones. *Antimicrob Agents Chemother* 1987; 31: 1079-84.
- Carcopino X, et al. Managing Q fever during pregnancy: the benefits of long-term cotrimoxazole therapy. *Clin Infect Dis* 2007; 45: 548-55.
- Therapeutic Guidelines: Antibiotic Version 13. Melbourne; Therapeutic Guidelines Limited; 2006.
- Assoc Prof Denis Spelman personal communication.
- Raoult D, et al. Treatment of Q fever endocarditis: comparison of 2 regimens containing doxycycline and ofloxacin or hydroxychloroquine. *Arch Intern Med* 1999; 159: 167-73.

Section 6

- Q-VAX®. Approved Product Information, 2008. CSL Biotherapies.
- Marmion BP, et al. Vaccine prophylaxis of abattoir-associated Q fever: eight years' experience in Australian abattoirs. *Epidemiol Infect* 1990; 104: 275-87.
- NHMRC. The Australian Immunisation Handbook 9th edition. 2008.
- Gidding HF, et al. Australia's national Q fever vaccination program. *Vaccine* 2009; 27: 2037-41.
- Ormsbee RA, Marmion BP. Prevention of *coxiella burnetii* infection; vaccine and guidelines for those at risk. In: 'Q Fever: The disease'. Marrie TJ: ed. CRC Press, Boca Raton, Florida. 1990: 225-48.
- Marmion BP. Q fever: the long journey to control by vaccination. *MJA* 2007; 186: 164-6.
- Marmion BP, et al. Vaccine prophylaxis of abattoir-associated Q fever. *Lancet* 1984; 2: 1411-4.
- Professor B Marmion personal communication.
- Adelaide Q Fever Research Group unpublished. Prof B Marmion personal communication.
- Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.

References

11. Izzo AA, Marmion BP, Worswick DA. Markers of cell-mediated immunity after vaccination with an inactivated, whole-cell Q fever vaccine. *J Infect Dis* 1988; 157: 781-9.
12. Ackland JR, *et al.* Vaccine prophylaxis of Q fever. A follow-up study of the efficacy of Q-Vax (CSL) 1985-1990. *Med J Aust* 1994; 160: 704-8.
13. Shapiro RA, *et al.* A randomized, controlled, double-blind, cross-over, clinical trial of Q fever vaccine in selected Queensland abattoirs. *Epidemiol Infect* 1990; 104: 267-73.
14. Gilroy N, *et al.* Abattoir-associated Q fever: a Q fever outbreak during a Q fever vaccination program. *Aust N Z J Public Health* 2001; 25: 362-7.
15. Chiu CK, Durrheim DN. A review of the efficacy of human Q fever vaccine registered in Australia. *N S W Public Health Bull* 2007; 18: 133-6.
5. Christie AB. In: *Infectious diseases: epidemiology and clinical practice*, 4th ed. Christie AB: eds. Edinburgh: Churchill Livingstone 1987; p 1098-115.
6. Marrie TJ. Epidemiology of Q fever. In: *Q fever. The disease*. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 49-70.
7. Stoker MG, Fiset P. Phase variation of the Nine Mile and other strains of *Rickettsia burnetii*. *Can J Microbiol* 1956; 2: 310-21.
8. Moos A, Hackstadt T. Comparative virulence of intra- and interstrain lipopolysaccharide variants of *Coxiella burnetii* in the guinea pig model. *Infect Immun* 1987; 55: 1144-50.
9. Mayer H, *et al.* Chemical and immunochemical studies on lipopolysaccharides of *Coxiella burnetii* phase I and phase II. *Adv Exp Med Biol* 1988; 228: 577-91.
10. Hoover TA, *et al.* Chromosomal DNA deletions explain phenotypic characteristics of two antigenic variants, phase 2 and RSA 514 (Crazy), of the *Coxiella burnetii* Nine Mile strain. *Infect Immun* 2002; 70: 6726-33.
11. Schramek S, *et al.* 3-C branched aldoses in lipopolysaccharide of phase 1 *Coxiella burnetii* and their role as immunodominant factors. *Eur J Biochem* 1985; 148: 455-461.
12. Izzo AA, Marmion BP. Variation in interferon-gamma responses to *Coxiella burnetii* antigens with lymphocytes from vaccinated or naturally infected subjects. *Clin Exp Immunol* 1993; 94: 507-15.
13. Capo C, *et al.* Subversion of monocyte functions by *Coxiella burnetii*: impairment of the cross-talk between alphavbeta3 integrin and CR3. *J Immunol* 1999; 163: 6078-85.
14. Dellacasagrande J, *et al.* alpha(v)beta(3) integrin and bacterial lipopolysaccharide are involved in *Coxiella burnetii*-stimulated production of tumor necrosis factor by human monocytes. *Infect Immun* 2000; 68: 5673-8.
15. Ormsbee RA, Marmion BP. Prevention of *Coxiella burnetii* infection; vaccine and guidelines for those at risk. In: 'Q Fever: The disease'. Marrie T: ed. CRC Press, Boca Raton, Florida. 1990: 225-48.
16. Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.
17. Peacock MG, *et al.* Serological evaluation of Q fever in humans: enhanced phase I titers of immunoglobulins G and A are diagnostic for Q fever endocarditis. *Infect Immun* 1983; 41: 1089-98.
18. Omsland A, *et al.* Host cell-free growth of the Q fever bacterium *Coxiella burnetii*. *Proc Natl Acad Sci U S A* 2009; 106: 4430-4.
19. Adelaide Q Fever Research Group unpublished. Prof Barrie Marmion personal communication.
20. Maurin M, Raoult D. Q fever. *Clin Microbiol Rev* 1999; 12: 518-53.
21. Hackstadt T, Williams JC. Biochemical stratagem for obligate parasitism of eukaryotic cells by *Coxiella burnetii*. *Proc Natl Acad Sci USA* 1981; 78: 3240-4.
22. Voth DE, Heinzen RA. Lounging in a lysosome: the intracellular lifestyle of *Coxiella burnetii*. *Cell Microbiol* 2007; 9: 829-40.
23. Zamboni DS, *et al.* *Coxiella burnetii* express type IV secretion system proteins that function similarly to components of the Legionella pneumophila Dot/Icm system. *Mol Microbiol* 2003; 49: 965-76.
24. Raoult D, *et al.* Natural history and pathophysiology of Q fever. *Lancet Infect Dis* 2005; 5: 219-26.
25. Capo C, *et al.* *Coxiella burnetii* avoids macrophage phagocytosis by interfering with spatial distribution of complement receptor 3. *J Immunol* 2003; 170: 4217-25.
26. Ghigo E, *et al.* *Coxiella burnetii* survival in THP-1 monocytes involves the impairment of phagosome maturation: IFN-gamma mediates its restoration and bacterial killing. *J Immunol* 2002; 169: 4488-95.
27. Meghari S, *et al.* Deficient transendothelial migration of leukocytes in Q fever: the role played by interleukin-10. *J Infect Dis* 2006; 194: 365-9.
28. Ghigo E, *et al.* Interleukin-10 stimulates *Coxiella burnetii* replication in human monocytes through tumor necrosis factor down-modulation: role in microbicidal defect of Q fever. *Infect Immun* 2001; 69: 2345-52.
29. Ghigo E, *et al.* Link between impaired maturation of phagosomes and defective *Coxiella burnetii* killing in patients with chronic Q fever. *J Infect Dis* 2004; 190: 1767-72.
30. Marmion BP, *et al.* Vaccine prophylaxis of abattoir-associated Q fever: eight years' experience in Australian abattoirs. *Epidemiol Infect* 1990; 104: 275-87.
31. Ackland JR, *et al.* Vaccine prophylaxis of Q fever. A follow-up study of the efficacy of Q-Vax (CSL) 1985-1990. *Med J Aust* 1994; 160: 704-8.
32. Waag DM. Immune response to *Coxiella burnetii* infection. In: *Q fever. The disease*. Volume 1. Marrie TJ: ed. CRC Press Inc 1990; p 107-23.
33. Dellacasagrande J, *et al.* IFN-gamma-mediated control of *Coxiella burnetii* survival in monocytes: the role of cell apoptosis and TNF. *J Immunol* 1999; 162: 2259-65.
34. Andoh M, *et al.* T cells are essential for bacterial clearance, and gamma interferon, tumor necrosis factor alpha, and B cells are crucial for disease development in *Coxiella burnetii* infection in mice. *Infect Immun* 2007; 75: 3245-55.
35. Marmion BP, *et al.* Long-term persistence of *Coxiella burnetii* after acute primary Q fever. *Q J Med* 2005; 98: 7-20.
36. Helbig KJ, *et al.* Variation in immune response genes and chronic Q fever. Concepts: preliminary test with post-Q fever fatigue syndrome. *Genes Immun* 2003; 4: 82-5.
37. Helbig KJ, *et al.* Immune response genes in the post-Q-fever fatigue syndrome, Q fever endocarditis and uncomplicated acute primary Q fever. *Q J Med* 2005; 98: 565-74.

Section 7

1. Marmion BP, *et al.* Vaccine prophylaxis of abattoir-associated Q fever. *Lancet* 1984; 2: 1411-4.
2. Q-VAX®. Approved Product Information, 2008. CSL Biotherapies.
3. Professor B Marmion personal communication.

Section 8

1. Q-VAX®. Approved Product Information, 2008. CSL Biotherapies.
2. Professor B Marmion personal communication.
3. Marmion BP, *et al.* Vaccine prophylaxis of abattoir-associated Q fever: eight years' experience in Australian abattoirs. *Epidemiol Infect* 1990; 104: 275-87.

Section 9a – Appendix 1

1. McCaul TF. The Developmental cycle of *Coxiella burnetii*. In *Q fever: The Biology*. Williams JC, Thompson HA: eds. CRC Press 1991; p 223-58.
2. McCaul TF, Williams JC. Developmental cycle of *Coxiella burnetii*: structure and morphogenesis of vegetative and sporogenic differentiations. *J Bacteriol* 1981; 147: 1063-76.
3. Professor B Marmion personal communication.
4. Williams JC. Infectivity, Virulence, and pathogenicity of *Coxiella burnetii* for various hosts. In *Q fever: The Biology*. Williams JC, Thompson HA: eds. CRC Press 1991; p 21-71.

Section 9b – Appendix 2

1. Turra M, *et al.* Diagnosis of acute Q fever by PCR on sera during a recent outbreak in rural south Australia. *Ann N Y Acad Sci* 2006; 1078: 566-9.
2. Adelaide Q Fever Research Group unpublished. Professor B Marmion personal communication.
3. Worswick D and Marmion BP. Antibody responses in acute and chronic Q fever and in subjects vaccinated against Q fever. *Med Microbiol* 1985; 19: 281-96.
4. Professor B Marmion personal communication.
5. Marmion BP, *et al.* Long-term persistence of *Coxiella burnetii* after acute primary Q fever. *Q J Med* 2005; 98: 7-20.

Q-VAX® Q Fever Vaccine & Q-VAX® Skin Test Product Information

Q-VAX® product information

Name of the medicine

Q Fever Vaccine.
Q Fever Skin Test.

Description

Q-VAX® is a purified suspension of formalin-inactivated, *Coxiella burnetii* prepared from the Phase 1 Henzlerling strain of the organism grown in the yolk sacs of embryonated eggs. Excess egg proteins are removed by fractionation and ultracentrifugation.

Q-VAX® Vaccine contains 25 µg of antigen in 0.5 mL of an aqueous solution. Thiomersal 0.01% w/v is added as a preservative.

Q-VAX® Skin Test contains 2.5µg of antigen per 0.5mL of aqueous solution. **Prior to administration**, Q-VAX® Skin Test is diluted with Sodium Chloride injection to ensure that 16.7ng (nanograms) of antigen is delivered per 0.1mL intradermal dose. (see DOSAGE AND ADMINISTRATION).

Pharmacology

Q fever is caused by *Coxiella burnetii*, an obligate, intracellular, Gram-negative coccobacillus. The *C. burnetii* is shed in the products of conception, and on the neonate of the infected animal. It may also be present in the udder and milk of infected animals and is passed on within their faeces. Infection is transmitted to humans by inhalation of infected airborne particles or dust during the handling or processing of these materials or by close proximity to infected animals when giving birth.

Early antibody response to the vaccine is predominantly with the IgM subclass; IgG antibodies appear later. Although the seroconversion rate is low (50-80%) and antibody levels are transient, cell mediated immunity develops. Clinical trials have demonstrated a high degree of efficacy (see CLINICAL TRIALS). As Q fever is often asymptomatic or misdiagnosed due to its non-specific nature, many abattoir workers develop immunity to Q fever without an obvious illness.

The duration of protective immunity following immunisation is unknown, but is believed to be in excess of five years.

Revaccination must never be undertaken due to the possibility of severe hypersensitivity reactions (see CONTRAINDICATIONS).

Clinical trials

A randomised, blind, controlled study comparing Q-VAX® and influenza vaccine for the prevention of Q fever amongst 200 workers in three Queensland abattoirs was undertaken, using sequential analysis for determining the efficacy of Q-VAX®. A statistically

significant difference in the incidence of symptomatic Q fever was noted 15 months after commencement of vaccination, with 7 cases in those given the control vaccine and no cases in those given Q-VAX®. At 15 months, 24% of those who had not been vaccinated and had not developed symptomatic infection had serological evidence of exposure to Q fever, indicating subclinical infection.

A retrospective cohort study in three South Australian abattoirs was undertaken to compare the incidence of Q fever in vaccinated and unvaccinated subjects between 1985 and 1990. There were two cases of Q fever amongst 2555 vaccinated employees compared with 55 cases in 1365 unvaccinated subjects. Both cases of Q fever in the vaccinated group occurred within two weeks of receiving the vaccine. For workers who were vaccinated, the mean duration of employment following vaccination was 1.9 years; 203 workers were employed for all five years of the study.

Protection against clinical infection over this period was demonstrated.

Although the dose in each of these studies was nominally 30 µg, one batch which contained only 20 µg in each dose was shown to be as effective. However, as with all vaccines, 100% effectiveness for generation of protective immunity against Q Fever cannot be guaranteed (see PRECAUTIONS).

Indications

Q-VAX® Vaccine is indicated for the immunisation of susceptible adults at identifiable risk of infection with Q fever.

Abattoir workers (and those closely associated with the meat industry), farmers, veterinarians, stockyard workers, shearers, animal transporters and many others exposed to cattle, sheep or goats or their products should be considered for vaccination.

Note also that Q fever has occurred among persons culling and processing kangaroos and that laboratory personnel handling potentially infected veterinary specimens, or visiting abattoirs, are at risk.

Q-VAX® Skin Test is indicated for the pre-screening of potential vaccine recipients for prior sensitisation to Q fever antigens.

It is essential to test for sensitisation to Q fever antigens using Q-VAX® Skin Test in every individual prior to immunisation (see PRECAUTIONS).

Contraindications

Q-VAX® should not be administered to:
– Persons who have a history of Q fever.
– Persons who have been previously vaccinated with Q fever vaccine.

– Persons who have a history of likely exposure followed by an illness strongly suggestive of Q fever.
– Persons with positive serology for Q fever antibody or a positive Q fever skin test.
– Persons with known hypersensitivity to egg proteins or any component of the medicinal product.

Precautions

Prior to immunisation, all potential vaccinees must have a serum antibody estimation and a skin test reported; administration of Q-VAX® to those who are already sensitised to Q Fever antigens can cause serious hypersensitivity reactions.

As with other injectable vaccines, including Q-VAX® Skin Test solution, appropriate medical treatment and supervision should always be available in case of anaphylactic reactions. Adrenaline should always be readily available whenever the injection is given.

Q-VAX® should never be administered intravenously.

There is no information available on the efficacy and safety of Q-VAX® in immunodeficient or immunosuppressed individuals.

Those who have a confirmed positive antibody test or a positive skin reaction must not be given Q-VAX® (see PREVACCINATION TESTING).

If the skin test is negative or equivocal and antibodies are present at low titres (reported as a borderline laboratory test result), it cannot be concluded that the subject has adequate protective immunity against Q fever. The low-level presence of antibodies may be non-specific or due to technical factors of the assay. The risk-benefit decision of being vaccinated or not should be individually assessed and discussed with the subject, in order to decide whether potential adverse events following vaccination outweigh the potential risk to that subject from Q-Fever infection and its associated complications.

It should be noted that a very small number of people may have had Q fever in the past and yet show no response to serological or skin testing. Such persons may have severe reactions to Q-VAX®. For this reason, subjects should be carefully questioned regarding the possibility of previous exposure to Q fever and the duration of such exposure.

Workers who are at risk of contracting Q fever should be immunised prior to commencement of work or as soon as possible after they commence work as the risk of infection is highest in the first few years.

Vaccination during the incubation period of Q fever does not prevent the onset of the disease.

Despite the significant efficacy of Q-VAX® in clinical trials, cases of Q fever following vaccination have

been reported (see CLINICAL TRIALS).

Prevaccination testing

Serology: The presence of antibodies to Q fever may be demonstrated by using the complement fixation test (CFT). Subjects in whom antibodies are unequivocally positive should not be given QVAX® (see PRECAUTIONS).

Skin test

Preparation: Skin Test solution should be prepared by diluting 0.5 mL of the Q-VAX® Skin Test in 14.5 mL of Sodium Chloride Injection (to a final volume of 15 mL). The diluted Q-VAX® Skin Test should be freshly prepared, stored at 4°C and used within six hours.

Administration: The dose administered for skin testing is 0.1 mL of the diluted Q-VAX® Skin Test.

This should be injected intradermally into the volar surface of the mid-forearm.

A positive reaction is indicated by any induration at the site of injection read seven days after the test dose. Any person with a positive reaction must not be vaccinated.

Use in pregnancy (Category B2)

Safety of use in pregnancy has not been established. Deferral of vaccination is recommended.

Use in lactation

No information is available.

Use in children

No information is available.

Interaction with other drugs

No information is available.

Adverse effects

Vaccination of already immune subjects may result in severe local or general reactions, with the possibility of local abscess formation.

Clinical trial data

In a clinical trial in South Australia the following adverse events were recorded amongst 464 persons who received Q-VAX®.

Reaction	Frequency of vaccine reactions (%)
Local:	
– Tenderness	48
– Erythema	33
– Induration/oedema	< 1
Systemic:	
– Headache	9
– Fever	0.2

Q-VAX® Q Fever Vaccine & Q-VAX® Skin Test Product Information

There was a single case report of abscess formation at the injection site.

Post-marketing data

A range of adverse reactions has been reported with clinical use of Q-VAX®. The reactions are summarised below and categorised by frequency according to the following definitions. Very common: $\geq 1/10$; common: $<1/10$ and $\geq 1/100$; uncommon: $<1/100$ and $\geq 1/1000$; rare: $<1/1000$ and $\geq 1/10,000$ and very rare: $<1/10,000$.

Blood and Lymphatic System Disorders

Very rare: Lymphadenopathy

Nervous System Disorders

Common: Headache

Very rare: Dizziness

Gastrointestinal Disorders

Uncommon: Nausea, vomiting and diarrhoea

Skin and subcutaneous tissue Disorders

Common: Delayed skin reaction (presenting up to 6 months after vaccination) at injection site (either vaccination and/or skin test site)

Uncommon: Hyperhidrosis

Musculoskeletal and connective tissue Disorders

Uncommon: Myalgia

Very rare: Arthralgia

General disorders and administration site conditions

Very common: Injection site inflammation (e.g. erythema, pain, warmth and swelling).

Uncommon: Injection site induration and/or oedema, pyrexia, malaise, fatigue

Rare: Injection site abscess formation, granuloma

Very rare: Chills, chronic fatigue syndrome

Dosage and administration

Q-VAX® Vaccine:

Q-VAX® Vaccine should not be administered until the results of serology and skin testing are known (see **PRECAUTIONS**). Q-VAX® should be given only to those who have no demonstrable evidence of sensitisation to Q Fever antigens.

The dose of Q-VAX® Vaccine is 0.5 mL given by subcutaneous [NOT INTRAMUSCULAR] injection.

The container should be gently shaken before use.

The vaccine should never be administered intravenously.

No information is available on paediatric use.

Revaccination must never be undertaken due to the possibility of severe hypersensitivity reactions.

Q-VAX® Skin Test:

Preparation: Skin Test solution should be prepared by diluting 0.5 mL of the Q-VAX® Skin Test in 14.5 mL of Sodium Chloride Injection (to a final volume of 15 mL). The **diluted** Q-VAX® Skin Test should be freshly prepared, stored at 4°C and used within six hours.

Administration: The dose administered for skin testing is 0.1 mL of the **diluted** Q-VAX® Skin Test.

This should be injected intradermally into the volar surface of the mid-forearm.

Overdosage

No information is available on overdosage.

Presentation and storage conditions

Q-VAX® Vaccine is available as a pre-filled syringe containing ≥ 25 µg of antigen, in 0.5 mL solution.

Q-VAX® Skin Test is available as a pre-filled vial containing ≥ 2.5 µg of antigen, in 0.5 mL solution.

Q-VAX® Skin Test must be diluted prior to use in pre-vaccination screening (see PRECAUTIONS).

Q-VAX® Vaccine and Skin Test should be protected from light and stored at 2° – 8°C. DO NOT FREEZE.

The Q-VAX® Vaccine syringe is supplied encased within a clear film wrapper. The Q-VAX® Skin Test vial is packaged with a plastic tear away cap covering the vial septum. The presence of the film wrapper and plastic cap provides assurance that the product has not been opened. Do not use if the film wrap or tear away cap is damaged or missing.

Name and address of sponsor

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PARKVILLE VIC 3052
AUSTRALIA

Poisons schedule of the medicine

S4

Date of approval

Date of TGA approval: **9 July 1999**

Safety Related Change: 17 November 2008

Date of Most Recent Amendment: 18 December 2008

Q-VAX® is a Registered Trademark of CSL Limited.

This product is not listed on the PBS.