

An update on Q fever

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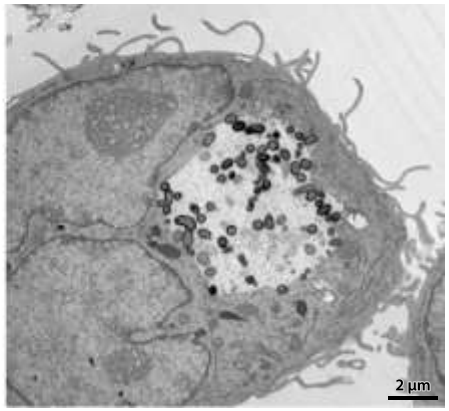
Overview

- History
- Microbiology
  - Axenic culture
  - Genomics
- Epidemiology
  - Endemic and outbreak patterns
  - Reservoirs
  - Transmission factors
- Laboratory diagnosis
- Clinical features
  - Acute and chronic infection
  - Post Q fever fatigue
- Pathophysiology
- Management
- Prevention

History

- 1935: *Davis* – isolation of filterable tick agent
- 1937: *Derrick* – 9 cases of febrile illness (5 meatworkers, 2 dairy farmers) – “Q (query) fever”
- 1937: *Burnet* – passage of organism to experimental animals - likely rickettsia
- 1938: *Cox*: “Nine Mile Agent” cultivated in embryonated eggs
- 1941: Dyer: *Rickettsia burnetii*

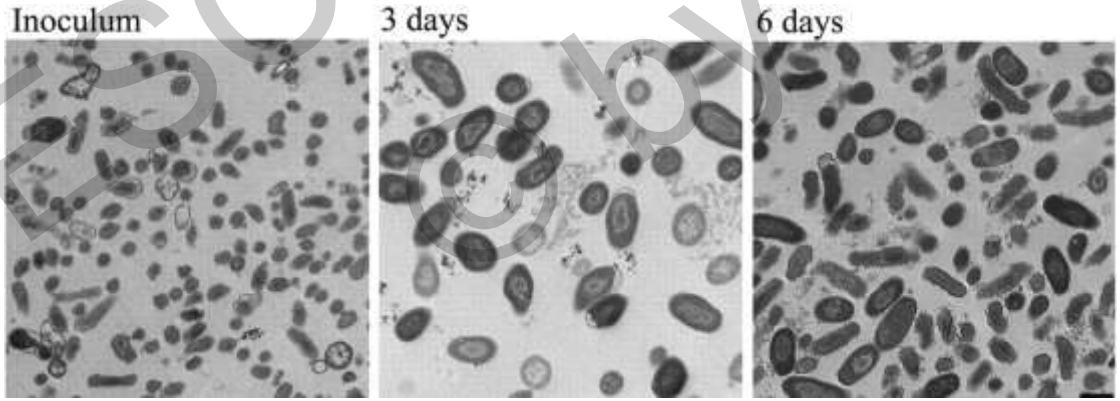


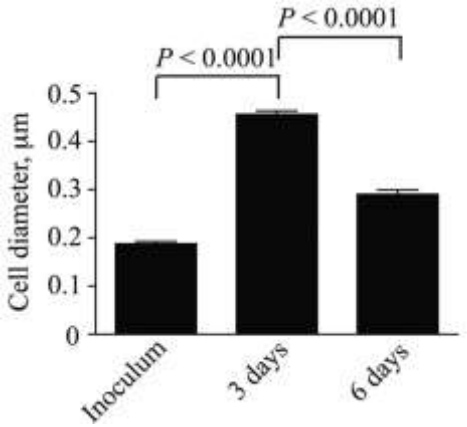
- 1990: *Shapiro:* Q fever vaccine
- 2003: *Seshadri*: Full genome sequence
- 2009: *Omsland*: Axenic (host cell free) culture

Derrick EH. “Q” fever, a new fever entity: clinical features, diagnosis and laboratory investigation. *Med J Aust* 1937; 2: 281-299.
Burnet FM, Freeman M. Experimental studies on the virus of “Q” fever. *Med J Aust* 1937; 2: 299-305.
McDade JE. Histoncal aspects of Q fever. In: Marrie T (ed) *Q Fever-The disease* Vol. I. CRC Press, Boca Raton Fl. 1990; 5: 21.
Shapiro RA, et al. A randomized, controlled, double-blind, cross-over, clinical trial of Q fever vaccine in selected Queensland abattoirs. Epidemiol Infect. 1990 Apr;104(2):267-73.
Seshadri R et al. Complete genome sequence of the Q-fever pathogen Coxiella burnetii. *Proc Natl Acad Sci U S A*. 2003 Apr 29;100(9):5455-60.
Omsland A, et al. Host cell-free growth of the Q fever bacterium Coxiella burnetii. *Proc Natl Acad Sci U S A.* 2009 Mar 17;106(11):4430-4.

Microbiology

- Obligate intracellular coccobacillus, *Gammaproteobacteria;* ~0.4 x 1µm, macrophage tropic
- Biphasic developmental cycle:
  - Large cell variant (LCV): metabolically active, exponential replication
  - Small cell variant (SCV): 0.2-0.5µm, condensed chromatin
    - environmentally stable (osmotic, mechanical, chemical, heat, desiccation)
    - upregulated genes of oxidative stress response, cell wall remodeling, and arginine acquisition
    - Heavily cross-linked peptidoglycans





Omsland A, et al. Host cell-free growth of the Q fever bacterium Coxiella burnetii. *Proc Natl Acad Sci U S A.* 2009 Mar 17;106(11):4430-4.

Microbiology

Axenic culture

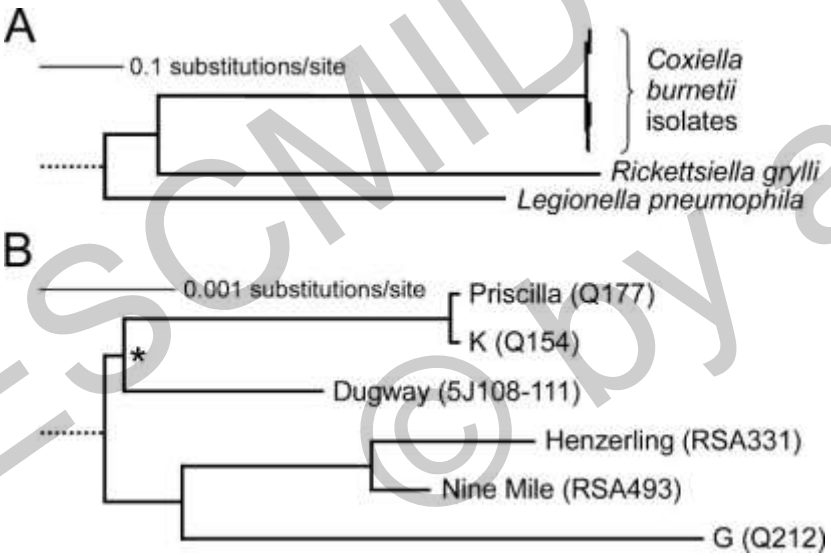
- Complex Coxiella Medium (CCM)
  - complex nutrient sources, chloride (140mM), and citrate (pH 4.75)
- In silico genomic analysis of metabolic deficiencies
- Acidified Citrate Cysteine Medium (ACCM)
  - Growth of *C. burnetii* in 2.5% O_2 to log 3
- ACCM2
  - methyl-cyclodextrin increased growth to log 5 (day 7)
  - isolation from animal tissue and genetic transformation was achieved with solid medium
- Genetic manipulation to inform host-pathogen interactions

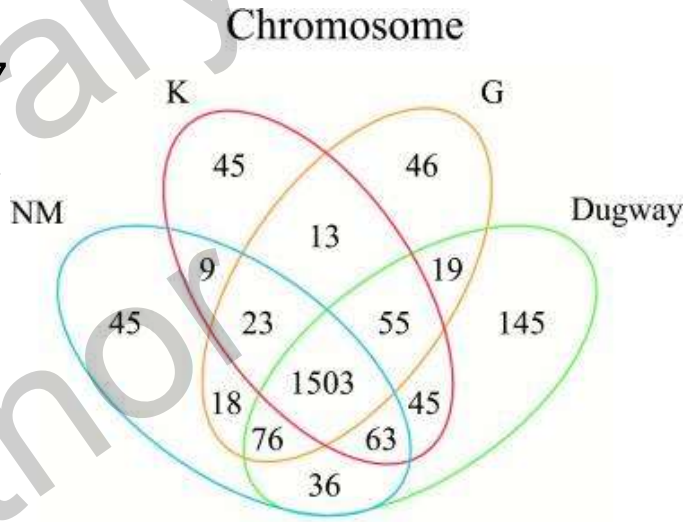
Omsland A et al Sustained axenic metabolic activity by the obligate intracellular bacterium Coxiella burnetii. *J Bacteriol* 2008. 190:3203–3212.
Omsland A, et al. Host cell-free growth of the Q fever bacterium Coxiella burnetii. *Proc Natl Acad Sci U S A.* 2009 Mar 17;106(11):4430-4
Omsland A et al. Isolation from animal tissue and genetic transformation of Coxiella burnetii are facilitated by an improved axenic growth medium. *Appl Environ Microbiol* 2011. 77:3720 –3725.
McClure EE, et al. Engineering of obligate intracellular bacteria: progress, challenges and paradigms. *Nat Rev Microbiol*. 2017 Sep;15(9):544-558.

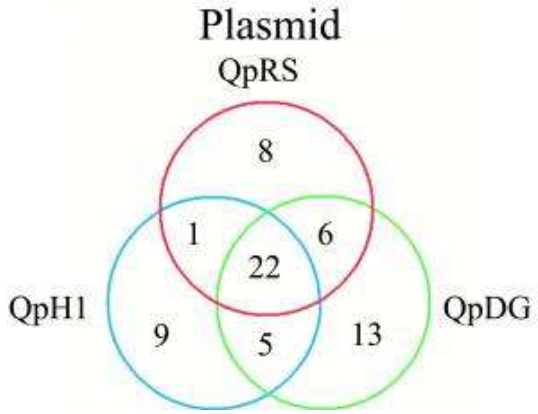
Microbiology

Genomics

- ~ 2Mb circular genome + QpH1 plasmid (37
- Closest human pathogen *L. pneumophila*
- ~ 2,000 ORFs, >100 unique
- Multiple pseudogenes







Seshadri R et al. Complete genome sequence of the Q-fever pathogen Coxiella burnetii. *Proc Natl Acad Sci U S A*. 2003 Apr 29;100(9):5455-60.
Beare PA et al. Comparative genomics reveal extensive transposon-mediated genomic plasticity and diversity among potential effector proteins within the genus Coxiella. *Infect Immun*. 2009 Feb;77(2):642-56.

Epidemiology

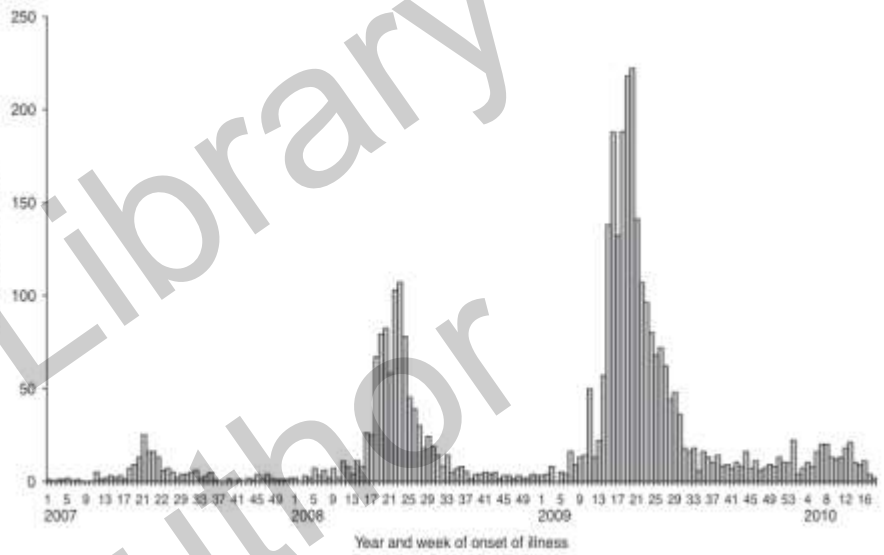
- Global distribution (except NZ, Samoa, French Polynesia)
- Endemic and outbreak patterns
- Highly under-recognised:
  - Generally non-notifiable
  - High sub-clinical: clinical ratio
  - Non-specific ‘flu-like’ illness manifestations
- Reservoirs
  - Goats, cattle, sheep (largely asymptomatic)
  - Native animals (e.g. kangaroos, bandicoots, three-toed sloths)
  - Ticks, amoebae
- Seroprevalence
  - Up to 25% in humans varying by country and rural region
  - 20-75% of domesticated livestock
  - Up to 25% of native species (possums, bandicoots, foxes, Spanish Ibex)

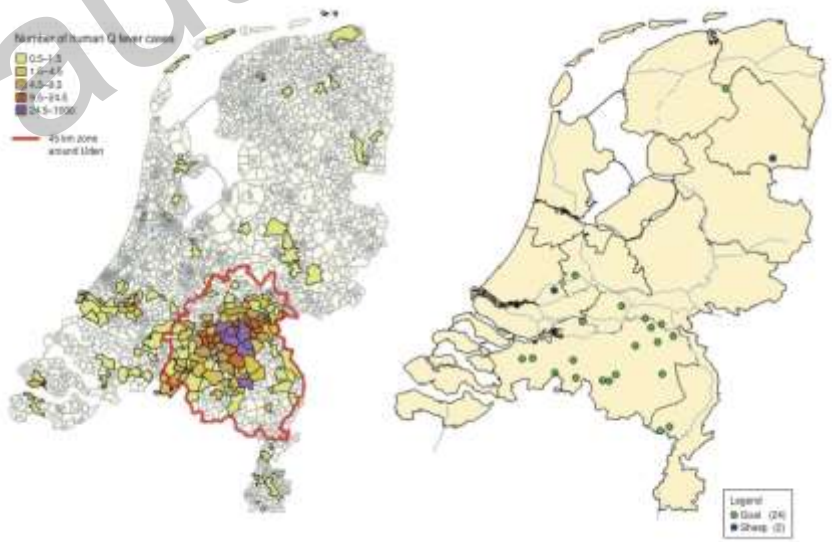
Eldin C et al. From Q Fever to Coxiella burnetii Infection: a paradigm change *Clin Microbiol Rev*. 2017 Jan;30(1):115-190.

Epidemiology

Transmission factors

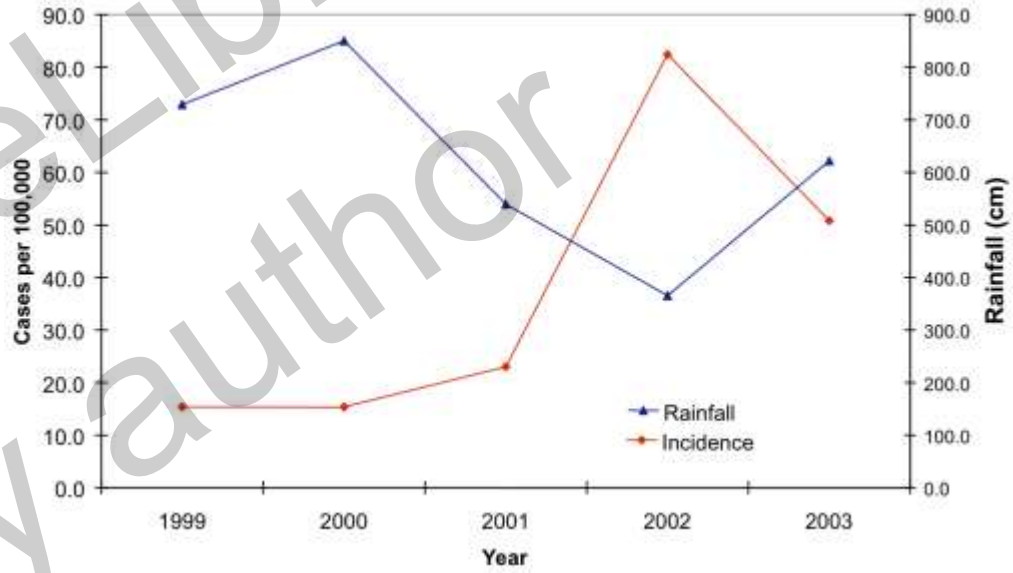
- Aerosol exposure to infected animals (placenta, abortion products, hides, wool, manure)
- Proximity to domesticated livestock
- Wind, season, and climate





Roest H et al. The Q fever epidemic in The Netherlands: history, onset, response and reflection. *Epidemiol Infect*. 2011 Jan;139(1):1-12.
Eldin C et al. From Q Fever to Coxiella burnetii Infection: a paradigm change *Clin Microbiol Rev*. 2017 Jan;30(1):115-190.

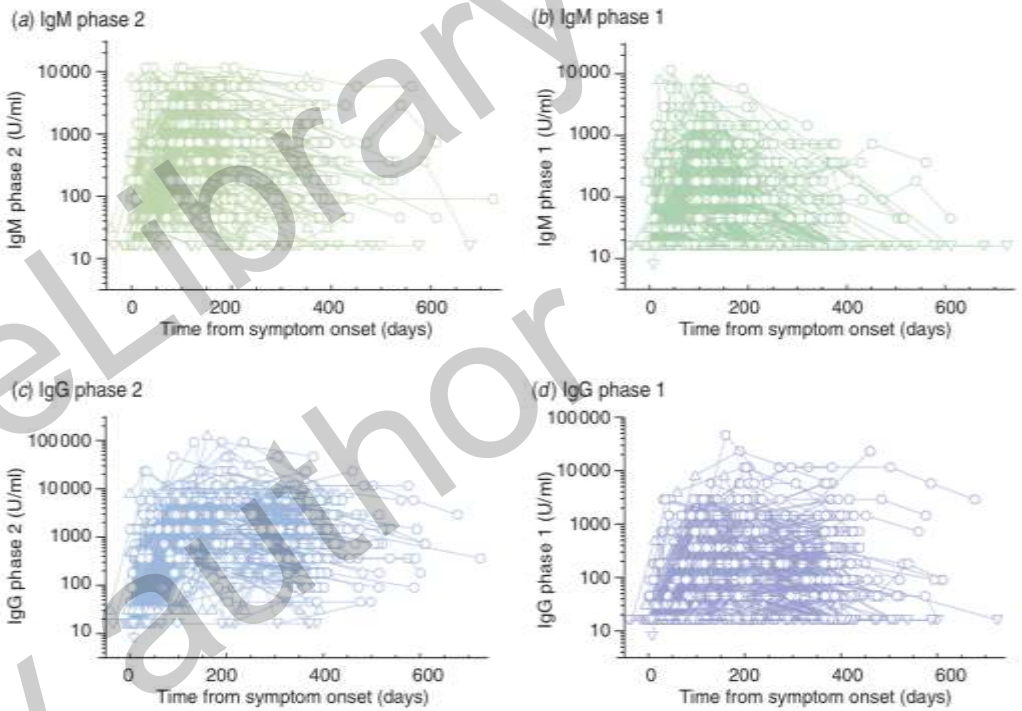
Epidemiology



Cameron B et al. Drought and Q Fever: The association between trends in the incidence of infection and rainfall in Rural Australia Ch 18 in T*he Principles and Practice of Q fever*. Simoes J, Anastacio SF, de Silva G (eds) Nova 2017

Laboratory diagnosis

- Serology
  - Phase I and II antigens
  - IFA, CFT and ELISA
  - Solitary IgM insufficient
- qPCR detection
  - IS1111 repetitive element
  - *ompA* gene
- Culture
  - BSL3
  - Shell vial
  - ACCM2
- Tissue
  - Immunohistochemistry
  - qPCR



Teunis PF et al. Time-course of antibody responses against Coxiella burnetii following acute Q fever. *Epidemiol Infect.* 2013 Jan;141(1):62-73.
Edlin C et al. From Q Fever to Coxiella burnetii Infection: a paradigm change *Clin Microbiol Rev*. 2017 Jan;30(1):115-190.

Clinical features

Acute Q fever

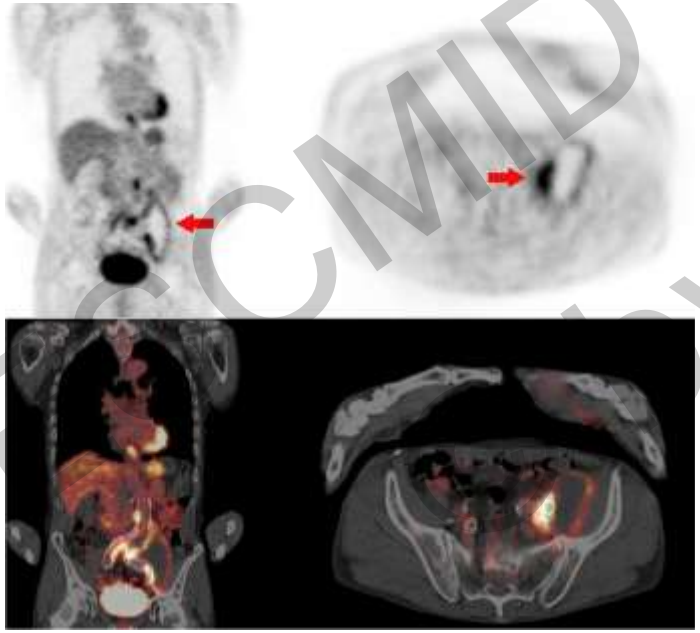
- Undifferentiated febrile illness (flu-like)
  - abrupt onset, high fever
  - myalgia, arthralgia, headache, drenching sweats
- Pneumonia
  - ‘atypical’ pattern – dry cough, headaches, biochemical hepatitis
  - relative leucopaenia, high CRP
  - varied Xray patterns
- Hepatitis
  - mixed picture LFT changes, anorexia
  - protracted course
- Other
  - carditis, meningo-encephalitis, lymphadenitis, etc

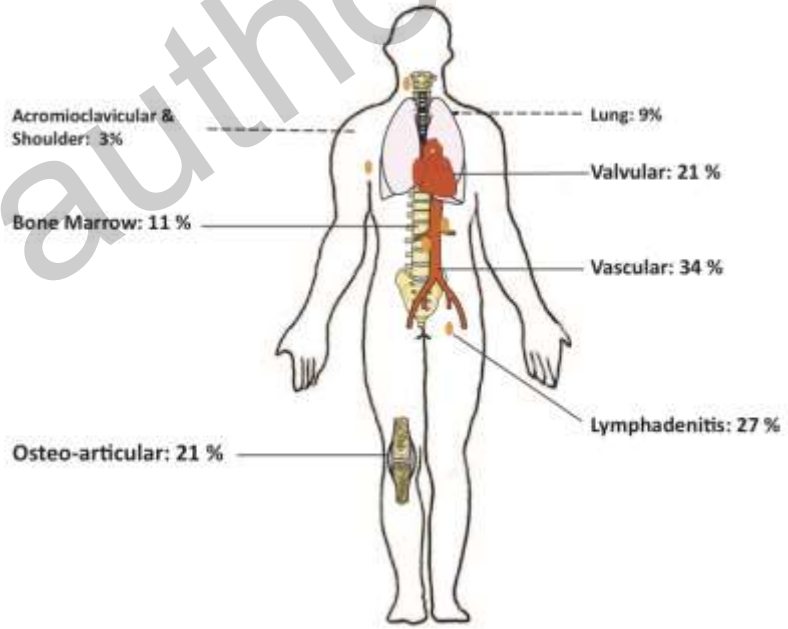
Edlin C et al. From Q Fever to Coxiella burnetii Infection: a paradigm change *Clin Microbiol Rev*. 2017 Jan;30(1):115-190.

Clinical features

Chronic localised (focal) Q fever

- endocarditis, osteomyelitis, hepatitis, other
- high titre >1:1024 phase I IgG
  - Modified Duke criteria for endocarditis
- PCR detection in blood or tissue
  - 18 FDG-PET/CT





Eldin C, et al.18F-FDG PET/CT as a central tool in the shift from chronic Q fever to Coxiella burnetii persistent focalized infection. *Medicine (Baltimore)*. 2016 Aug;95(34):e4287;

Barten DG, et al Localizing chronic Q fever: a challenging query. *BMC Infect Dis.* 2013 Sep 3;13:413..

Clinical features

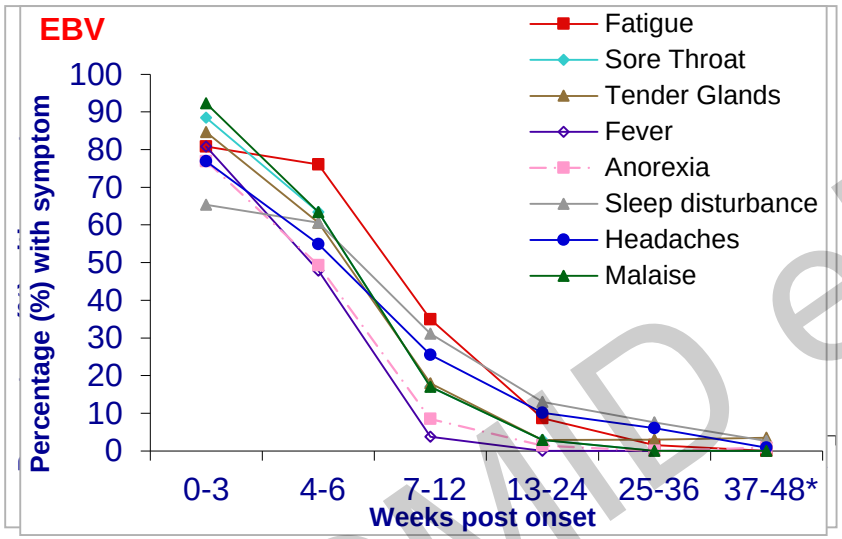
Post Q fever fatigue (QFS)

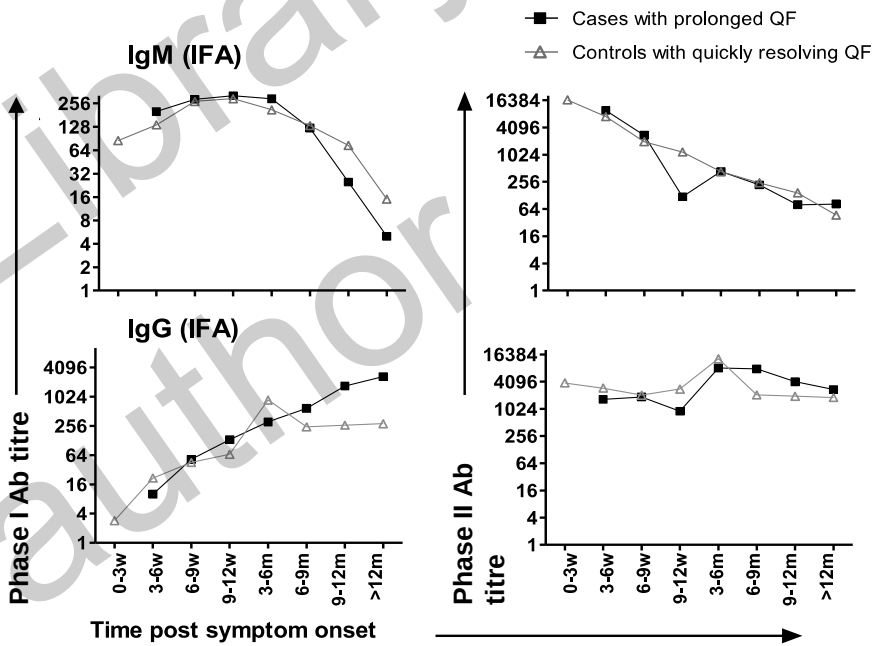
- Diagnostic criteria for chronic fatigue syndrome
  - persistent disabling fatigue (>6 months)
  - no alternative medical or psychiatric diagnosis (Hx, Exam, Ix)
- Chronic localised Q fever excluded
- No symptom-free interval from acute Q fever
- Concordant with other post-infective fatigue syndromes
  - Epstein-Barr virus, Ross River virus
- No abnormal persistence of the pathogen
- Unknown pathophysiology

Hopper B, et al. The natural history of acute Q fever: a prospective Australian cohort. QJM. 2016 Oct;109(10):661-668.
Hickie I, et al. Post-infective and chronic fatigue syndromes precipitated by viral and non-viral pathogens. BMJ. 2006 Sep 16;333(7568):575.

Clinical features

Post Q fever fatigue





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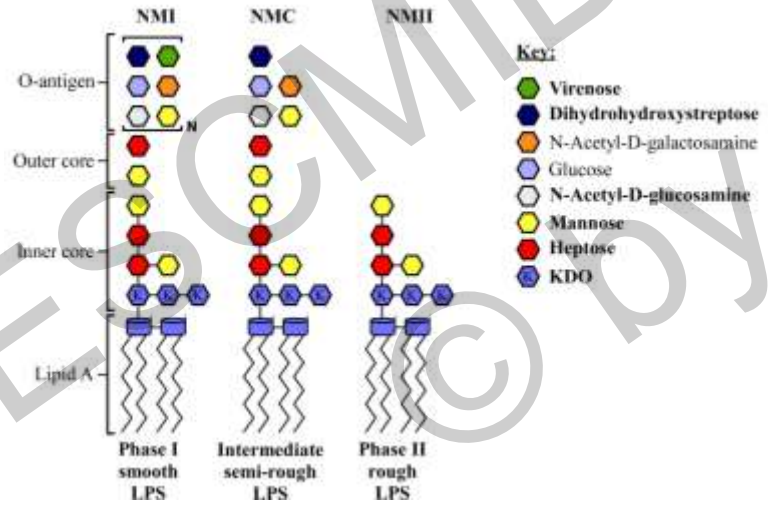
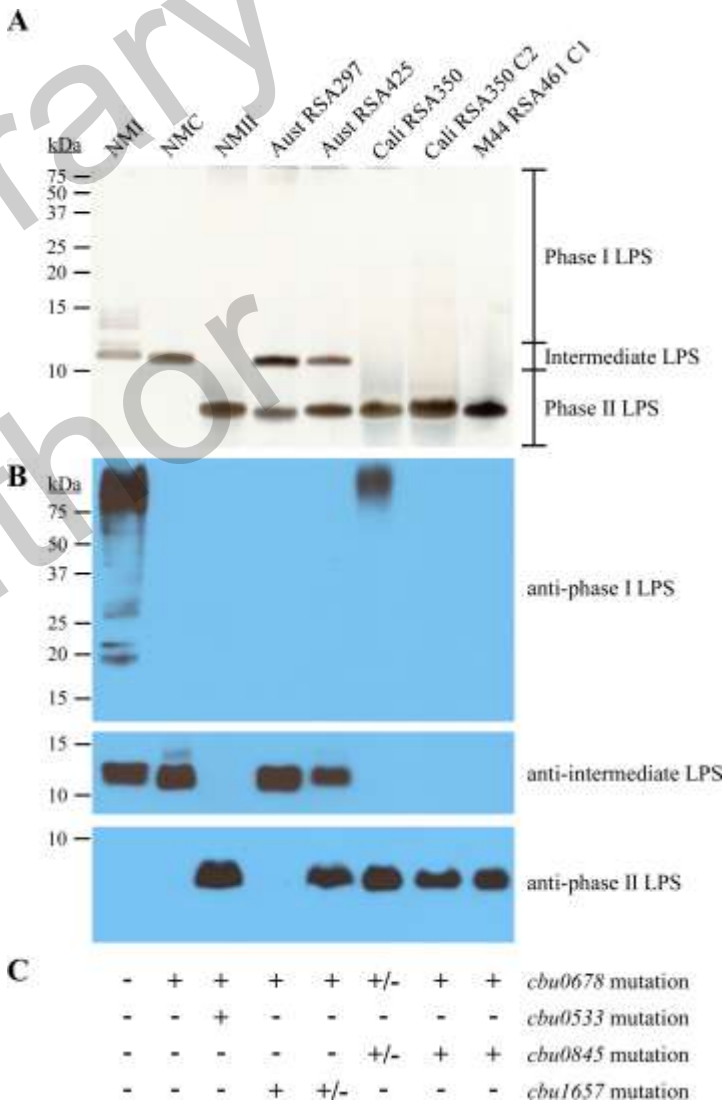
Hickie I, et al. Post-infective and chronic fatigue syndromes precipitated by viral and non-viral pathogens. BMJ. 2006 Sep 16;333(7568):575.

Pathophysiology

Acute Q fever – determinants of illness patterns

- Role of the organism – virulence factors
  - LPS (Phase I, Phase II)



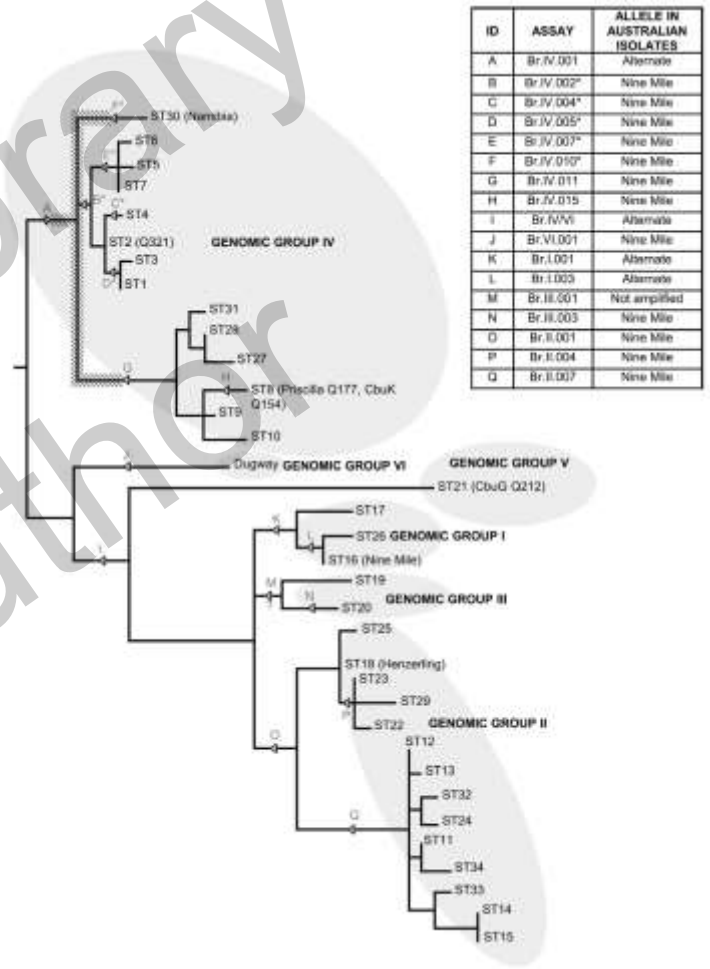


Beare PA, et al. Genetic mechanisms of *Coxiella burnetii* lipopolysaccharide phase variation. *PLoS Pathog* 2018; 14(3): e1006922.

Pathophysiology

Acute Q fever – determinants of illness patterns

- Role of the organism – virulence factors
  - RFLP and PFGE typing: six genomic groups
    - Associations with acute (I, II, III) or chronic (IV, V) disease
  - MST and MLVA
  - SNP analysis



ID	ASSAY	ALLELE IN AUSTRALIAN ISOLATES
A	Br.IV.001	Alternate
B	Br.IV.002*	Nine Mile
C	Br.IV.004*	Nine Mile
D	Br.IV.005*	Nine Mile
E	Br.IV.007*	Nine Mile
F	Br.IV.010*	Nine Mile
G	Br.IV.011	Nine Mile
H	Br.IV.015	Nine Mile
I	Br.IV/VI	Alternate
J	Br.VI.001	Nine Mile
K	Br.I.001	Alternate
L	Br.I.003	Alternate
M	Br.III.001	Not amplified
N	Br.III.003	Nine Mile
O	Br.II.001	Nine Mile
P	Br.II.004	Nine Mile
Q	Br.II.007	Nine Mile

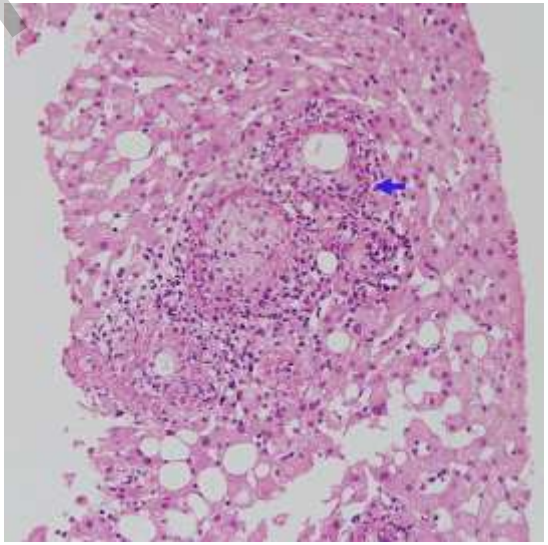
Beare PA et al. Genetic diversity of the Q fever agent, Coxiella burnetii, assessed by microarray-based whole-genome comparisons. *J Bacteriol.* 2006 Apr;188(7):2309-24.

Vincent G et al. Novel genotypes of Coxiella burnetii identified in isolates from Australian Q fever patients. *Int J Med Microbiol*. 2016 Sep;306(6):463-70

Pathophysiology

Acute and chronic localised Q fever – determinants of illness patterns

- Role of the host
  - High subclinical: clinical ratio in acute infection
  - Older age (adults vs children), male gender (2.5:1), pregnancy
  - Pre-existing valvular lesions (endocarditis) – esp. bicuspid aortic
  - Cellular immunity
    - Pro- and anti-inflammatory cytokines (IFN-γ, IL-12, IL-10)
    - M1 (pro-inflammatory) / M2 macrophage polarisation
    - Sex genes

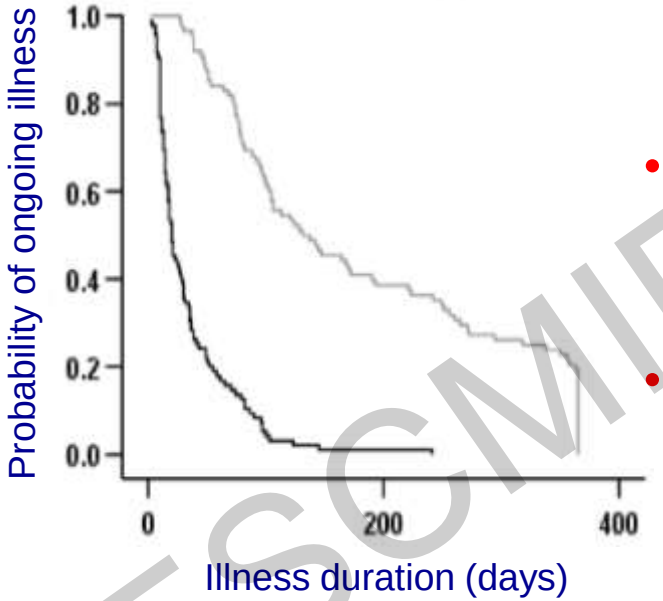


Capo C, Mege JL. Role of innate and adaptive immunity in the control of Q fever. Adv Exp Med Biol. 2012;984:273-86..
Edlin C et al. From Q Fever to Coxiella burnetii Infection: a paradigm change *Clin Microbiol Rev*. 2017 Jan;30(1):115-190.

Pathophysiology

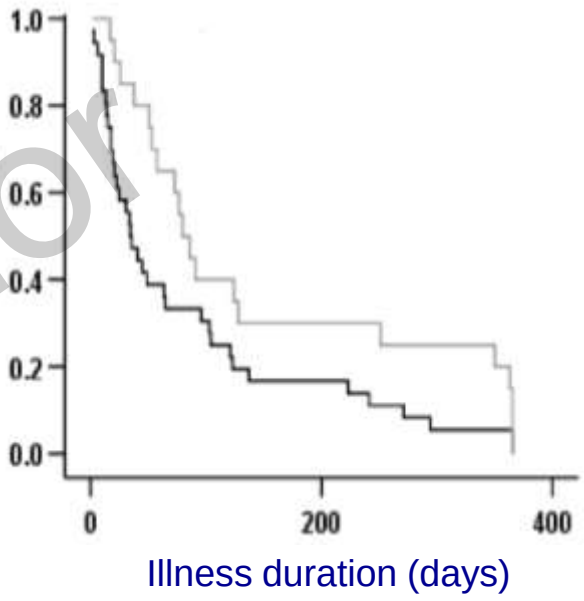
Post Q fever fatigue - determinants of illness patterns

Acute illness severity



- *High* severity: IFN-γ +874 T/A
  - OR 2.9; *p*=0.004
- *Low* severity: IL-10 -592 C/A
  - OR 1.9; *p*=0.03)
- High severity: combined IFN-γ / IL-10
  - OR 6.; *p*=0.001

High risk IFN-γ / IL-10 genotype



Vollmer-Conna U, et al Cytokine polymorphisms have a synergistic effect on severity of the acute sickness response to infection. *Clin Infect Dis.* 2008 Dec 1;47(11):1418-25; Piraino B et al. Genetic associations of fatigue and other symptom domains of the acute sickness response to infection. *Brain Behav Immun*. 2012 May;26(4):552-8

Management

Acute Q fever

- No randomised controlled trials
- Early (<3d) doxycycline (200mg) x 14 days shortened fever duration
  - fluoroquinolones, macrolides, co-trimoxazole as alternatives
- Screening for risk factors for chronic localised infection?
  - TTE, anti-cardiolipin, FDG-PET/CT
- Follow-up of uncomplicated infection?

Chronic localised Q fever

- No randomised controlled trials
- Doxycycline (200mg) + hydroxychloroquine >18 months
- Follow-up – serology, imaging (TOE, FDG-PET/CT)

Post Q fever fatigue

- Qure study: CBT effective / doxycycline ineffective

Million M et al. Evolution from acute Q fever to endocarditis is associated with underlying valvulopathy and age and can be prevented by prolonged antibiotic treatment. *Clin Infect Dis*. 2013 Sep;57(6):836-44.

Raoult D, et al Treatment of Q fever endocarditis: comparison of 2 regimens containing doxycycline and ofloxacin or hydroxychloroquine. *Arch Intern Med.* 1999;159(2):167–173;

Keijmel SP et al Effectiveness of long-term doxycycline treatment and cognitive-behavioral therapy on fatigue severity in patients with Q fever fatigue dyndrome (Qure Study): A randomized controlled trial. Clin Infect Dis. 2017 Apr 15;64(8):998-1005

Prevention

- No proven effective personal protective equipment
  - N95 mask, BSL3
- Immunisation
  - Qvax™ (CSL Ltd, Australia)
  - Formalin-inactivated Henzerling strain, Phase 1
  - Pre-vaccination screening – serology and DTH skin testing

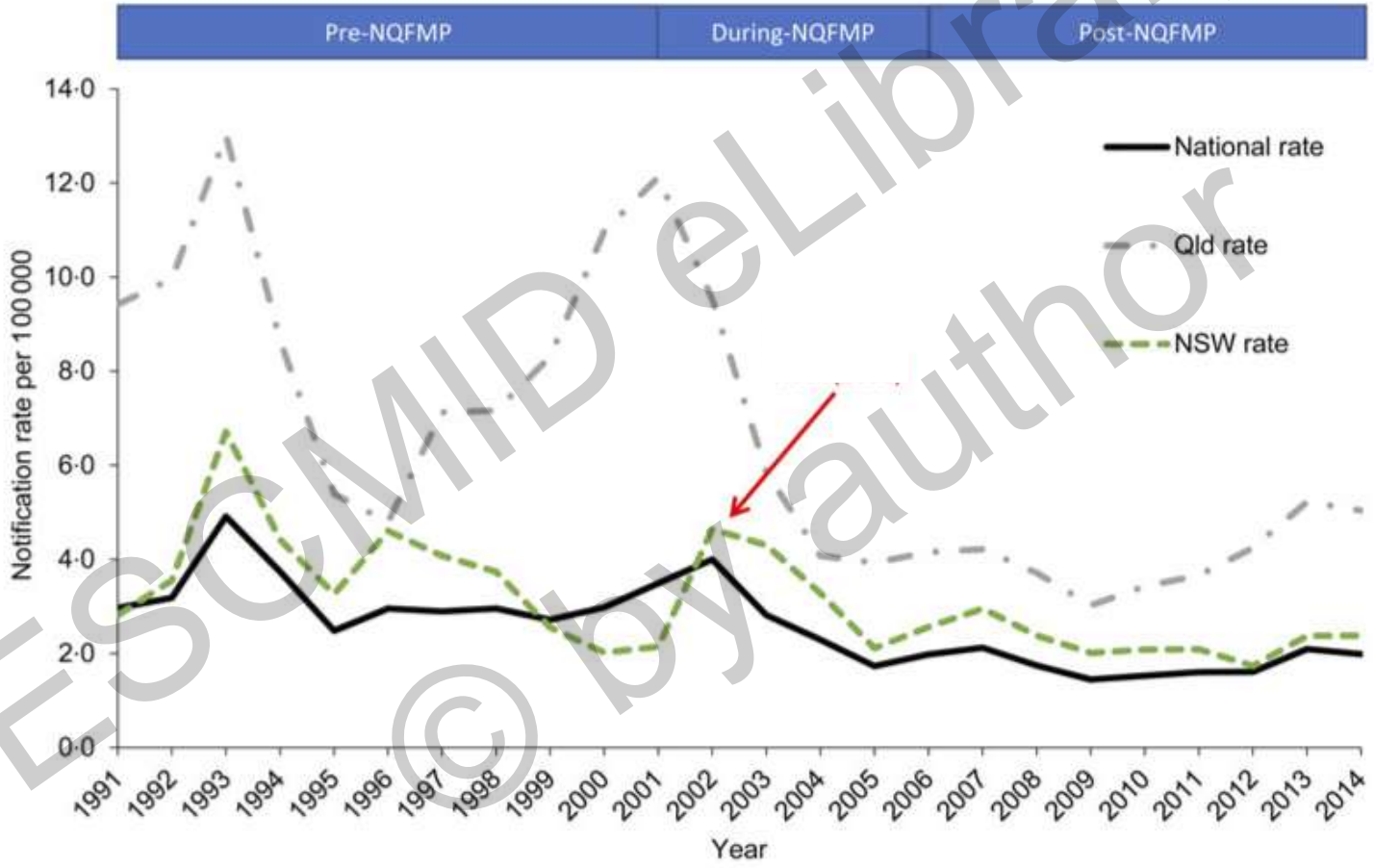
Abattoir	Q fever vaccine (15)		Influenza vaccine (01)		Unvaccinated cases*
	Vaccinees	Cases	Vaccinees	Cases	
Kilcoy	41	0	30	5	8
Beenleigh	21	0	28	0	2
Regional Metropolitan	36	0	44	2	5
Total	98	0	102	7	15

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

Shapiro RA et al A randomized, controlled, double-blind, cross-over, clinical trial of Q fever vaccine in selected Queensland abattoirs. Epidemiol Infect. 1990 Apr;104(2):267-73.

Prevention

Australian national targeted immunisation – NQFMP (2002-6)



Sloan-Gardner TS et al. Trends and risk factors for human Q fever in Australia, 1991-2014. Epidemiol Infect. 2017 Mar;145(4):787-795.
Sellens E et al. Q Fever Knowledge, Attitudes and Vaccination Status of Australia's Veterinary Workforce in 2014. PLoS One. 2016 Jan 12;11(1):e0146819.

Conclusions

What is known:

- Q fever is an emerging zoonotic infection worldwide
- Vaccine preventable

What is unknown, but needs further investigation:

- Global burden of disease
- Pathogen virulence determinants
- Host determinants of illness severity and course
- Standardisation of diagnostics
- Randomised trials of treatments
- Improved vaccine

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- Beth Hopper
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