FS101016: Q fever risk to human health from the consumption of contaminated unpasteurised milk and milk products

Deliverable D03/01. Risk pathways and data requirements for *C. burnetii* infection in humans due to consumption of raw milk and cheeses made from raw milk

Original submitted: 29th April 2013
Revised: 17th June 2013

Prepared for the Food Standards Agency by Paul Gale & Emma Snary

Risk Analysis Workgroup
Epidemiology, Surveillance and Risk Group
Animal Health and Veterinary Laboratories Agency

Contents

1 Introduction

Q fever is a zoonotic widespread disease caused by the bacterium *Coxiella burnetii* and is present in cattle, sheep and goats. *It* is an emerging disease that can cause considerable morbidity and serious long-term complications in humans. The main route of transmission to humans is via the inhalation of aerosols from the parturient (birth) products of infected animals. It is known that meat, milk and milk products can be contaminated, however, the link to clinical disease in humans is unclear (EFSA, 2010). Risk assessment is an important tool in assessing the risk posed by food borne pathogens and contaminants. Therefore within project FS101016, risk pathways are being developed to assess the risk to human health from consumption of unpasteurised milk and cheese contaminated with *C. burnetii* in the UK. The agreed risk question is:

What is the probability of human infection with Q fever due to the consumption of a serving of unpasteurised milk or milk products?

Within this report, the risk pathways are presented for *C. burnetii* in unpasteurised goat and cows' milk and also cheese (hard and soft) produced from goat and cows' milk. Risk pathways identify the steps that need to happen for a certain event to occur and, for this situation, will consider the entire farm-to-consumption pathway. Using the risk pathway as guidance, a table of data requirements for the various model inputs required to quantitatively assess the risk of human Q fever infection has been generated. Using the data collected within the Hazard Identification and Risk Profile, a brief review of the available data has been given for each model input. A traffic light system has been adopted to identify the quantity and quality of the published data available to populate the risk pathway; Red – No data, Amber – Some data, but deficiencies, Green – good data. Specific industry data relating to the processing of cheese are coloured blue as it will be highly dependent on the type of cheese being manufactured.

Three pathways have been presented here:

(1) Risk pathway for the probability and levels of *C. burnetii* in Bulk Tank Milk (BTM) per herd;

(2) Risk pathway for the probability of human infection with *C. burnetii* due to the consumption of raw milk;

(3) Risk pathway for the probability of human infection with *C. burnetii* due to the consumption of cheese made from raw milk.

The pathways are generic and can be parameterised for cow's or goats' milk/cheese. Although given separately, the pathways are inter-connectable so the entire farm-to-consumption pathway is described. Thus for the full farm-to-consumption pathway for raw milk pathways (1) and (2) are combined and for raw cheese, pathways (1), (2) and (3) are combined.

2 Pathway and data requirements for occurrence of *Coxiella burnetii* in bulk tank milk at the herd level

The risk pathway for numbers of *C. burnetii* in bulk tank milk (BTM) at the level of the individual herd is shown inFigure 1. The pathway is generic, i.e. applicable to both cattle and goat herds, but would need to be parameterised differently for the two livestock species. The data available for cattle and goat farms are set out in Table 1. Survival of the *C. burnetii* is not considered within this pathway because decay is not likely over the short timescales on the farm and growth will not occur. The probability an animal is lactating on a given day reflects the number of days per year that individuals are milked.

From Table 1 it can be seen that there are data gaps and data deficiencies associated with the microbiological data for *C. burnetii*. In comparison, the availability of data for herd management (e.g. herd size, volume of milk produced) is better although some of these parameters are still reliant on expert opinion.

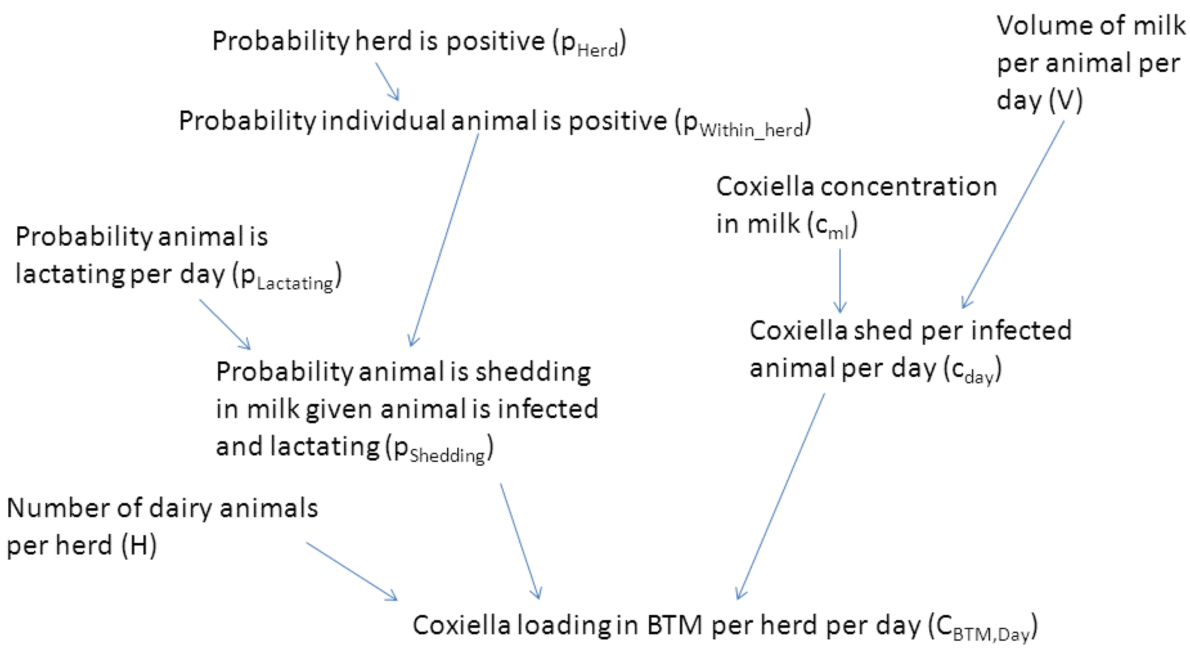

Figure 1: Schematic diagram for the probability and levels of *C. burnetii* in BTM per cattle/goat herd

Table 1: Data requirements and availability for estimating probability and levels of *C. burnetii* in BTM (per herd).

Description	Parameter	Cattle	Goats	Justification for grading
Probability herd is positive	P_{Herd}	• 0.21 to 0.484 (McCaughey et al 2010, Paiba et al. 1999). • 0.697 for herds in south-west England (Valergakis et al. 2012)	• 0.0297 (Lambton et al. (unpublished))	Seroprevalence data only
Probability animal is positive given herd is positive	P_{Within_Herd}	• 0.089 (Spain) (Astobiza et al. (2012)) • 0.389 (France) (Rodolakis et al. 2007)	• 0.299 (UK) (Lambton et al. (unpublished)) • 0.308 (France) (Rodolakis et al. 2007)	Not UK data Seroprevalence data only.
Probability animal is lactating[1]	$P_{Lactating}$	• 0.85 (UK Agriculture, 2013). • Range: 0.73-0.95. Most likely 0.83. (South Africa) (ARC 2013)[2]	• 0.83 (US) (Dairy Goat Journal 2012)[3]. • Mean 0.825. range 0.8-0.85 (Adkin et al. 2010[4])	
Probability animal is shedding *C. burnetii* in milk given animal is lactating and infected	$P_{Shedding}$	• 0.92 (Rodolakis et al. 2007)	• 0.73 (Rodolakis et al. 2007); • 0.31 to 0.38 (Rousset et al. 2009)	Detected by PCR. Viability not addressed.
Volume of milk (per animal per day)	V	• 23.5- 27.5[5] litres/day (DairyCo, 2013) for a 305 day cycle.	• 6-12lb/day (US) (Richardson 2013) • 6-8lb/day (US) Dairy Goat Journal 2012 • 2 – 2.2 litres (UK) (Adkin *et al.* 2010[4])	
C. burnetii concentration in milk	c_{ml}	• Mean, 620, Max 10,000 guinea pig ID_{50}/ml (Enright et al. 1957). PCR data for *C. burnetii* DNA/ml (Guatteo *et al* 2007; Valergakis et al. 2012)		Units for bovine milk are guinea pig ID_{50} and therefore requires dose-response data for humans in same units. Viability of PCR data is not known. Multiple copy numbers.
Number of dairy animals per	H	• Average herd sizes: England	• Mean 683, range 1-4090	

herd		– 77; Wales – 80; Scotland – 120; N. Ireland – 80.(EBLEX 2012)[6] • Data from FSA for 81 herds registered as unpasteurised milk producers: Average = 88 (range 1-300)	(Sheep and Goat Inventory, 2009) • Data from FSA for 25 herds registered as unpasteurised milk producers: Average = 46 (range 2-500)	

[1] Most dairy herds are all year round calving to ensure continuity of milk supply to market but there will be variation and some herds may be batch calving for heifers entering the herd. The assumption is the all year round calving pattern.

[2] The lactation period is usually 300 to 305 days (43 weeks) with limits of 265 to 340 days (38 to 49 weeks).

[3] The average dairy goat has a 10-month lactation period.

[4] Industry opinion from dairying goat owners collected by telephone questionnaire in 2009.

[5] Raw-Milk-Facts (2013) state that Holstein-Friesian produce 12,700 kg per cycle (305 days) –i.e. 41.6 kg/day. However it is considered that most herds are not achieving this.

[6] Unpublished data are also available from Defra funded project OD2031.

3 Pathway and data requirements for *Coxiella burnetii* infection due to the consumption of raw milk.

The risk pathway for infection with *C. burnetii* due to the consumption of raw milk is shown in Figure 2. It considers both farm gate sales of unpasteurised milk and also the selling of milk at farmers' markets and by other distributors. The pathway also links to the risk pathway for cheese (see Figure 4). As before, the pathway is generic, i.e. applicable to both cows' and goats' milk, however a number of parameters are still specific to the livestock type, e.g. consumption patterns. For sheep, goat and buffalo raw milk the regulations are less strict compared to cows' milk (FSA, 2009). In particular, sheep, goat and buffalo milk can be sold indirectly to the consumer. However, due to the small-scale nature of this production and therefore its artisan character it is assumed that the production/packaging and retail is still mostly carried out by the farmer. Tanker collections of milk, as might be the situation for pasteurised milk, are not considered. This assumption was verified by the discussions with the Specialist Cheesemakers Association, who have contact with raw milk producers.

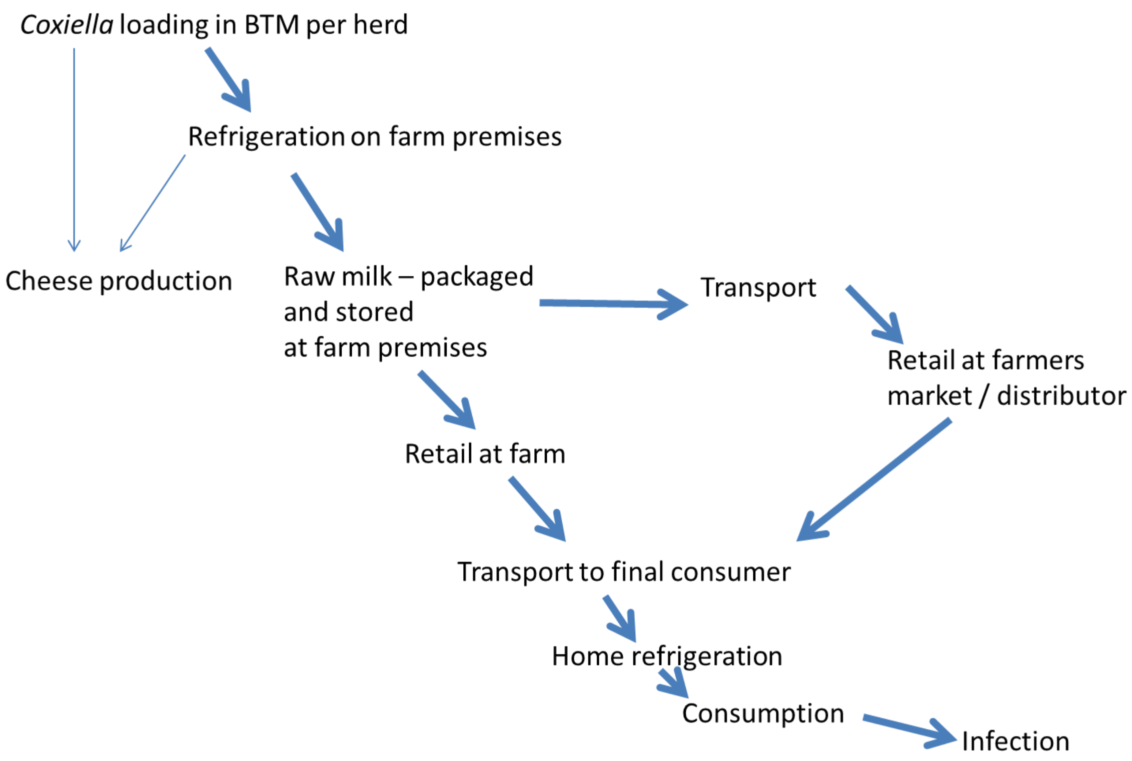

Figure 2: Generic risk pathway for human *C. burnetii* infection due to the consumption of unpasteurised milk. Each stage is broken down into input parameters in Table 2.

Table 2 lists the data that would be required for the development of risk assessment using the pathway given in Figure 2. Detailed data on the temperatures have not been obtained from the industry because they are unlikely to be of importance for *C. burnetii* which, being an obligate intracellular pathogen, will not grow outside the host cell. Furthermore there are no data on how temperature affects survival although lower temperatures typically promote survival of microorganisms. Significant data gaps are (1) lack of data on the survival/decay of *C. burnetii* in milk and (2) lack of data for the human dose-response of *C. burnetii* in milk.

Table 2: Data requirements and availability for estimating probability of *C. burnetii* infection due to the consumption of a serving of raw milk

Parameter	Notation	Data	Justification for grading
Proportion of herds that process cheese from non-refrigerated milk.	$\rho_{non\text{-}chilled,}$	10% of raw cheese makers (Specialist Cheesemakers Association) [1]	Expert opinion
Proportion of herds that sell raw milk at the farm gate.	$\rho_{off\text{-}farm,m}$	• FSA study reported that 65% (from 62 farms) sell milk/cream from farm gate. • Goats: 10% of herds process their milk on-farm (Adkin *et al.* 2010[2]) but will also include farms that produce pasteurised milk.	Expert opinion. Very limited number of farmers.
Proportion of herds that process cheese on the farm.	$\rho_{off\text{-}farm,c}$	Maximum of 5% (Specialist Cheesemakers Association)	Expert opinion.
Time and temperature decay rate for *C. burnetii* in milk	D(t, T)	No data[3]	
Refrigeration on farm in BTM			
Time	$t_{ref,f1}$	If to be used for cheese making, hauliers collect milk either every day, every other day or, rarely, every 3 days (Specialist Cheesemakers Association). For raw milk, storage time in the BTM is likely to be minimal as limited shelf life.	Expert opinion
Temperature	$T_{ref,f1}$	<8°C if collected the same day as milking. If	

		collected less frequently <6°C (Anon 2013a) 1-4°C (Specialist Cheesemakers Association) [4]	Very limited number of farms.
Bottling and storage of milk at farm			
Time	$t_{BS,f}$	To be collected from industry	Likely to be minimal.
Temperature	$T_{BS,f}$	<5°C (Specialist Cheesemakers Association) [4]	Very limited number of farms.
Transport conditions (farm - retail)			
Time	$t_{BTM,d}$	No data	
Temperature	$T_{BTM,d}$	Recommended <10°C at arrival to dairy (Anon 2013a)	
Retail conditions (farm, farmers market, distributor)			
Time	t_{retail}	Maximum time will be dependent on the use-by date. Shelf life is 3 days (Specialist Cheesemakers Association)[4].	Very limited number of farms.
Temperature	T_{retail}	• Backroom coldstore: Mean 3.3°C, maximum 16.6°C. Retail display: Mean 4.0°C, maximum 14.4°C. (FAO 2002) • <5°C on/off farm (Specialist Cheesemakers Association) [4].	Data are old (1999) and for the USA. Full data set not given. Very limited number of farms.
Transport conditions (retail – home)			
Time	$t_{r\text{-}h}$	• Mean 65 min, maximum >120 min (FAO 2002). • Local buyers more likely so limited travel time (Specialist Cheesemakers Association) [4].	Data are old (1999) and for the USA. Full data set not given. Very limited number of farms.
Temperature	$T_{r\text{-}h}$	• Mean 10.3°C, maximum 36.6 (FAO 2002). • Local buyers more likely so	Data are old (1999) and for the USA. Full data set not given. Very limited number of farms.

		temperature may not change greatly (Specialist Cheesemakers Association) [4].	
Home refrigeration			
Time	$t_{home,c}$	• Will be dependent shelf life, which is assumed to be 3 days (Specialist Cheesemakers Association) [4].	Very limited number of farms.
Temperature	$T_{home,c}$	Mean 4.0°C, maximum 21.1°C (FAO 2002)	Data are old (1999) and for the USA. Full data set not given.
Serving size of raw milk	S_{milk}	Mean of 127 g/person/day[5]	
Probability of infection from contaminated raw milk(dose-response)	P_{inf}	No data	

[1] Milk from the morning milking may be used immediately, i.e. without chilling, to produce cheese. It may be combined with refrigerated milk from the previous evening.
[2] Industry opinion from dairying goat owners collected by telephone questionnaire in 2009.
[3]The time between milking and consumption of the milk is relatively short. Since the milk is refrigerated for much of that time it is unlikely there will be any decay of the *C. burnetii*. Since *C. burnetii* survived in milk (dried 37°C) for 30 – 60 days (Babudieri and Moscovici 1950) and survived in sterile milk at room temperature for 125 days (Zubkova 1957). However, these data are not sufficient for risk assessment purposes. It is assumed that there will be no decay in refrigerated raw milk over a couple of days.
[4] Enquiries kindly made by the Specialist Cheesemakers Association (3 farms).
[5]Department of Health (2011) Whole milk consumption for consumers of 19 to 64 years. Not specified whether raw or pasteurized, or hot/cold at point of consumption

4 Pathway and data requirements for *Coxiella burnetii* infection due to the consumption of raw cheese.

The process of cheese-making is set out in Figure 3 and has been split into soft and hard cheeses[1]. The data associated with the cheese industry are highly specific to the type of cheese being produced and therefore, where available, broad ranges have been identified.

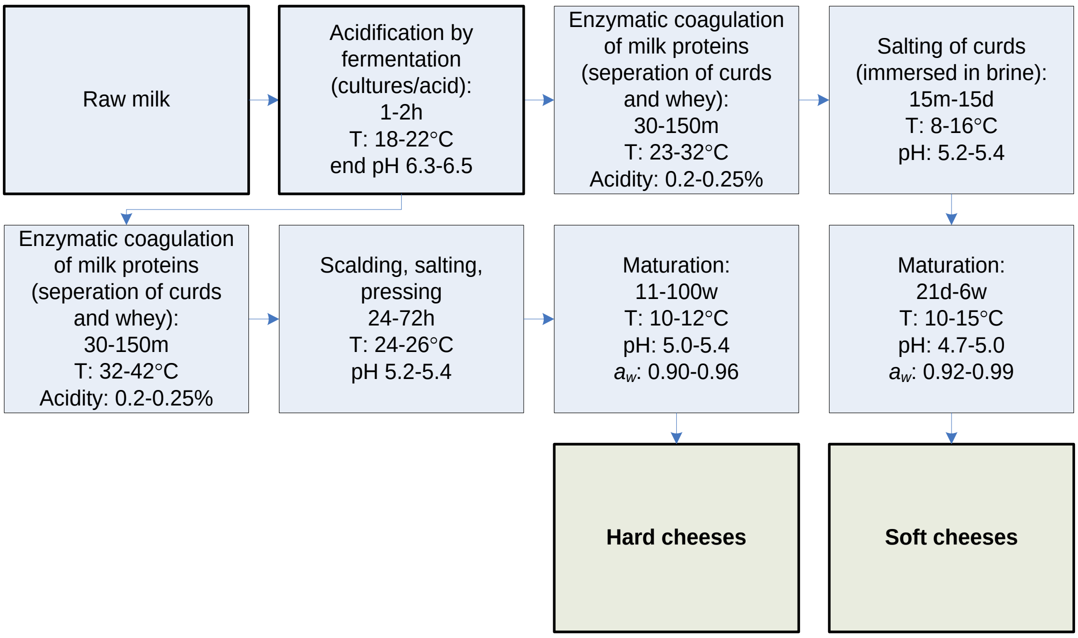

Figure 3: The process of cheese-making)

The risk pathway from raw milk to cheese includes these steps and is set out in Figure 4. The likely factors to consider in manufacture of different cheeses, with respect to bacterial pathogens such as *Escherichia coli* O157 and *Listeria monocytogenes* include the rate of acid development, titratable acidity of the fresh curd, salt concentration, pH changes during maturation and maturation time (Banks 2006; FSA 2013). The data requirements and availability for the risk pathway are set out in Table 3.

A potentially important factor not considered by Banks (2006) is that 95% of the water content and hence water-soluble components is removed as whey, while 95% of the fat content and other proteins is coagulated into the curds (Anon 2013b). Thus it could be that either 95% of the *C. burnetii* is lost with the whey, or alternatively that 95% of the *C. burnetii* precipitates into the curds which go on to become cheese. This is a major data gap. However, evidence (Enright 1961) that fat content affects sensitivity to pasteurisation in some milk products suggests an association between *C. burnetii* and fats. Furthermore

[1] Figure 3 has been discussed and reviewed by members of the Specialist Cheesemakers Association.

the amount of whey is to some extent affected by salt content and impacts on moisture content of the cheese which differ between soft and hard cheeses.

As described earlier, the industry data relating to the processing of cheese are coloured blue as they will be highly dependent on the type of cheese being manufactured. The data provided are thought to be generic, but there are a range of parameters used in cheese making and it is impossible to ensure that the values suggested here encompass all possible variations.

Similar to the raw milk risk pathway there are no data for the decay of *C. burnetii* during cheese making conditions (i.e. for different time, temperature and pH/acidity combinations) and no data on the dose-response for *C. burnetii* in cheese. However, it should be noted that unlike for raw milks (Loftis et al. 2010), viable *C. burnetii* have not been isolated from PCR-positive cheese samples (Hirai et al 2012; Eldin et al. 2013).

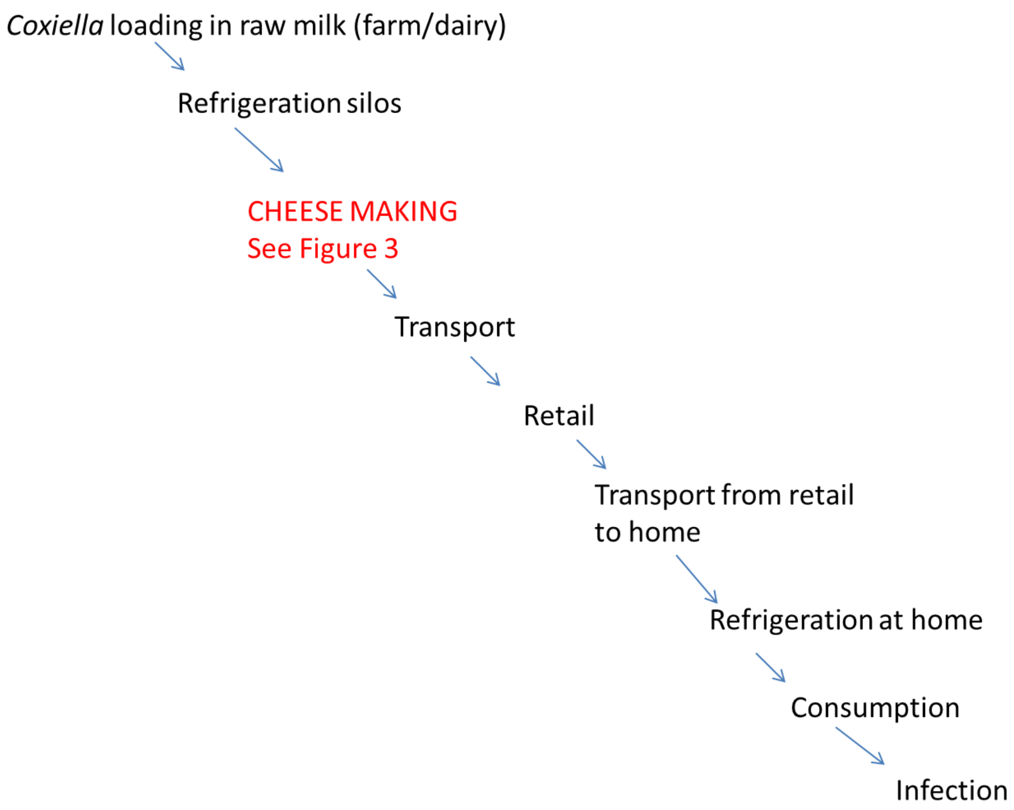

Figure 4: Generic risk pathway for human *C. burnetii* infection due to the consumption of unpasteurised cheese. Each stage is broken down into input parameters in Figure 3 or Table 3.

Table 3: Data requirements and availability for estimating probability of *C. burnetii* infection due to the consumption of a serving of cheese made from raw milk

Parameter	Notation	Data	Justification for grading
Time, temperature and pH/acidity dependent decay rate for *C. burnetii* in cheese	D(t, T, pH)	No data[1,2]	
Refrigeration of raw milk			
Time	$t_{ref,d}$		
Temperature	$T_{ref,d}$	4 - 7°C	
pH/Acidity	$pH_{ref,d}$	6.8[4]	
Cheese production – for time/temp/pH/a_w conditions see Figure 3.			
Transport conditions (cheese dairy – retail)			
Time	$t_{d\text{-}r}$	No data	
Temperature	$T_{d\text{-}r}$	Typically 0 - 7°C (TIS 2013)	
Retail conditions			
Time	t_{retail}	Maximum time will be dependent on the use-by date.	
Temperature	T_{retail}	Backroom coldstore: Mean 3.3°C, maximum 16.6°C Retail display: Mean 4.0°C, maximum 14.4°C. (FAO 2002)	Data are old (1999) and for the USA. Full data set not given.
Transport conditions (retail – home)			
Time	$t_{r\text{-}h}$	Mean 65 min, maximum >120 min (FAO 2002)	Data are old (1999) and for the USA. Full data set not given.
Temperature	$T_{r\text{-}h}$	Mean 10.3°C, maximum 36.6 (FAO 2002)	Data are old (1999) and for the USA. Full data set not given.
Refrigeration at home			
Time	$t_{home,c}$	Will be partly dependent on use-by date.	
Temperature	$T_{home,c}$	Mean 4.0°C, maximum 21.1°C (FAO 2002)	Data are old (1999) and for the USA. Full data set not given.
Serving size	S_{cheese}	Mean of 25 g/person/day[3]	
Probability of infection from contaminated raw cheese (dose-response)	P_{inf}	No data	

[1]No suitable data are available in the published literature, but it is thought to be unlikely to cause inactivation of *C. burnetii* due to the time, temperature and pH conditions.
[2] Some qualitative data suggest *C. burnetii* survive better at neutral pH than low pH. Thus *Coxiella* retains better viability in cheese at neutral pH than at pH 5.0 (Robert Heinzen pers comm). This is based on their experience of freezing *Coxiella* in acidic media, and also the information showing poor viability in soured milk but viability in cheese for 30-40 days. However, it should be noted that the two experiments are not directly comparable and give no data on decay rates or starting loadings.
[3]Department of Health (2011) Cheese consumption for consumers of 19 to 64 years. Not specified whether raw or pasteurised

5 Conclusions.

Three risk pathways have been presented here: (1) probability and levels of *C. burnetii* in BTM per herd; (2) human infection from consumption of raw milk and (3) human infection from consumption of raw cheese. The pathways are generic and can be parameterised for cow's or goats' milk/cheese.

For each pathway, the data requirements have been identified and the availability of data assessed (Tables 1-3). This work has identified a lack of data (quantitative or otherwise) that are readily available for the parameterisation of a farm-to-consumption risk assessment for *C. burnetii*. In particular, the absence of data for the survival of *C. burnetii* in milk or cheese and dose-response is highlighted. As a consequence of this, the project team have concluded that the development of a farm-to-consumption risk assessment for *C. burnetii* is not possible as the uncertainties associated with any risk estimate would be vast. However, given the better availability of data at the farm level, it is our intention to develop a simple exposure assessment for *C. burnetii* through consumption of a serving of unpasteurised cows' milk. The outputs of the baseline exposure model, developed by combining the pathways in Figure 1 and 2 will be the

(1) probability of a human being exposed to *C. burnetii* due to the consumption of a serving of unpasteurised cows' milk; and
(2) dose of *C. burnetii* ingested through consumption of a single serving of contaminated cows' milk at the time of consumption.

Cows' milk was selected due to the availability of data (from Enright et al. 1957) for the levels of *C. burnetii* in milk (albeit in terms of guinea pig infective dose 50% units). It should be noted that, unfortunately, data gaps/deficiencies will remain (e.g. survival data) and assumptions will still have to be made as is always the case in any risk assessment. Once the baseline exposure assessment has been developed a scenario analysis will be carried out to investigate the impact on human exposure to *C. burnetii* if a Q fever outbreak occurs on a farm that produces raw milk.

6 References

Anon (2013a) Milk storage and temperature control (https://www.coolmilk.com/files/Milk%20storage%20and%20temperature%20control%20sheet%20Embedded%20v2.pdf)

Anon (2013b) Innovate with dairy - What exactly are curds and whey? (http://www.innovatewithdairy.com/Pages/Whatexactlyarecurdsandwhey.aspx)

ARC (2013) General concepts regarding dairy cattle (http://www.arc.agric.za/home.asp?PID=1&ToolID=2&ItemID=1927)

Arricau Bouvery, N., Souriau, A., Lechopier, P. and Rodolakis, A. (2003) Experimental Coxiella burnetii infection in pregnant goats: excretion routes. Vet Res 34, 423-433.

Astobiza, I., Barandika, J.F., Hurtado, A., Juste, R.A. and Garcia-Perez, A.L. (2010) Kinetics of Coxiella burnetii excretion in a commercial shepp flock after treatment with oxytetracycline. Vet Journal, 184, 172-175.

Banks, J.G. (2006) Risk Assessment of L. monocytogenes in UK retailed cheese'. Report for Food Standards Agency-funded study B12006. (http://www.food.gov.uk/science/research/foodborneillness/microriskresearch/b12programme/B12projlist/b12006/)

Dairy goat journal (2012) (http://www.dairygoatjournal.com/issues/84/84-6/Tim_King.html)

DairyCo 2013. Kingshay Dairy Costings – National. Available at: http://www.dairyco.org.uk/market-information/farming-data/kingshay-dairy-costings/kingshay-dairy-costings-national/. Last accessed 25th April 2013.

Dairy Co Technical Information (2012) (http://www.dairyco.org.uk/technical-information/animal-health-welfare/mastitis/working-arena-prevention-of-infection/dry-periods-resting-cows/)

Dairy sheep fact sheep (2013) (http://www.dbicusa.org/documents/Dairy%20Sheep%20Fact%20Sheet.pdf)

Department of Health (2011) National diet and nutrition survey. (http://webarchive.nationalarchives.gov.uk/20130107105354/http:/www.dh.gov.uk/en/Publicationsandstatistics/Publications/PublicationsStatistics/DH_128166)

EBLEX 2012. UK Yearbook 2012 – Cattle. Available at: http://www.eblex.org.uk/documents/content/markets/m_uk_yearbook12_cattle240812.pdf. Last accessed 25th April 2013.

Eldin, C., Angelakis, E., Renvoise, A. and Raoult, D. (2013) Coxiella burnetii DNA, but not viable bacteria, in dairy products in France. American Journal of Tropical Medicine and Hygiene 88, 765-769.

Enright, J.B., Sadler, W.W. and Thomas, R.C. (1957) Pasteurization of milk containing the organism of Q fever. American Journal of Public Health, 47, 695-700.

Enright, J. B. (1961). The Pasteurization of Cream, Chocolate Milk and Ice Cream Mixes Containing the Organism of Q fever. *Journal of Milk and Food Technology 24*(11), 351-355.

FAO (2002) Risk assessments of Salmonella in eggs and broiler chickens 2. Section 6.2.5 Retail, distribution and storage (http://www.fao.org/docrep/005/Y4392E/y4392e0n.htm#bm23)

FSA (2009). Raw drinking milk and raw cream control requirements in the different countries of the UK (http://food.gov.uk/business-industry/guidancenotes/dairy-guidance/rawmilkcream)

FSA (2013). Information on UK raw milk cheeses. Document provided by FSA to the FS11016 Project Team.

Guatteo, R., Beaudeau, F., Joly, A., Seegers, H. (2007a). Coxiella burnetii shedding by dairy cows. Vet. Res. 38, 849–860

Hirai, A., Nakama, A., Chiba, T and Kai, A. (2012) Development of a Method for Detecting Coxiella burnetii in Cheese Samples. Journal of Veterinary Medical Science 74, 175-180.

Loftis AD, Priestley RA, Massung RF (2010). Detection of *Coxiella burnetii* in commercially available raw milk from the United States. *Foodborne Pathog and Dis*; 7:1453–56.

Mullan, W.M.A. (2005) Role of cheese starters. [On-line]. Available from: http://www.dairyscience.info/index.php/cheese-starters/225-role-of-starters.html . Accessed: 22 April, 2013.

Raw-Milk-Facts (2013) Major dairy cow breeds (http://www.raw-milk-facts.com/dairy_cow_breeds.html)

Richardson, C.W. (2013) Oklahoma cooperative Extension Service - Let's compare dairy goats and cows (http://oklahoma4h.okstate.edu/litol/file/animal/dairy/N-424_web.pdf)

Sheep 201 (2013) – a beginner's guide to raising sheep (http://www.sheep101.info/201/dairysheep.html)

TIS (2013) Transport Information Service for German marine insurers (http://www.tis-gdv.de/tis_e/ware/milchpro/kaese/kaese.htm)

UK Agriculture 2013. Dairy Production Cycle Available at: http://www.ukagriculture.com/production_cycles/dairy_production_cycle.cfm. Last accessed 25th April 2013.

Valergakis GE, Russell C, Grogono-Thomas R, Bradley AJ and Eisler MC (2012). *Coxiella burnetii* in bulk tank milk of dairy cattle in south-west England. *Veterinary Record* 171(6): 156.