

# Guideline for the interpretation of the results of *Coxiella burnetii* (Q Fever) bulk tank milk analysis in ruminants



Q Fever is a very widespread zoonosis (see our freely available Q-Facts: '[When to suspect Q Fever \(\*Coxiella burnetii\* infection\) in cattle?](#)' and '[When to suspect Q Fever \(\*Coxiella burnetii\* infection\) in small ruminants?](#)'). Infected dairy cows and goats may shed *C. burnetii* in milk for an extended period. In sheep, excretion in milk is also possible. Bulk tank milk (BTM) is a useful matrix to assess the *C. burnetii* status of a dairy herd/flock by antibody (ELISA) or agent detection (PCR). ELISA provides information on past exposure or vaccination of the lactating animals, while PCR (genome detection) demonstrates the possible presence of *C. burnetii* shedding animals<sup>[1,2,3,4,5,6,7]</sup>.



## PURPOSE OF THIS FACTSHEET

To define the circumstances that may justify *Coxiella burnetii* diagnostic tests on bulk tank milk (BTM), and to guide selection of the relevant methods (PCR and/or ELISA) according to the practitioner's objectives.



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*"In an emergency context, as the one experienced since the largest human Q Fever outbreak (the Netherlands – 2007–2010), the systematic application of real-time PCR tests on bulk tank milk every month has proven to be a rapid and effective monitoring system to highlight the circulation of Coxiella burnetii in wide regions. Mandatory tests on BTM represent a cost-effective tool for surveillance programs at the service of Public Health authorities"*



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*"Bulk tank milk PCR should not be used to diagnose abortions, however this is a very relevant analysis for monitoring excretion dynamics especially in cattle and goat herds over time. In addition, using ELISA on tank milk is an inexpensive method for identifying herds with a rather favourable status."*



**THE MAJOR EXCRETION ROUTES FOR *C. BURNETII* IN RUMINANTS ARE PARTURITION PRODUCTS, VAGINAL MUCUS, AND MILK.**

## WHAT'S THE POINT OF ANALYSING BULK TANK MILK?

Below are the summarised recommendations of the EU Q Fever Committee to ruminant practitioners. The following points detail current thinking and supporting arguments.

INDICATION	RELEVANCE OF BULK TANK MILK PCR	RELEVANCE OF BULK TANK MILK ELISA
ABORTION DIAGNOSIS	NOT RELEVANT	NOT RELEVANT
<i>COXIELLA BURNETII</i> SUSPICION UPON REPRODUCTIVE FAILURE IN CATTLE (EXCL. ABORTION)	RELEVANT TOGETHER WITH INDIVIDUAL SEROLOGY ON COWS PRESENTING REPRODUCTIVE DISORDERS (LOOK FOR SEROCONVERSION)	NEED FURTHER STUDIES
FOLLOW-UP OF CONTROL MEASURES (VACCINATION INCL.)	RELEVANT WHEN REPEATED OVER TIME	RELEVANT WHEN REPEATED OVER TIME NOT RELEVANT AFTER VACCINATION PLAN (NO DIVA* ELISA)
DETECTION OF WITHIN-HERD <i>C. BURNETII</i> CIRCULATION/ HERD STATUS DETERMINATION	RELEVANT (EXCEPT IN SHEEP IF USED ALONE)	RELEVANT

\*DIVA: Differentiation of Infected from Vaccinated Animals.

## GENERAL CHARACTERISTICS OF THE EXCRETION OF *COXIELLA BURNETII* BY INFECTED DOMESTIC RUMINANTS

The major excretion routes for *C. burnetii* in ruminants are parturition products, vaginal mucus, milk and faeces (Figure 1).

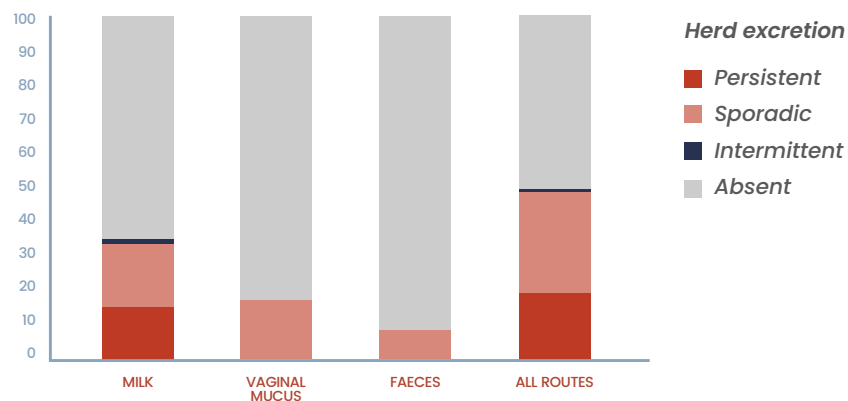


Fig 1. Excretion pathways and profiles of cows in herds infected by *Coxiella burnetii*<sup>[6]</sup>

In all ruminant species, excretion peaks at the time of abortion and/or parturition, but it can also occur at all physiological stages, as seen for example in the follow-up of excretion kinetics in dairy sheep herds in France<sup>[9]</sup>. And it of course occurs in asymptomatic animals, which are the largest part of the infected animals within an infected herd (Figure 2).

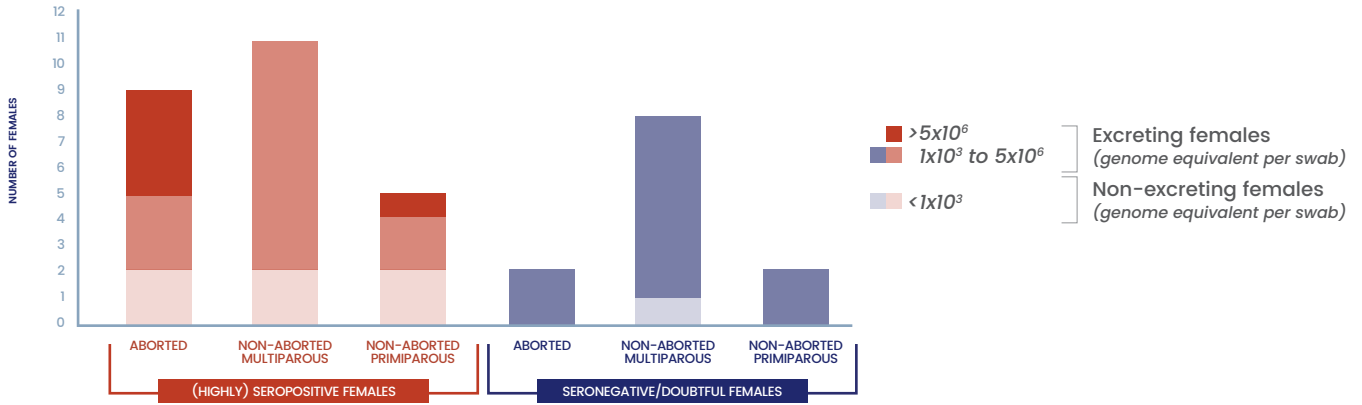


Fig 2. Bacterial loads detected in the vaginal mucus of ewes (having aborted or not) infected by *C. burnetii*<sup>[10]</sup>

**NOTE:** Bacterial excretion in milk and vaginal mucus does not always take place simultaneously. This is why testing the BTM to investigate the cause of an abortion is not relevant (no shedding in the milk at the time of abortion). A BTM qPCR is useful for detecting circulation of *C. burnetii* within a herd, and to monitor its dynamics over time, especially in cattle and goats. In sheep, excretion is less frequent in this matrix.

## CHARACTERISTICS OF THE HUMORAL RESPONSE DETECTABLE IN MILK

A good qualitative concordance exists between anti-*Coxiella burnetii* antibodies in blood and milk (Figure 3).

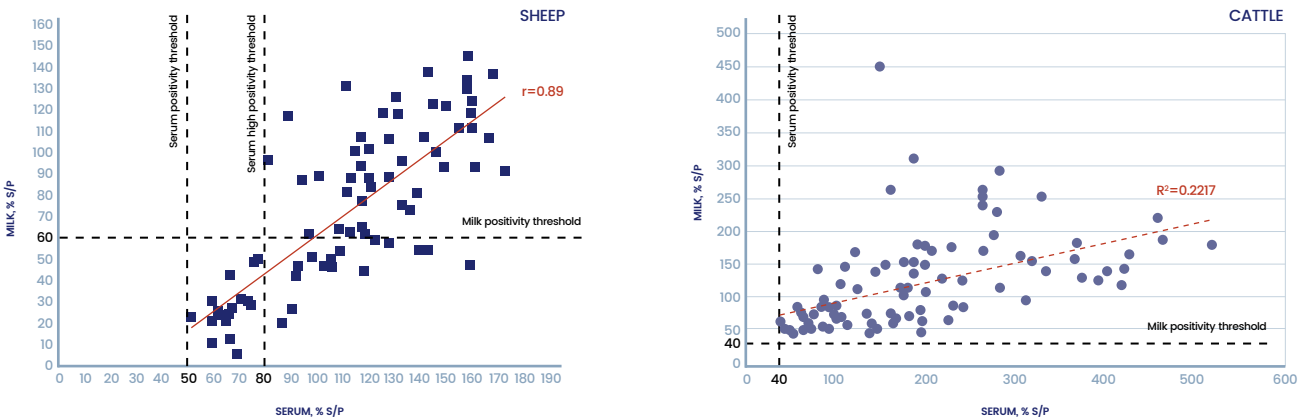


Fig 3. Concordance of the detection of anti-*Coxiella burnetii* antibodies in blood and milk of sheep (left<sup>[10]</sup>) and cattle (right<sup>[8]</sup>).

## INFORMATIVE VALUE OF qPCR AND ELISA

Available ELISA tests are unable to discriminate between vaccinated and infected animals (no DIVA ELISA). A linear association exists between the prevalence of excreting cows and the genomic load in BTM (Figure 4). However, the existence of heavy shedders limits its use for this purpose.

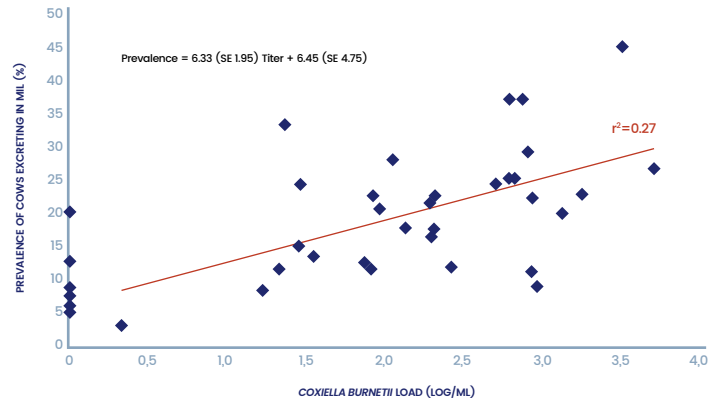


Fig 4. Correlation between the value of a qPCR applied to bulk tank milk and the prevalence of cows excreting *C. burnetii* in milk<sup>[11]</sup>.

Regarding ELISA testing on BTM, the correlation between the level of antibodies measured in milk and the proportion of seropositive lactating animals (herd prevalence) has been poorly documented.

In **cows**, the correlation between antibody levels in BTM and herd prevalence was explored in a single study, and was found moderate (Figure 5)<sup>[12]</sup>. This infers that in the case of a negative or low-positive ELISA result on BTM, a low prevalence herd (<20%) can be expected.

In **sheep**, one study<sup>[13]</sup> reports that two thirds of the flocks with a seroprevalence > 30% had a positive BTM qPCR and only one in five herds with a seroprevalence < 30% had a positive BTM qPCR. These findings can be explained, as mentioned above, by the low number of infected ewes actually shedding *C. burnetii*.

No data are available in **goats**.

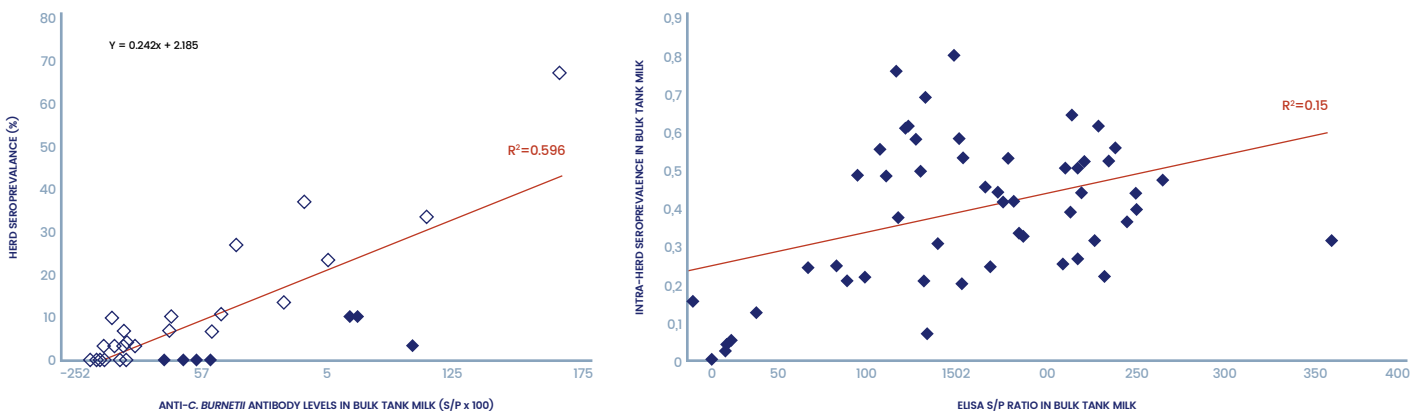


Fig 5. Correlation between the antibody titer measured BTM by ELISA and the seroprevalence of seropositive sheep (left) and seropositive cattle (right).

However, BTM is a valid matrix for anti-*C. burnetii* antibodies detection by ELISA in non-vaccinated herds/flocks, and can be used to assess the status of a given dairy herd, and to explore epidemiological risk factors of infection at regional level<sup>[14]</sup>.

Finally, when a practitioner aims at identifying whether high shedders are present within a herd, repeated serological testing could be a reliable tool as a French study evidenced high and persistent serological titres in cows with a persistent shedding pattern<sup>[11]</sup>.

**NOTE:** The ELISA applied to bulk tank milk cannot accurately estimate the proportion of seropositive animals in a herd (herd seroprevalence), but it can be used to assess and assign overall herd status.

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## EUROPEAN Q FEVER COMMITTEE

The Q FEVER COMMITTEE, co-chaired by Professors Raphaël Guatteo and George Valiakos, was created in July 2024 with the support of Ceva Santé Animale.

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