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## Post-mortem detection of *Coxiella burnetii* in eight ewes from a flock recently confronted with Q fever clustered human cases sampled 89 to 229 days post-lambing

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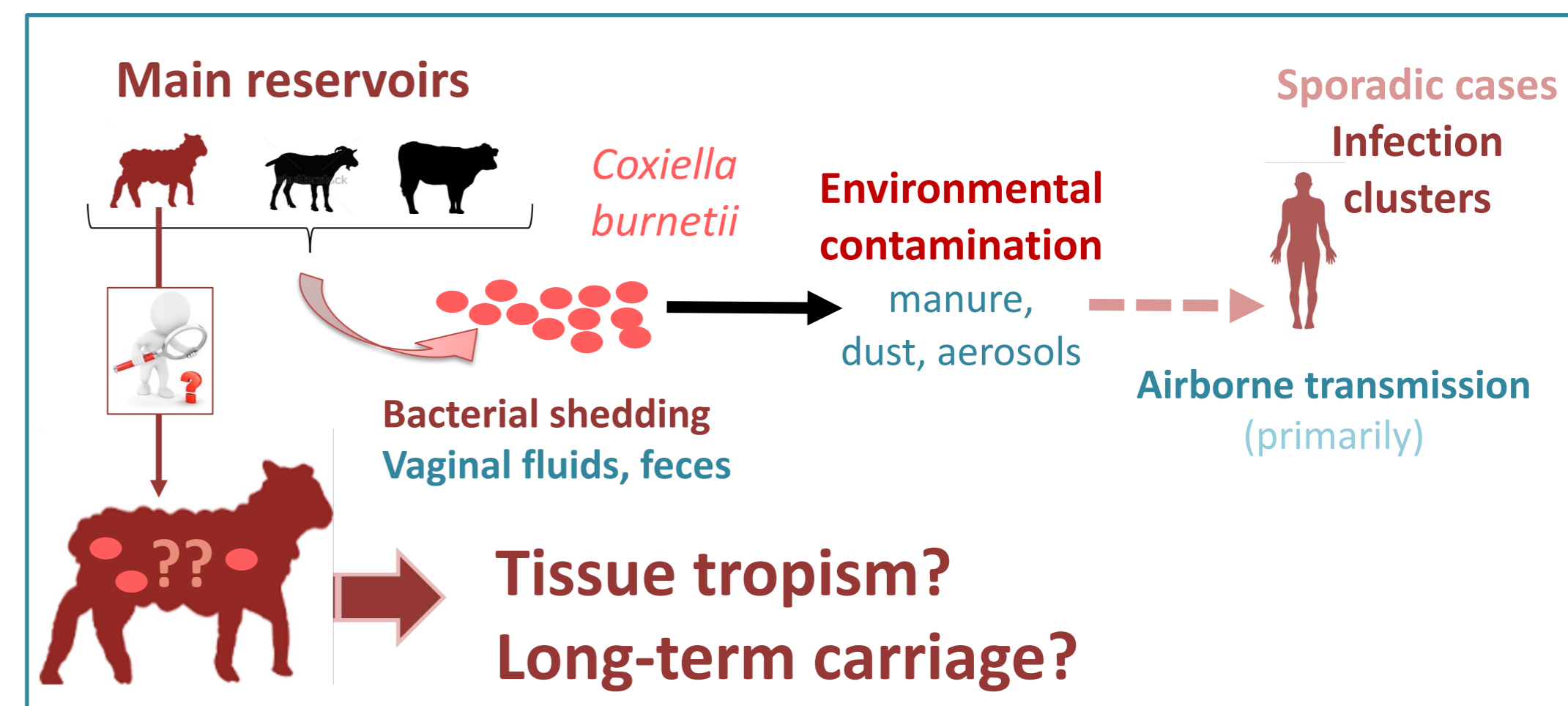
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# Post-mortem detection of *Coxiella burnetii* in 8 ewes from a flock recently confronted with Q fever clustered human cases sampled 89 to 229 days post-lambing



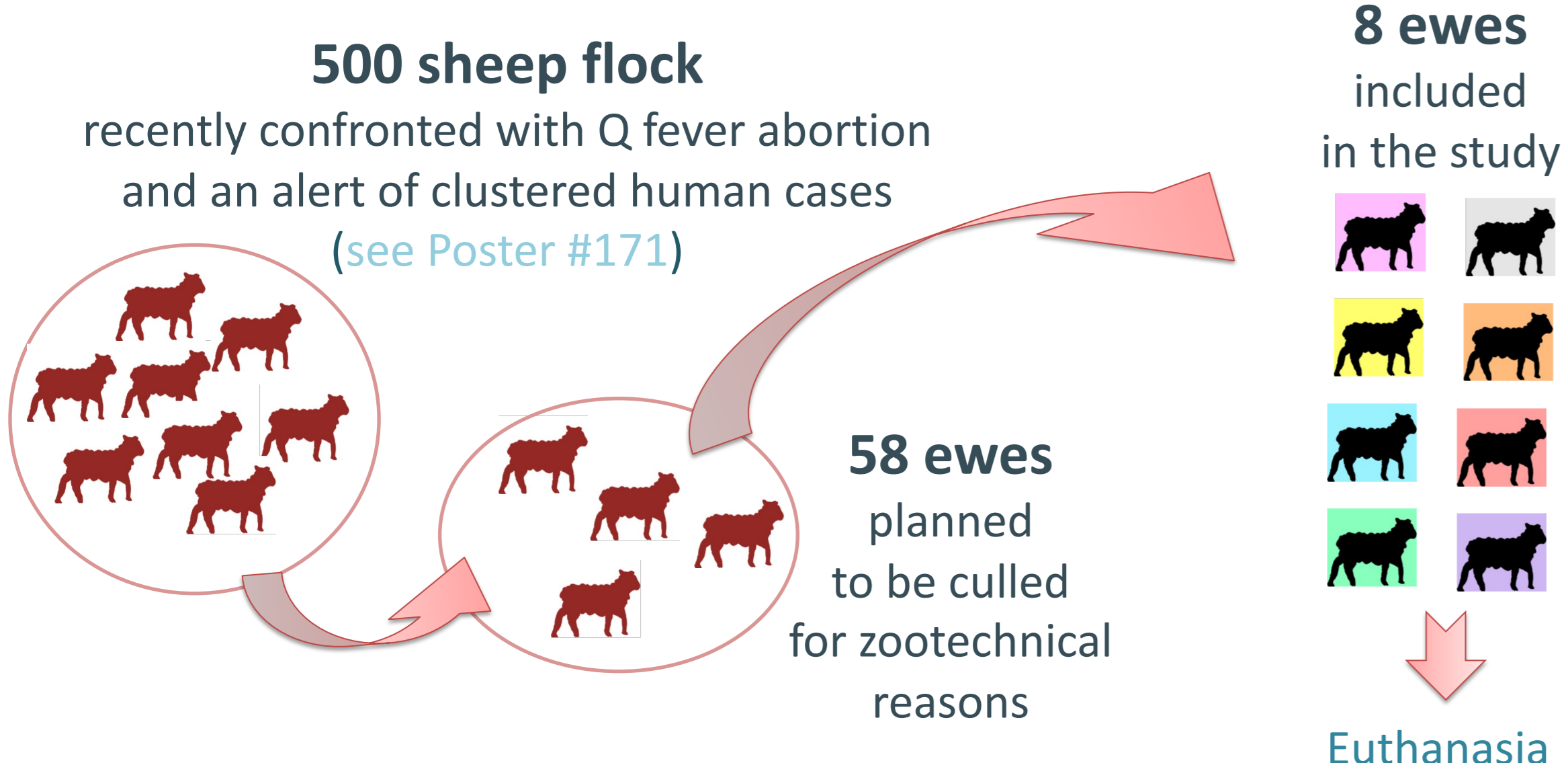
Elsa Jourdain<sup>1</sup>, François Schelcher<sup>2</sup>, Marie-Noelle Lucas<sup>2</sup>, Mathilde Poivre<sup>3</sup>, Jérôme Lafon<sup>4</sup>, Séverine Barry<sup>1</sup>, Aurélie Couesnon<sup>5</sup>, Richard Thiéry<sup>5</sup>, Elodie Rousset<sup>5</sup>

Q fever is a zoonotic disease caused by *Coxiella burnetii*. Domestic ruminants, including sheep, provide a major primary reservoir of exposure for humans. Bacterial shedding is massive in clinically affected flocks through the vaginal fluids and feces of infected ewes, secondarily contaminating bedding areas and other indoor and outdoor environments<sup>1,2</sup>. However, knowledge on tissue tropism and long-term carriage by individual ewes remains scarce.



**OBJECTIVE:** to investigate *C. burnetii*'s tissue dissemination in ewes more than 3 months post-lambing

## METHODS



- **Histopathology**
- **Serology:** PRIOCHECK commercial test
- **qPCR:** in-house method targeting IS1111 → limit of quantification (LOQ) estimated at 200 bacteria per swab or milliliter of suspension

Collection of diverse tissues, swabs and body fluids (see Table)

Protocol approved by Vet AgroSup's ethic committee (no 1929)

## Description of the 8 selected ewes

|   |   |   |
|---|---|---|
| <ol style="list-style-type: none"> <li>Parity</li> <li>Reason for culling</li> <li>Lambing circumstances</li> <li>Post-partum vaginal shedding</li> <li>qPCR test "Day x" post-partum <ul style="list-style-type: none"> <li>Vaginal: vaginal shedding</li> <li>Nasal: anterior nasal shedding</li> </ul> </li> </ol> | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Mastitis</li> <li>Normal lambing</li> <li>Not tested</li> <li>Day 64 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: 6.5E+2</li> <li>Nasal: negative</li> </ul> </li> </ol>        | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Udder shape</li> <li>Normal lambing</li> <li>2.3E+9 (Day 4 pp<sup>§</sup>)</li> <li>Day 122 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: 7.5E+2</li> <li>Nasal: Not tested</li> </ul> </li> </ol> |
| <ol style="list-style-type: none"> <li>Primiparous</li> <li>Milk somatic cells</li> <li>Normal lambing</li> <li>Not tested</li> <li>Day 158 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: 2.8E+2</li> <li>Nasal: negative</li> </ul> </li> </ol>   | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Old ewe</li> <li>Normal lambing</li> <li>Not tested</li> <li>Day 64 pp <ul style="list-style-type: none"> <li>Vaginal: negative</li> <li>Nasal: 3.7E+2</li> </ul> </li> </ol>                     | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Old ewe</li> <li>Normal lambing</li> <li>Not tested</li> <li>Day 177 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: negative</li> <li>Nasal: negative</li> </ul> </li> </ol>                        |
| <ol style="list-style-type: none"> <li>Multiparous</li> <li>Low milk production</li> <li>1 stillborn lamb</li> <li>Not tested</li> <li>Day 182 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: 1.3E+3</li> <li>Nasal: negative</li> </ul> </li> </ol>  | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Udder shape</li> <li>1 stillborn lamb</li> <li>Not tested</li> <li>Day 203 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: negative</li> <li>Nasal: &lt;LOQ</li> </ul> </li> </ol> | <ol style="list-style-type: none"> <li>Multiparous</li> <li>Udder shape</li> <li>1 stillborn lamb</li> <li>Not tested</li> <li>Day 202 pp<sup>§</sup> <ul style="list-style-type: none"> <li>Vaginal: negative</li> <li>Nasal: 3.9E+2</li> </ul> </li> </ol>                    |

<sup>§</sup> pp: post-partum

## RESULTS

Time-frame between lambing and necropsy ranged from 89 to 229 days

|                                   | Histopathology  | Serology   | qPCR analyses   |         |     |     |     |     |     |  |
|-----------------------------------|---|--|---|---------|-----|-----|-----|-----|-----|--|
|                                   | No macroscopic nor microscopic lesion relevant to diagnostic purposes | All serologies positive or near positivity threshold | Low levels of <i>C. burnetii</i> DNA detected in 1 to 6 samples depending on the considered ewe (see Table) |         |     |     |     |     |     |  |
| Ewe number                        | days post-lambing at necropsy   | 1  | 2   | 3       | 4   | 5   | 6   | 7   | 8   |  |
|                                   |   | 89   | 152   | 181     | 205 | 205 | 208 | 227 | 229 |  |
| Shedding (swabs)                  | Vaginal   | -  | +   | -       | -   | +   | -   | +   | -   |  |
|                                   | Anterior nasal  | -  | -   | 6.5E+3* | +   | -   | +   | +   | -   |  |
|                                   | Deep nasal  | -  | -   | -       | -   | -   | +   | +   | -   |  |
| Body fluids                       | Milk  | -  | -   | +       | -   | -   | -   | -   | -   |  |
|                                   | Serum   | -  | -   | -       | +   | -   | -   | -   | -   |  |
| Organs (swabs or crushed tissues) | Bone marrow   | -  | -   | -       | -   | -   | -   | +   | -   |  |
|                                   | Uterus  | -  | -   | -       | -   | -   | -   | +   | -   |  |
|                                   | Ovary   | -  | -   | -       | -   | -   | -   | +   | -   |  |
|                                   | Bladder   | -  | -   | +       | -   | -   | -   | -   | -   |  |
|                                   | Mammary LN  | -  | +   | -       | -   | -   | -   | -   | +   |  |
|                                   | Peyer's patches   | -  | -   | +       | -   | -   | -   | -   | -   |  |
|                                   | Cardiac valves  | +  | +   | -       | -   | -   | -   | -   | -   |  |

Table : qPCR results obtained from the samples collected at necropsy (LN: lymph node)

No detection in: tonsils, bronchi, lungs, spleen, kidney, ileus lymph node, colon, feces and wool

+: DNA detection with bacterial load <LOQ (tardive Ct-value but curve shape typical of specific amplification)

- : no DNA detection \* : bacterial load in GE per swab

## DISCUSSION & CONCLUSION

All ewes included in the study presented evidence of past or current infection by *C. burnetii* without displaying any macroscopic or microscopic lesion potentially-associated to infection

*C. burnetii* DNA was detected in diverse tissues and body fluids including not only the genital tract but also the respiratory tract as well as cardiac valves, bone marrow and Peyer's patches

⚠ with one exception, bacterial loads were too low to be precisely quantified (<LOQ=200 bacteria per swab or milliliter)



These results in sheep

Previous data from experimental infection in goats<sup>3,4</sup>



...raise questions about

- the reality of long-term bacterial carriage by small ruminants coming from flocks with *C. burnetii* circulation
- the occurrence of potential sub-clinical effects due to the infection of organs other than those of the genital tract

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## References

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