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Background

- Footrot accounts for over 80% of lameness in England and is estimated to cost the UK sheep industry approximately £80 million per annum
- Footrot is an infectious dermatitis of the interdigital skin of sheep. The causal agent is *Dichelobacter nodosus*
- *Fusobacterium necrophorum* is closely associated with the disease but its role in disease pathogenesis is not fully understood
- Recent research suggests that *D. nodosus* drives disease pathogenesis, and that *F. necrophorum* is a secondary pathogen



Witcomb, et al., 2014; Witcomb 2012

Table 1 Potential reservoirs of *F. necrophorum* in sheep and their environment

Site	<i>F. necrophorum</i> % detected
Faeces (n=20)	0%
Soil (n=20)	0%
Grass	No data
Healthy feet (n=54)	59%
Oral cavity (n=35)	74%

Methods

- DNA extraction – chemical and mechanical lysis, purification on hydroxyapatite spin column
- Quantitative PCR (qPCR) targeting *rpoB* gene of *F. necrophorum*
- Multiple locus variable number tandem repeat analysis (MLVA)

Purdy, et al., 1996; Witcomb, et al., 2014



Pilot study

Aims:

- To optimise sampling strategies for sheep and their environment
- To identify sites in the environment of sheep where *F. necrophorum* can be detected
- To determine if methods allow detection of variability in load of *F. necrophorum* over time and with disease



Methods:

- Swabs collected from 5 ewes and 5 lambs every 2 weeks for 2 months
- Soil, grass and faeces collected from pasture
- Lesions scored for interdigital dermatitis and footrot
- Samples analysed using qPCR, and MLVA at three loci

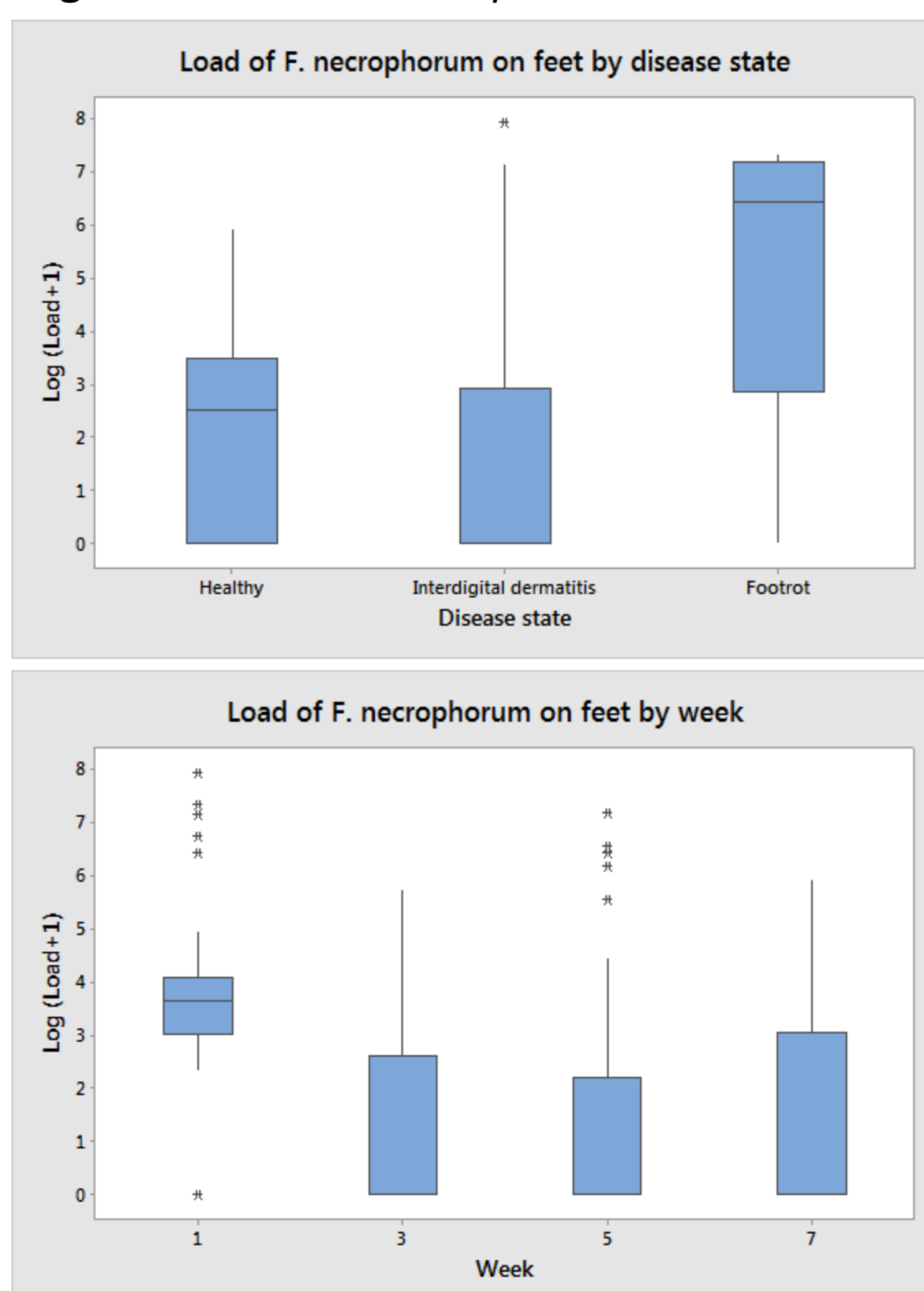
Results

- *F. necrophorum* detected on 50% of foot swabs (n=152)
- *F. necrophorum* detected at all sites in the environment of sheep (oral cavity n=38, soil n=88, grass n=24, faeces n=40) (Table 2)
- Load of *F. necrophorum* varies over time and with disease state (Figure 2)

Table 2 N° of environment samples with detectable *F. necrophorum* by week

Week	Samples with detectable <i>F. necrophorum</i> (%)			
	Oral cavity	Soil	Grass	Faeces
1	90	27	0	0
3	90	5	17	10
5	60	0	0	0
7	75	0	0	0

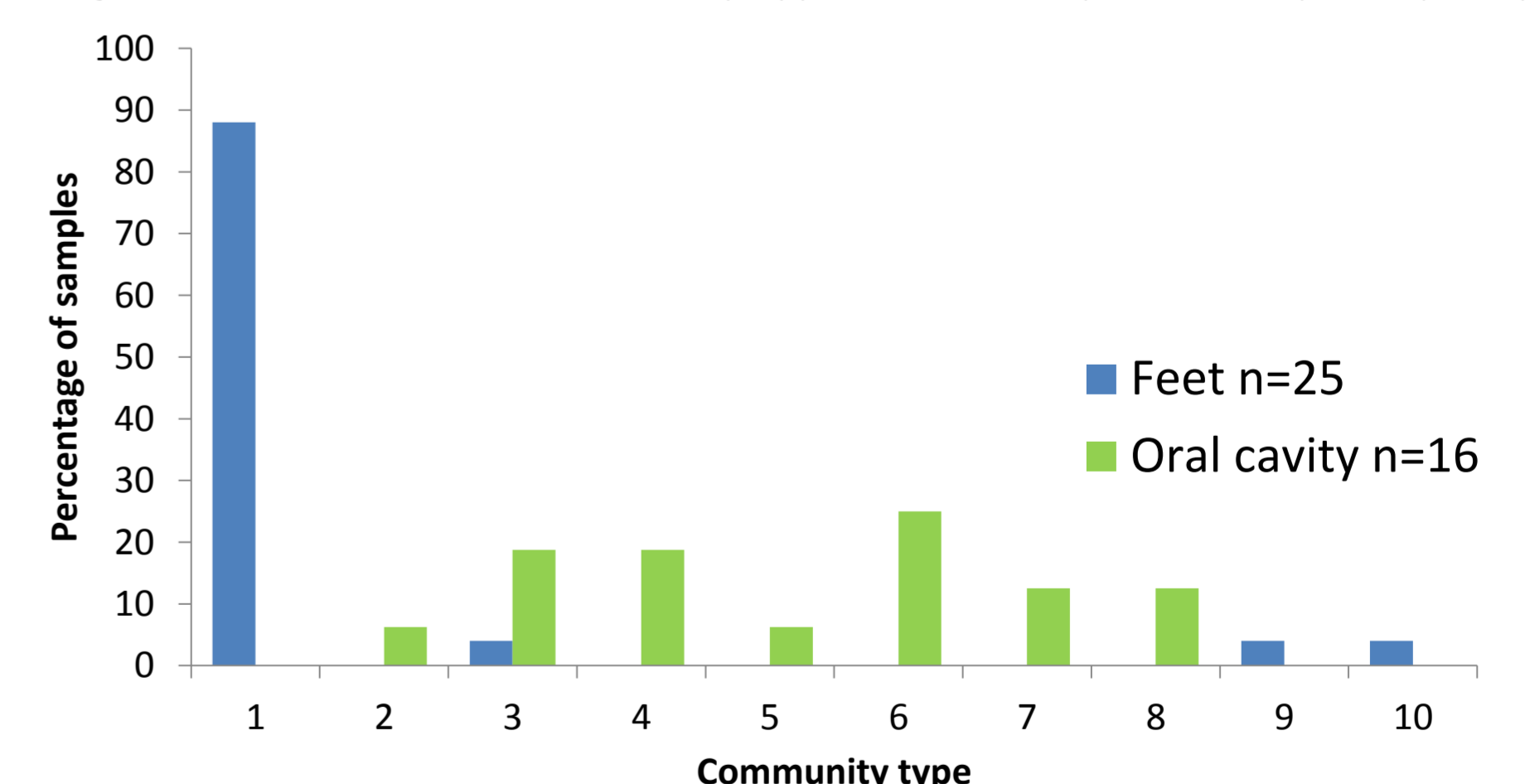
Figure 2 Load of *F. necrophorum* on foot swabs



MLVA

- A variety of strains of *F. necrophorum* are present on swabs from feet and mouths
- Multiple strains of *F. necrophorum* can be detected in one sample

Figure 3 Variation of community type of *F. necrophorum* by sample type



Conclusions

- *F. necrophorum* can be detected in all sample types but not all samples
- Pilot data demonstrate variability in load of *F. necrophorum* over time and with disease
- Pilot data demonstrate differences in communities of *F. necrophorum* between foot and oral cavity swabs

Future work

- Longitudinal study sampling sheep and their environment over 4 months
- MLVA of *F. necrophorum* isolates

References

- Purdy, K.J., Embley, T.M., Takii, S., Nedwell, D.B., 1996. Rapid extraction of DNA and rRNA from sediments by a novel hydroxyapatite spin-column method. *Applied and Environmental Microbiology* 62, 3905-3907.
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