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**Epidemiological studies of  
enteropathogens of newborn calves in  
New Zealand dairy farms**

Julanda Al Mawly

2014

**Epidemiological studies of  
enteropathogens of newborn calves in  
New Zealand dairy farms**

*A thesis presented in partial fulfilment of the requirements for the degree of*  
**Doctor of Philosophy**

Institute of Veterinary, Animal and Biomedical Sciences  
Massey University  
Palmerston North, New Zealand

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July 2014

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## ABSTRACT

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**J. Almawly (2014). Doctoral thesis, Massey University, Palmerston North, New Zealand.**

This PhD thesis comprises an overview of the literature pertinent to the principles of calf rearing in dairy farms, and the major infectious and non-infectious causes of neonatal calf diarrhoea (Chapter 1 and 2), followed by accounts of four epidemiological studies of neonatal calf diarrhoea in New Zealand dairy farms (Chapters 3-6). The first study assessed the utility of halofuginone lactate for the prevention of cryptosporidiosis in the presence of co-infection (Chapter 3), and the following two studies investigated the prevalence of the common enteropathogens of calves, and risk factors for neonatal calf diarrhoea in New Zealand dairy farms. The final study describes a molecular analysis of *Cryptosporidium* parasites isolated from calves, and a genetic comparison with human *C. parvum* clinical isolates collected by diagnostic laboratories in the same regions.

The results of the first study highlight the limitations of the use of halofuginone lactate for the prevention of cryptosporidiosis of calves in the presence of co-infection. The prevalence study provides epidemiologically robust estimates of the national prevalence of calf enteropathogens in dairy farms and the risk factor analysis identified a number of potential risk factors for neonatal calf diarrhoea pertaining to infection status, colostrum management, infrastructure and even human resource management. Finally, the molecular analysis of *Cryptosporidium* indicate that *C. parvum* is the predominant species cycling in newborn calves in New Zealand. The significant genetic similarities between human and bovine *C. parvum* observed in this project support the model considering young calves as amplifiers of potentially zoonotic *C. parvum* in New Zealand.

This project provides new data on the prevalence of the enteropathogens of newborn calves and the risk factors for neonatal calf diarrhoea in dairy farms, which can be used by the New Zealand industry to target interventions aimed at improving animal health, welfare and productivity. This PhD project represents the first large scale epidemiological study of neonatal calf diarrhoea performed in New Zealand and to the author's knowledge, one of the most comprehensive national studies, worldwide.

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## GENERAL PREFACE

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The initial objective of this PhD project was to study the epidemiology, molecular epidemiology and control of cryptosporidiosis in newborn calves in New Zealand. The first study aimed at evaluating the efficacy of halofuginone lactate for disease prevention. The study was performed on a dairy farm in Taranaki, which had been selected in view of the presence of cryptosporidiosis and an absence of rotavirus, and *Salmonella* among winter calves before the spring calving season. However, faecal specimens submitted for analysis at the beginning of the study tested positive also for rotavirus, *Salmonella* and *Giardia* spp. This result, initially considered a drawback, allowed a study of the efficacy of halofuginone lactate in the presence of co-infections with other enteropathogens, which has been previously poorly characterised. Furthermore, this diagnosis prompted an expansion of the scope of this PhD project, to a more comprehensive study of neonatal calf diarrhoea, including studies of the aetiology and risk factor for this important condition.

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## LIST OF ABBREVIATIONS

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BRV	Bovine rotavirus
BCV	Bovine coronavirus
K99	Enterotoxigenic <i>E. Coli</i> K99
<i>C. parvum</i>	<i>Cryptosporidium parvum</i>
spp	Species
HL	Halofuginone lactate
KM	Kaplan–Meier analysis
ANOVA	Analysis of variance
EHEC	Enterohaemorrhagic <i>E. Coli</i>
EPEC	Enteropathogenic <i>E. Coli</i>
EIEC	Enteroinvasive <i>E. Coli</i>
DAEC	Diffusely adherent <i>E. Coli</i>
ETEC	Enterotoxigenic <i>E. Coli</i>
stx	Shiga toxin
bp	Base-pairs
CI	Confidence interval
GP60	Glycoprotein (or 60-kDa glycoprotein)
HSP70	70 kDa Heat Shock Protein gene
IVABS	Institute of Veterinary, Animal and Biomedical Sciences
MU	Massey University
OPG	Oocysts per gram of faeces
TON	Total oocysts number
PCR	polymerase chain reaction
RFLP	Restriction fragment length polymorphism
UV	Ultra violet
XLD	Xylose Lysine-dehydroxycolate
18S rRNA	Small subunit 18S ribosomal RNA
PRU	Protozoa Research Unit
NZDB	New Zealand <i>Cryptosporidium</i> sequence database
LR	logistic regression
SNP	Standardised national prevalence
OR	Odds ratio

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## LIST OF PUBLICATIONS

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**Almawly J.**, Prattley D., French N.P., Lopez-Villalobos N., Hedgespeth B., Grinberg, A., 2013. Utility of halofuginone lactate for the prevention of natural cryptosporidiosis of calves, in the presence of co-infection with rotavirus and *Salmonella* Typhimurium. *Vet. Parasitol.* 197, 59-67.

**Almawly J.**, Prattley D., French N.P., Lopez-Villalobos N., Hedgespeth B., Grinberg A. Is the anti-cryptosporidium effect of halofuginone lactate preserved in the presence of co-infection with rotavirus and *Salmonella* Typhimurium in calves. Submitted poster in IDReC Symposium 2012, Palmerston North, New Zealand.

**Almawly J.**, Prattley D., French N.P., Grinberg A. Field trial of the utility of full and half dosage regimens of halofuginone lactate for the prevention of calf cryptosporidiosis. Presentation at the IV International *Giardia* and *Cryptosporidium* conference held in Wellington in 2011.

**Almawly J.**, Grinberg A., Prattley, D., Moffat J., French N. P. (2014). Prevalence of endemic enteropathogens of calves in New Zealand dairy farms. *N Z Vet J.* 19, 1-18.

**Almawly J.**, Grinberg A., Prattley, D., Moffat J., Jonathan, M., French N. P. (2014). Risk factors analysis for neonatal calf diarrhoea and enteropathogens shedding in New Zealand dairy farms (submitted to *The Veterinary Journal*).