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***Salmonella* Brandenburg in sheep meat
in New Zealand**

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***Salmonella* Brandenburg in sheep meat in New Zealand –
Preliminary studies to support a risk assessment approach**

**A Thesis presented in partial fulfillment of the requirements for the degree
of Masters of Veterinary Sciences in Veterinary Public Health**

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Abstract

Abortion and death of ewes caused by a particular strain of *Salmonella* Brandenburg is an animal disease problem that is unique to the South Island of New Zealand. Like most *Salmonella* serovars, this organism is zoonotic and has caused cases in occupationally exposed people. As *Salmonella* are primarily recognised as agents of foodborne disease, the potential for foodborne transmission must be acknowledged, although human cases attributed to consumption of sheep meat have not yet been reported. *Salmonella* Brandenburg has an additional concern for New Zealand's sheep meat industry owing to the possibility that contamination of sheep meat products could compromise market access. In 1995, the Sanitary Phytosanitary Agreement of the World Trade Organisation specified that scientific risk analysis was required before countries could refuse to import animal or plant materials on the basis of risks to animal, plant, or human health. This thesis presents initial microbiological studies of the prevalence and concentration of *Salmonella* Brandenburg on sheep meat carcasses that were conducted in conjunction with other projects designed to address the *Salmonella* Brandenburg issue using a modern risk assessment approach.

The microbiological studies (Chapters 3 and 4) are preceded by two introductory discussions that provide the context for the project. Chapter 1 presents an overview of national and international regulatory approaches to food safety, foodborne diseases and protection of consumer health relevant to meat and meat products. A selective review of literature on *Salmonella* focuses on *Salmonella* in sheep and on aspects most relevant to food safety. Chapter 2 summarises information on published quantitative microbiological risk assessments (QRA) conducted using the guidelines developed by the Codex Alimentarius Commission to apply QRA to microbiological foodborne hazards. A conceptual framework is presented for developing a QRA for *Salmonella* Brandenburg in sheep meat that covers all sectors of the food supply chain from animal production to the point of consumption. Following the precedent of previous QRA efforts, the food supply chain is divided into a series of five modules: animal production; transport and lairage; slaughter and processing; retail and distribution; and consumer. For each module, key outputs (prevalence and concentration of *Salmonella* in animals or product at various points in the supply chain), and their likely determinants, are identified. The specific objective of the microbiological studies conducted was to estimate the prevalence and

concentration of *Salmonella* on sheep carcasses from animals originating from farms that had experienced *Salmonella* Brandenburg disease and other farms from the same region that had no history of this disease.

Prior to undertaking the field studies, it was necessary to conduct some methodological studies to evaluate the effect of sample handling procedures on the results obtained with quantitative bacteriology. Chapter 3 presents three controlled laboratory experiments with swab samples taken from meat contaminated experimentally with the epidemic strain of *Salmonella* Brandenburg. The Most Probably Number (MPN) method was used to quantify counts of *Salmonella* Brandenburg per 100cm² area of meat swabbed. In each experiment, control samples were processed immediately, and treatment samples were subjected to different periods and conditions of storage. Treatments were chosen to emulate anticipated conditions that would be required for the field studies due to logistic constraints. The three storage protocols evaluated were:

Experiment 1: Storage of swabs diluted in buffered peptone water (BPW) for 48h at 4⁰C

Experiment 2: Storage of swabs diluted in BPW for 5 days at 4⁰C

Experiment 3: Storage of swabs for 24h at 4⁰C before dilution in BPW, followed by storage for a further 48h at 4⁰C.

Differences in counts between control and treatment samples were not tested statistically, owing to the small samples sizes, but were numerically less than one log difference in all experiments. In 2 of the 3 experiments, counts for stored samples were in fact numerically greater than for samples processed immediately. These results suggested that carcass swabs contaminated with *Salmonella* could be stored under the specified conditions without affecting the results of quantitative bacteriology using the MPN method.

Chapter 4 presents a study undertaken to obtain initial qualitative and quantitative estimates of the presence of *Salmonella* organisms on sheep carcasses sampled at 3 points in the processing chain (i.e. slaughter floor, cooler, and boning room). Slaughtered sheep (ewes and lambs) were sourced from six farms in the Central Otago/Southland region of the South Island where *Salmonella* Brandenburg disease is endemic. Three farms (case farms) were selected based on the occurrence of an outbreak of *Salmonella* Brandenburg

disease during the spring of 2000. Three non-case farms from the same region were also sampled. As the disease epidemics are temporally clustered in July and August, well before lambs are sent for slaughter, sampling was replicated after an interval of approximately 2 months to assess likely temporal variation in risk of carcass contamination. For comparative purposes, samples from sheep carcasses were also collected from 6 groups of sheep slaughtered at 2 plants in the North Island where salmonellosis due to *Salmonella* Brandenburg infection in sheep has not been reported. A total of 1417 carcasses were sampled in the study and initially tested by BAX[®] test. Of these, 1214 samples were sourced from the 3 case and 3 non-case farms supplying the South Island plant. The remaining 203 carcasses were sampled at the 2 North Island plants. A total of 138 (11.3%) of the 1214 samples collected in the South Island plant tested positive for the presence of *Salmonella* Brandenburg. No positive findings were obtained from the samples collected in the North Island plants. The vast majority (130 or 94%) of the 138 positive samples was obtained in the first period of sampling, indicating a substantial decline in risk of carcass contamination in the period between the first and second sampling. These findings indicated that the prevalence of carcass contamination with *Salmonella* Brandenburg was markedly elevated in the region where sheep flocks experienced abortion outbreaks caused by the organism. Although clinical *Salmonella* Brandenburg enteric disease has not been reported in lambs, the first sampling revealed that overall prevalence of contamination was higher (33%) for lamb carcasses than ewe carcasses (10%) from the same farms. While the prevalence of lamb carcass contamination was comparable for both case and non-case farms, the prevalence of ewe carcass contamination was strongly clustered and only 2 samples were positive from non-case farms. Estimates of the prevalence of contamination were influenced by the location of sampling carcasses (e.g. slaughter floor, cooler), but estimates of bacterial numbers on positive carcasses were generally similar regardless of class of stock, time of sampling, or sampling location in the plant. No positive samples were obtained from swabs of primary cuts in the boning room. Collectively these findings suggest that the emergence of *Salmonella* Brandenburg infection of sheep in the South Island may have considerable implications for product safety and public health. A strong case can be made for more research to better characterise the potential risks and to explore potential risk mitigation strategies. While the data obtained in this study have provided valuable insights into several important aspects of the issue, due to logistic and other constraints they have

considerable shortcomings with respect to the requirements of the formal QRA. These shortcomings were discussed and evaluated in terms of representativeness and suitability for quantitative risk assessment.

Chapter 5 presents an extension of the conceptual framework for a QRA outlined in Chapter 2, by integrating the data obtained from the bacteriological study, as well as data from other sources. Major data gaps are identified and suggestions are presented with respect to options for ongoing research to advance understanding and management of *Salmonella* Brandenburg in New Zealand sheep meat. More extensive and representative surveys are required to obtain more reliable data on farm, and within-farm, prevalence of infection as well as more extensive and representative longitudinal studies of the prevalence and concentration of the organism during slaughter and processing. It is considered that more systematic surveys at the time of apparent highest risk would be a more reliable means of assessing potential exposure of consumers than predictive microbiology.

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“Never give up, ... there is always hope”

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List of Abbreviations

ALOP	Appropriate Level Of Protection
BSE	bovine spongiform encephalopathy
CAC	Codex Alimentarius Commission
CCP	Critical Control Point
CCFH	Codex Committee on Food Hygiene
ESR	The New Zealand Institute of Environmental Sciences & Research Limited
EU	European Union
FAO	Food and Agriculture Organisation
FSO	Food Safety Objective
GATT	General Agreement on Tariffs and Trade
GHP	Good Hygiene Practices
GMP	Good Manufacturing Practices
HACCP	Hazard Analysis and Critical Control Point
MPN	Most Probable Number method
NMD	New Zealand National Microbiological Database
NZFSA	New Zealand Food Safety Authority
OIE	Office International des Epizooties (World Organisation for Animal Health)
S. Brandenburg	<i>Salmonella enterica</i> subsp. <i>enterica</i> (Brandenburg)
S. Brandenburg - QRA project	Multisectorial quantitative risk assessment project administered by NZFSA and funded primarily by Meat New Zealand over a 3 year period. Sectors include NZFSA, primary producers, the meat processing industry, field veterinarians, Ministry of Health, local health authorities, science providers (Massey University, AgResearch, ESR, LABNET), animal remedy industry
SPS	Sanitary and Phytosanitary Agreement
QRA	Quantitative Risk Assessment
VCJD	variant Creutzfeldt-Jacob disease
UK	United Kingdom
USA	United States of America
USDA	United States Department of Agriculture
USDA-FSIS	United States Department of Agriculture Food Safety and Inspection Services
WHO	World Health Organisation
WTO	World Trade Organisation

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