



# LIVE SHEEP TRADE INITIATIVES

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Live Export

## ABSTRACT

Mortalities of sheep during shipping continue to be a significant threat to an export trade worth approximately \$200 million to Australia. A major research effort has been conducted to determine the reasons for deaths and ways of reducing the losses. The present project is a continuation of this research which, recently, has focussed on farm groups of sheep. Most of the deaths during export were shown to be from relatively few high mortality lines. Death rates by farm group were as high as 30%. Also, the mortality category (high or low) for sheep from the same farm was repeatable in different years. Hence, any future research should focus on the reasons why some farm groups suffer higher death rates than others. Part of this project included a preliminary metabolic study which found differences between sheep from high and low mortality farms. Commercial exporters may choose to monitor the mortality of farm groups of sheep and use that information to influence their buying decisions. Those farmers who may be identified as having sheep that suffer high mortalities will be urgently seeking advice on how to manage the problem to ensure that they can continue to sell sheep to the export trade.

### **EXECUTIVE SUMMARY**

#### **Background and Industry Context**

Exports of live sheep continue to make a major contribution to the Australian economy. This is particularly so for Western Australia which provided 75% of exported sheep in 1995 worth in excess of 150 million dollars. At the national level, the welfare of sheep during export by sea has attracted the most consistent public attention of all animal welfare issues. The concern of the community has the potential to threaten the future of the trade. Substantial improvements in welfare have been made during the last decade although deaths during the shipping phase continue to be significant. Annual death rates in recent years were around 2% from loading onto the ship to the end of discharge in the Middle East.

A major research effort has been conducted to determine the reasons for deaths and ways of reducing the losses. The causes of death have been clearly defined and were the same in studies on different voyages. Although the causes of death were the same in different voyages, death rates followed a seasonal pattern and were higher in the second half of the year. In addition to the seasonal pattern, there was considerable variation in mortality for different farm groups of sheep which was up to 30% between loading and discharge in a recent study. In an epidemiological study of sheep deaths, half of all deaths aboard ship occurred in 25% of 133 lines of sheep. In another study, 57% of all deaths occurred in 5 (16%) of 31 lines. The 5 lines contributed only 21% of the sheep loaded.

The latter evidence supports the contention that death rates for individual farm groups do not occur by chance and, by deduction, there must be reasons for the skewed mortality distribution. Research to date has indicated that death rates by farm group may be reduced by treatment before export, that there are metabolic differences between high and low mortality farms and that mortality is repeatable for farm groups of sheep exported in different years. The present project was split into four activities that were designed to pursue these issues.

#### **Project Objectives**

- 1. To test whether treatment with vitamin B12 or ivermectin reduces the death rate of adult wethers exported live by sea.
- 2. To determine whether macrocyclic lactones increase feed intake and reduce mortality in adult wethers exported live by sea.
- 3. To investigate metabolic differences between high and low mortality farms.
- 4. To establish whether high death rates are repeatable in successive consignments of sheep from the same farm, over two years.

#### Major Results and Conclusions

Treatment with vitamin B12 did not reduce death rates during shipping. The voyage mortality rate was 2.95% and 2.62% for the control and vitamin B12 treated groups (P > 0.05). The ivermectin group had 43% fewer deaths (2.84%) than the controls (4.99%; P = 0.09). Results of this trial suggested that ivermectin was the likely cause of 43% fewer deaths in treated sheep in the 1989 research voyage, in which several treatments were combined, as the same reduction in deaths was achieved in both studies.

During simulated shipping, macrocyclic lactone treatment (ivermectin or moxidectin) was not associated with a significant increase in chaff intake during the first 7 days after treatment. Nor was there any difference in the proportion of feeders in any group. In trial 2, we chose to restrict the comparison to ivermectin and controls using shipper pellets instead of chaff. Again, there was no response to the treatment. Mean feed intake data indicated that internal parasite burdens, at the moderate levels in our sheep, did not adversely affect appetite during simulated shipping. There was no evidence that ivermectin stimulated appetite. There was no difference in mortalities in the research voyage. We concluded that there was no evidence that macrocyclic lactones increased feed intake in adult Merino wethers, either by an effect dependent or independent of anthelmintic activity. We considered that macrocyclic lactones could not be recommended as a blanket treatment to reduce mortalities in sheep exported live.

Sheep from two farms, with different mortality rates during actual export, responded differently to both metabolic and stress challenges. Insulin resistance was indicated by the increased basal and glucose stimulated secretion rates of insulin observed in the sheep from the high mortality farm. Obese ruminants have tissue resistance to the glucoregulatory effects of insulin. In our study the insulin resistance did not relate well to condition score or liveweight but this may reflect limitations in using condition score as an indicator of body fat. There was a 30% greater cortisol release in the sheep from the high mortality farm. This may reflect the sheep 'over responding' to a challenge which in turn could lead to adrenal exhaustion.

We concluded that mortality was repeatable and that the likely mortality category (high or low) of a line of sheep can be predicted with considerable confidence. In addition to the evidence for repeatability, there was a strong association between regional location and mortality rate. The association was supported by the spatial analyses, the comparison of average death rates by zone and the comparison of the proportions of high mortality farms by zone. The results showed that there were more high death rate farms and higher death rates in the longer growing season zones of the southwest of Western Australia. We considered that this was consistent with earlier published studies on the factors leading to mortalities. Follow-up studies are indicated to further investigate the reasons for differences in mortality rates with a view to determining practical strategies to reduce the problem.

#### Implications for Industry

Results of this project have not provided any simple solutions to the mortality problem. However, it was only the vitamin B12/ivermectin studies that had the potential to provide a solution that could be implemented immediately. The repeatability study was an interim step leading to further studies, if indeed repeatability was demonstrated to occur, on the reasons why mortality by farm group may be the same in successive years. The metabolic study was preliminary in nature and provided some indication of differences between high and low mortality groups of sheep.

The potential impact of the finding of regional differences in mortality rates is that export buyers could change their pattern of farm selection to avoid higher mortality areas. At present this could done by favouring sheep from the shorter growing season zones. Should export companies choose to record mortalities by farm group, they could develop their own database of the performance of farm groups of sheep and, with our conclusion about repeatability, alter their buying pattern for farms that they determine to be high mortality. Those farmers who may be identified as having sheep that suffer high mortalities will be urgently seeking advice on how to manage the problem to ensure that they can continue to sell sheep to the export trade.

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# 1. BACKGROUND AND INDUSTRY CONTEXT

Exports of live sheep continue to make a major contribution to the Australian economy. This is particularly so for Western Australia which provided 75% of exported sheep in 1995 worth in excess of 150 million dollars. At the national level, the welfare of sheep during export by sea has attracted the most consistent public attention of all animal welfare issues. The concern of the community has the potential to threaten the future of the trade. Substantial improvements in welfare have been made during the last decade although deaths during the shipping phase continue to be significant. Annual death rates in recent years were around 2% from loading onto the ship to the end of discharge in the Middle East.

A major research effort has been conducted to determine the reasons for deaths and ways of reducing the losses. The causes of death have been clearly defined and were the same in studies on different voyages. Although the causes of death were the same in different voyages, death rates followed a seasonal pattern and were higher in the second half of the year. In addition to the seasonal pattern, there was considerable variation in mortality for different farm groups of sheep which was up to 30% between loading and discharge in a recent study. In an epidemiological study of sheep deaths, half of all deaths aboard ship occurred in 25% of 133 lines of sheep. In another study, 57% of all deaths occurred in 5 (16%) of 31 lines. The 5 lines contributed only 21% of the sheep loaded.

The latter evidence supports the contention that death rates for individual farm groups do not occur by chance and, by deduction, there must be reasons for the skewed mortality distribution. Research to date has indicated that death rates by farm group may be reduced by treatment before export, that there are metabolic differences between high and low mortality farms and that mortality is repeatable for farm groups of sheep exported in different years. The present project was split into four activities that were designed to pursue these issues.

### 2. PROJECT OBJECTIVES

- 1. To test whether treatment with vitamin B12 or ivermectin reduces the death rate of adult wethers exported live by sea.
- 2. With reference to adult Merino wethers:
  - a. To determine whether macrocyclic lactones increase feed intake due to an effect independent of anthelmintic activity, in the first and second halves of the year.
  - b. If a pharmacological effect is identified, to determine the effect of dose rate of macrocyclic lactones on feed intake.
  - c. To evaluate macrocyclic lactones as a treatment to reduce death rates in high mortality lines during actual export.

- 3. To investigate metabolic differences between high and low risk farms by:
  - a. Measuring the response to chronic stress using an ACTH challenge to evaluate adrenal cortex function.
  - b. Investigating the difference in glucose clearance rates between and within farms.
  - c. Describing the relationship between body condition score and source (farm) on plasma insulin concentrations.
- 4. To establish whether high death rates are repeatable in successive consignments of sheep from the same farm, over two years.

### 3. PROJECT ACTIVITIES

Separate project activities were designed specifically to satisfy the objectives listed in section 3.2 above. Detailed technical reports were submitted for each activity and are listed in the publications section 3.6 below. A brief summary of each activity is presented in this report. Copies of the detailed technical reports can be obtained from the authors.

#### 3.1. Vitamin B12 and Ivermectin Study

Several treatments were applied to adult wethers in a feedlot before a voyage in August 1989 (DAW 027). There was a 21% reduction in the proportion of non-feeders in the treatment group at the end of the feedlot period and a 43% reduction in mortality during the voyage (P < 0.05). It was concluded that either or both ivermectin and vitamin B12 caused the reduction in feedlot non-feeders and heavily influenced mortality. Anecdotal evidence suggested that ivermectin may stimulate appetite, in addition to its effect on internal parasites, by an unknown mechanism. Deficiency of vitamin B12 results in an inability to metabolise propionic acid and is accompanied by a loss of appetite and sometimes death. Consequently, we sought to test whether treatment with vitamin B12 or ivermectin reduces the death rate of adult wethers exported live by sea.

Four of 7 farms included in a study in 1993 (DAW 054) were available for investigation. Voyage mortality, in 1993, exceeded 22% in sheep from one of these farms (farm 1) and was between 3% and 5% for the other 3 farms. Sheep from the 3 lower mortality farms were systematically divided into 2 equal groups and tagged. On the remaining (high mortality) farm, sheep were systematically divided into 3 equal groups and tagged. Sheep from one treatment group on each farm were treated with a 1 mL subcutaneous injection of Young's Vitamin B12<sup>®</sup> (1.8 mg hydroxycyanocobalamin, 0.2 mg cyanocobalamin per mL; Young's Animal Health Pty Ltd). Sheep in the second group (controls) received a subcutaneous injection of 1 mL normal saline. Sheep in the third group on the high mortality farm were given a 25 mL oral drench of Ivomec<sup>®</sup> (0.8 g/L ivermectin; MSD Agvet), sufficient to treat a 100 kg wether. All sheep were treated between 5 and 14 days before trucking to the feedlot where they were held in the same shed. Blood samples were collected from each group before trucking to the ship. Sheep that died aboard ship were necropsied to determine the cause of death.

The voyage mortality rate was 2.95% and 2.62% for the control and vitamin B12 treated groups (P > 0.05). The mortality rate was 4.99% and 2.84% for the control and ivermectin groups from farm 1. The ivermectin group had 43% fewer deaths than the controls (P = 0.09). Sheep from farm 1 had the highest mortality rate. Plasma vitamin B12 concentrations, from blood samples collected before trucking to the ship, exceeded marginal levels (0.3 to 0.5  $\mu$ g/L) except in 2 sheep, 1 from farm 5 (0.38  $\mu$ g/L) and the other from farm 3 (0.14  $\mu$ g/L) which is considered deficient (< 0.3  $\mu$ g/L). All liver vitamin B12 concentrations (n = 17) were adequate (> 0.04 mg/kg). The cause of death was determined for 92.1% of sheep that died; inanition was diagnosed as the primary cause of death for 94.3%, salmonellosis for 4.3% and obstructive urolithiasis for 1.4%.

Results of this trial suggested that ivermectin was the likely cause of 43% fewer deaths in treated sheep in the 1989 research voyage, the same reduction in deaths that was achieved in the present study. However, vitamin B12 may also have contributed to some degree; there was a trend towards lower death rates in the 2 higher mortality farm groups. However, any response to treatment with vitamin B12 is unlikely to be due to the correction of a deficiency of vitamin B12 in exported sheep.

The mortality rate for farm 1 was considerably lower than in 1993 but again it was the highest mortality line of those selected for study. The poor seasonal conditions in the southwest of Western Australia in 1994 resulted in lower body condition in the State's sheep flock. The low death rate in sheep from farm 6 (0.5%) and the lower death rate in sheep from farm 1, in 1994 compared to 1993, provided further support for the hypothesis that feed restriction can reduce mortality.

Treatment of sheep with ivermectin was associated with a 43% reduction in mortalities aboard ship in a research voyage in 1989 and in the present study. There was no evidence that a deficiency in vitamin B12 was a factor contributing to deaths aboard ship but treatment was associated with a trend to lower death rates. Further research is warranted on the mechanism by which ivermectin reduces mortality in sheep exported live by sea.

#### 3.2. Macrocyclic Lactone Studies

It was recommended from the above activity that ivermectin be further investigated as a potentially cost effective and practical method of reducing mortality. There was a need to determine the mechanism of any effect of ivermectin, in order to formulate the most appropriate method of applying the treatment. Hence the aims of this study were to determine pharmacological explanations for lower death rates in treated sheep and to examine the effect of ivermectin on high mortality lines during actual export. Treatments to reduce mortalities will be of particular benefit to producers who are identified as having sheep which suffer high death rates when exported live.

A series of 4 animal house trials was conducted in 1995 to examine feed intake responses to various treatments. The animal house trials were followed by a research voyage to assess the effect of ivermectin on high mortality lines. All sheep selected for the animal house trials were purchased from a farm with a history of high mortalities in mature (5 year old) wethers exported in 1994. Trials 1 and 2 were conducted in autumn, trial 3 in winter and trial 4 in spring. The research voyage departed Fremantle in November 1995.

In the first animal house trial, macrocyclic lactone treatment (ivermectin or moxidectin) was not associated with a significant increase in chaff intake during the first 7 days after treatment. Nor was there any difference in the proportion of feeders in any group. In trial 2, we chose to restrict the comparison to ivermectin and controls using shipper pellets instead of chaff. Again, there was no response to the treatment.

While the evidence from trials 1 and 2 did not support a pharmacological benefit of ivermectin we considered that other appetite stimulants may be effective. In trial 3, we evaluated the potential use of brotizolam, an appetite stimulant registered for use in cattle in New Zealand, but reported to be effective in sheep. Our data indicated a transient and minor increase in feed intake in 3 of 13 sheep on day 1; feed intake for the 3 sheep was lower on day 2. However, the number of non-feeders after 4 days in the shed was similar for each group, suggesting that any effect on appetite was in sheep that were going to eat later during the shed phase.

It is well recognised that internal parasites reduce feed intake. Trial 4 examined the effect of ivermectin treatment on feed intake in the second half of the year in sheep given standard parasite burdens. The mean feed intake data indicated that internal parasite burdens, at the moderate levels in our sheep, did not adversely affect appetite during simulated shipping. There was no evidence that ivermectin stimulated appetite.

We concluded that there was no evidence that macrocyclic lactones increased feed intake in adult Merino wethers, either by an effect dependent or independent of anthelmintic activity. The lack of benefit applied to treatments administered in the first and second halves of the year. That there was no appetite benefit in the animal house studies was consistent with the lack of a difference in mortalities in the research voyage. We considered that macrocyclic lactones could not be recommended as a blanket treatment to reduce mortalities in sheep exported live.

#### 3.3. Metabolic Study of High and Low Mortality Farms

Most deaths aboard ship are concentrated in relatively few lines of sheep. In an earlier study (DAW 054), the mortality rate by farm group of sheep ranged from 2% to more than 29%. The reasons for differences between mortality rates by farm may be additional to the established effects of season, age and fatness on death rate during export. The range in mortality rates between farms suggested that sheep from high and low mortality farms may respond differently to stress and metabolic challenge. An inherent assumption in this study was that farms could be identified as high or low mortality based on previous records. However, as evidence for repeatability of mortality was still being examined at the time of this study, it was acknowledged that the decision to proceed was pre-emptive. Identification of hormonal and metabolic differences between farm groups of sheep at high and low risk of mortality would provide a model for investigating the failure to eat syndrome. An understanding of the mechanisms predisposing sheep to the failure to eat syndrome may enable the development of practical strategies to minimise inappetence thereby decreasing mortality.

Two farms were selected, one classified as high mortality (HM) and the other as low mortality (LM). Thirty sheep were systematically selected from a line of wethers scheduled for export within 3 weeks. Half of the selected sheep on each farm was randomly allocated to glucose

tolerance test (GTT) or ACTH challenge treatment groups. Fifty systematically selected sheep were fasted overnight for collection of blood samples the following day.

Sheep in the ACTH treatment group were injected intramuscularly with 4 mg of dexamethasone phosphate to suppress endogenous cortisol production. Each sheep in the ACTH treatment group was injected with 16 ug ACTH and the GTT sheep were infused with 200 mg/kg of dextrose (50 % dextrose, Baxters). Blood samples were taken before and after the ACTH and glucose infusions. Glucose concentrations in the plasma were determined using a Cobas Mira Automatic analyser and concentrations of insulin and cortisol in the plasma were determined by radioimmunoassay. Single sample means were compared using Student's t-test. Cortisol, glucose and insulin results were compared using analysis of variance at each time interval.

The line mortality was 7% and 3.5% for the HM and LM farms respectively. The fasting insulin and cortisol concentrations were higher in the sheep from the HM farm (P < 0.05). Sheep from the HM farm had a significantly greater insulin response to the glucose challenge and insulin concentrations were approximately 40% higher in sheep from the HM farm 15-60 minutes after glucose infusion. Mean plasma glucose concentration was significantly lower in sheep from the HM farm after 40 minutes. The response to ACTH challenge was significantly greater in sheep from the HM farm. Cortisol levels in both groups returned to basal levels after 3 hours.

Sheep from two farms, with different mortality rates during actual export, responded differently to both metabolic and stress challenges. Insulin resistance was indicated by the increased basal and glucose stimulated secretion rates of insulin observed in the sheep from the HM farm. Obese ruminants have tissue resistance to the glucoregulatory effects of insulin. In our study the insulin resistance did not relate well to condition score or liveweight but this may reflect limitations in using condition score as an indicator of body fat. There was a 30% greater cortisol release in the sheep from the high mortality farm. This may reflect the sheep 'over responding' to a challenge which in turn could lead to adrenal exhaustion.

This was a preliminary study to determine whether there are metabolic differences in sheep from high and low mortality farms. The difference between the sheep from these farms suggested a basis for a hypothesis that sheep from high mortality farms may have a degree of insulin resistance and that they may respond to stress with increased levels of cortisol production. These metabolic differences may in turn lead to a greater tendency to inappetence in the feedlot. Higher insulin levels were found in the high intake sheep in the pen study in DAW 054 and the highest levels were found in the persistent non-feeder. An understanding of how to manipulate the metabolism of these sheep could lead to a treatment to reduce mortality during shipping.

#### 3.4. Repeatability of Mortality Study

In 2 separate epidemiological studies at least half of all deaths aboard ship occurred in 25% and 16% of the total number of lines of sheep. Also, death rates in sheep from 2 farms were similarly high in different years. These results indicated that death rates for individual farm groups do not occur by chance and pointed to a need to determine if mortality is repeatable. If mortality is repeatable, then research can focus on farms with high and low death rates to determine differences. Those farmers who may be identified as having sheep that suffer high mortalities will be urgently seeking advice on how to manage the problem to ensure that they can continue to sell sheep to the export trade.

Truckloads of sheep were identified using existing farm tags or individually numbered pink tags either before or upon arrival at export feedlots. All such tagged sheep were held as a separate group in the feedlot and loaded onto one deck aboard ship. During the voyage, the tag numbers from dead sheep were recorded by the ship's Officers. Details of the property of origin were entered into a database and mortality data added after each voyage. Line identification was conducted in the second half of each year only, as this is the period when death rates are higher. Additional mortality data were provided from research voyages conducted from 1985 to 1993 where mortalities were recorded by farm group.

For each consignment, the mortality figure was assigned its percentile ranking, within the month and year of loading, to remove any effects of season and year. Farms were classified as high or low mortality based on their percentile ranking (range 0 to 1) in relation to the selected cut-off points 0.25, 0.33, 0.5, 0.67 and 0.75. The proportion of high mortality farms in years 1 and 2 were compared by Pearson's chi-square analysis for each cut-off point. Farms with location data were used to examine for spatial clustering of high mortality farms. Differences in mortality between rainfall and growing season zones were analysed using the Kruskal-Wallis one-way nonparametric analysis of variance.

A total of 479 lines were monitored in the study of which 474 had property of origin information. Farm line mortality rates ranged from nil to 28.2%. Of the 479 farms monitored, 189 (39.5%) had mortality rates between nil and less than 1% and 96.2% of consignments had death rates below 10%. Half of all deaths were from 14.2% of the consignments.

There was a significant association (P < 0.05) between the category of mortality (high or low) in the first and second years of monitoring for 4 of the 5 percentile ranking cut-off points. The strength of association was greatest when the 25th percentile was used as the cut-off point and not significantly different when the 75th percentile was chosen (P = 0.0946). The overall P values from the spatial analyses indicated that there was clustering in the mortality database. The Kruskal-Wallis one-way nonparametric analysis of variance indicated that there were significant differences (P < 0.001) in mortality grouped by either rainfall or growing zone.

We concluded that mortality was repeatable and that the likely mortality category (high or low) of a line of sheep can be predicted with considerable confidence. In addition to the evidence for repeatability, there was a strong association between regional location and mortality rates. The association was supported by the spatial analyses, the comparison of average death rates by zone and the comparison of the proportions of high mortality farms by zone. The results showed that there were more high death rate farms and higher death rates in the longer growing season zones of the southwest of Western Australia. We considered that this was consistent with earlier published studies on the factors leading to mortalities. Follow-up studies are indicated to further investigate the reasons for differences in mortality rates with a view to determining practical strategies to reduce the problem.

#### 4. IMPACT ON MEAT AND LIVESTOCK INDUSTRY - NOW AND WITHIN 5 YEARS

Results of this project have not provided any simple solutions to the mortality problem. However, it was only the vitamin B12/ivermectin studies that had the potential to provide a solution that could be implemented immediately. The repeatability study was an interim step leading to further studies, if indeed repeatability was demonstrated to occur, on the reasons why mortality by farm group may be the same in successive years. The metabolic study was preliminary in nature and provided some indication of differences between high and low mortality groups of sheep.

The potential impact of the finding of regional differences in mortality rates is that export buyers could change their pattern of farm selection to avoid higher mortality areas. At present this could done by favouring sheep from the shorter growing season zones. Should export companies choose to record mortalities by farm group, they could develop their own database of the performance of farm groups of sheep and, with our conclusion about repeatability, alter their buying pattern for farms that they determine to be high mortality.

### 5. CONCLUSIONS AND RECOMMENDATIONS

- When administered before export, neither vitamin B12 nor macrocyclic lactones reduced mortalities in sheep during shipping. The benzodiazepine derivative brotizolam did not improve appetite in sheep held under simulated shipping conditions. **Recommendation:** Treatment of sheep before export with vitamin B12, brotizolam or macrocyclic lactones is not recommended.
- 2. There were metabolic differences in sheep from high and low mortality farms. The differences may explain the cause of inappetence during shipping. **Recommendation:** Any further studies to investigate the cause of the failure to eat syndrome should include examination of the metabolic differences between sheep from high and low mortality farms.
- 3. Sheep from farms in the longer growing season zones of the southwest of Western Australia suffered higher death rates than those from the shorter growing season zones. **Recommendation:** Any further studies to investigate the cause of the failure to eat syndrome should include examination of the reasons for the regional variation in mortality.
- 4. Mortality by farm group was repeatable for consignments exported in different years. Recommendation: Any further studies to investigate the cause of the failure to eat syndrome should be based on comparisons of high and low mortality farms to determine differences.

### 6. PUBLICATIONS

- Dynes RA, Fry JM, Higgs ARB, Adams NR, Richards RB and Norris RT (1995) A metabolic study of high and low mortality farms. Final Technical Report. CSIRO Division of Animal Production and Agriculture Western Australia.
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