

**The impact of some management stressors on the acute phase protein  
haptoglobin in beef cattle**

**Lysandra Lyn Slocombe**

Bachelor of Science, University of Melbourne, 1999

Bachelor of Science (Honours), University of Melbourne, 2000

Graduate Diploma of Education, Australian Catholic University, 2001

Graduate Certificate in Religious Education, Australian Catholic University, 2001

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Co-operative Research Centre for Cattle and Beef Quality

CSIRO Livestock Industries, Armidale, NSW

and

School of Rural Science and Agriculture, The University of New England

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**Declaration**

The research reported in this thesis is original, except where clearly acknowledged in the text. This thesis, or part thereof, has not been submitted for a higher degree to any university. All sources used in the preparation of this thesis have been cited in the bibliography, and all assistance noted in the acknowledgements.



*Lysandra Slocombe*

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## List of Major Abbreviations

ACTH	Adrenocorticotrophic hormone
APP	Acute Phase Protein
Au	Australian dollars
CPK	Creatine phosphokinase
CRP	C-reactive protein
CV	Coefficient of Variation
EDTA	Ethylenediamine tetra-acetic acid
Hb	Haemoglobin
Hb-Hp	Haemoglobin-Haptoglobin Complex
Hct	Haematocrit
Hp	Haptoglobin
HPA	Hypothalamic-Pituitary-Adrenal axis
IFN	Interferon
IgG	Immunoglobulin G
IL	Interleukin
IL-1	Interleukin-1
IL-6	Interleukin-6
LDH	Lactic dehydrogenase
MCH	Mean Corpuscular Haemoglobin
MCHC	Mean Corpuscular Haemoglobin Concentration
MCV	Mean Corpuscular Volume
NFI	Net Feed Intake
PBS	Phosphate Buffered Saline
PCV	Packed Cell Volume
RBC	Red Blood Cells
RPM	Revolutions per minute
SAA	Serum amyloid A
SGOT	Serum glutamic oxalacetic transaminase
SGPT	Serum glutamic pyruvic transaminase
Th	T-helper cells
TMB	Tetramethylbenzidine
TNF	Tumour necrosis factor

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TNF- $\alpha$       Tumour necrosis factor -  $\alpha$   
WBC          White Blood Cell Count

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## Abstract

Assessment of the response of cattle to stressors has previously relied on behavioural changes and physiological responses, in particular cortisol concentrations. This thesis examines the impact of some management practices on the acute phase protein, haptoglobin, in beef cattle. Haptoglobin is readily measured in plasma using a biochemical assay of the peroxidase activity generated by the binding of haptoglobin to haemoglobin. It was found that the accuracy of this method was reduced by haemolysis occurring at the time of blood sampling. The assay presented in this thesis used the peroxidase nature of the haemoglobin-haptoglobin complex in the presence of hydrogen peroxide to catalyse a redox reaction which results in a chromogen changing colour. The assay is highly repeatable (92%) and robust. A correction equation was developed to adjust estimated haptoglobin values for the effect of haemolysis caused at the time of blood collection. The correction equation incorporated measures of plasma haemoglobin concentration and endogenous peroxidase activity present in the sample.

Two sites commonly used in cattle for collection of blood samples are the jugular vein and coccygeal vessels. The impact of blood collection site on haematological variables and haptoglobin was examined. Total white blood cell count, lymphocytes, basophils, red blood cells and platelets differed significantly ( $P < 0.05$ ) between the two sites. Basophil counts had a higher concentration in blood collected at the tail. The other variables that significantly differed were lower in the tail vessels compared to the jugular vein. Haptoglobin concentrations did not differ ( $P > 0.05$ ) between blood samples from the two sites.

The effect of the production stressors; weaning, transport; intensive management; muscle biopsy and social re-grouping on haptoglobin were analysed. Haptoglobin was significantly increased by weaning ( $F=4.227$ ,  $P=0.048$ ), transportation ( $t=-2.449$ ,  $P=0.016$ ) and intensive management ( $t=-3.191$ ,  $P=0.002$ ). Preliminary ranking of the stressors based on their haptoglobin concentrations from greatest to least stress was weaning  $>$  transport  $\geq$  feedlot environment  $\gg$  social re-grouping = muscle biopsy. Correlation analyses between haptoglobin and various variables revealed that there were strong correlations between haptoglobin immediately following transport and net-feed intake ( $r=0.594$ ), average daily gain ( $r=-0.381$ ), and feed conversion rate ( $r=0.545$ ). Haptoglobin was also found to be an indicator of non-inflammatory (disease or tissue injury) stress due to increased haptoglobin concentrations not being attributed to inflammation or infection.

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These studies indicate that haptoglobin can provide an alternative measure of the production stressors, weaning, transportation and intensive management and may be a possible measure of psychological stress. Like all measures of stress haptoglobin needs to be used in conjunction with other stress measures to ensure that the correct conclusion may be drawn about the stressor. This is particularly evident for haptoglobin as it is also a measure of inflammation.