

MASTER OF PHILOSOPHY

THESIS

Individual differences in coping style influence acute
endocrine and neurobiological responses to psychosocial
stress.

Louise Marie Masters

B Biomed Sci

March 2009

Statement of Originality

This work contains no material, which has been accepted for the award of any other degree or diploma in any university or other tertiary institution to the best of my knowledge and belief. The work embodied in this thesis does form the basis of a scholarly work of which I am a joint author, and I have included a statement endorsed by my supervisor clearly outlining the extent of my contribution to the joint publication. I give consent to this copy of my Thesis being made available for loan and photocopying, when deposited in the University Library, subject to the provisions of the Copyright Act 1968.

Candidate signature: Louise Masters

Acknowledgement of authorship/collaboration

I hereby certify that the work embodied in this Thesis is the result of original research, the greater part of which was completed subsequent to admission to candidature for the degree and comprises the major contribution of behavioural testing and analysis, tissue collection and processing, immunohistochemistry, hormone detection assays, brain imaging, Fos detection, statistical analysis, and academic writing to the joint scholarly work.

Supervisor signature: Trevor Day

Acknowledgements

Many thanks.

Trevor Day. Words are inadequate to describe my experience with my supervisor.

Robert Dielenberg. A valuable guide in model development, and a voice that brought a difference to our work.

Peter Dunkley. For advice I am grateful to have heard.

Rohan Walker. Help with RIAs, behavioural assays, statistics, and writing the scholarly publication on which we are co-authors.

My family and friends, who have my heartfelt gratitude.

NHMRC provided funds for this research.

Table of Contents

CHAPTER 1.....	12
GENERAL INTRODUCTION.....	12
The impact of stress on health.....	12
Limiting the impact of stress	13
Individual differences in responses to stress	14
Approaches to the study of individual differences in neural mechanisms that underpin responses to stress.....	15
i) Should studies be clinical or pre-clinical?	15
ii) Which neural mapping techniques are suitable for identifying critical brain circuitry?	16
iii) Which stressor(s) should be used to assess individual differences in responses to stress?	17
A pre-clinical model of psychosocial stress - the resident/intruder model .	18
Individual differences in coping style	20
Using the concept of coping style to explain individual differences in the response to social conflict interactions	23
Neural activation patterns associated with stress	24
Neural activation patterns associated with acute social conflict	27
Neural regions of potential importance to the adoption of different coping strategies	32
Impact of coping style on conflict-induced neural activation patterns	35
Study aims and general approach	37
CHAPTER 2.....	39
Impact of coping behaviour adopted during social conflict on acute HPA axis response.....	39

Table of Contents

INTRODUCTION	39
METHODS	41
Study design	41
Animals and housing	42
Defensive burying test	43
Surgery	43
Resident/intruder model preparation	44
Resident training and selection	45
Intruder preparation	46
Social conflict interaction protocol	46
Social conflict behaviour observations	46
Data analysis	50
RESULTS	51
Behaviours displayed during social conflict interaction episodes	51
Relationship between social conflict behaviour and plasma corticosterone	52
Relationship between defensive burying, social conflict interaction behaviours, and post-conflict interaction plasma corticosterone levels.	54
Analysis of corticosterone responses based on social conflict behaviour 'cut-off' criteria	55
DISCUSSION	57
High levels of fight and guard behaviours are associated with a reduced adrenocortical response to social conflict interaction.	58
Previous reports concerning the effect of coping behaviour on responses to social conflict interaction	59

Table of Contents

Failure of the defensive burying test to predict the adrenocortical response to social conflict.	60
Observation of guard behaviour during social conflict interaction.	61
Limitations of the current study	62
Conclusions	62
CHAPTER 3.....	64
Impact of coping behaviours adopted during a social conflict interaction on neural activation	64
INTRODUCTION	64
METHODS	66
Animals, housing, defensive burying, surgery, and social conflict.....	66
Tissue fixation and sectioning.....	66
Immunohistochemistry	67
Brain regions and Fos immunoreactivity quantification.	68
Data analysis	70
RESULTS	71
Relationship between social conflict behaviour and <i>c-fos</i> activation	71
Differences between 'high fight', 'low fight', and control groups	72
Differences between 'high guard', 'low guard', and control groups	74
DISCUSSION	76
Effect of social conflict on patterns of neural activation in the forebrain	77
Conclusion	81
CHAPTER 4.....	82
General Discussion	82
Overview of the current findings	82

Table of Contents

Broader implications of the current findings.....	83
Future directions	85
REFERENCES	87

Synopsis

Thesis Synopsis

The psychosocial stress of social conflict contributes to the development of depression and anxiety in those individuals vulnerable to its effects, yet the factors that contribute to vulnerability remain unclear. Researchers investigating factors such as behaviour and physiology have used the animal resident/intruder social conflict model whereby a young male rodent (intruder) is placed into the home cage of an older male (resident) that is trained to attack and defeat all intruders. Findings reported previously have shown that defeated intruders displayed medium to longer-term stress-related changes in behaviour and physiology, with considerable variability in the severity of these changes reported from one individual to another. Interestingly, a reduction in severity of behavioural and physiological changes was associated most significantly with intruders that deployed 'active coping' behaviours during the social defeat interaction than animals that deployed 'passive coping'. However, these findings do not describe the short-term effects, raising the question; does coping style also influence the short-term stress response?

We investigated the relationship between coping behaviour adopted by intruders during a 10 minute social conflict culminating in defeat and both acute peak plasma corticosterone (CORT) stress hormone levels and number of cells expressing Fos protein in eight brain regions. Our investigations revealed that higher levels of fight and guard behaviours were associated with lower peak plasma CORT levels compared to ready submission, and that higher levels of fight were associated with fewer numbers of Fos-ir cells in prefrontal cortex (PFC), amygdala (Am), and paraventricular nucleus (PVN) brain regions. In general terms, these findings indicate that coping behaviour deployed *during*

Synopsis

social conflict influences the endocrine and neurobiological elements of the acute phase of the HPA axis response to psychosocial stress. Intruders that deploy an 'active' coping style including fight behaviours display significantly smaller physiological and neurobiological alterations in the acute response than intruders that deploy a 'passive' coping style during social conflict. These results demonstrate that the vulnerability to the effects of psychosocial stress are ameliorated by actively engaging with the perpetrator rather than passively taking the attack, and that adopting the behaviour fight is most protective. Further elucidation of the neural mechanisms that underpin the reduction in stress-induced effects is warranted.

Abbreviations

Abbreviation	Definition
ABC	Avidin:Biotinylated enzyme Complex
ACEC	Animal Care and Ethics Committee
ACTH	adrenocorticotrophic hormone
aHyp	anterior hypothalamus
AHipA	amygdalohippocampal area
Am	amygdala
ANOVA	analysis of variance
AUC CORT	area under the curve for plasma corticosterone
AVP	arginine vasopressin
Base CORT	plasma corticosterone levels prior to defeat encounter
BNST	bed nucleus of the stria terminalis
BSA	bovine serum albumin
CA	catecholamine
CeA	central amygdala
CG	cingulate gyrus
CN	cuneiform nucleus
CORT	corticosterone
CRF	corticotrophin-releasing factor
DAB	diaminobenzidine
dBNST	dorsal bed nucleus of the stria terminalis
dmH	dorsomedial hypothalamus
dR	dorsal raphe
fMRI	functional magnetic resonance imaging
h	hour
HPA	hypothalamic-pituitary-adrenal
IEG	immediate early gene
IL PFC	infralimbic medial prefrontal cortex
ITF	inducible transcription factor
LC	locus coeruleus
IHyp	lateral hypothalamus
IPAG	lateral periaqueductal grey
IPOA	lateral preoptic area
LS	lateral septum
MeA	medial amygdala
mPFC	medial prefrontal cortex
mPOA	medial preoptic area
mR	medial raphe
mRNA	messenger ribonucleic acid
NSRI	noradrenaline serotonin reuptake inhibitor
NTS	nucleus of the solitary tract
OT	oxytocin
PACAP	pituitary adenylate cyclase-activating polypeptide
PAG	periaqueductal grey
PBS	phosphate buffered saline
Peak CORT	maximum plasma corticosterone level 15 or 30 min after onset of the defeat encounter
PFC	prefrontal cortex
PL PFC	prelimbic medial prefrontal cortex
PVN	paraventricular nucleus

Abbreviations

S	septum
s	second
SEM	standard error of the mean
SHypN	septo hypothalamic nucleus
SNS	sympathetic nervous system
SON	supraoptic nucleus
SSRI	selective serotonin reuptake inhibitor
TH	tyrosine hydroxylase
vBNST	ventral bed nucleus of the stria terminalis
VLM	ventrolateral medulla
vIPAG	ventrolateral periaqueductal grey
vLS	ventral lateral septum
vmHyp	ventromedial hypothalamus