MATERNAL OVERNUTRITION AND THE REGULATION OF ENERGY BALANCE AND APPETITE BEFORE AND AFTER BIRTH

Beverly Sara Mühlhäusler B.Sc. (Hons)

Discipline of Physiology

School of Molecular and Biomedical Science

The University of Adelaide

South Australia

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“The Road goes ever on and on
Down from the door where it began.
Now far ahead the Road has gone,
And I must follow, if I can,
Pursuing it with eager feet,
Until it joins some larger way
Where many paths and errands meet.
And whither then? I cannot say.”

-J.R.R. Tolkien, The Fellowship of the Ring

DECLARATION

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Beverly S Muhlhausler

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PUBLICATIONS ARISING FROM THIS THESIS

1. BS Mühlhäusler, CT Roberts, BSJ Yuen, E Marrocco, H Budge, ME Symonds, JR McFarlane, KG Kauter, P Stagg, JK Pearse, IC McMillen (2003) Determinants of fetal leptin synthesis, fat mass and circulating leptin concentrations in well nourished ewes in late pregnancy. *Endocrinology*, **144**:4947-4954.

2. BS Mühlhäusler, IC McMillen, G Rouzaud, PA Findlay, EM Marrocco, SM Rhind, CL Adam (2004) Appetite regulatory neuropeptides are expressed in the sheep hypothalamus before birth, *Journal of Neuroendocrinology*,**16**: 502-507.

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4. BS Mühlhäusler, CL Adam, PA Findlay, JA Duffield, JR Farlane, KG Kauter, IC McMillen, Programming of postnatal appetite regulation: maternal overnutrition increases hypothalamic expression of POMC in the postnatal lamb (*FASEB Journal*, in press)

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1. IC McMillen, CL Adam, BS Mühlhäusler (2005) Early origins of obesity: programming the appetite regulatory system *Journal of Physiology*, **565**:9-17.

2. IC McMillen, BS Mühlhäusler, JA Duffield, BSJ Yuen (2004) Fetal nutrition and the regulation of leptin synthesis and secretion before birth, *Proceedings of the Nutrition Society*, **63**: 405-412.

3. BSJ Yuen, PC Owens, BS Mühlhäusler, CT Roberts, ME Symonds, Keisler DH, McFarlane JR, Kauter KG, Evens (2003) Leptin alters the structural and functional characteristics of adipose tissue before birth. *FASEB Journal*. **17**: 1102-1104.

4. IC McMillen, JA Duffield, BS Mühlhäusler Perinatal programming of adult obesity (book chapter) in *Perinatal Programming* by Hodgson & Coe

5. IC McMillen, LJ Edwards, JA Duffield, BS Mühlhäusler The regulation of leptin synthesis and secretion before birth: implications for the early programming of adult obesity. (Reproduction, in press).

6. BS Mühlhäusler, CT Roberts, JR McFarlane, KG Kauter, IC McMillen (2002) Fetal leptin is a signal of fat mass independent of maternal nutrition in ewes fed at or above maintenance energy requirements. *Biology of Reproduction*, **67**:493- 499.

COMMONLY USED ABBREVIATIONS

A B C

ad libitum	to any desired extent
AGRP	agouti related protein
ANOVA	analysis of variance
ARC	arcuate nucleus
ATP	adenosine triphosphate
BAT	brown adipose tissue
BMI	body mass index
bp	base pairs
cDNA	complementary deoxyribonucleic acid
cAMP	cyclic AMP
CART	cocaine- and amphetamine-regulated transcript
CNS	central nervous system
CoA	Coenzyme A

D E F G

d	day(s)
DNA	deoxyribonucleic acid
dsDNA	double stranded deoxyribonucleic acid
DMH	dorsomedical hyopthalamus
EDTA	ethylenediamine tetraacetic acid
ELISA	enzyme linked immunosorbent assay
FABP	fatty acid binding protein
FAS	fatty acid synthase
FFA	free fatty acids
g	gravitational acceleration
GDP	guanosine diphosphate
GDM	gestatational diabetes mellitus
GH	growth hormone
G3PDH	glyceraldehyde-3-phosphate

H I J K L

h	hour(s)
Hb	arterial haemoglobin content
HPA axis	hypothalamo-pituitary-adrenal axis
HSL	hormone sensitive lipase
i.c.v.	intracerebroventricular
IDM	infant of the diabetic mother
IGFs	insulin-like growth factors
IGF-I	insulin-like growth factor I
IRS	insulin receptor substrate
i.v.	intravenous
JAK	Janus Kinase
LPL	lipoprotein lipase
LH	lateral hypothalamic area

M N O

mRNA	messenger ribonucleic acid
min	minute(s)
α-MSH	alpha-melanocortin stimulating hormone
MCH	melanocortin concentrating hormone
MCR	melanocortin receptor
ME	medien eminence
NE	noradrenaline
NEFA	non-esterified free fatty acids
NPY	neuropeptide Y
O_2 content	arterial oxygen content
ObR $_{(a-e)}$	leptin receptor isoform (a-e)

P Q R S

PCO_2	arterial partial pressure of carbon dioxide
PO_2	arterial partial pressure of oxygen
POMC	proopiomelanocortin

PAT	perirenal adipose tissue
PVN	paraventricular nucleus
rRNA	ribosomal ribonucleic acid
RT-PCR	reverse transcription polymerase chain reaction
SEM	standard error of the mean
SPSSX	statistical package for social sciences on a vax mainframe computer
SCAT	subcutaneous adipose tissue
SNS	sympathetic nervous system
SSC	saline sodium citrate
STAT	signal transducer activator of transcription

T U V W X Y Z

TG	triacylglycerol (triglyceride)
UCP-1	uncoupling protein 1
VMH	ventromedial hypothalamus
WAT	white adipose tissue

ABSTRACT

Based on a large series of epidemiological studies, it has been proposed that exposure to an increased nutrient supply before birth increases the risk of developing obesity in postnatal life. The physiological mechanisms underlying the association between increased nutrition before birth and later obesity are, however, poorly understood. This thesis has investigated the impact of an increased fetal nutrient supply on the programming of key systems within the appetite-regulating network and/or the adipocytes before and after birth.

The studies in this thesis have demonstrated that plasma concentrations of the adipostatic hormone leptin are directly related to adiposity and the size of adipose cells in fetuses of ewes fed at or above maintenance energy requirements, which suggests that leptin may act as a peripheral signal of fat mass before birth. It has also been demonstrated that the components of the central network for appetite regulation are expressed in the hypothalamus of the fetal sheep from at least 110 d gestation (term = 150 ± 3 d gestation), and that the expression of the appetite-regulating neuropeptides is responsive to signals of increased nutrient supply before birth.

This thesis has also demonstrated that an increase in maternal nutrition in late pregnancy results in increases in both food intake and glucose concentrations in the lamb in early postnatal life and in a significant increase in subcutaneous adiposity on postnatal day 30. Importantly, increased maternal nutrition resulted in an altered relationship between signals of increased fat mass and nutrition and expression of a central appetite-inhibitory neuropeptide, CART, in the lamb hypothalamus. It was also demonstrated that there was an interaction between the prenatal and postnatal nutritional environments in the determination of lipogenesis in the early postnatal period.

The findings presented in this thesis provide evidence that programmed changes to the sensitivity of the appetite-regulating neuropeptides to signals of increased adiposity and nutritional status in early postnatal life are an important part of the physiological pathway through which exposure to an increased nutrient supply before birth may lead to an increased risk of obesity in later life.