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The effects of prolactin on prolactin receptor gene
expression and wool growth in Romney ewes.

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Abstract

The effect of exogenous prolactin on prolactin receptor (PRLR) gene expression and wool growth in pregnant and non-pregnant Romney ewes was assessed. Three experiments were performed where exogenous prolactin was administered by subcutaneous injection (daily for 18 days) or constant infusions (for 3, 9 or 18 days) and endogenous prolactin secretion was altered by exposing ewes to long day or short day photoperiods. Prolactin administration started a week after mating (in autumn), or in non pregnant ewes in mid-spring. Blood samples were collected for measurement of circulating prolactin by radioimmunoassay, skin biopsies were collected for the quantification of PRLR long (PRLR-L) and PRLR short form (PRLR-S) mRNA expression using real-time PCR assay. Wool patch samples were clipped monthly for assessing wool growth.

Constant prolactin infusion of more than 3 days activated a positive feedback mechanism for PRLR-L synthesis, resulting in a sustained elevation PRLR-L mRNA expression for up to 38 days after infusion was over. This was associated with short- and long-term stimulation of wool growth in the pregnant Romney ewe. The main increase in wool production happened after parturition. This positive effect on wool growth by prolactin treatment was related to the length of prolactin treatment. A 3 day infusion resulted in a smaller degree of enhancement compared to the 9 days and 18 days. The biggest impact on wool growth was observed in one of the 18 days infused group, which resulted in a 25% increase in clean fibre production when compared to the pregnant group. The expression of PRLR-S mRNA was not associated with an elevation of prolactin levels. Daily injections neither increased PRLR-L mRNA expression nor increased wool growth, demonstrating that a constant and moderate increase in prolactin levels is necessary to stimulate PRLR synthesis.

Data obtained in these trials also suggests that other reproductive hormones may influence PRLR expression and wool growth. The non-pregnant groups showed steady

levels of PRLR-L mRNA expression, which could be associated with changes in hormonal levels due to the reproductive cycle. Seasonal molecules could also interfere with the system, as prolactin manipulation in non-pregnant ewes exposed to an artificial short day environment during spring time showed a different pattern of PRLR-L and PRLR-S mRNA expression and no wool growth effect.

A mathematical model of prolactin/PRLR interaction was shown to be a good predictor of short-term PRLR gene expression, as its simulations agreed with our biological data. However, the inclusion of other gestational and seasonal hormones may be necessary if the model is to be used for simulations of long-term PRLR expression and wool growth during pregnancy and lactation.

Overall, these results suggest that seasonal wool growth can be manipulated via prolactin, which increases PRLR-L mRNA expression resulting in enhancement of wool growth. However, there is a minimum period of constant prolactin elevation necessary to activate this positive feedback mechanism, Also there is a window of opportunity where this mechanism can be manipulated. This window is most likely associated with the animals interpretation of photoperiod, which also regulates the reproductive seasonality and therefore, could as well interact with prolactin in the regulation of PRLR mRNA expression and seasonal wool growth. This observation could lead to the development of products, suitable for on farm conditions, to enhance wool production.

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List of Abbreviations:

aa	Amino acids
BSA	Bovine serum albumin
CFW	Clean fleece weights
CIDR	Controlled internal drug release
CT	Cycle threshold
DEPC	Diethyl Pyrocarbonate
DP	Dermal papilla
DPAR	Pre-development assay reagent
DTT	Dithiothreitol
ECD	Extracellular domain
EDTA	Ethylenediaminetetraacetic acid
FFD	Fleece fibre diameter
FW	High fleece weight Romney
FY	Fleece yield
GABA	Gamma aminobutyric acid
GAP	Gonadotrophin releasing hormone-associated peptide
GAPDH	Glyceroldehyde-3-phosphate dehydrogenase
GFW	Greasy fleece weight
GH	Growth hormone
GHR	Growth hormone receptor
hCG	Human chorionic gonadotropin
ICD	Intracellular domain
IRS	Inner root sheath
JAK 2	Janus kinase 2
LD	Long photoperiod
LIC	Livestock Improvement Corporation
MAPK	Mitogen-activated protein kinase
MCR	Metabolic clearance rate
ND	Natural photoperiod
NRS	Normal rabbit serum
ORS	Outer root sheath
PBS	Phosphate buffered saline

PMSG	Pregnant mare serum gonadotropin
PRLR	Prolactin receptors
PRLR-L	Prolactin receptor long form
PRLR-S	Prolactin receptor short form
PVN	Paraventricular nucleus
RIA	Radioimmunoassay
RT	Reverse transcriptase reaction
SAS	Sheep anti-rabbit serum
SCG	Superior cervical ganglion
SCN	Suprachiasmatic nucleus
SD	Short photoperiod
SOCS	Suppressor of cytokine signalling
STAT	Signal transducer and activator of transcription
TE	Tris/EDTA
TMD	Transmembrane domain
TRH	Thyrotropin-releasing hormone
UBE1	Ubiquitin activating enzyme 1
VIP	Vasoactive intestinal peptide
WRONZ	Wool research organization of New Zealand (Inc)
NIDDK	National institute of diabetics & digestive & kidney diseases