Neural pathways regulating feed intake in ruminants, impacts of disease and relationship to reproduction

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Auburn University, AL USA Appetite control in the hypothalamus is regulated by a complex array of molecules

A complex set of signals is sent to the brain for processing based on food available, nutrients in the digestive system, circulating nutrients and amount of stored energy.



Hypothalamic nuclei regulating feeding - integrating nutrient and endocrine signals

Cross section of brain



PVN--Paraventricular nucleus LH--Lateral hypothalamus DMH--Dorsomedial hypothalamus VMN--Ventromedial nucleus ARC--Arcuate nucleus

Chaillou et al, Eur J Neurosci. 12:4515, 2000 Qi et al. Endocrinology 151: 2106-2116, 2010

Neurotransmitter Regulators of Appetite

Orexigenic

- Neuropeptide Y
- Agouti related protein
- Melanin concentrating hormone
- Orexin

Anorexigenic

- Cocaine and amphetamine regulated transcript
- α-Melanocyte stimulating hormone
- Oxytocin
- Corticotropin releasing hormone
- Thyrotropin releasing hormone

Hypothalamus and Appetite Regulation in Farm Animals



Neuropeptide Y Agouti Related Protein Melanin Concentrating Hormone Orexin α -Melanocyte Stimulating Hormone

Frontal x-ray of sheep brain showing injection radio-opaque dye into lateral ventricle

Agouti Related Protein (AgRP)

Agouti Related Protein (AgRP) found in the Arcuate Nucleus (ARC) Is AgRP a physiological regulator of appetite in ruminants?

- Is AgRP synthesized in the hypothalamus areas regulating feed intake?
- Is AgRP synthesis regulated by a physiological stimulus to feed intake?
- AgRP should increase feed intake when administered into the lateral ventricle of the sheep brain.

Effect of Fasting on AgRP neuron activation in the hypothalamic arcuate nucleus.

Wagner et al. Neuroendocrinology 80:210,2004.

Control



AgRP (cytoplasm; brown stain) colocalization with c-Fos (nucleus; black stain) in the arcuate nucleus of sheep. C-Fos is an index of neuronal activation.

Effect of fasting on AgRP neuron activation in the arcuate nucleus.



Effect of AgRP on Feed Intake in *ad libitum* Fed Sheep



AgRP (2 nmol) differed from controls at 4,6,12 Hours P<0.05. AgRP at 0.2 nmol did not differ from controls.

Wagner et al. Neuroendocrinology 80:210,2004.

AgRP-conclusions

- AgRP stimulates feed intake in sheep.
- AgRP is synthesized in the hypothalamus.
- AgRP is regulated by a physiological appetite stimulus.
- Therefore, AgRP is an endogenous appetite regulator in sheep.



NPY located in the Arcuate Nucleus (ARC)

Effect of ICV administration of NPY on feed intake





Decreased LH Thomas et al. Domest Anim Endocrinol 16:159, 1999.

Variable		Dose of NPY	
	0 µg	50 µg	500 µg
Mean LH (ng/ml)	4.7 ± 1.0	2.9 ± 0.5 *	$2.7\pm0.4*$
Pulse frequency/4 hr	1.6 ± 0.2	1.3 ± 0.5	0.8 ± 0.4
Pulse amplitude (ng/ml)	7.7 ± 2.4	$\textbf{3.5} \pm \textbf{1.4**}$	$2.1\pm0.9^{\boldsymbol{**}}$
Area under the curve	1162 ± 241	$674 \pm 107^{**}$	642 ± 82^{stst}

Abbreviations: LH, luteinizing hormone; NPY, neuropeptide Y; TCV, third cerebroventricular.



ORX located in the Dorsomedial Hypothalamus (DMH) and Lateral Hypothalamus (LH)

Effects of orexin B on feed intake.

Feed intake in pigs (i.m.)



Feed intake in sheep (i.c.v.)



Dyer et al. Domest Anim Endocrinol 16:145, 1999.

Sartin et al. J Anim Sci, 79:1573, 2001.

Melanin Concentrating Hormone (MCH)

Melanin Concentrating Hormone found in the Lateral Hypothalamus (LH) and Dorsomedial Hypothalamus (DMH)

Effect of ICV administration of MCH and NPY on appetite in sheep



Effect at 12 and 24 h.

Whitlock et al. Domest Anim Endocrinol 28:224, 2005.

Orexigenic Neurotransmitters



- ORX-orexin
- MCH-melanin concentrating hormone
- NPY-neuropeptide-Y
- AgRP-agouti related protein

α -Melanocyte Stimulating Hormone (α -MSH)

α-MSH neurons regulating
appetite are found in the ARC of
the hypothalamus

POMC gene expression and α -MSH expression due to fasting



In situ hybridization of POMC mRNA in the ARC nucleus. No effect of fasting. Henry et al. Neuroendocrinology, 2001



Dual labeling immunohistochemistry for c-Fos and α -MSH in the ARC Sartin et al. J Anim Sci, 2008

High Cortisol Response to Adrenocorticotrophic Hormone Identifies Ewes with Reduced Melanocortin Signalling and Increased Propensity to Obesity



Anorexigenic Neurotransmitters



- ORX-orexin
- MCH-melanin concentrating hormone
- MSH- α-melanocyte stimulating hormone
- POMCproopiomelanocortin



Signals from outside the brain

Satiety signals are leptin from fat cells, hepatic oxidation signals and from the GI system there are cholecystokinin (CCK), ghrelin, bombesin, glucagon, glucagon-like peptide-1 (GLP-1), GLP-2, apolipoprotein A-IV, amylin, somatostatin, enterostatin, and peptide YY(3–36) (PYY_{3–36}) Feed intakes (g) in intact and hepatically vagotomized sheep during 3h intraportal sodium propionate or saline (control) infusions

	Intact	Intact	Vagotomy	Vagotomy
Treatment	Control	Sodium Propionate	Control	Sodium Propionate
Feed Intake	300+ 52	87 + 25**	323 + 66	332 + 68

** P<0.01 compared to other treatments (n=6, 5, 7, 6, respectively)

Anil and Forbes. Quarterly J Exp Physiol 73:59, 1988.

Effects of glucose and acetate on feed intake appear inconsistent. Allen et al. J Anim Sci 87:3317, 2009.





Hormonal regulation of feed intake

Adipose signals-Leptin GI signals- Cholecystokinin (CCK) Ghrelin

Leptin

- Leptin is a product of the Ob gene and is synthesized and released from the fat cell of most animals including ruminants.
- Leptin increases plasma GH and LH.



Effect of ICV infusion on feed intake in well fed lambs (P<0.001) but had no effect in fasted lambs



Morrison et al. J Endocrinol 168:317, 2001





Elevated leptin reduces food intake at the hypothalamus.

Hormones and hypothalamic nuclei regulating feed intake



Disease and appetite control

-Plasma Leptin as a disease mediator of appetite inhibition-AgRP to recover appetite

Effect of Endotoxin on Plasma Leptin Concentrations

- Plasma leptin is not affected by parasitism, even with severe reductions in feed intake.
- Greer et al., Br J Nutr 102: 954, 2009; Zaralis et al., Br J Nutr 101:499, 2009
- Salmonella in pigs does not alter circulating leptin.
- Jenkins et al, Domest Anim Endocrinol, 2004.



Soliman et al. Jpn J Vet Res. 50:107 2002. Daniel et al. J Anim Sci, 81:2590, 2003

MSH signaling through the MC4R and disease

- Cachexia consists of relative anorexia, increased metabolic rate, and pathological wasting of lean body mass.
- Central MSH activation of the melanocortin-4 receptor reproduces all features of cachexia. Thus an antagonist might have a role in modulating appetite during disease.





Does endotoxin alter POMC expression in sheep?

Effect of LPS on POMC grains per cell in sheep hypothalamus



Tissue was collected at 60 min after LPS

Does blocking the MC4R prevent the effects of endotoxin?

Effect of AgRP on endotoxin (LPS)- induced cachexia in sheep

Sartin et al. J Anim Sci 86:2557-2567, 2008.



Effect of treatment: (P=0.0271) Time: (P<0.001 Treatment by time: (P=0.0081).

LPS reduced feed intake At 12, 24 and 48 h (P<0.05) compared to control and to AGRP+LPS.

AGRP+LPS did not differ from saline feed intake at any time.

AgRP-Conclusions

- AgRP is a MC4-R antagonist.
- AgRP prevents LPS reductions in feed intake.

 Therefore LPS actions to reduce feed intake may be mediated by (altered α-MSH release and/or acetylation) inhibition of appetite at the MC4-R. Can IV administered pharmaceuticals be developed to improve appetite in disease models

Small molecule MC4R antagonists

Effect of an IV MC4R antagonist (6h)



Relationship between reproductive and appetite control

General relationship between appetite and reproduction

- When reproduction is stimulated appetite is inhibited.
- When appetite is stimulated, reproduction is inhibited.
- Do reproductive regulators alter feed intake?

Effect of Kisspeptin and GnIH on Appetite & Metabolic Systems

Kisspeptin and GnIH are major regulators of GnRH and LH

Effect of Kisspeptin on NPY Neuron Activation



Effect of ICV NPY infusion on plasma GH concentrations in sheep

- GH AUC after i.c.v. injection and infusion of CSF, NPY or Interleukin-1receptor antagonist on GH AUC.
- SAL/CSF- and SAL/NPYtreated sheep (**P<0.05)*.



Effect of ICV Kp-10 on GH release in sheep



Effect of an NPY antagonist (Bibo 3304) on Kp-10 stimulated GH



GnIH on feeding and metabolic gene expression



NPY

f

POMC

Neuropeptide mRNA

GHRH

GALP



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