

Metabolic influences on hypothalamic-pituitary-ovarian function in the pig

P. J. Booth

A.F.R.C. Research Group on Hormones and Farm Animal Reproduction, University of Nottingham, Sutton Bonington, Loughborough, Leics LE12 5RD, UK

Keywords: pig; metabolism; hypothalamus; ovary; pituitary

Introduction

A relationship between nutrition and reproduction has been known to exist for a considerable time, although the physiological mechanisms mediating these effects are only beginning to be elucidated. Body weight, fatness or body composition have all been suggested to play an important role in the long-term control of reproductive function. However, there is now evidence to indicate that nutritional modulation of the reproductive axis can occur in the absence of any change in body weight or composition and that factors regulating the metabolic status of an animal may provide a link between nutrition and reproduction. These physiological signals appear to involve metabolic hormones and substrates.

This review will briefly describe the influence of nutrition on plasma substrate concentrations, metabolic hormones and neurotransmitter activity and the mechanisms by which these physiological signals affect the activity of the hypothalamo-hypophysial-ovarian axis. Subsequently, this paper will review experiments in pigs and other species which have directly investigated the mechanisms by which manipulation of nutrition or metabolic status influence reproductive function during the prepubertal period, in sexually mature animals and also during the post-partum period.

Mechanisms mediating nutritional effects on reproduction

Insulin and metabolic substrates

Nutritional modulation of the plasma concentrations of insulin, glucose, amino acids and free fatty acids (Riis, 1983) provide signals to the brain that influence food intake (Oomura, 1976; Fernstrom, 1983), energy balance and body weight regulation (Porte & Woods, 1981). It is conceivable that these centres may be functionally linked to proximate hypothalamic centres known to be involved in reproductive function. Although direct effects of insulin on LHRH synthesis and release have not been described, the normal function of insulin-receptive tuberoinfundibular neurones in relaying signals to hypothalamic loci may be disturbed in the diabetic condition and may thereby affect LHRH neurones and gonadotrophin release (Rossi & Bestetti, 1981). Glucose is the primary energy substrate of the brain and acts as a precursor to a host of metabolites, some of which are neurotransmitters that have been reported to affect gonadotrophin secretion. These include gamma-aminobutyric acid, glutamate and aspartate. Plasma substrates and insulin also affect pituitary function; Adashi *et al.* (1981) demonstrated that insulin augmented basal and LHRH-stimulated release of LH and FSH from rat pituitary cells *in vitro*, a process that is energy dependent (Sen *et al.*, 1979). At the ovarian level insulin enhances amino acid transport into bovine granulosa cells (Allen *et al.*, 1979), stimulates glucose uptake and utilization (Otani *et al.*, 1985) and low-density lipoprotein metabolism (Veldhuis *et al.*, 1986) and regulates growth and development

of pig granulosa cells (Channing *et al.*, 1976; May & Schomberg, 1981). Insulin also potentiates FSH-stimulated LH receptor induction and progesterone production by pig granulosa cells (May & Schomberg, 1981).

The growth hormone–insulin-like growth factor axis

The plasma concentrations of growth hormone (GH) and insulin-like growth factor-I (IGF-I) are responsive to plane of feeding and dietary composition. In the ovary, GH augments FSH-stimulated LH receptor induction and progesterone biosynthesis by pig granulosa cells (Jia *et al.*, 1986). These actions may be mediated by GH-stimulated IGF-I production in these cells (Hammond *et al.*, 1985). IGF-I stimulates mitogenesis (Baranao & Hammond, 1984), low-density lipoprotein metabolism (Veldhuis *et al.*, 1987) and enhances FSH-stimulated processes such as LH receptor induction and progesterone and oestrogen biosynthesis by pig granulosa cells (Maruo *et al.*, 1988).

Prolactin

Plasma prolactin concentrations in pigs are related to the protein content of a meal (McMurtry *et al.*, 1983) and rise during the postprandial period (Armstrong *et al.*, 1986). The action of prolactin at the ovary (reviewed by McNeilly *et al.*, 1982) is permissive at low concentrations, whereas hyperprolactinaemia inhibits the reproductive axis (reviewed by McNeilly, 1980).

Thyroid axis

Nutrition modulates the blood concentrations of thyroxine, triiodothyronine and reverse triiodothyronine (Dauncey *et al.*, 1983) and in so doing may influence reproduction. At the ovarian level, thyroid hormones in the presence of insulin increase lipid accumulation and stimulate spontaneous and gonadotrophin-induced progesterone production and morphological luteinization of pig granulosa cells (Channing *et al.*, 1976). Similarly, thyroxine and triiodothyronine augment FSH-stimulated processes such as differentiation, LH receptor induction and oestradiol production by pig granulosa cells (Maruo *et al.*, 1987).

Adrenal axis

Plasma glucocorticoid concentrations generally increase during nutritional deficits (Dubey *et al.*, 1986) although exceptions are documented (Britt *et al.*, 1988). Administration of adrenocorticotrophic hormone (ACTH) or hydrocortisone blocks ovulation and the preovulatory LH surge in gilts (Barb *et al.*, 1982). There is also indirect evidence to suggest that adrenal steroids inhibit hypothalamic GnRH release in pigs (Fonda *et al.*, 1984). In pig granulosa cells, cortisol inhibits cell maintenance but, in the presence of insulin, enhances progesterone secretion and spontaneous and gonadotrophin-induced luteinization (Channing *et al.*, 1976). Also, glucocorticoids inhibit FSH-dependent LH receptor induction, aromatase activity and oestrogen production in rat granulosa cells (Schoonmaker & Erickson, 1983).

Catecholamines and the sympathoadrenal system

The catecholamines play a role in the control of gonadotrophin release in the pig (Parvizi & Ellendorff, 1982). As the precursor of these neurotransmitters is tyrosine and the uptake of this amino acid into the brain is dependent upon the ratio of plasma tyrosine to other large neutral amino acids, nutrition can directly regulate the activity of this neurotransmitter system (Fernstrom, 1983).

The sympathoadrenal system consists of the sympathetic nervous system and the adrenal medulla. The activity of both systems is affected by diet. Although the influence of nutritional modulation of sympathoadrenal activity on the gonad has not been investigated, catecholamines have been shown to affect ovarian function (Webley *et al.*, 1988).

Serotonin and acetylcholine neurotransmitter systems

The blood concentration of choline apparently determines the synthesis of the neurotransmitter acetylcholine in the brain (Hirsch & Wurtman, 1978). Like the dietary control of brain catecholamine neurotransmitter synthesis, brain concentrations of serotonin are also controlled by substrate limited pathways (Fernstrom, 1983). Therefore, nutrition directly controls these neurotransmitter systems, both of which are known to be involved in the regulation of gonadotrophin secretion.

Brain-gut peptides

Nutritionally-induced changes in opioid tone may influence gonadotrophin release as reported by Dyer *et al.* (1985) who discovered that fasting activated an opioid pathway that inhibited LH secretion. In pigs, infusion of the opioid antagonist naloxone increased LH secretion in only one-third of nutritionally-induced anoestrous gilts (Armstrong & Britt, 1987). Preliminary investigations in our laboratory (J. R. Cosgrove, P. J. Booth & G. R. Foxcroft, unpublished observations) suggest that naloxone does not increase LH secretion in ad libitum or food-restricted prepubertal gilts.

Apart from opiates, other brain-gut peptides may influence reproductive function. Indeed, cholecystokinin, gastrin, neurotensin and gastric inhibitory peptide which are released into the circulation after feeding have all been demonstrated to affect LH secretion. The physiological significance of these changes on gonadotrophin release awaits investigation.

Studies on metabolic regulation of reproduction in domestic mammals

Conceptual aspects of regulation of puberty

It has been suggested that the onset of puberty is linked to the attainment of a critical body weight, a minimum lean to fat ratio and to a minimum percentage of body fat. Alternatively, metabolic mass and food intake or its correlated metabolic rate may be the triggering mechanisms (see Frisch, 1984). Body weight and hence fatness has significant effects on steroid metabolism (Frisch, 1984; Kirkwood & Aherne, 1985) which may be of importance during puberty and also in the adult. In pigs, Kirkwood & Aherne (1985) concluded from the wealth of data available that the initiation of puberty is dependent upon the attainment of a minimum threshold age and weight.

Cameron *et al.* (1985a) investigated the physiological mechanisms by which developmental changes are linked to sexual maturation by comparing the plasma profiles of circulating hormones and substrates in fed and fasted adult and prepubertal male monkeys: they concluded that the transition from the fed to the fasted state occurred more rapidly in prepubertal monkeys and that the different temporal responses of metabolic hormones and substrates between adults and juveniles were probably related to the greater glucose production rate, higher metabolic rate, smaller body reserves and the growth requirement of the juvenile. Cameron *et al.* (1985b) suggested that the dynamic fluctuations of plasma hormones and substrates that occurred during the post-prandial and postabsorptive periods provide signals to the brain that link metabolic status to the activation of the reproductive system. They speculated that prepubertal animals are "suspended in a fasting state" in which the LHRH pulse generator is effectively starved of substrates and the appropriate hormonal signals suitable for its activation (Steiner *et al.*, 1983).

Nutritional and metabolic manipulation of sexual maturation

To investigate the effect of blood-borne substrates on gonadotrophin release, a sustained intravenous infusion of glucose alone or glucose plus a mixture of essential amino acids was given to castrated juvenile monkeys (Steiner *et al.*, 1983; Cameron *et al.*, 1985a). The results (Fig. 1) showed that, in those monkeys that responded to the combined glucose and amino acid infusion, plasma LH concentration increased within 1 week. However, the sustained infusion of glucose alone had no stimulatory influence on plasma LH concentrations (Steiner *et al.*, 1983).

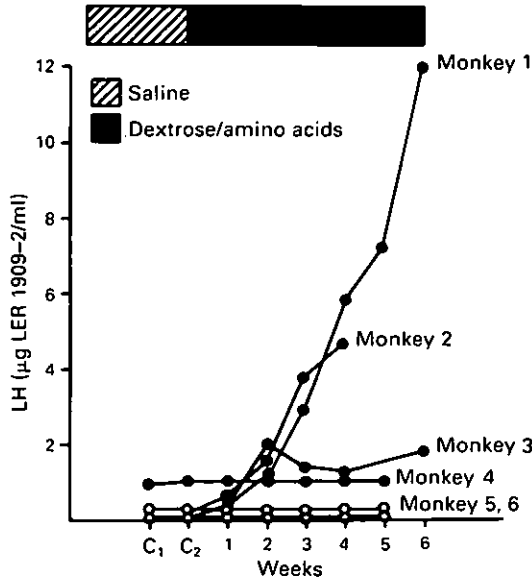


Fig. 1. Plasma LH concentrations in samples collected weekly from 6 prepubertal macaques receiving intravenous infusions of saline (2 control weeks; C) followed by chronic intravenous infusions of dextrose and amino acids (4–6 weeks). Plasma LH increased significantly ($P < 0.05$) in Monkeys 1, 2 and 3 during dextrose/amino acid infusion. Open circles represent values below the limit of assay detectability. (Reproduced from Cameron *et al.*, 1985a.)

Foster *et al.* (1988) have also attempted to discover the mechanism by which growth and nutrition affect gonadotrophin secretion by using the nutritionally-restricted prepubertal lamb as the experimental model. Plasma LH concentrations were basal during the time of dietary restriction but LH pulse frequency dramatically increased within 48 h after realimentation. If the re-fed lambs were returned to the restricted diet, LH pulse frequency could still be maintained by a constant parenteral infusion containing glucose and amino acids (Fig. 2).

In an attempt to elucidate further the mechanisms by which nutrition influences reproduction, Booth (1990) investigated the role of metabolic state on circulating hormones and blood glucose in relation to sexual maturity in prepubertal gilts. Sexual maturity and metabolic state were ultimately compared in littermates at 85 kg body weight and at identical ages after allocation to one of two feeding regimens at 75 kg; these involved either twice-daily feeding to appetite up to 85 kg and then maintenance feeding until slaughter (Group 4), or maintenance feeding from 75 kg and then feeding to appetite to reach 85 kg at the time of study (Group 5) (Fig. 3). Other groups of littermates were studied at the start of treatment (Group 1), and when dietary intakes were changed (Groups 2 and 3). At the time of slaughter the ad libitum-fed animals (Group 5) showed significantly advanced development compared to those in Group 4 in terms of uterine weight, follicular development and the peak LH and FSH responses to an LHRH challenge ($P < 0.01$ and $P < 0.05$, respectively), in the absence of any change in backfat depth or eye muscle area. Furthermore, total plasma

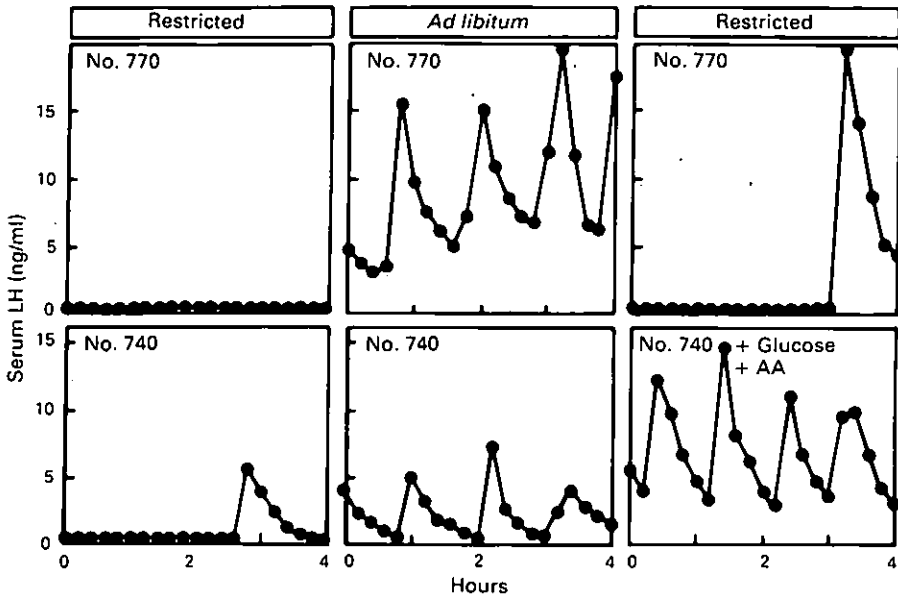


Fig. 2. Circulating LH concentrations in an ovariectomized lamb (No. 770) on low nutrition, after 14 days of realimentation, and 7 days after return to the restricted diet, and in an ovariectomized lamb (No. 740) treated as above except that glucose and amino acids (AA) were infused intravenously for 7 days after the return to the restricted diet. (From Foster *et al.*, 1988.)

IGF-I ($P < 0.001$) and basal ($P < 0.001$) and postprandial ($P < 0.001$) free triiodothyronine concentrations were significantly higher in Group 5 than in Group 4 gilts. Re-feeding induced a significant increase in postprandial insulin concentrations (Group 3 vs Group 5; $P < 0.05$). The maintenance feeding regimen reduced basal plasma glucose concentrations between Groups 1 and 3 ($P < 0.02$) and Groups 2 and 4 ($P < 0.05$) although no significant differences were established between Groups 4 and 5. These results therefore support other suggestions that changes in metabolic status may mediate short-term nutritional effects on reproductive function in the absence of major changes in body composition.

To explore further the physiological mechanisms involved, a second experiment was designed in which the temporal relationships between plasma LH, metabolic hormones and glucose concentrations were investigated. Growth-matched prepubertal littermate gilts were fed a restricted diet designed to maintain body weight at 75 kg for 7 days (Days 1–7). From Day 8 to Day 14, 1 gilt of each pair was fed twice daily to appetite while the other continued to receive the restricted food regimen (Fig. 4). Feeding to appetite increased episodic LH secretion over the 24-h sampling period on Day 8 ($P < 0.001$) and during a 6-h period of sampling on Day 14 ($P < 0.01$); furthermore, the first significant ($P < 0.05$) increase in episodic LH frequency occurred during the first 6 h of sampling on Day 8 (Fig. 5). Total plasma IGF-I concentrations gradually increased over the 7-day sampling period in animals fed to appetite. In the same gilts, meal ingestion induced a rapid rise in plasma insulin to approximately twice the concentrations observed in the postprandial period in the restricted-fed littermates. Furthermore, in the animals fed to appetite, elevated plasma insulin concentrations were sustained between morning and evening feeds, in contrast to maintenance-fed gilts in which values fell to preprandial concentrations within 6 h of feeding (Fig. 6). In conclusion, the observed advancement of utero-ovarian maturation accompanying the induced anabolic state in gilts fed to appetite is probably mediated, at least in part, by enhanced gonadotrophic stimulation; this ovarian response may also be mediated by the effects of elevated plasma concentrations of insulin and IGF-I present in the realimented condition.

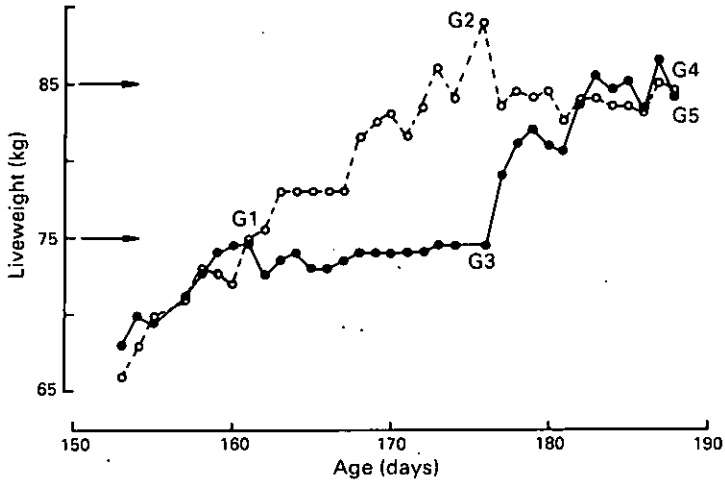


Fig. 3. Typical growth profiles of a littermate pair of prepubertal gilts during the period of dietary manipulation. (From Booth, 1990.)

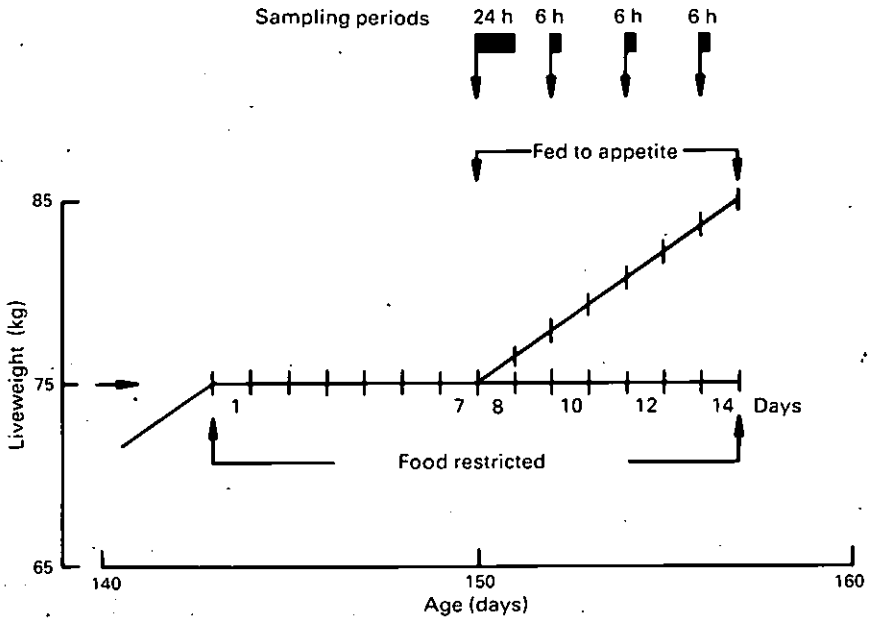


Fig. 4. Experimental protocol for the study of acute effects of alimentation and metabolic state on the endocrine status of prepubertal gilts. (From Booth, 1990.)

The possible involvement of the insulin-glucose system as a mediator of the rapid nutritional enhancement of LH secretion was evaluated in a third experiment in which maintenance feeding was again imposed for 7 days on trios of growth-matched littermate gilts of 75 kg body weight. On Day 8, gilts from each set were allocated to one of three treatment groups: gilts in Group A were fed to appetite at 09:00 h; those in Group M were given continued maintenance feeding and those in

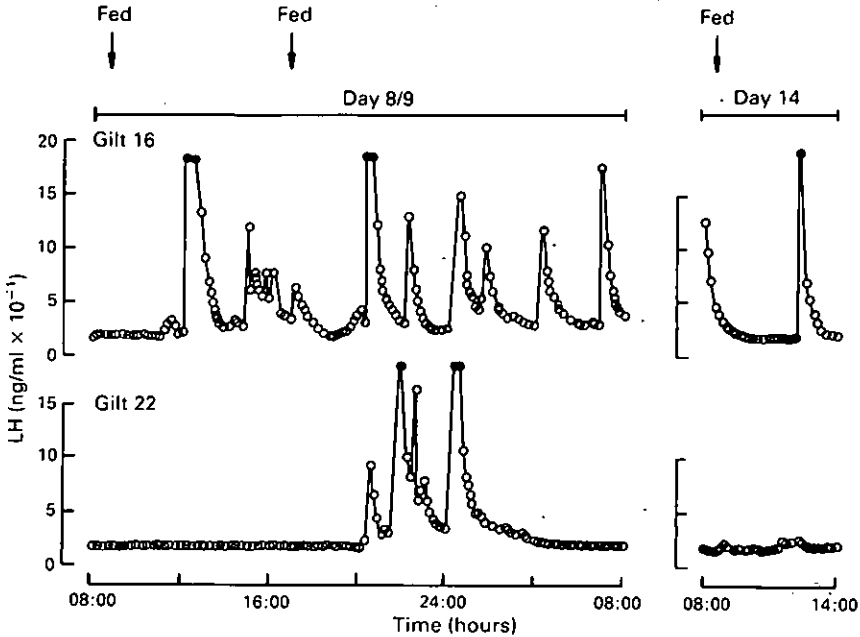


Fig. 5. Typical LH profiles of one littermate pair of prepubertal gilts on Days 8/9 and 14 during a period of dietary restriction of 14 days (Gilt 22) and following realimentation (twice daily feeding to appetite) from Day 8 after 7 days of dietary restriction (Gilt 16). A maturational diurnal pattern of increased episodic LH secretion beginning at approximately 20:00 h is evident in the maintenance-fed gilt. Closed circles represent values below the sensitivity, or the upper limit of detection, of the assay (P. J. Booth & G. R. Foxcroft, unpublished observations).

Group G were given maintenance feeding plus intravenous glucose administration from 09:00 to 14:00 h. Glucose (50 g/100 ml saline), equivalent in energy to the mean difference in food weight ingested between Groups A and M, was administered as 175 ml bolus injections at 09:00, 09:20 and 09:40 h, followed by 40-ml injections at 10-min intervals from 10:00 until 14:00 h. Littermates in Groups A and M received identical injections of saline. Postprandial plasma insulin ($P < 0.05$), mean ($P < 0.05$) and maximum ($P < 0.05$) plasma LH concentrations and episodic LH frequency ($P < 0.01$) were greater in gilts in Groups A and G than in their Group M littermates (Fig. 7). These results demonstrate that glucose administration to restricted-fed gilts and the associated plasma insulin rise induces a rapid increase in episodic LH secretion similar to that observed in response to re-alimentation. The mechanisms mediating this acute enhancement of LH secretion (and therefore GnRH release) have yet to be elucidated. McCann & Hansel (1986) reported that re-feeding of fasted adult heifers induced a rapid increase in plasma LH concentration that occurred in the absence of any change in plasma insulin or glucose concentrations. This may suggest that insulin and glucose played no role in mediating this effect, although plasma glucose concentrations *per se* do not necessarily reflect glucose flux rates. Alternatively, the enhanced secretion of LH may be mediated by the release of brain-gut peptides or by vagal afferents arising from splanchnic tissue that are sensitive to gut fill or nutrient absorption (Novin, 1985); however, such mechanisms would not be expected to mediate the effects of nutrients administered parenterally on reproduction. A rapid, food ingestion-induced, metabolic/neuroendocrine pathway regulating GnRH secretion has been proposed by Bronson & Manning (1988), who reported that food-restricted prepubertal rats show evidence of LH pulsing for several hours following their daily meal, an effect not observed when the meal was missed.

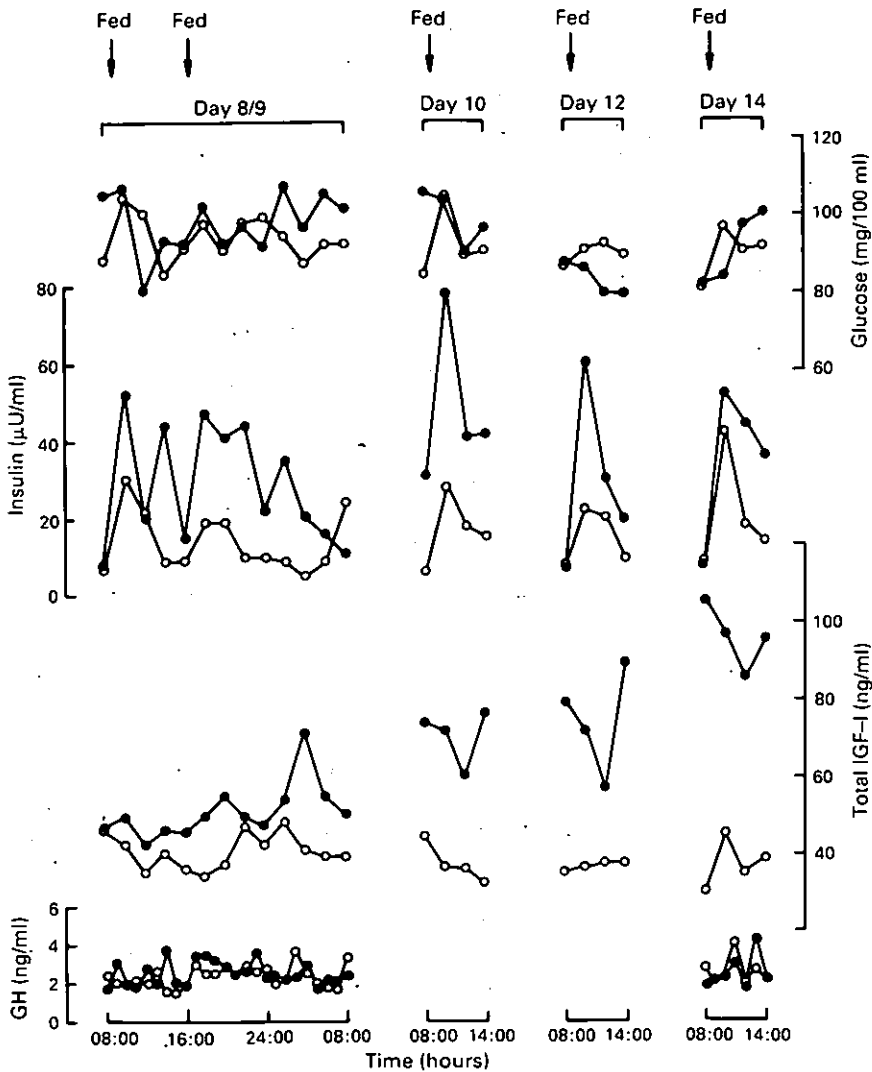


Fig. 6. Plasma concentrations of glucose, insulin, total IGF-I and growth hormone for the same littermates for which data are presented in Fig. 5 during the same experiment (O, Gilt 22, restricted fed; ●, Gilt 16, fed to appetite) (P. J. Booth & G. R. Foxcroft, unpublished observations).

Nutritional and metabolic manipulation of reproduction in the adult

Armstrong & Britt (1987) studied the effect of long-term dietary energy restriction and subsequent realimentation on the reproductive axis and on the metabolism and endocrinology of sexually mature gilts. They reported that animals receiving a restricted diet became acyclic, having lost 15 and 25% of mean body weight and back fat depth, respectively. Mobilization of fat and protein in restricted-fed gilts was associated with lower basal plasma insulin concentrations and a general tendency for higher preprandial free fatty acids and blood urea nitrogen concentrations compared to gilts receiving the control diet. During the postprandial period dietary restriction only transiently reduced free fatty acid concentrations, decreased the plasma insulin rise and induced a greater increase in plasma GH concentrations compared with control gilts. However, postprandial glucose concentrations were not reduced and, surprisingly, basal concentrations were higher in

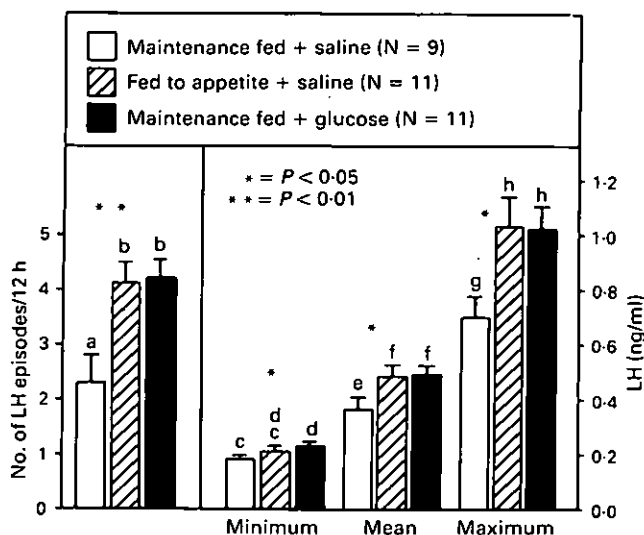


Fig. 7. Analysis of 12-h LH profiles on Day 8 for animals receiving the twice daily maintenance diet plus saline infusion, the maintenance diet plus glucose infusion and the ad-libitum diet (feeding to appetite at 09:00 h) plus saline infusion, after a 7-day period of dietary restriction. Minimum, mean and maximum LH characteristics were calculated using the sliding window technique of Shaw & Foxcroft (1985). Values are mean \pm s.e.m. Values with the same superscripts are not significantly different. (From Booth, 1990.)

restricted than in control-fed animals. Dietary restriction had no chronic effect on plasma cortisol concentrations. These metabolic adaptations were accompanied by reduced LH pulse frequency and follicular development in restricted-fed gilts. Metabolic variables were measured 2 weeks after re-alimentation, by which time no significant differences were evident between control- or restricted-fed gilts, and yet gilts did not become cyclic until the 6th week after re-alimentation. Plasma LH concentrations increased linearly over this 6-week period. These results suggest that (i) the plasma concentrations of the metabolic variables measured had no direct influence on the GnRH pulse generator, (ii) the response to these metabolic changes was very slow, or (iii) other metabolic factors were involved.

The effect of energy and insulin administration on reproduction in pigs has been studied by Cox *et al.* (1987). In their experimental design gilts received a high energy or control diet and short- or long-acting insulin or acted as controls. In the two experiments performed, insulin had a variable effect on plasma LH concentrations but consistently increased ovulation rate, which suggests that insulin had a direct stimulatory effect on ovarian function (Table 1). Further studies (Britt *et al.*, 1988) have demonstrated that insulin administration reduced follicular atresia and increased plasma oestradiol concentrations during the follicular phase. Stimulation of ovarian function may have been mediated directly by insulin or indirectly by elevated concentrations of plasma GH observed in insulin-treated gilts. Plasma glucose concentrations bore no relationship to reproductive status: long- and short-acting insulin induced considerable differences in glycaemia but both increased ovulation rate compared to controls.

Nutritional and metabolic manipulation of reproduction *post partum*

To investigate relationships between metabolism and the occurrence of post-weaning anoestrus, Armstrong *et al.* (1986) fed primiparous sows either an ad-libitum or a restricted diet during lactation. Their results demonstrate that diet had no effect on plasma LH or oestradiol concentrations or subsequent reproductive performance, although body condition and metabolism

Table 1. Effect of energy intake and insulin on number of corpora lutea in gilts (from Cox *et al.*, 1987)

	Insulin (IU/kg body wt)		Energy mean
	0	0.1	
Energy			
Control	13.4 ± 1.5	14.6 ± 1.3	14.0 ± 1.3
High	15.8 ± 1.5	19.4 ± 1.2	17.6 ± 0.9*
Insulin mean	14.6 ± 1.0	17.0 ± 0.9†	

Values are mean ± s.e.m.

*Main effect of energy ($P < 0.01$).

†Main effect of insulin ($P < 0.05$).

were affected. Weight and backfat changes during lactation were identical between oestrous and anoestrous sows but backfat and heartgirth changes during lactation were related to the weaning to oestrus interval in those sows that exhibited oestrus within 8 days after weaning. In sows that exhibited oestrus, plasma LH pulse frequency was greater before weaning compared with anoestrous sows, but at 6 h after weaning the situation was reversed (Armstrong & Britt, 1984). Basal concentrations of prolactin and the preprandial prolactin to insulin ratio were greater during late lactation in anoestrous sows than in sows exhibiting oestrus. Plasma profiles of preprandial glucose and free fatty acid concentrations were also different during lactation between oestrous and anoestrous sows. Armstrong *et al.*, (1986) speculated that altered energy metabolism during lactation could contribute to post-weaning reproductive performance although the mechanism by which this was effected could not be precisely determined. Furthermore, litter size in sows that exhibited a post-weaning oestrus was related to plasma glucose and insulin concentrations (Armstrong & Britt, 1984).

In comparison with the above data, Baidoo (1989) allotted either a high or low food intake to sows during lactation and induced a significant reduction in the length of the remating interval in animals fed the high food intake. Measured between Days 2 and 28 of lactation, weight loss ($P < 0.01$) and backfat loss ($P < 0.01$) were greater, whereas GH ($P < 0.05$) and cortisol ($P < 0.05$) increased to a greater extent in sows receiving the low food intake. During the same period plasma insulin concentrations increased to a greater extent ($P < 0.05$) in sows allotted the high food intake, but plasma prolactin decreased to a similar extent in both treatment groups. Although changes in LH, FSH and oestradiol were similar between the two groups, it is possible that altered metabolic status during lactation was responsible for the subsequent reproductive performance.

Conclusions

The literature reviewed in this paper has described nutritional modulation of reproductive function, the time scale of which may be hours, days or weeks. The available evidence suggests that these responses could be mediated by a host of metabolic signals: these include vagal signals rising from the alimentary canal, brain-gut hormones, other metabolic hormones, or plasma substrate concentrations or fluxes. As parenterally infused nutrients influence gonadotrophin secretion, one should conclude that gastrointestinal signals contribute little to this effect, although the study performed by McCann & Hansel (1986) suggests that this may not always be true. Other metabolic signals modulating gonadotrophin secretion include altered oestrogen metabolism and therefore steroid feedback as a consequence of long-term changes in body weight or composition. The effects of undernutrition on reproductive dysfunction appear to be located primarily at the hypothalamic

level as pulsatile administration of GnRH can induce gonadotrophin release and follicular development in restricted-fed prepubertal rats (Bronson & Manning, 1988), nutritionally anoestrous gilts (Armstrong & Britt, 1987) and lactating and anoestrous weaned sows (Britt *et al.*, 1985). The experimental evidence presented in this paper indicates that energy metabolism provides one of the fundamental links between nutrition and reproduction. Insulin-regulated glucose metabolism is probably a signal of major importance sensed by the brain. Although the absolute concentrations of plasma glucose and insulin are important, under certain conditions they are not correlated to reproductive activity: experimental or pathological alteration of glucose metabolism (e.g. diabetes, insulin-induced hypoglycaemia or glucose infusion) all suggest that the rate of glucose utilization influences the activity of the LHRH pulse generator. Apart from insulin and glucose, other nutritionally responsive substrates and metabolic hormones have been reported to influence reproduction. However, the paucity of information documenting nutrition–metabolism–reproduction interactions does not permit any firm conclusions regarding the relative contribution of these factors to the control of reproductive function. It is evident, however, from the data presented in this paper that metabolic status has a profound effect on hypothalamic–pituitary–ovarian function in the pig.

References

- Adashi, E.Y., Hsueh, A.J.W. & Yen, S.S.C. (1981) Insulin enhancement of luteinizing hormone and follicle-stimulating hormone release by cultured pituitary cells. *Endocrinology* **108**, 1441–1449.
- Allen, W.R., Nilsen-Hamilton, M., Hamilton, R.T. & Gospodarowicz, D. (1979) Serum-dependent regulation of α -aminoisobutyric acid uptake in bovine granulosa cells. *J. cell Physiol.* **98**, 491–502.
- Armstrong, J.D. & Britt, J.H. (1984) Effect of energy restriction during lactation on reproductive performance, energy metabolites and endocrine changes in primiparous sows. *Proc. 10th Int. Congr. Anim. Reprod. & A.I., Urbana-Champaign* Abstr. 157.
- Armstrong, J.D. & Britt, J.H. (1987) Nutritionally-induced anestrus in gilts: metabolic and endocrine changes associated with cessation and resumption of estrous cycles. *J. Anim. Sci.* **65**, 508–523.
- Armstrong, J.D., Britt, J.H. & Kraeling, R.R. (1986) Effect of restriction of energy during lactation on body condition, energy metabolism, endocrine changes and reproductive performance in primiparous sows. *J. Anim. Sci.* **63**, 1915–1925.
- Baidoo, S.K. (1989) *The effect of weight and body fat loss on the reproductive performance and endocrinological status of the lactating and postweaning sow*. Ph.D. thesis, University of Alberta.
- Baranao, J.L.S. & Hammond, J.M. (1984) Comparative effects of insulin and insulin-like growth factors on DNA synthesis and differentiation of porcine granulosa cells. *Biochem. Biophys. Res. Commun.* **124**, 484–490.
- Barb, C.R., Kraeling, R.R., Rampacek, G.B., Fonda, E.S. & Kiser, T.E. (1982) Inhibition of ovulation and LH secretion in the gilt after treatment with ACTH or hydrocortisone. *J. Reprod. Fert.* **64**, 85–92.
- Booth, P.J. (1990) *Physiological mechanisms mediating nutrition-reproduction interactions in the gilt*. Ph.D. thesis, University of Nottingham.
- Britt, J.H., Armstrong, J.D., Cox, N.M. & Esbenshade, K.L. (1985) Control of follicular development during and after lactation in sows. *J. Reprod. Fert., Suppl.* **33**, 37–54.
- Britt, J.H., Armstrong, J.D. & Cox, N.M. (1988) Metabolic interfaces between nutrition and reproduction in pigs. *Proc. 11th Int. Congr. Anim. Reprod. & A.I., Dublin*, Vol. 5, pp. 117–125.
- Bronson, F.H. & Manning, J. (1988) Food, energy expenditure and puberty in female rats. *Proc. 11th Int. Congr. Anim. Reprod. & A.I., Dublin*, Vol. 5, pp. 109–116.
- Cameron, J.L., Hansen, P.P., McNeill, T.H., Koerker, D.J., Clifton, D.K., Rogers, K.V., Bremner, W.J. & Steiner, R.A. (1985a) Metabolic cues for the onset of puberty in primates. In *Adolescence in Females*, pp. 59–78. Eds J. Givens, C. Flamingi & S. Venturoli. Year Book Medical Publishers, Chicago.
- Cameron, J.L., Koerker, D.J. & Steiner, R.A. (1985b) Metabolic changes during maturation of male monkeys: possible signals for onset of puberty. *Am. J. Physiol.* **249**, E385–E391.
- Channing, C.P., Tsai, V. & Sachs, D. (1976) Role of insulin, thyroxine and cortisol in luteinization of porcine granulosa cells grown in chemically defined media. *Biol. Reprod.* **15**, 235–247.
- Cox, N.M., Stuart, M.J., Althen, T.G., Bennett, W.A. & Miller, H.W. (1987) Enhancement of ovulation rate in gilts by increasing dietary energy and administering insulin during follicular growth. *J. Anim. Sci.* **64**, 507–516.
- Dauncey, M.J., Ramsden, D.B., Kapadi, A.L., Macari, M. & Ingram, D.L. (1983) Increase in plasma concentrations of 3,5,3'-triiodothyronine and thyroxine after a meal, and its dependence on energy intake. *Horm. Metab. Res.* **15**, 499–502.
- Dubey, A.K., Cameron, J.L., Steiner, R.A. & Plant, T.M. (1986) Inhibition of gonadotropin secretion in castrated male rhesus monkeys (*Macaca mulatta*) induced by dietary restriction: analogy with the prepubertal hiatus of gonadotropin release. *Endocrinology* **118**, 518–525.
- Dyer, R.G., Mansfield, S., Corbert, H. & Dean, A.D.P. (1985) Fasting impairs LH secretion in female rats by activating an inhibitory opioid pathway. *J. Endocr.* **105**, 91–97.

- Fernstrom, J.D. (1983) Role of precursor availability in control of monoamine biosynthesis in brain. *Physiol. Rev.* **63**, 484–546.
- Fonda, E.S., Rampacek, G.B. & Kraeling, R.R. (1984) The effect of adrenocorticotropin or hydrocortisone on serum luteinizing hormone concentrations after adrenalectomy and/or ovariectomy in the prepubertal gilt. *Endocrinology* **114**, 268–273.
- Foster, D.L., Ebling, F.J.P., Vannerson, L.A., Bucholtz, D.C., Wood, R.I. & Micka, A.F. (1988) Modulation of gonadotrophin secretion during development by nutrition and growth. *Proc. 11th Int. Congr. Anim. Reprod. & A.I., Dublin*, Vol. 5, pp. 100–108.
- Frisch, R.E. (1984) Body fat, puberty and fertility. *Biol. Rev.* **59**, 161–188.
- Hammond, J.M., Baranao, J.L.S., Skaleris, D., Knight, A.B., Romanus, J.A. & Rechler, M.M. (1985) Production of insulin-like growth factors by ovarian granulosa cells. *Endocrinology* **117**, 2553–2555.
- Hirsch, M.J. & Wurtman, R.J. (1978) Lecithin consumption increases acetylcholine concentrations in rat brain and adrenal gland. *Science, NY* **202**, 223–224.
- Jia, X.C., Kalmijn, J. & Hsueh, A.J.W. (1986) Growth hormone enhances follicle-stimulating hormone-induced differentiation of cultured rat granulosa cells. *Endocrinology* **118**, 1401–1409.
- Kirkwood, R.N. & Aherne, F.X. (1985) Energy intake, body composition and reproductive performance of the gilt. *J. Anim. Sci.* **60**, 1518–1529.
- Maruo, T., Hayashi, M., Matsuo, H., Yamamoto, T., Okada, H. & Mochizuki, M. (1987) The role of thyroid hormone as a biological amplifier of the actions of follicle-stimulating hormone in the functional differentiation of cultured porcine granulosa cells. *Endocrinology* **121**, 1233–1241.
- Maruo, T., Hayashi, M., Matsuo, H., Ueda, Y., Morikawa, M. & Mochizuki, M. (1988) Comparison of the facilitative roles of insulin and insulin-like growth factor I in the functional differentiation of granulosa cells: in vitro studies with the porcine model. *Acta endocr., Copenh.* **117**, 230–240.
- May, J.V. & Schomberg, D.W. (1981) Granulosa cell differentiation in vitro: effects of insulin on growth and functional integrity. *Biol. Reprod.* **25**, 421–431.
- McCann, J.P. & Hansel, W. (1986) Relationships between insulin and glucose metabolism and pituitary-ovarian functions in fasted heifers. *Biol. Reprod.* **34**, 630–641.
- McMurtry, J.P., Steele, N.C., Bereskin, B., Richards, M.P. & Rosebrough, R. (1983) Dietary protein influences metabolic hormone secretory profiles in growing pigs. *J. Anim. Sci.* **57** (Suppl. 1), 199, Abstr. 163.
- McNeilly, A.S. (1980) Prolactin and the control of gonadotrophin secretion in the female. *J. Reprod. Fert.* **58**, 537–549.
- McNeilly, A.S., Glasier, A., Jonassen, J. & Howie, P.W. (1982) Evidence for direct inhibition of ovarian function by prolactin. *J. Reprod. Fert.* **65**, 559–569.
- Novin, D. (1985) The nature and integration of signals controlling food intake. *West J. med.* **143**, 208–211.
- Oomura, Y. (1976) Significance of glucose, insulin and free fatty acid on the hypothalamic feeding and satiety neurons. In *Hunger: Basic Mechanisms and Clinical Implications*, pp. 145–157. Eds D. Novin, W. Wyrwicka & G. Bray. Raven Press, New York.
- Otani, T., Maruo, T., Yukimura, N. & Mochizuki, M. (1985) Effect of insulin on porcine granulosa cells: implications of a possible receptor mediated action. *Acta endocr., Copenh.* **108**, 104–110.
- Parvizi, N. & Ellendorff, F. (1982) Further evidence on dual effects of norepinephrine on LH secretion. *Neuroendocrinology* **35**, 48–55.
- Porte, D., Jr & Woods, S.C. (1981) Regulation of food intake and body weight by insulin. *Diabetologia* **20**, 274–280.
- Riis, P.M. (1983) Adaptation of metabolism to various conditions: nutritional and other environmental conditions. In *Dynamic Biochemistry of Animal Production*, pp. 319–357. Ed. P. M. Riis. Elsevier, Amsterdam.
- Rossi, G.L. & Bestetti, G. (1981) Morphological changes in the hypothalamic-hypophyseal-gonadal axis of male rats after twelve months of streptozotocin-induced diabetes. *Diabetologia* **21**, 476–481.
- Schoonmaker, J.N. & Erickson, G.F. (1983) Glucocorticoid modulation of follicle-stimulating hormone-mediated granulosa cell differentiation. *Endocrinology* **113**, 1356–1363.
- Sen, K.K., Azhar, S. & Menon, K.M.J. (1979) Evidence for the involvement of an energy-dependent process in gonadotropin-releasing hormone-stimulated luteinizing hormone release by rat anterior pituitary. *Endocrinology* **105**, 1158–1161.
- Shaw, H.J. & Foxcroft, G.R. (1985) Relationships between LH, FSH and prolactin and reproductive activity in the weaned sow. *J. Reprod. Fert.* **75**, 17–28.
- Steiner, R.A., Cameron, J.L., McNeill, T.H., Clifton, D.K. & Bremner, W.J. (1983) Metabolic signals for the onset of puberty. In *Neuroendocrine Aspects of Reproduction*, pp. 183–227. Ed. R. L. Norman. Academic Press, New York.
- Veldhuis, J.D., Nestler, J.E., Strauss, J.F., Gwynne, J.T., Azimi, P., Garmey, J. & Juchter, D. (1986) Insulin regulates low density lipoprotein metabolism by swine granulosa cells. *Endocrinology* **118**, 2242–2253.
- Veldhuis, J.D., Nestler, J.E., Strauss, J.F., III, Azimi, P., Garmey, J. & Juchter, D. (1987) The insulin-like growth factor, somatomedin-C, modulates low density lipoprotein metabolism by swine granulosa cells. *Endocrinology* **121**, 340–346.
- Webley, G.E., Luck, M.R. & Hearn, J.P. (1988) Stimulation of progesterone secretion by cultured human granulosa cells with melatonin and catecholamines. *J. Reprod. Fert.* **84**, 669–677.