Nutritional status of donor cows: insulin related strategies to enhance embryo development

Claire Ponsart, G. Gamarra, S. Lacaze, Andrew A. A. Ponter

Anim. Reprod., v.11, n.3, p.195-198, Jul./Sept. 2014

Nutritional status of donor cows: insulin related strategies to enhance embryo development

C. Ponsart[1,2,6], G. Gamarra[1,2], S. Lacaze[3], A.A. Ponter[4,5]

[1]ANSES, Animal Health Laboratory, Maisons-Alfort, France.
[2]UNCEIA, R&D, Paris, France.
[3]MIDATEST, Domaine de Sensacq, Denguin, France.
[4]Université Paris Est, Ecole Nationale Vétérinaire d'Alfort, UMR Maisons-Alfort, France
[5]INRA, UMR 1198 Biologie du Développement et Reproduction, Domaine de Vilvert, Jouy-en-Josas, France.

Abstract

Nutritional and metabolic status of domestic ruminant females is linked with reproductive success. Diet can influence ovarian activity via effects at various levels of the hypothalamus-pituitary-ovarian axis. Changes in the plane of nutrition can affect follicular growth by inducing changes in plasma metabolites and metabolic hormones, such as insulin and IGF1. This paper will review different results from *in vivo* and *in vitro* feeding approaches describing a programmed sequence in circulating insulin concentrations. The stimulatory effect of insulin and IGF1 on follicle growth has been previously demonstrated, especially on small follicle growth prior to superovulation. Thus, *in vivo* feeding strategies have been recently tested to enhance embryo development. It has been shown that the interaction between the gonadotropin content of the superstimulatory preparation with the nutritional program of the donor cow needs to be considered when aiming to optimize the success of ovarian superstimulatory protocols. Moreover, some practical feeding strategies such as short term dietary propylene glycol could improve *in vitro* embryo production in superovulated growth-restricted heifers. To conclude, different diets or dietary supplements may improve fertility and embryo quality by inducing a programmed sequence in circulating insulin concentrations.

Keywords: bovine, feeding, *in vitro*, *in vivo*, metabolic status.

Introduction

It has long been recognized that the nutritional and metabolic status of domestic ruminant females are associated with reproductive success, but the underlying mechanisms remain poorly understood (Gutierrez *et al*., 1997; Gong *et al*., 2002b; Adamiak *et al*., 2005). Several authors have proposed that metabolic signals, such as circulating concentrations of insulin, growth hormone (GH), leptin and the insulin-like growth factor system (IGF) interact at the central level to modulate the release of gonadotrophins (Garnsworthy *et al*., 2008). The quality or competence of harvested oocytes appears to be the most important determinant of embryo yields (Seneda *et al*., 2001) and this depends on the physiological and reproductive status of the donor animal, which are influenced by age, health and nutrition (Majerus *et al*., 1999; Armstrong, 2001; Boland *et al*., 2001). When working with young healthy cattle, the variability in embryo development rates *in vitro* may be influenced by the diet of the donor prior to oocyte recovery (Boland *et al*., 2001).

Lessons from *in vitro* approaches

A large number of *in vitro* studies have demonstrated the direct action of metabolic factors on granulosa and theca cells (Webb *et al.*, 1999a, b; Lucy, 2000) and numerous experiments have also studied the direct effects of the diet on follicular variables following spontaneous or superovulation. *In vitro* studies have highlighted the fact that insulin and IGF1 are important mediators of follicular development, steroidogenesis, oocyte maturation and embryonic development (Gong *et al*., 1993).

In vivo feeding strategies to enhance embryo development

Diet can influence ovarian activity via effects at various levels of the hypothalamus-pituitary-ovarian axis. Changes in the plane of nutrition can affect follicular growth (Gutierrez *et al.*, 1997; Gong *et al.*,2002a) by inducing changes in plasma metabolites and metabolic hormones, such as insulin and IGF1 (Armstrong *et al.*, 2001) and/or in hormones and growth factors in follicular fluid (Landau *et al.*, 2000; Matoba *et al.*, 2014). Diet can also affect oocyte morphology (O'Callaghan *et al.*, 2000), oocyte developmental capacity and embryo production.

It has been shown that over-feeding can be harmful for the developmental quality of oocytes and embryos produced *in vivo* and *in vitro* (Mantovani *et al*., 1993; Papadopoulos *et al*., 2001; Freret *et al.*, 2006; Santos *et al*., 2008).

In addition, restricted feeding can have a positive effect on oocyte quality (Lozano *et al*., 2003) and the production of blastocysts *in vitro* (Armstrong *et al*., 2001; Freret *et al*., 2006). These results have been linked to the level of metabolites and hormones

[6]Corresponding author: claire.ponsart.vet@gmail.com
Received: July 14, 2014
Accepted: July 19, 2014

involved in the regulation of energy metabolism: in particular, an increase in the concentration of insulin and IGF1 is associated with high energy intake in dairy heifers (Freret *et al*., 2006). Webb *et al.* (2004) reviewed the stimulatory effect of insulin and IGF1 on follicle growth and Freret *et al*. (2006) showed that an increase in insulin concentrations during a short time period has a positive effect on small follicle growth prior to superovulation.

However, in the heifer, it has been shown that feeding regimens which increase insulin (hyperinsulinaemia) negatively influence the quality of oocytes (Adamiak *et al*., 2005; Freret *et al*., 2006). These findings support the idea that it may be possible to modulate insulin concentrations transiently to improve reproductive success, i.e. increase insulin during the phase of follicle growth (Scaramuzzi *et al.*, 2006) and then return to pre-stimulated levels just before ovulation so as not to have a negative effect on oocyte quality. To further support this hypothesis, Garnsworthy *et al.* (2009) modified circulating insulin levels (high and low) in dairy cows post-partum via the diet. They showed that a diet causing high insulin concentrations between calving and the first post-partum rise in progesterone followed by a diet causing low insulin until 120 days post-partum improved pregnancy rate compared with the other sequences of dietary treatment to modify insulin (high-high, low-high and low-low). Two experiments were recently performed using a Latin square design with treatments arranged as a 2×2 factorial: feed restriction (FR; 25% reduction in dry matter intake) compared with ad libitum (AL) feeding, combined with high (H) *versus* low (L) LH in the last 4 injections of the superstimulatory protocol (Bender *et al*., 2014). As expected, FR decreased circulating insulin concentrations (26.7 *vs*. 46.0 μIU/mL). Fertilization rates were higher for the AL-L (89.4%) and FR-H (80.1%) treatments compared with the AL-H (47.9%) and FR-L (59.9%) treatments. In addition, the number of degenerate embryos was decreased for AL-L (1.3) and FR-H (0.4) treatments compared with the AL-H (2.6) and FR-L (2.3) treatments. Thus, cows with either too low (FR-L) or too high (AL-H) insulin and LH stimulation had lower embryo production after superstimulation because of reduced fertilization rate and increased percentage of degenerate embryos. Therefore, the interaction between the gonadotropin content of the superstimulatory preparation with the nutritional program of the donor cow needs to be considered when aiming to optimize the success of ovarian superstimulatory protocols.

Insulin related feeding strategies

Exogenous insulin administration increased the recruitment of follicles in response to gonadotropin in gilts (Cox *et al*., 1987) and also rescues follicles from atresia and therefore increases the number of ovulatory follicles (Matamoros *et al*., 1991). The roles of the IGF system and insulin appear particularly critical. Increased energy decreases IGFBP2 and IGFBP4 expression in small follicles. IGFBP2 has been shown to inhibit the actions of IGF in different cell lines and is present at high concentrations during the post-partum period (McGuire *et al.*, 1995). Lowered IGFBP2 may in turn increase the bioavailability of systematically derived IGF1 and locally produced IGF2 in these follicles. On the contrary, in case of strong NEB, Wathes *et al.* (2008) have reported an increased expression of IGFBP2 in the liver whereas transcripts for IGFBP3 to 6 were down regulated. Together with the reduced IGF1 production, these changes are likely to lower IGF1 availability and receptivity for follicular cells leading to impaired follicular growth.

Other methods exist to modulate insulin secretion, for example the administration of dietary supplements which affect metabolism such as, propylene glycol (PG). PG, also known as 1,2-propianediol, is a 3-carbon compound ($C_3H_8O_2$) derived from propylene and has been used since the 1950's in the treatment of ketosis in post-partum dairy cattle (Maplesden, 1954). PG increases plasma glucose and insulin and decreases non esterified fatty acids (NEFA) and β-hydroxybutyrate (BHB). The latter two effects are seen in underfed animals (Nielsen and Ingvartsen, 2004). In a recent study on heifers we showed that short term dietary propylene glycol was associated with raised levels of glucose, insulin and stimulated follicle growth, causing an increase in the number of small follicles (diameter 1-4 mm) during the first days of an estrous cycle (Gamarra *et al*., 2014a). A second experiment aimed to test whether the daily oral administration of propylene glycol could improve *in vitro* embryo production in superovulated growth-restricted heifers (600 g/day) differing in their Anti-Mullerian Hormone (AMH) profiles (Gamarra *et al*., 2014b). Sixteen Holstein heifers were grouped according to AMH concentrations: low (L = 1-80 pg/mL; n = 7) or high (H: >150 pg/mL; n = 9). Administration of PG significantly increased the number of: small follicles (2-3 mm) and total follicles (2-8 mm) on day 2 of the cycle in all heifers. PG improved *in vitro* embryonic development rate (total number of embryos/number of fertilized oocytes) in all heifers compared to the control.

Taken together, these findings open-up the possibility of improving fertility and embryo quality by using diets or dietary supplements which induce a programmed sequence in circulating insulin concentrations.

References

Adamiak SJ, Mackie K, Watt RG, Webb R, Sinclair KD. 2005. Impact of nutrition on oocyte quality:

cumulative effects of body composition and diet leading to hyperinsulinemia in cattle. *Biol Reprod,* 73:918-926.

Armstrong DG, McEvoy TG, Baxter G, Robinson JJ, Hogg CO, Woad KJ. 2001. Effect of dietary energy and protein on bovine follicular dynamics and embryo production in vitro: associations with the ovarian insulin-like growth factor system. *Biol Reprod,* 64:1624-1632.

Bender RW, Hackbart KS, Dresch AR, Carvalho PD, Vieira LM, Crump PM, Guenther JN, Fricke PM, Shaver RD, Combs DK, Wiltbank MC. 2014. Effects of acute feed restriction combined with targeted use of increasing luteinizing hormone content of follicle-stimulating hormone preparations on ovarian superstimulation, fertilization, and embryo quality in lactating dairy cows. *J Dairy Sci,* 97:764-778.

Boland MP, Lonergan P, O'Callaghan D. 2001. Effect of nutrition on endocrine parameters, ovarian physiology and oocyte and embryo development. *Theriogenology,* 55:1323-1340.

Cox NM, Stuart MJ, Althen TG, Bennett WA, Miller HW. 1987. Enhancement of ovulation rate in gilts by increasing dietary energy and administering insulin during follicular growth. *J Anim Sci*, 64:507-516.

Freret S, Grimard B, Ponter AA, Joly C, Ponsart C, Humblot P. 2006. Reduction of body weight gain enhances in vitro production in overfed superovulated dairy heifers. *Reproduction,* 131:783-794.

Gamarra G, Ponsart C, Lacaze S, Le Guienne B, Deloche M-C, Monniaux D, Ponter A. 2014a. Short term dietary propylene glycol supplementation affects circulating metabolic hormones, progesterone concentrations and follicular growth in dairy heifers. *Livest Sci*, 162:240-251.

Gamarra G, Ponsart C, Lacaze S, Le Guienne B, Humblot P, Deloche M-C, Monniaux D, Ponter A. 2014b. Effect of dietary propylene glycol on ovarian follicle growth, the superovulatory response and in vitro embryo production after ovum pick-up in growth-restricted heifers differing in their plasma AMH profiles. *Reprod Fertil Dev*. (accepted for publication).

Garnsworthy PC, Sinclair KD, Webb R. 2008. Integration of physiological mechanisms that influence fertility in dairy cows. *Animal*, 2:1144-1152.

Garnsworthy PC, Fouladi-Nashta AA, Mann GE, Sinclair KD, Webb R. 2009. Effect of dietary-induced changes in plasma insulin concentrations during the early post partum period on pregnancy rate in dairy cows. *Reproduction,* 137:759-768.

Gong JG, McBride D, Bramley TA, Webb R. 1993. Effects of recombinant bovine somatotrophin, insulin-like growth factor-I and insulin on the proliferation of bovine granulosa cells in vitro. *J Endocrinol,* 139:67-75.

Gong JG, Armstrong DG, Baxter G, Hogg CO, Garnsworthy PC, Webb R. 2002a. The effect of increased dietary intake on superovulatory response to FSH in heifers. *Theriogenology,* 57:1591-1602.

Gong JG, Lee WJ, Garnsworthy PC, Webb R. 2002b. The effect of dietary induced increases in circulating insulin concentrations during the early post partum period on reproductive function in dairy cows. *Reproduction,* 123:419-427.

Gutierrez CG, Oldham J, Bramley TA, Gong JG, Campbell BK, Webb R. 1997. The recruitment of ovarian follicles is enhanced by increased dietary intake in heifers. *J Anim Sci*, 75:1876-1884.

Landau S, Braw-Tal R, Kaim M, BorA,Bruckental I. 2000. Preovulatory follicular status and diet affect the insulin and glucose content of follicles in high-yielding dairy cows. *Anim Reprod Sci*, 64:181-197.

Lozano JM, Lonergan P, Boland MP, O'Callaghan D. 2003. Influence of nutrition on the effectiveness of superovulation programmes in ewes: effect on oocyte quality and post - fertilization development. *Reproduction,* 125:543-553.

Lucy MP. 2000. Regulation of ovarian follicular growth by somatotropin and insulin growth factors in cattle. *J Dairy Sci*, 83:1635-1647.

Majerus V, De Roover R, Etienne D, Kaidi S, Massip A, Dessy F, Donnay I. 1999. Embryo production by ovum pick up in unstimulated calves before and after puberty. *Theriogenology*, 52:1169-1179.

Mantovani R, Enright WJ, Keane MG, Roche JF, Bolan MP. 1993. Effect of nutrition and dose of follicle stimulating hormone (FSH) on superovulatory response in beef heifers. *In*: Proceedings of the 9th AETE Meeting, 1993, Lyon, France. Lyon: AETE. abstr. 234.

Maplesden DC. 1954. Propylene glycol in the treatment of ketosis. *Can J Comp Med Vet Sci*, 1:287-293.

Matamoros IA, Cox NM, Moore AB. 1991. Effects of exogenous insulin and body condition on metabolic hormones and gonadotropin-induced follicular development in prepuberal gilts. *J Anim Sci,* 69:2081-2091.

Matoba S, Bender K, Fahey AG, Mamo S, Brennan L, Lonergan P, Fair T. 2014. Predictive value of bovine follicular components as markers of oocyte developmental potential. *Reprod Fertil Dev*, 26:337-345.

McGuire MA, Dwyer DA, Harrell RJ, Bauman DE. 1995. Insulin regulates circulating insulin-like growth factors and some of their binding proteins in lactating cows. *Am J Physiol*, 269: E723-E730.

Nielsen NI, Ingvartsen KL. 2004. Propylene glycol for dairy cows a review of the metabolism of propylene glycol and its effects on physiological parameters, feed intake, milk production and risk of ketosis. *Anim Feed Sci Technol,* 115:191-213.

O'Callaghan D, Yaakub H, Hyttel P, Spicer LJ, Boland MP. 2000. Effect of nutrition and superovulation on oocyte morphology, follicular fluid composition and systemic hormone concentrations in ewes. *J Reprod Fertil*, 118:303-313.

Papadopoulos S, Lonergan P, Gath V, Quinn KM, Evans AC, O'Callaghan D, Bolan MP. 2001. Effect of

diet quantity and urea supplementation on oocyte and embryo quality in sheep. *Theriogenology*, 55:1059-1069.

Santos J, Cerri R, Sartori R. 2008. Nutritional management of the donor cow. *Theriogenology*, 69:88-97.

Scaramuzzi RJ, Campbell BK, Downing JA, Kendall NR, Khalid M, Muñoz-Gutiérrez M,Somchit A. 2006. A review of the effects of supplementary nutrition in the ewe on the concentrations of reproductive and metabolic hormones and the mechanisms that regulate folliculogenesis and ovulation rate. *Reprod Nutr Dev*, 46:339-354.

Seneda M Relationship between follicle size and ultrasound-guided transvaginal oocyte recovery. *Anim Reprod Sci*, 67:37-43.

Wathes DC, Fenwick MA, Liewellyn S, Cheng Z, Fitzpatrick R, McCarthy SD, Morris DG, Patton J, Murphy JJ. 2008. Influence of energy balance on gene expression in the liver and reproductive tract of lactating cows and consequent effects on fertility. *In*: Szenci O, Bajcsy A (Ed.). Proceedings of the 25th World Buiatrics Congress, 2008, Budapest, Hungary: Factors affecting reproductive performance in the cow. Budapest: WAB. pp. 158-171.

Webb R, Campbell BK, Garverick HA, Gong JG, Gutierrez CG, Armstrong DG. 1999a. Molecular mechanisms regulating follicular recruitment and selection. *J Reprod Fertil Suppl*, 54:33-48.

Webb R, Gosden RG, Moor RM. 1999b. Factors affecting folliculogenesis in ruminants. *Anim Sci*, 68:257-284.

Webb R, Garnsworthy PC, Gong J-G, Armstrong DG. 2004. Control of follicular growth: local interactions and nutritional influences. *J Anim Sci*, 82:63-74.