



# Using different carbohydrates in prenatal life, growing and mid-lactation can affect lactation persistency in first lactating sheep

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**Abstract.** Improving milk production by using digestible fiber rich by-products would increase production efficiency and reduce environmental impact of sheep production. We hypothesized that prenatal and early life exposure to either starch from corn grain or highly digestible fiber from beet pulps and soy hulls would affect sheep response to the same feeds in mid-lactation. Two groups of 12 Sarda ewes each were exposed during the last two months of the prenatal life to two different isoproteic diets (one per group) and then fed the same diet during their growing phases and their first pregnancy. The diets were one glucogenic, being rich in starch from corn grains (S; 21% of starch and sugars, DM basis), the other lipogenic, being rich in digestible fiber from soy hulls and beet pulps (F; 9% of starch plus sugars, DM basis). In early lactation (until 54 DIM) all the 24 ewes received the same glucogenic S diet to support the lactation peak. From 55 to 160 days in milk (DIM) the ewes with S and F background, homogeneous per yield and BCS, were split to S or F total mixed rations (with 24%, S, and 13%, F, of starch plus sugars, DM basis), creating 4 groups of 6 ewes each: SS, SF, FS and FF based on the dietary sequence applied between prenatal life and parturition and between mid-lactation to the end of the experiment. Milk yield and composition were measured each 10 days and BCS every two weeks. Milk yield and composition were compared by using the PROC MIXED procedure of SAS for repeated measurements. At 55 DIM, milk production was (mean  $\pm$  SE)  $1.592 \pm 0.096$  kg/d per ewe and BCS was  $2.85 \pm 0.15$ . The S diet provided during early life and until parturition favored the production of fat and protein corrected milk (FPCMY) and a higher persistency on ewes fed the F diet compared to those fed the S diet during their mid lactation ( $1.483 \pm 0.180$  vs.  $1.143 \pm 0.198$  kg/d for SF vs. SS, respectively;  $P < 0.05$ ), whereas ewes with F background had no different milk persistency with both mid-lactating diets ( $1.269 \pm 0.198$  vs  $1.198 \pm 0.180$  kg/d for FS and FF, respectively). BCS did not differ among groups at 160 DIM. On ewes exposed to S early in life, the use of F in mid-lactation was significantly associated to higher FPCMY compared to S ewes after 110 DIM ( $P = 0.05$ ). The positive effects of the exposure to glucogenic diets in early life suggest to investigate the hormonal and metabolic mechanisms underlying these results.

**Keywords.** Starch – Digestible fiber – Fetal programming – Dairy.

## ***Influence de l'utilisation de différents hydrates de carbone pendant la vie prénatale, la croissance et la mi-lactation sur la persistance de la lactation chez les brebis en première lactation***

**Résumé.** L'amélioration de la production de lait en utilisant des sous-produits riches en fibre digestible augmenterait l'efficacité de la production et réduirait l'impact environnemental de la production ovine. Notre hypothèse est que l'exposition pendant la vie prénatale et le premier âge à, soit de l'amidon provenant de grains de maïs, soit de la fibre hautement digestible de pulpe de betteraves et d'enveloppes de soja influencerait la réponse des ovins à ces mêmes aliments à la mi-lactation. Deux groupes de 12 brebis sardes chacun ont été exposés pendant les deux derniers mois de vie prénatale à deux régimes isoprotéiques différents (un par groupe) et ont reçu ensuite le même régime pendant leurs phases de croissance et leur première gestation. Les régimes étaient, l'un glucogène, étant riche en amidon provenant de grains de maïs (S; 21% d'amidon et de sucres, sur base MS), l'autre lipogène, étant riche en fibre digestible provenant d'enveloppes de soja et de pulpe de betterave (F; 9% d'amidon et de sucres, sur base MS). Lors des premiers stades de lactation, jusqu'à 54 JEL (jours en lait) les 24 brebis ont toutes reçu le même régime glucogène S pour soutenir le pic de lactation. De 55 à 160 JEL les brebis à alimentation première S et F, homogènes pour le rendement et la note d'état corporel, ont été

réparties entre les rations totales mélangées S ou F (avec 24% S, et 13% F, d'amidon et de sucres, sur base MS), créant ainsi 4 groupes de 6 brebis chacun: SS, SF, FS et FF basés sur la séquence de régime appliquée entre la vie prénatale et le vêlage et entre la mi-lactation jusqu'à la fin de l'expérience. La production de lait et sa composition ont été mesurées tous les 10 jours et la note d'état corporel toutes les deux semaines. La production de lait et sa composition ont été comparées par la procédure PROC MIXED de SAS pour des mesures répétées. À 55 JEL, la production de lait était de (moyenne  $\pm$  ET)  $1,592 \pm 0,096$  kg/j par brebis et la note d'état corporel était de  $2,85 \pm 0,15$ . Le régime S distribué pendant le premier âge et jusqu'au vêlage favorisait la quantité de lait corrigé pour la matière grasse et la protéine (FPCMY) et une plus forte persistance chez les brebis recevant le régime F comparées à celles alimentées au régime S pendant leur mi-lactation ( $1,483 \pm 0,180$  vs.  $1,143 \pm 0,198$  kg/j pour SF vs. SS, respectivement;  $P < 0,05$ ), tandis que les brebis à alimentation première F ne montraient pas de différences pour la persistance du lait avec les deux régimes de mi-lactation ( $1,269 \pm 0,198$  vs  $1,198 \pm 0,180$  kg/j pour FS et FF, respectivement). La note d'état corporel ne différait pas entre groupes à 160 JEL. Sur les brebis recevant S pendant le premier âge, l'utilisation de F à la mi-lactation était significativement reliée à un FPCMY plus élevé en comparaison aux brebis S après 110 JEL ( $P = 0,05$ ). Les effets positifs de l'exposition aux régimes glucogènes lors du premier âge suggèrent que des recherches sont à mener sur les mécanismes hormonaux et métaboliques sous-jacents à ces résultats.

**Mots-clés.** Amidon – Fibre digestible – Programmation foetale – Laitier.

## I – Introduction

Production efficiency in dairy sheep farms can be reached by increasing yearly production levels per head either achieving high lactation peaks and/or high lactation persistency. Improving milk production using digestible fibrous substrates from industrial by products increases efficiency and reduces environmental impact of small ruminant productions. Cannas *et al.*, (2002) highlighted that the shape of the lactation curve might be influenced with nutritional strategies. In particular in early lactation, glucogenic diets with high starch contents help to support lactose production and milk yield, then in mid-late lactation, switching to lipogenic diets by substituting the most part of the starch with digestible fiber from soyhulls or beet pulp help to maintain lactation persistency (Cannas *et al.*, 2004). Decreases of glucogenic substrates, from mid to late lactation, should limit the peripheral insulin response reducing the accumulation of body reserves and favoring the nutrient utilization by the mammary gland, which is not sensitive to insulin (Sasaki, 2002). Several authors, using high digestible fiber versus high starch diets in mid lactation, observed increases in milk production and reduction of body reserves deposition (Cannas *et al.*, 2004; Lunesu *et al.*, 2016; 2017). Furthermore several authors have observed important effects of maternal nutrition on offspring characteristics, including production performances and metabolism. The maternal environmental, moreover the metabolic substrates to which the fetus is exposed during pregnancy, influences epigenetic processes and developmental changes which may have consequences later in life (Poore *et al.*, 2014). Prenatal diet can affect body weight, milk yield and milk composition in the subsequent first generation (Blair *et al.*, 2010) or reproductive organs and reproduction performances (Rhind *et al.*, 2001). Both in humans and small ruminants has been observed that overnutrition or undernutrition during prenatal life can impair the glucose metabolism in adult life (Paliy *et al.*, 2014; Husted *et al.*, 2008). In previous works studying prenatal lipogenic diets mainly considered the fat content, whereas few studies focused the effect of the type of carbohydrate on the offspring metabolism of dairy ruminants (Poore *et al.*, 2014). Following the evidences observed by Cannas *et al.*, (2004; 2007) and Lunesu *et al.*, (2016) on lactation persistency and also the findings from studies on fetal programming of glucose metabolism this work hypothesized that the exposures to starch or high digestible fiber early in life might affects the sheep response to carbohydrates in adult life.

## II – Materials and methods

Twenty-four Sarda ewes that lambed in the same week ( $\pm 3$  days from the expected date of parturition) were selected from a larger group of animals that were exposed to different carbohydrates from their prenatal life until the first lambing. The 24 sheep were divided in two groups of 12 sheep each that were exposed to two different isoproteic diets (one per group) during the last two months of prenatal life. In this phase their mothers were kept indoor and the offered diets consisted of a common basis of ryegrass hay and a pelleted concentrate: one glucogenic, being rich in starch and sugars (diet S), the other lypogenic with low starch content (diet F) (Table 1); the glucogenic was formulated with high amounts of corn grains whereas in the lypogenic one the most part of the corn grain was substituted with digestible fiber from soy hulls and beet pulps (Table 1). After birth the lambs were fed with only maternal milk, then from 20 to 60 days of age were gradually weaned with a pelleted starter that was initially composed, for both groups, with a mix of the two prenatal concentrates. After weaning, during the growing phase until the first parturition, the lambs were fed with diets similar to those offered in prenatal life (S and F, respectively) but accounting for the higher protein requirements (Table 1). In early lactation (until 54 DIM) all the 24 ewes received a glucogenic (S) diet based on corn meal, soybean meal, finally chopped ryegrass hay and chopped straw and offered as total mixed ration. The choice of using the same S diet, in early lactation for all groups, assumed that glucogenic substrates in this phase are needed to support metabolic efforts until the lactation peak. From 55 to 160 days in milk (DIM) the ewes with S and F background, homogeneous per yield and BCS, were split to S or F total mixed rations (with 24%, S, and 13%, F, of starch plus sugars, DM basis), creating 4 groups of 6 ewes each: SS, SF, FS and FF based on the dietary sequence applied between prenatal life and parturition and between mid-lactation to the end of the experiment. Milk yield and composition were measured each 10 days, BCS every two weeks and intake every 20 days. Milk production and composition and fat and protein corrected milk yield (FPCMY; Pulina *et al.*, 2004) were compared by using the PROC MIXED procedure of SAS for repeated measurements considering as fixed effects the mid-lactation diets (2 levels: S and F for glucogenic and lypogenic diets, respectively) nested within the nutritional background (2 levels: growing and prenatal diets: S and F), and the sheep as random effect.

**Table 1. Feeds used for the experimental diets in the last two months of prenatal life (last 75 days of gestation), growing phases (from weaning to first lambing) and lactation (from lambing until 160 days)**

| Offered feeds              | Hay  | Prenatal concentrate |        | Growing concentrate |        | Lactation total mixed ration* |        |
|----------------------------|------|----------------------|--------|---------------------|--------|-------------------------------|--------|
|                            |      | Diet S               | Diet F | Diet S              | Diet F | Diet S                        | Diet F |
| Dry matter, % of as fed    | 90.0 | 87.5                 | 86.4   | 86.4                | 86.4   | 88.3                          | 88.7   |
| Crude protein, % of DM     | 11.0 | 17.6                 | 17.9   | 20.1                | 19.1   | 17.2                          | 16.4   |
| NDF, % of DM               | 63.1 | 15.7                 | 32.9   | 20.2                | 40.2   | 41.3                          | 50.2   |
| ADF, % of DM               | 49.9 | 6.2                  | 27.9   | 8.6                 | 27.9   | 23.4                          | 31.9   |
| ADL, % of DM               | 3.9  | 1.2                  | 1.7    | 0.9                 | 1.7    | 3.8                           | 4.9    |
| Ether extract, % of DM     | 1.1  | 2.9                  | 3.2    | 2.8                 | 3.0    | 2.7                           | 2.0    |
| Ashes, % of DM             | 11.3 | 8.5                  | 10.3   | 4.4                 | 5.9    | 8.4                           | 8.6    |
| NFC, % of DM               | 26.7 | 55.3                 | 35.7   | 52.6                | 31.8   | 30.5                          | 22.9   |
| Starch and sugars, % of DM |      | 44.5                 | 15.4   | 42.3                | 13.7   | 23.8                          | 12.8   |
| Forage: concentrate ratio  |      | 45/55                | 45/55  | 40/60               | 40/60  |                               |        |

\* TMR was fed ad libitum; during milking each ewe received 160 gr/d of DM of pelleted mix (17% of CP, 24% of NDF, 4% of EE and 9% of ashes).

### III – Results and discussion

Animal performances at 54 days of lactation before to switch to mid-lactation diets, mean  $\pm$  SE of milk production was equal to  $1.592 \pm 0.323$  kg/d per ewe, whereas BCS was  $2.85 \pm 0.15$ . On average for the first lactation, the nutritional background did not influence significantly the milk production (Table 2). Even though it was observed a significant effect on daily milk yield for the interaction between nutritional background and mid-lactation diet and in particular the diet F favored higher milk yield in animals with glucogenic background ( $P < 0.001$ ; Table 2). Milk composition was not significantly affected by prenatal diet or mid-lactation diet, except than for lactose content, which resulted very low in the FF group (Table 2). It perhaps indicates heavy shortages of glucose for lactose synthesis in the group FF fed with lypogenic diets after a lypogenic background. It has to be noticed that milk fat and protein contents were also low in respect to the standards of the Sarda breed and it was partially attributed to a negative effect of the small TMR particle sizes on animal rumination. High starch background (S) favored higher FPCMY on ewes fed low starch from 55 to 160 DIM ( $1.483$  vs.  $1.231 \pm 0.150$  kg/d for SF vs. SS, respectively). The shape of the lactation curve of the SF group showed higher FPCMY persistency than SS group (Figure 1). At the opposite, for lypogenic background (F) FPCMY persistency was similar among FS and FF groups (Table 2; Figure 1). On ewes exposed to glucogenic background (S) the use of digestible fiber in mid-lactation was significantly associated to maintain FPCMY after 110 DIM ( $P < 0.05$ ; Figure 1). The observed differences among SS and SF were numerically high, whereas tested statistical significances were quite poor, it was attributed to the high variation among individuals and to the low number of ewes included in each group. FPCMY at 150 DIM was  $1.242 \pm 0.196$  vs.  $0.886 \pm 0.214$  kg/d ( $P < 0.05$ ) for SF vs. SS ewes, and  $0.947 \pm 0.214$  vs.  $0.770 \pm 0.196$  for FA and FF ewes ( $P > 0.20$ ). Differences among BCS were very small and not significant among the experimental groups, BCS at 160 DIM was 3.00, 2.96, 2.94,  $3.02 \pm 0.10$  for SS, SF, FS and FF, respectively.

**Table 2. Animal performances in mid and late lactation. Reported values of animal performance and milk composition were calculated as average from 55 days from lambing to end of lactation**

| Background diets:<br>(prenatal and growing) | Glucogenic,<br>high starch (S) |               |                   | Lipogenic, high<br>digestible fiber (F) |               |                   | Statistics                     |                               |                                     |
|---|--------------------------------|---------------|-------------------|---|---------------|-------------------|--------------------------------|-------------------------------|-------------------------------------|
|   | Starch<br>(SS)                 | Fiber<br>(SF) | Mean+SE<br>n = 12 | Starch<br>(FS)                          | Fiber<br>(FF) | Mean+SE<br>n = 12 | Background<br>(B) <sup>a</sup> | Lactation<br>(L) <sup>a</sup> | Interaction<br>(B X L) <sup>a</sup> |
| Mid lactation diet                          |                                |               |                   |   |               |                   |                                |                               |                                     |
| Milk, gr/d                                  | 1225                           | 1606          | 1434 $\pm$ 53     | 1365                                    | 1253          | 1309 $\pm$ 57     | NS                             | *                             | ****                                |
| Fat, %                                      | 6.10                           | 5.96          | 6,02 $\pm$ 0.08   | 5.91                                    | 6,04          | 6.0 $\pm$ 0.09    | NS                             | NS                            | NS                                  |
| Protein, %                                  | 5.59                           | 5.45          | 5.51 $\pm$ 0.05   | 5.43                                    | 5.69          | 5.56 $\pm$ 0.06   | NS                             | NS                            | **                                  |
| Lactose, %                                  | 4.75                           | 4.95          | 4,86 $\pm$ 0.04   | 4.78                                    | 4.51          | 4,65 $\pm$ 0.05   | ****                           | NS                            | ****                                |
| FPCM, gr/d                                  | 1231                           | 1483          | 1376 $\pm$ 39     | 1269                                    | 1198          | 1233 $\pm$ 48     | **                             | NS                            | ***                                 |

<sup>a</sup> Statistical differences within levels of tested effects were equal to NS = not significant \* =  $P < 0.01$ ; \*\* =  $P < 0.05$ ; \*\*\* =  $P < 0.01$ ; \*\*\*\* =  $P < 0.001$ .

The observed results of this research confirmed that feeding high digestible fiber diets from mid to late lactation favors lactation persistency as exposed in previous studies (Cannas *et al.*, 2013; Lunesu *et al.*, 2016). In addition this research suggests that ewe response to starch and fiber in terms of milk yield is highly dependent on the nutritional background.

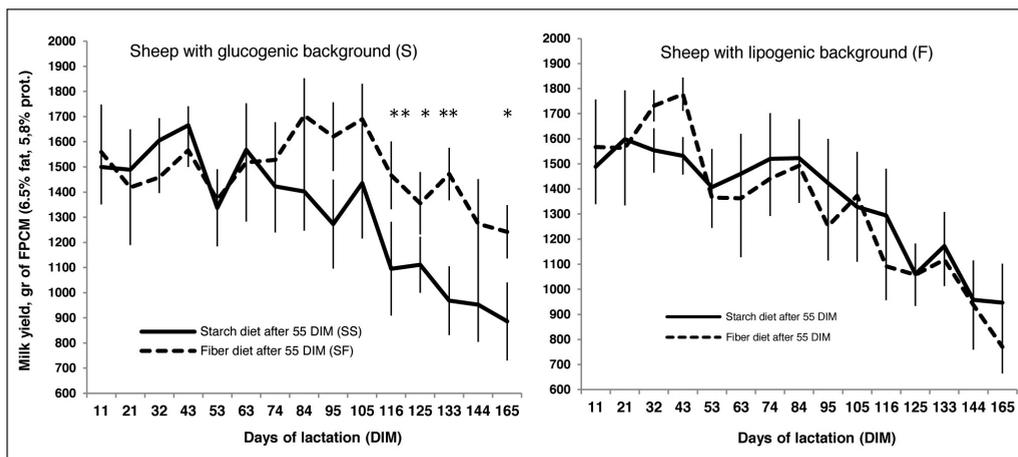


Fig. 1. Production of fat and protein corrected milk (FPCM) during the first lactation of the experimental ewes. In the first 55 days of lactation all the groups were fed with the glucogenic (S) diet. \* and \*\* indicates that observed FPCM of the two groups were significantly different for  $P < 0.1$  and  $P < 0.05$ .

## IV – Conclusion

The use of glucogenic diets from high starch content or lipogenic diets from high digestible fiber used early in life caused different animal response to amilaceous or fibrous carbohydrates after the first lactation peak. Ewes with glucogenic backgrounds in uterine land growing phases showed higher lactation persistency when the mid lactation diet included low starch and high digestible fiber content. This effect was higher after 110 days of lactation. Otherwise ewes with lipogenic background did not show differences in milk persistency attributable to the dietary carbohydrates used from mid to late lactation. The positive effects obtained with the exposure to glucogenic diets early in life suggest to deeply investigating the hormonal and metabolic mechanisms underlying these results, including insulin sensitivity and energy homeorhesis in mid-lactating dairy sheep.

## Aknowledgements

This work was carried out within the project “Permanent effects of starch and fiber supplied during uterine and postnatal life of dairy sheep on gastrointestinal microbiota and energy partitioning between milk production and fat deposition” funded by the Italian Ministry of University and Research (Future in Research Program, 2013).

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