Iranian Journal of Applied Animal Science

Animal Factors Condition Milk Performance and Quality of Grazing Dairy Cows

Review Article

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Received on: 13 Apr 2013
Revised on: 18 May 2013
Accepted on: 1 Jun 2013
Online Published on: Mar 2014

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ABSTRACT

The base of this review is to consider the relevant role that animal factors (potential milk yield, body weight, body condition score, state of lactation, parity and fertility) play on milk performance (considering the energy balance and the rumen function across the full lactation curve of animals) and milk quality (milk protein content, milk fat content, milk lactose content, vitamins, minerals, immunoglobulins, cells in milk, pathogenic bacteria and inhibitors) of grazing dairy cows (taking into account the substitution rate and the milk response at pasture of animals under different supplementation regimes). All this, looking for high individual milk production and paying attention to grassland management as the key point for sustainability of grazing dairy systems in order to maintain high pasture dry matter intake in the swards with high grass quality (high pasture levels of crude protein, water soluble carbohydrates and digestibility of organic matter but low pasture levels of acid and neutral detergent fibers) all over the grazing season to satisfy cow needs at all times thought the lactation curve of dairy cows. It is also necessary to consider the use of supplementation at pasture (normally at the peak of lactation, when cows are generally in a state of negative energy balance and the provision of silage / concentrate can help to alleviate this situation) to keep the animals within the desired levels of milk production. It seeks, thereby, to achieve an efficient conversion of grass into milk, in a competitive and profitable way taking into consideration the current context in which pasture-based milk production systems are developed in mostly of temperate regions worldwide and focusing our interest in the milk premium price got by dairy farmers in a humid area as Galicia (NW Spain) when we consider the quantity and the quality of the milk delivered to the dairy industry.

KEY WORDS body condition score, body weight, energy balance, milk quality, potential milk yield, rumen function.

INTRODUCTION

Efficient pasture based milk production systems and constraints

Grazing dairy systems have decreased considerably over the last 30 years in many European countries (Bourgeois, 2002) and the number of dairy cows which are kept indoors for all or part of the herbage growing season has increased considerably (Van Den Pol-van Dasselaar *et al.* 2008). Nevertheless, it is now time to take into consideration that due to economic, environmental and animal welfare constraints, the abolition of EU milk quotas by 2015 it is envisaged that a larger proportion of the milk produced in temperate regions will be produced from grazed pastures (Dillon, 2006). Considering the fact that grass is the cheapest source of nutrients available for feeding ruminants, it should be the basis of profitable, consumer acceptable and sustainable milk production systems in humid regions (Roca-Fernández, 2011) where climatic conditions are adequate for grass growth (Mayne and Peyraud, 1996). Despite differences both between and within countries on the seasonality of grass growth and expected yield, high utilization of fresh herbage in the diet of cows helps to reduce the feeding costs of milk production. Low-inputs grazing systems have also social and environmental values related to the preservation of the rural landscape and the promotion of a clean, animal welfare friendly and transferring a good image for dairy cattle production to consumers (Peyraud and Gonzalez Rodríguez, 2000).

The comparisons made at world level show that milk production systems maximizing grassland utilization appear to be highly competitive. A study of the international competitiveness of using available farm resources as grazed grass found that the total cost of milk production is negatively related to the proportion of grazed grass in the diet of dairy cows Dillon *et al.* (2005). These data showed that increasing the proportion of grazed grass in a system that already entails a high proportion of grazed grass (such as happen in countries with high tradition for grazing grass as United Kingdom and Ireland) will have a greater benefit in reducing milk production costs than other system with a low proportion of grazed grass (such as happen in countries with a high inputs milk production systems as Denmark and The Netherlands). These relationship also highlighted that the average milk production cost is reduced by 1 cent €/liter per 2.5% increase in grazed grass in the cows' diet. Models indicate that in Ireland, early grazing will increase profitability per cow and day for each extra day at grass, through higher animal performance and lower feed costs (Kennedy *et al.* 2005). Similarly, in the Netherlands, the more grass the cows eat at pasture the larger is the farmer's income. Nevertheless, the level of herbage utilization on many of the Irish dairy farms is low and may be increased through increasing stocking rate (SR) and applying modern grazing management strategies at farm level (Kennedy *et al.* 2005). In the past, high performance from pasture-based milk production systems was based on applying high SR accompanied by high herbage utilization, but compromising animal performance. However, increased emphasis on issues like product quality and nitrogen (N) leaching, soil compaction, greenhouse-gas emissions and animal welfare, pasture-based milk production systems in the future will require higher per-animal productivity (Dillon, 2006). Therefore, the efficient utilization of grazed grass will need the development of grazing management systems designed to maximize daily pasture dry matter intake (PDMI) per cow, while maintaining a greater sward quality over the grazing season (Mayne and Peyraud, 1996; Roca-Fernández *et al.* 2012). In a recent review, Roca-Fernández (2013) has considered the important role that sward factors play on PDMI of grazing dairy cows. The implication is that most of the grazing systems designed to maximize cow performance are inefficient in utilization of grass per hectare (ha) and this is a key point of research that need to be improved for future development of sustainable milk production systems in humid areas looking for an efficient conversion of grass into milk and profitability of grazing dairy systems (Roca-Fernández, 2011). The PDMI may be maximized by adhering to sward important characteristics such as maintaining a high proportion of green leaf within the grazing horizon while allocating an adequate daily herbage allowance (DHA) (Dillon, 2006; Roca-Fernández *et al.* 2011; Roca-Fernández *et al.* 2012). Increasing the green leaf proportion at the base of the sward through appropriate grazing management strategies implemented in early spring may play an important role in increasing PDMI and making grazing management easier (Roca-Fernández, 2011). Van Vuuren and Van den Pol van Dasselaar (2006) used data of Bruinenberg *et al.* (2002), Bargo *et al.* (2003), Butler *et al.* (2003), Ribeiro-Filho *et al.* (2005) and Tas *et al.* (2005) to calculate that when fed a grass only diet, a maximum PDMI of 110 to 120 g per (kg of body weight)$^{-0.75}$ can be expected. This amount is considered enough to meet relatively high energy and protein requirements from dairy cows.

During forty years the selection of dairy cows has been almost exclusively oriented toward high genetic merit (HGM) dairy cows on milk production (Delaby *et al.* 2010). Today, HGM Holstein-Friesian dairy cows are able to produce more than 10000 kg milk per lactation in high-input farming systems, but they cannot produce such amount of milk from grazing alone (Dillon, 2006). Consequently, pasture-based milk production systems prevent HGM dairy cows from fully expressing their milk potential despite high amounts of concentrate being provided (González-Rodríguez *et al.* 2010). Several trials, however, have shown that relatively high milk production (i.e. 7400 kg per lactation) is achievable with HGM dairy cows in pasture-based milk production systems with only 350 kg of concentrate at least under areas well suited to produce high yields of grass over a prolonged grazing season (Buckley *et al.* 2000; Kennedy *et al.* 2002; Horan *et al.* 2005; Delaby *et al.* 2010; Roca-Fernández *et al.* 2011; Roca-Fernández *et al.* 2012a). Likewise, in the future, the dairy cow genotype must be compatible with the system of milk production; the prediction of the phenotypic performance of dairy cattle must be based on the knowledge of the genotype, the environment and its interaction (Peyraud *et al.* 2010).

There are several constraints that limit pasture-based milk production systems and the decrease on the number of grazing dairy cows in the last times can be explained by different reasons (Roca-Fernández, 2011). Firstly, the convenience of managing confined dairy herds, particularly with continuous calving pattern, fed with supplements in order to control their diet and balance and the variability of the fresh herbage production (Peyraud *et al.* 2010). Secondly, the lower milk yield (MY) per cow has largely contributed to reduce the use of many suitable areas for grazing. In addition, the increasing use of automated milking systems makes grazing more difficult, although the combination of grazing and milking robot is now possible without penalizing MY per cow (Wiktorsson and Spörndly, 2002). Increased herd size may be another reason for which the decreasing of grazing can be explained in many dairy farms, at least for countries in northern of European countries, where pressure for land use and SR are very high (Van Den Pol van Dasselaar *et al.* 2008).

Many farms are also limited by land area and fragmentation (Roca-Fernández, 2011). Van Hung *et al.* (2007) defined land fragmentation whereby a single farm has a number of parcels of land which are not adjoining each other. In a pasture-based milk production system land area fragmentation is an important constraint on the use of grazing systems in dairy farms. With land becoming the limiting factor in a post-quota scenario alternative uses for this land such as the outsourcing of replacements to outside land blocks or the contract rearing of dairy heifers will be explored for potential increases in efficiency (Shalloo *et al.* 2007). A study by Jha *et al.* (2005) on the effects of land fragmentation in India found that it had a negative effect on technical efficiency leading to inefficient use of resources in agriculture and increased costs. If land fragmentation means that more labor and other resources are used than is necessary and that these resources can be used more effectively elsewhere, there is an overall economic gain from reduced levels of land fragmentation (Van Hung *et al.* 2007).

Animal factors affecting milk performance

In this review, the focus will be done on the relevant role that animal factors play on milk performance and quality of grazing dairy cows for future development of sustainable milk production systems in humid areas.

Potential milk yield (MY)

There has been great genetic progress in the last times with the selection of high milk yield (MY) per cow (Roca-Fernández, 2011). Internationally, genetic selection among cattle breeders for higher MY has resulted in an average production gains of 1 to 2% per year (Dillon *et al.* 2006). This genetic selection has been generally accompanied with an increased proportion of North American Holstein-Friesian genes with high MY per cow, lower weight and greater MY response to concentrate supplementation (Rauw *et al.* 1998). Furthermore, genetic selection for increased MY is associated with lower milk fat and protein, higher MY post-calving and at peak and lower persistency of lactation compared to animals of lower MY potential.

As MY and dry matter (DM) intake are correlated, selection for high MY should produce cows with higher intake potential (Van Arendon *et al.* 1991). There are also indications that improving DM intake pre-partum has a positive residual effect on DM intake post-partum which can lead to improvements in MY. Improving body condition score at calving has beneficial effects on post-partum MY. Beneficial effects on reproductive performance as a direct result of improving diet pre-calving are difficult to find but BCS at calving has been shown to effect conception rates to first service and, therefore, nutrition in the pre-calving period should indirectly affect reproductive performance.

Journet and Demarquilly (1979) reported average milk responses (MR) from 0.4 to 0.6 kg of milk per kg of concentrate. However, Peyraud and Delaby (2001) found that the MR to concentrate was higher than previously reported research in the literature published after 1990, which can be attributed to the increase in genetic merit of dairy cows. A greater response to supplementation may be expected in high genetic merit (HGM) dairy cows because they partition more nutrients to milk production and lose more body weight in early lactation than low genetic merit (LGM) dairy cows (Kellaway and Porta, 1993). Stage of lactation influences lactational responses to concentrate supplements (Dixon and Stockdale, 1999; Roca-Fernández *et al.* 2012). In early lactation, cows partition more nutrients toward milk production; thus, milk response (MR) to supplementation may be higher than in late lactation, when more nutrients are directed to body weight (Kellaway and Porta, 1993). The MR to concentrate supplementation of grazing cows supplemented with 3 kg DM/day concentrate was 0.7, 0.4, 0.5 and 0 kg of milk per kg of concentrate when they were between 86 to 114, 115 to 133, 134 to 187 and 188 to 243 days in milk (DIM), respectively (O'Brien *et al.* 1999). Summarizing five experiments with supplement DM intake from 0 to 7 kg/day, the marginal MR was 1.3, 1.1 and 0.7 kg of milk per kg of supplement in early, mid and late lactation, respectively (Stockdale *et al.* 1987).

Body weight (BW)

Output from cows is frequently expressed as a measure of MY (i.e. volume, weight of solids, energy, etc.) and no references are made to body weight (BW) change. Nevertheless, maintenance requirements are related to cow BW (Jarrige, 1989), and thus, total energy demands are related to cow BW. Cow size, as described by BW, has been shown to have a major influence on intake (Stockdale, 2000). It is reported that under grazing conditions, daily intakes of herbage are less than 3.0% of BW but this may increase to 3.25% of BW in high producing cows (Leaver, 1985). Peyraud *et al.* (1996) stated that pasture dry matter intake (PDMI) increases by 1.0 to 1.5 kg OM per 100 kg of BW. Stakelum and Connolly (1987) reported that for each 100 kg increase in BW daily PDMI increased by 2.2 kg. As

MY and feed consumption are closely correlated, and the use of North American Holstein-Friesian genetics has been widespread all over the world in the past 15 years, dairy cows with higher MY and higher intake potential have been produced. Kennedy *et al.* (2003) reported that HGM cows were heavier and had a higher total dry matter intake (TDMI) (552 kg and 18.3 kg DM/cow/day, respectively) than their medium genetic counterparts (548 kg and 17.4 kg DM/cow/day). Kertz *et al.* (1991) also reported that BW loss is greater in multiparous than primiparous cows. Mature animals reach their lowest BW between week's five to seven of lactation, it then progressively increases until week eighteen. Primiparous animals, however, reach their lowest BW at week four of lactation and remain at that level until week twelve.

Muller and Fales (1998) reported that lactation curve of grazing cows declined more rapidly in the spring and cows gained less body condition score (BCS) (0.3 gain in BCS) in the 6-month study than the benchmark values for confinement feeding. The authors also reported that Hoffman *et al.* (1993) fed grain at rates of 1 kg to 3 kg milk or 1 kg to 4.2 kg milk and that these rates did not result in MY differences between the groups. However, BW gains and BCS were lower for cows fed the lower amount of grain. Rook *et al.* (1994) concluded that mean BW was significantly lower at 4.0 cm sward height (SH) than at 6.0 or 8.0 cm. Supplementation had no effect on mean BW at 4.0 or 6.0 cm but increased it at 8.0 cm SH. In a further experiment supplementation also increased mean BW at 8.0 cm but not at 6.0 cm. The mean BW change pattern was maintained by cows on the 6.0 and 8.0 cm swards but those on the 4.0 cm sward suffered substantial BW loss.

Fulkerson *et al.* (2001) compared two genetic merit dairy cows groups (high, HGM and medium, MGM) fed three different levels of concentrate feeding on a predominantly pasture-based feeding system. The low, medium and high concentrate feeding levels were 0.34 t/cow, 0.84 t/cow and 1.7 t/cow, respectively. There were no significant differences between levels of feeding within level of genetic merit. However, dairy heifers on the low concentrate feeding level lost more BW and the heifers in the high concentrate feeding level lost the least. There was no significant difference in BW losses in the multiparous dairy cows in the post-calving period.

Body condition score (BCS)

BW alone is not a good indicator of body reserves as a cow of a given weight could be large and thin or small and fat; additionally BW is subject to variations in gut fill. For example, Stockdale (1999) reported that cows that lost BCS consistently through spring and summer increased their BW throughout. It has been found that as BCS at calving rises, the rate of increase in food intake after calving decreases and the delay between peak MY and time of maximum food intake becomes greater (Garnsworthy and Jones, 1987). Kellaway and Porta (1993) reported that when thin dairy cows were able to take advantage of their increased appetite in early lactation by being given access to *ad libitum* feed of high digestibility, the advantage of having dairy cows in good BCS was much less. These authors also established that this may be possible with a balanced complete diet but this is very unlikely with grazing dairy cows. They found, however, that concentrate supplementation in a grazing system had no effect on energy balance (EB) up to 60 days in milk (DIM), yet the inclusion of concentrate in the diet did result in greater BCS gain in mid to late lactation. In general, dairy cows selected from within a pasture-based system (New Zealand strain) maintained higher BCS throughout lactation (Roca-Fernández, 2011).

The need for specific levels of body reserves in dairy systems, where pasture and supplements rather than total mixed rations (TMR) are fed, is not clear, and this is partly because the information on the influence of BCS at calving on the milk production system of grazing cows is scarce (Stockdale, 2001). Therefore, there is a need to better define this relationship for pasture fed cows. The response of animals at different BCS to additional feeding, particularly through the use of concentrates, needs to be established since most farmers now use supplements to complement pasture in the diet of early lactation cows. Gibb *et al.* (1992) assessed body tissue changes, by serial slaughter procedures, in 54 multiparous cows offered grass silage *ad libitum* plus 3, 6 or 9 kg of concentrates. Mean BW at calving was 606 kg and by lactation week 8 this had declined to 563 kg in those cows remaining. Body fat loss accounted for over 0.85 of the combined loss of body fat and protein. Following calving, both the 6 and 9 kg groups had similar rates of body fat loss which were greater than those in the 3 kg group. However, body fat repletion recommenced on the 6 and 9 kg diets in lactation week 8, while body fat loss continued on the 3 kg diet for a further 3 weeks. Maximum fat loss on any one diet was 42 kg and body protein loss over the same period was only 8.7 kg.

However, over the first 8 weeks of lactation cows on average increased the net weights of liver and gut tissues by over 0.5 of the immediate post-calving weights, and the largest increases were observed in the tissues of the stomach and the intestines. With this demand for extra protein to support tissue hypertrophy at a time when total dietary intake is limiting, it is not surprising that the cow is unwilling to sacrifice a net loss of body protein and this may provide a partial explanation for why milk protein content declines during this stage of the lactation cycle (DePeters and Cant, 1992).

Stage of lactation

In seasonal calving dairy systems the effects of stage of lactation are usually confounded with those of season, i. e. the effects of variation in photoperiod, climate and weather conditions (Knapp and Grummer, 1990; Davison *et al.* 1996; Aharoni *et al.* 1999), and variations in the supply and nutritive characteristics of herbage (Papalois *et al.* 1996; Auldist *et al.* 1998). Stage of lactation of a dairy cow, when considered separately from the effects of nutrition and / or season, significantly affects both MY and milk composition (Roca-Fernández, 2011) through the effect of significant changes in the cow's physiological state (Butler, 2000; Holmes *et al.* 2002). Several experiments have studied the lactation curves of dairy cows (Tekerli *et al.* 2000; Holmes *et al.* 2002; Horan *et al.* 2005; Roca-Fernández, 2011). Typical lactation curves of spring calving dairy cows show a peak or maximum daily yield occurring at 4 to 8 weeks post-calving, followed by a daily decrease in MY until the cow is dried-off, or production is naturally terminated (Keown *et al.* 1986). Additionally, Olori *et al.* (1997) reported that decline in daily yield, due to pregnancy, began from the first month of gestation. Peak MY and the associated peak in the cow's energy requirements occur before the potential for maximum dry matter intake is reached so that cows in early lactation experience a period of negative energy balance (Butler, 2000). The extent and duration of this period depends upon the genetic merit of the cow (Westwood *et al.* 2000), the BCS of cows at calving and nutrition post-calving (Stockdale, 2004). Where nutritional and / or seasonal effects are minimised, milk production and / or seasonal effects are minimised, milk production usually peaks 35-45 days post-partum and declines after about 60 days post-partum (Thomas and Rook, 1983; Butler, 2000; Holmes *et al.* 2002). However, the shape of the lactation curve and the rate of decline in milk production can be changed considerably by varying the cow's energy intake. The concentration of fat and protein in milk declines after calving, reaching its nadir when cows are 40-60 days post-partum. This decline is due to dilution as MY increases with increasing production of lactose by the mammary gland and milk fat and protein production tends to peak at the same time as MY (Thomas, 1983; Holmes *et al.* 2002). Beyond 40-60 days post-partum, the concentration of fat and protein in milk increases until the end of lactation, although milk fat and protein yield declines over the same period (Butler, 2000; Holmes *et al.* 2002; Delaby *et al.* 2009). There appears to be no effect of stage of lactation on the responsiveness of milk fat concentration to nutrition (Stockdale *et al.* 1987; Coulon and Rémond, 1991).

Nevertheless, stage of lactation can affect milk fat composition as a result of the large variation in the dairy cow's energy balance (Roca-Fernández *et al.* 2012a). The responsiveness of milk protein concentration to level of energy intake varies with stage of lactation. Based on 66 experiments, Coulon and Rémond (1991) found that the change in milk protein concentration with increases in the intake of metabolic energy (ME) was larger when dairy cows were in mid- to late lactation than when in early lactation (0.05 *vs.* 0.03 g protein/kg for every MJ ME). Changes in the rates of synthesis of both the casein and whey proteins by mammary secretory cells do not vary significantly with stage of lactation so that, with the exception of very late lactation, the ratio of casein to whey does not vary significantly with stage of lactation (Kefford *et al.* 1995; Rowney and Christian, 1996; Auldist *et al.* 1998; Coulon *et al.* 1998). Kefford *et al.* (1995) reported, however, an interaction between the effects of stage of lactation and of nutrition on milk protein composition and on the cheddar cheese-making properties of milk. In this study, cows in mid or late lactation were offered 1 of 2 diets consisting of hay, silage and cereal grain (high quality diet) or hay, silage and straw (low quality diet). Dry matter intakes for the high and low quality diets were 14.7 and 11.3 kg/cow/day, respectively. The low quality diet resulted in milk with a lower ratio of casein to whey for cows in late lactation, but not for cows in mid-lactation. There was no effect of stage of lactation on the ratio of casein to whey in milk from cows fed the high quality diet.

Parity

The milk production of a herd is affected by its lactation number profile (Roca-Fernández, 2011). Several authors have investigated the effect of parity on MY (Ray *et al.* 1992; Peyraud *et al.* 1996; Tekerli *et al.* 2000; Horan *et al.* 2005). Upon examination of total lactation MY, Ray *et al.* (1992) found that first parity dairy cows yielded 92% of the total MY of second lactation cows. Third lactation cows had a MY 5% higher than that of the second lactation cows while there was no difference in cows greater than fourth parity. Peyraud *et al.* (1996) also confirmed that multiparous cows had a higher MY and DM intake than their primiparous cows (+6.3 and 3.2 kg/day, respectively). Tekerli *et al.* (2000) reported lower peak and total lactation MY for first lactation cows but higher persistency. This indicates that the secretory tissue in primiparous cows takes longer to reach its peak activity than in multiparous cows (Rao and Sundaresan, 1979).

Fertility

Reproductive function can be influenced by more than simply a deficiency of nutrients (Staples *et al.* 1992). Recommendations for high producing cows during the first 12 weeks of lactation for CP content in the diet are 17-18% of the DM. Ferguson and Chalupa (1989) reported that fertility was reduced in mature cows (fourth or greater lactation) consuming diets high in crude protein (CP) content (19 vs. 16%). However, first lactation cows increased conception rates (65 *vs.* 36%) when fed diets of 16% CP.

But not only conception rate was observed to be affected by feeding high levels of CP, the excess of nitrogen (N) in the rumen will be absorbed and transformed into urea in the liver. This will induce an increase of blood urea nitrogen (BUN) and milk urea nitrogen (MUN), which may result in energy wastage and ammonia toxicity. Visek (1984) observed that urea concentrations in uterine secretions may be harmful to spermatozoa.

Energy status and age of the animal are other important factors affecting fertility (Roca-Fernández, 2011). Cows in negative energy balance may be the most susceptible to reproductive problems (Staples *et al.* 1992). These considerations suggest that grazing may create reproductive problems in high producing dairy cows. However, a study conducted by Washburn and White (1997) contradict this assumption. Data of four seasonal sets of cows suggests that neither first service conception rate (51 *vs.* 50%) nor overall conception rate (all services) differ between confinements and pasture groups, respectively. Pregnancy rates for 75-day breeding windows were 60% for Holsteins in pasture and 56% in confinement. It can be concluded that the management system had little effect on measures of reproductive efficiency. Delaby *et al.* (2010) also reported that reproduction performance shows little variation in relation to feeding strategies.

Energy balance and rumen function

Energy balance (EB)

In the context of the post-partum dairy cow, energy balance (EB) is defined as the difference between the requirement of energy for body maintenance, tissue growth and milk production (energy required) and the intake of utilizable energy (energy ingested) (Roca-Fernández, 2011). Dairy cattle undergo an energy deficit in early lactation because maximum milk production is attained prior to maximum feed consumption (Bauman and Currie, 1980) due to increases in appetite and in absorptive capacity of the digestive tract usually occur more slowly than the increase in MY. Hence, a shortfall in nutrients is faced in early lactation and has to be met by the use of reserves of adipose tissue lipids, proteins and skeletal muscle and minerals from bone. It is established that animals in early lactation have higher energy requirements than can be supported by dietary intake and are in negative EB (Canfield and Butler, 1990). High producing cows experience a variable period of negative EB during early lactation that is characterized by the loss of BW and the mobilization of body tissue reserves (Beam and Butler, 1999). Cows may experience this negative EB for first 12 weeks of lactation (Bauman and Currie, 1980; Butler *et al.* 1981). The ability to achieve concurrent reciprocal adaptations in the metabolic activity of different tissues is a major factor in the efficient nutrient partitioning and it is based on a hierarchy of nutrient utilization, known as homeorhesis (Bauman and Currie, 1980). Within their ranking, reproduction ranks below maintenance, production and growth, suggesting that reproduction is under severe metabolic pressure at this time after calving. Reproduction seems to be affected by both the acute and the sub-acute nutrient deficiencies, induced by the rising demand for the nutrients and metabolites, which is rapid to be compensated for by the bodies' homeostatic mechanisms (Britt, 1994).

According to Villa-Godoy *et al.* (1988), at least 80% of dairy cows experience negative EB in early lactation. Hernández Urdaneta *et al.* (1976) showed that to maximize energy intake in early lactation, the forage to concentrate ratio needs to be reduced, thereby, reducing the negative EB. Holter *et al.* (1990) reported that body fat balance was strongly negative in groups of both fat and thin cows in the first few weeks post-calving. This was especially true for the fat cows at 6 weeks post-calving, which also had peak MY at this stage. Apparently in this study the group with the fattest reserves mobilized most body reserves for milk synthesis. According to Jarrige (1989), cows with a peak MY of 30 to 35 kg/day can tolerate an average energy deficit of 2.5 UFL/day over the first 7 to 8 weeks of lactation with most of this occurring in the first 3 to 5 weeks. This trend is similar to that found by Sutter and Beever (2000) for animals offered a hay based diet where EB declined from a maximum of 63.8 MJ of ME/day in the first week of lactation to 20.7 MJ of ME/day in week 8 of lactation. Butler *et al.* (1981) found that cows reached positive balance by day 80 post-calving on average. Villa-Godoy *et al.* (1988) stated that greater than 50 days post-calving would be required for cows to reach positive EB.

Several authors (Butler *et al.* 1981; Butler and Smith, 1989; Staples *et al.* 1990; Zurek *et al.* 1995; Beam and Butler, 1997; Beam and Butler, 1998) have demonstrated that the extent and the duration of negative EB after calving are the most important indicators determining when a cow will resume normal ovarian activity. It is known that negative EB affects the timing of first ovulation and the resumption of normal ovarian cyclicity, and the negative EB after calving has also been shown to be critical for the expression of oestrus behaviour (Schopper and Claus, 1986).

Two important factors have been linked to the number of days between calving and normal ovarian activity of healthy dairy cows: amount of nutrients delivered for the dairy cows' metabolism (body tissue metabolism and DM intake) and quantity of milk produced (Marion *et al.* 1968;

Whitmore *et al.* 1974). Attempts have been made to link the interval between calving and first ovulation to metabolic status. A significant, positive relationship has been observed (Butler *et al.* 1981) between EB over the first weeks post-partum and the interval to first observation oestrus while at the same time others were unable to relate mean negative EB with duration of post-partum anoestrus. In several studies conducted by different authors (Butler *et al.* 1981; Canfield *et al.* 1990; Canfield and Butler, 1991), it has been reported that first ovulation occurred 10 and 14 days after daily EB nadir, respectively. At ovulation, EB was still negative but in all cases was decreasing.

Rumen acclimatization

The rumen is a dynamic ecosystem, which needs time to adjust to the changes in the level and composition of the diet that occur at calving. Simple and complex carbohydrates are digested by rumen microbes and converted into volatile fatty acids (VFA). These VFA, which consist mainly of acetic, propionic and butyric acids, are the primary energy source for ruminants. When large amounts of forage are fed, the formation of acetic acid predominates (60-70% of total) with lesser amounts of propionic (15-20%) and butyric (5-15%) acids occurring (Roca-Fernández, 2011). In this case, the supply of acetate may be adequate to maximize milk fat production, but the amount of propionate produced in the rumen may limit the amount of milk produced because of limited supply of glucose (especially in early lactation). However, when grain feeding is increased or when finely ground forages are fed, the proportion of acetic acid may decrease to 40%, while the amount of propionic acid may increase to 40%. Such a change in VFA production generally is associated with a reduction in milk fat. In addition, excess propionate relative to acetate makes the cow use the available energy for fatty tissue deposition (BW gain) rather than milk synthesis. Thus, excess concentrates in the ration may lead to fat cows. Continued feeding of this type of ration may have a detrimental effect on the health of the cow, which is more likely to have a difficult calving and to develop fatty liver or ketosis. On the other hand, not enough concentrate in the ration limits energy intake, MY and milk protein production.The benefit of adding concentrate to the pre-partum diet is to adapt the ruminal tissues and the ruminal microbial population to the type of diet that will be fed after calving (Goff and Horst, 1997).

Concentrate feeding increases the length of the ruminal papillae, which are the structures that absorb the VFA produced from ruminal fermentation. Thus, increasing pre-partum energy intake, by increasing the intake of fermentable carbohydrate may provide benefits beyond the addition of energy. A reduction in the length of the rumen papillae reduces the absorptive capacity of the VFA in the rumen mucosa by a 50% in the dry non lactating period. Dirksen *et al.* (1985) stated that cows that are fed high energy diets pre-calving have a more adapted mucosa for higher intake of concentrates post-calving and better absorption of VFA. Research has shown that rumen papillae elongate in the presence of increased concentrations of VFA (Dirksen *et al.* 1985), thereby, increasing the absorptive capacity of the rumen. The VFA, as propionic and butyric, produced in the rumen during bacterial decomposition are responsible for the development of ruminal mucosa in calves and for the structure of the mucosa in older animals (Brownlee, 1956; Sander *et al.* 1959; Sakata and Tamate, 1978; Sakata and Tamate, 1979). Development of the ruminal papillae is essential to minimize ruminal VFA accumulation, reduction in ruminal pH, and the likelihood of acidosis when high concentrate lactation diets are introduced post-partum. The normal pH range in the rumen of adult cattle is between 5.5 and 7. This may drop to 5 or less after the animal has consumed feed high in fermentable carbohydrates, which may expose the animal to sub-clinical acidosis.

Development of ruminal papillae takes 4 to 6 weeks according to Dirksen *et al.* (1985). Therefore, concentrate must be increased during the pre-partum period to benefit the cow post-partum. The ruminal papillae reach their maximum size 6 to 8 weeks after parturition. However, by introducing a high energy diet low in fibre and high in fermentable carbohydrates even 2 weeks prior to calving resulted in maximal papillae size being reached 3 to 4 weeks after calving (Mayer *et al.* 1986). The ruminal mucosa, therefore, must be considered a metabolic organ functioning between the content of the rumen and the bloodstream (Dirksen *et al.* 1985).

The freshly calved cow if abruptly switched to a high energy lactation diet is at risk of developing rumen acidosis because the rumen bacteria producing lactate respond rapidly to the higher starch diets and produce high amounts of lactate. The bacterial population that metabolizes lactate responds slowly to a change in diet, requiring 3 to 4 weeks to reach levels that effectively prevent lactate from accumulating in the rumen (Goff and Horst, 1997). The acidity of lactate is 10 times stronger than that of propionate, acetate or butyrate so its presence has a greater effect on rumen pH than VFA. Lactate and VFA are only absorbed in the free acid state by the rumen epithelium. Because lactate is more acidic than VFA, lactate is absorbed more slowly from the rumen. Poorly developed rumen epithelia of the un-adapted cows are not able to absorb the lactate and VFA quickly enough to prevent the build up of organic acids within the rumen. This causes rumen pH to fall to the point at which the protozoa and many of the bacteria are killed or are inactive (Goff and Horst, 1997).

Rumen fermentation patterns

Cows grazing good quality, young, lush pastures (with high contents of CP, which is rapidly degraded in the rumen) contain higher rumen concentrations of the total ammonia N (NH_3N) than cows fed a typical TMR diet. Data reported by Beever (1993) revealed that of the NH_3N produced in the rumen, only 30% was incorporated into microbial N and much of the remainder was absorbed across the rumen wall. Ammonia absorbed will be converted into urea by the liver, a conversion that costs the animal about 12 kcal/g of N (Van Soest, 1994). Most rumen microbes depend on carbohydrates as sources of energy (Hoover and Stokes, 1991). Thus, under a grazing situation, adding fermentable carbohydrates should promote the microbial utilization of excess ammonia. Furthermore, synchronization of carbohydrate and protein degradation in the rumen should improve NH_3N utilization. Therefore, different amounts and / or types of energy sources will have different effects in rumen fermentation patterns. Diurnal variations of rumen pH, total VFA and NH_3N concentrations have been barely studied. Van Vuuren *et al.* (1986) studied the influence of level and composition of concentrate supplements on rumen fermentation patterns of six rumen cannulated Dutch-Friesian lactating dairy cows. Cows grazed pastures based on 80 to 90% perennial ryegrass and were supplemented either with a high starch or a low starch diet (25.8 or 1.5% DM of starch) in two different amounts (7 or 1 kg/day), given in two equal portions at milking (06.00 and 16.00 h). No difference was found in rumen pH between treatments, except for the 08.00 h samples, which were the lowest for cows supplemented with 7 kg of the high starch concentrate. The lowest pH occurred at 24.00 h, after the pm milking, when it varied from 6.2 to 5.2. Total VFA and NH_3N concentrations had an inverse pattern compared to rumen pH. Maximum concentrations were at 24.00 h for all treatments. Van Vuuren *et al.* (1986) speculated that this could have been due to a higher PDMI during late afternoon and early evening and due to the higher sugar concentrations in the herbage DM at that particular time of the day. Total VFA were not different among treatments; however, NH_3N and iso-acid concentrations were higher on cows supplemented with 1 kg of concentrate. In the same study the effects of feeding concentrates were less pronounced and, therefore, amount and composition of concentrate mixtures apparently did not influence patterns of VFA concentration and pH value (Van Vuuren *et al.* 1986). In another study, Berzaghi *et al.* (1996) compared rumen patterns of lactating grazing Holstein cows either supplemented with 5.4 kg of corn or not supplemented. No difference was observed in rumen pH between supplemented and unsupplemented cows. However, total VFA concentrations and propionate as a % of total VFA tended to be higher in cows supplemented with 5.4 kg of corn, leading to lower acetate to propionate ratio in the same treatment.

Substitution rate and milk response at pasture

Successful pasture-based milk production systems pivot on balancing cows feed requirements with seasonal and annual fluctuations in pasture production (Roca-Fernández, 2011). In order to maximise cow production from grazing systems, it is necessary to reach an efficient utilization of grazed grass for feeding cows and the development of appropriate grazing management systems designed to maximize daily PDMI (Roca-Fernández *et al.* 2011), while maintaining high sward quality over the grazing season PDMI (Roca-Fernández *et al.* 2012a) by keeping high pasture levels of crude protein, water soluble carbohydrates and digestibility of organic matter and low levels of acid and neutral detergent fibers in the swards (Roca-Fernández, 2013).

Most grazing studies indicate that one of the key factors influencing substitution rate (SRt) at pasture, defined as kg reduction in PDMI per kg increase in supplement DM intake, is herbage availability. Responses to supplementation at pasture are dependent upon the effects of supplementation on PDMI. When daily herbage allowance (DHA) is high, supplementation results in a large substitution effect with a small increase in TDMI, SRt is around 0.6 kg per kg increase in supplement resulting in milk response (MR) between 0.4 and 0.6 kg milk per kg increase in concentrate DM intake (Journet and Demarquilly, 1979; Leaver, 1985; Mayne, 1991).

Nevertheless, Bargo *et al.* (2002) and Horan *et al.* (2005) have obtained an efficiency of over 1 kg of milk per 1 kg DM of concentrate. Meijs (1981) and Hijink *et al.* (1982) have also reported that the SRt increases slightly with the amount of concentrate when fresh grasses are fed indoors; but grazing trials failed to obtain this response (Meijs and Hoekstra, 1984; Kibon and Holmes, 1987; Opatpatanakit *et al.* 1993). The reduction in PDMI is essentially mediated by a reduction of 10-20 min. per kg concentrate DM in time spent grazing (Combellas *et al.* 1979; Kibon and Holmes, 1987). Kellaway and Porta (1993) suggested that long-term factors should be considered in an economic evaluation of supplementation at pasture. These factors include an increase in stocking rate (SR), improvement in herbage utilization, positive effects on BCS and reproduction, increase in lactation length and positive effects on milk composition. In contrast, when DHA is reduced, SRt is lower and the response in milk and daily intake is increased. When grazing cows are offered concentrate, PDMI is reduced as a result of herbage substitution, unless DHA is severely restricting intake.

SRt is also positively correlated with herbage digestibility (Grainger and Mathews, 1989) and this explains why production responses to supplementation are lower in spring and higher later in the season, and on swards with a lower proportion of green leaf. Similarly, because mean PDMI does not differ between strip-grazing and rotational grazing, the efficiency of supplement is independent of the grazing management (Hoden *et al.* 1987). A higher response to concentrate is achieved when a low DHA is offered or when the pre-grazing SH is low (Wilkins *et al.* 1995). Bargo *et al.* (2002) found a 2 kg/day decrease in PDMI at a low DHA (26.7 kg DM/cow/day) and a decrease of 4.4 kg/day when a high DHA (48.9 kg DM/cow/day) was offered in conjunction with concentrate (7 kg DM/cow/day). Robaina *et al.* (1998) showed a higher response to concentrate when animals in mid-late lactation were offered a low compared to a high DHA. Hoden *et al.* (1991) informed an increase in milk production response from 0.5 to 0.8 kg milk per kg DM of concentrate with increasing SR from 2.3 to 3.0 cows/h. On high DHA, the response of MY reaches a plateau after 4 kg whereas on low DHA there is a linear response up to 6 kg of concentrates. Moreover, when feeding concentrate, the decrease in acetate to propionate ratio in the rumen is more pronounced on high (2.6 to 1.9) than on low DHA (2.7 to 2.5) because the cows have access to a more leafy diet, which is more rapidly fermented. Hence the effect of concentrate on milk fat content is higher on high than on low DHA.

Higher SRt and lower MR are usually observed with forage supplements due to higher forage fill value, with high DHA. Nonetheless, there is a wide variation in responses to supplementary feeding at pasture, depending on grazing conditions, production potential of the grazing animal and level of supplementation. Thus, it appears that there is scope for substantial improvement in response to supplementary feed inputs at grazing providing that concentrate inputs should be defined according to the grazing conditions which determine the EB of the unsupplemented cows. High yielding cows have a greater nutrient demand and this is reflected by an increased PDMI when DHA is high, with incremental increases in PDMI when DHA is high, will incremental increases in intake ranging from 200 to 300 g organic matter (OM) (Peyraud *et al.* 1996) to 460 g OM (Stakelum, 1993) per kg increase in milk. As MY increases, the incremental increase in PDMI tends to decrease, as a result of behavioral constraints and, thus, the increase in intake provides only half to two thirds of the net energy lactation (NEL) requirement per kg of additional milk produced for high yielding dairy cows. Consequently, high yielding cows produce a greater response to concentrate supplementation, with Hoden *et al.* (1991) observing daily responses of 0.55, 0.77 and 0.84 kg milk per kg DM of concentrate for cows yielding at turnout 25, 30 and 35 kg milk/day, respectively.

MR is inversely related to SRt affected by DHA, with high PDMI when low levels of concentrate are fed and with MY when high levels of concentrate are fed. From a review of the literature up until the early 1990s, average SRt published were around 0.6, resulting in an efficiency of 0.4 to 0.6 kg of milk per kg of concentrate DM (Journet and Demarquilly, 1979; Meijs, 1981; Leaver, 1985; Stakelum *et al.* 1988). However, most of these studies were carried out with low to moderate yielding dairy cows in the region of 15 to 25 kg per cow per day. Since the late 1990s, lower SRt and higher efficiencies have been observed than those published previously, with an average substitution rate of 0.40, resulting in an efficiency of 0.92 kg of milk per kg of concentrate (Delagarde *et al.* 2011). The higher MR to concentrate supplementation with HGM cows may be attributed to greater nutrient partition to milk production than with LGM cows. Grazing studies conducted with high producing dairy cows have shown an inconsistent relationship between the amounts of supplement and the MR and SRt (Roca-Fernández, 2011).

Kellaway and Porta (1993) have suggested that SRt increases with the amount of concentrate. Peyraud and Delaby (2001), however, reported that in the range of 2 to 6 kg DM/day, amount of concentrate had no consistent effect on SRt. Over four studies, three studies showed a negative relationship between MR and SRt. In contrast, Dillon *et al.* (1997) reported results from 2 years showing reductions in SRt and MR for cows grazing ryegrass pasture when the amount of supplementation was increased from 2 to 4 kg DM/cow/day.

The marginal MR to increasing amounts of concentrate has been described as curvilinear; i.e., the marginal increase in milk per kilogram of concentrate decreases as the amount of concentrate increases (Kellaway and Porta, 1993). Marginal MR decreased above 3 to 4 kg DM/cow/day of concentrate in some studies, but this is not consistent and occurred when pasture quality and quantity were not limiting and with dairy cows of moderate genetic merit (Peyraud and Delaby, 2001).

Grazing studies evaluating the effect of DHA on SRt and MR of high producing dairy cows reported that SRt increased and MR decreased as DHA increased (Meijs and Hoekstra, 1984; Stockdale and Trigg, 1985; Stakelum, 1986a; Stakelum 1986b; Grainger and Mathews, 1989; Stockdale, 1999; Robaina *et al.* 1998; Bargo *et al.* 2002). Many of these studies were conducted with low producing cows supplemented with less than 5 kg DM/cow/day of concentrate; only the study of Bargo *et al.* (2002) reported high producing cows fed more than 7 kg DM/cow/day of concentrate.

When stratifying the treatments in those studies as either low DHA (<25 kg DM/cow/day; range: 7.6 to 25 kg

DM/cow/day) or high DHA (>25 kg DM/cow/day; range: 25 to 42.3 kg DM/cow/day), SRt averaged 0.20 kg pasture/kg concentrate (range: 0 to 0.31 kg pasture per kg concentrate) at low DHA and 0.62 kg pasture per kg concentrate (range: 0.55 to 0.69 kg pasture/ kg concentrate) at high DHA. Considering the study effect as random, a significant regression was found between SRt (kg pasture per kg concentrate) and DHA (kg DM/cow per day):

SRt= −0.55 (SE 0.13) + 0.05 (SE 0.009) PA − 0.0006 (SE 0.0002)

DHA2 (r^2=0.94) and a negative relationship between MR (kg milk/kg concentrate) and SRt (kg pasture/kg concentrate) was found:

MR= 1.71 (SE 0.29) − 2.01 (SE 0.66)

SRt (r^2= 0.43), indicating that the lowest the SRt the highest the MR expected. This agrees with Stockdale (2000), who summarized data from 20 grazing experiments and reported that MR was negatively related to SRt.

Under good grazing conditions, giving conserved forages as a buffer feed result in SRt over 0.9 and very low MR or even a decrease in MY (Bryant and Donnelly, 1974; Leaver, 1985). Thus, conserved forages must be provided only during periods of grass shortage or in the areas where availability of grass is not sufficient.

Therefore, TDMI increases and the response to supplementary forage are much higher in summer than in spring (Phillips and Leaver, 1985). In Galicia (NW Spain), the maize and ryegrass crops have showed a greater silage production than the grass only systems that allow improving the SR from 2.1 to 2.7 cows per ha while maintaining individual milk performances (Mosquera-Losada and González-Rodríguez, 1998).

The provision of supplementary feed in addition to grass silage normally results in a reduction in silage intake, with typical SRt ranging between 0.3 and 0.7 kg reduction in silage DM intake per kg increase in concentrate DM intake. Mayne and Peyraud (1996) have shown that SRt is highly correlated with the intake of silage as a sole feed, although this relationship is also influenced by supplement type. Consequently, with high intake potential silages, increases in supplement feed level will result in higher SRt and lower production responses than those obtained with low intake silages.

More consideration is now being given to formulating supplementary feeds which act as true complementary feeds with high quality grass silage. Another forage supplement for lactating dairy cows under grazing systems is maize silage. Holden *et al.* (1995) conducted a study to determine the effects of supplementation of high producing Holstein cows with 2.3 kg/day of corn silage DM in addition to grain (1 kg of grain DM per 4 kg of milk) based on milk production at the start of the trial. Cows supplemented with maize silage had significant lower PDMI than the control group (11.5 vs. 14.2 kg DM). No significant difference occurred in TDMI (22.5 *vs.* 22.7 kg DM). Maize silage supplementation also showed a positive effect on milk production when the amount of pasture offered was low (Stockdale, 1994). However, where DHA was adequate, supplementation with corn silage reduced PDMI and resulted in similar TDMI and similar milk production (Holden *et al.* 1995). A study conducted by Burke *et al.* (2008) which offered autumn calving dairy cows a low DHA (14.6 kg DM/cow/day >4cm) in combination with 4 kg DM of either maize silage or concentrate during the early spring period reported a SRt of < 0.5 which ensured a high level of herbage utilization and a high response to offered supplement. The effects of feeding concentrate on cow performance in relation to SR were reviewed by Peyraud and Delaby (2001). Efficient response of one kg of milk to one kg of concentrate is now reached when the amount of concentrate per cow does not exceed 6 kg/day. Moreover, the efficiency of supplementation at grazing appears to be closely related to EB of the cows, and it increases when PDMI is restricted through increased SR, with economic returns depending of the concentrate to milk price ratios and most often positive. Therefore, feeding concentrate can be a very efficient tool to maintain a high SR and good sward management, which allows the control of post-grazing SH while achieving high MY per cow and per ha with high economic returns. The effects of concentrate type on MR at pasture are extremely variable. Delaby and Peyraud (1999) estimated that milk production response reached a plateau at 4 kg DM/cow/day of concentrate when DHA was high; whereas when herbage was restricted there was a linear response up to 6 kg DM/cow/day of concentrate. The interaction between level of concentrate supplementation and DHA on milk production response can be substantial. SR increases with increasing pasture availability, from 0 for high GP to 0.6-0.8 for low GP (Stakelum *et al.* 1988; Stockdale, 2000; Peyraud and Delaby, 2001). The efficiency and the SR, is influenced by a large range of factors such as DHA, herbage composition, concentrate feeding level, concentrate composition and potential MY of the cows evaluated (Bargo *et al.* 2002). Delaby and Peyraud (1994) observed that energy source in the concentrate (starch or fibre and rate of degradation) had little effect on milk output and composition when moderate levels (2-4 kg DM/cow/day) of concentrates are fed.

Compared with wheat, fibre concentrate slightly increased fat content (+1.3 g/kg) and decreased protein content (-0.5 g/kg) (Delaby and Peyraud, 1994). The nature of energy does not appear to affect the SRt when fresh grass is fed indoors. In pasture, PDMI was shown to be about 1 kg higher when cows are supplemented with 5 kg DM/cow/day of high fibre concentrate compared with 5 kg DM/cow/day of high starch concentrate (Kibon and Holmes, 1987), probably because net energy content is lower for fibrous than for starch concentrate. On more severe grazing conditions there is no effect of the source of energy on PDMI and MY (Kibon and Holmes, 1987; Delagarde *et al.* 1999). The implication of this result is that there is little improvement to be expected by modifying the nature of energy at grazing when low concentrate amounts are given. However, the effect of the energy source becomes more pronounced when more than 8 kg DM/cow/day of concentrate are fed by grazing dairy cows. To avoid metabolic health problems such as acidosis or sub clinical acidosis, it is not recommended to supplement more than 10 kg DM/cow/day (or >50% of the total diet DMI). At that limit, decreased marginal MR traditionally observed when supplementation is increased did not occur with high producing cows. Another factor that needs to be considered is the sward quality. The neutral detergent fiber (NDF) was >50% in several studies, suggesting that high fibre intake may allow for feeding high amounts of concentrate.

Milk quality

Milk and dairy products are important components of western diets. The composition of raw bovine milk determines the nutritional value and the technological properties of milk and dairy products and it also conditions the farmers' milk price (Roca-Fernández, 2011). Therefore, the composition of milk is of great importance for the milk producers and the dairy industry. Milk of many species is consumed by humans (i.e. goat, sheep, cows and buffalo) but bovine milk is economically the most important. Milk composition varies with breed, health status, lactation stage and parity of the animal.

Furthermore, milk composition depends on feeding practices and genetic characteristics of the animal (Fox and McSweeney, 1998; Roca-Fernández et al. 2012b). New findings also highlighted the importance of considering seasonal variation in milk composition across the year (Heck *et al.* 2009). In the last decades, significant progresses were made in order to improve MY and composition of bovine milk, mainly in relation to milk protein and milk fatty acids (FA) profile by both breeding and feeding practices (Roca-Fernández et al. 2012b)

Milk chemical composition

Milk is mainly considered a product of the mammary gland secretion, and it is a complex and nutritious fluid that contains five main components such as water, lipids, proteins, sugars and minerals (Ling *et al.* 1961; Jenness, 1974). Milk is defined as an emulsion of fat globules and a suspension of casein micelles (casein, Ca and P), all suspended in an aqueous phase which contains solubilized lactose, whey proteins and some minerals. It also contains immunoglobulins, hormones, growth factors, cytokines, nucleotides, peptides, polyamines, enzymes and other bioactive peptides with potential antithypertensive, antithrombotic and antimicrobial activities (Park *et al.* 2007). The casein micelles and the fat globules gave milk most of its physical characteristics, and gave taste and flavor to dairy products such as butter, cheese, yogurt, etc. The main role of milk is to provide nourishment and protection for the mammalian young cattle. However, milk is also highlighted as an important food source for humans (Jenness, 1974). Milk is a highly perishable product that should be cooled to about 4 $^{\circ}$C as soon as possible after collection. Extremes of temperature, acidity or contamination by microorganisms can rapidly decrease its quality.

Water

Cow milk is about 88% water and the amount of water in milk is regulated by the amount of lactose synthesized by the secretory cells of the mammary gland. The water that goes into the milk is delivered to the mammary gland by the blood. The addition of water to milk can be detected by different methods. These methods are based on changes in freezing point of the milk or on changes on refraction of light of the whey component of milk after precipitation and removal of casein and milk fat (Mabrook and Petti, 2003).

Milk fat

Fat is the major source of energy in milk and it is used by the mammalian newborn for accumulating body adipose tissue. Fat in milk is present as fat globules ranging from 0.1 to 15 µm in diameter. A thin membrane whose properties are different from both milk fat and plasma covers these fat globules or droplets. The fat globule membrane helps to stabilize the fat globules in an emulsion within the aqueous environment of the milk. Milk fat plays an important role in the structure, mouth feel, flavor and stability of milk, butter, dairy products and a host of foods in which milk or milk components are added as functional ingredients. Certain of these functional properties have made butterfat a desirable ingredient in processing and food formulations. For example, butterfat is an essential component in the structure and texture of ice-cream and pastries (Pomeranz, 1985). Nevertheless, many of the properties of milk fat are less than optimal for food uses (O'Donnell, 1989). Milk fat is the most variable component in bovine milk in both their concentration and chemical composition, whether inter- or intra-species differences are considered (Gibson, 1989). Milk lipid composition is influenced by environmental and physiological factors, including age, stage of lactation, gestation length and diet. Some intra-species differences reflect inherited variation (Jenness, 1974). Milk lipids belong to various lipid classes, but triacylglycerides account for 97-98% of lipid in milk (Iverson and Oftedal, 1995). Other lipids classes include di- and mono-acyglycerols, phospholipids, free cholesterol, cholesterol esters and fatty acids (Garton, 1963). Normally, fat makes up from 35 to 60 g/kg of milk, varying between breeds and with feeding practices. A ration too rich in concentrates that do not elicit rumination in the cow may result in milk with a depressed percentage of milk fat (20 to 25 g/kg).

Milk protein

It consists for a large part (±90%) of the six main milk proteins à-lactalbumin (α-LA), ß-lactoglobulin (ß-LG), caseins (αS1-CN, α-S2-CN, ,-CN and ,-CN). The other part of the protein fraction (±10%) consists of minor proteins like bovine serum albumin, ,-caseins, proteose peptones, immunoglobulins, lactoferrine, lactoperoxidase and a large number of other proteins that occur in very low concentrations. The concentration of protein in milk of dairy cows varies from 30 to 40 g/kg. This amount varies with the breed of the cow and in proportion to the amount of fat in the milk (Roca-Fernández *et al*. 2013). There is a close relationship between the amount of fat and the amount of protein in milk therefore, the highest the fat, the highest the protein. the higher the fat, the higher the protein. The protein falls into two major groups (Jenness, 1974), based on their behavior at pH 4.6: caseins (80%) and whey proteins (20%). The caseins are the proteins that precipitate and the whey proteins are the proteins that remain soluble at this pH. Historically, this classification followed the process of cheese making, which consists of separating the casein curd from the whey after the milk has clotted under the action of rennin or rennet (a digestive enzyme collected from the stomach of calves). The behavior of the different types of caseins in milk when treated with heat, different pH (acidity) and different salt concentrations provide the characteristics of cheeses, fermented milk products and different forms of milk (condensed, dried, etc.). Occasionally, infants or young children are allergic to milk because their bodies develop a reaction to the proteins in the milk. The allergy causes rash, asthma and/or gastrointestinal disorders (colic, diarrhea, etc.). In cases of allergies, goat milk is often used as a substitute; however, sometimes hydrolyzed casein milks must be used. Most of the caseins in milk exist in colloidal particles called casein micelles. In milk of most mammalian species, there are 3-4 caseins; the different caseins are distinct molecules but are similar in structure. Casein is composed of several similar proteins, which form a multi-molecular granular structure called a casein micelle. The main function of the micelle is thought to be the supply large amount of insoluble calcium phosphate to the mammalian young but its properties have also a major influence on the technological properties of the milk. The micellar structure of milk casein is an important part in the mode of milk digestion in the stomach and the basis of many of the milk products industries such as cheese.

Milk fatty acids

It was probably Booth *et al*. (1935) who for the first time established the presence of conjugated fatty acids (FA) in milk fat. They reported that when cows were turned out to pasture after winter, the FA of milk fat showed greatly increased absorption in the ultraviolet region at 230 nm. Moore (1939) concluded that absorption at 230 nm was the result of two conjugated double bonds. Hilditch and Jasperson (1941) and Hilditch and Jasperson (1945) suggested that conjugated unsaturation occurred with polyunsaturated FA of 18C chains. Bartlett and Chapman (1961) found a constant relationship between *trans*-C and conjugated unsaturation in a large C18:1 number of butter samples as determined by differential infrared spectroscopy, which prompted them to suggest a sequence of reactions that would help to explain the biohydrogenation of conjugated linoleic acid (CLA) in the rumen. Riel (1963) showed a 2-fold increase in milk fat conjugated dienes during summer when cows were grazing on pasture compared with winter when cows were fed TMR. Parodi (1977) determined that the conjugated double bonds were cis-9 and trans-11 of C. Conjugation at other positions was found C18:2 later. Jensen (2002) has catalogued approximately 400 different FA in milk fat; most of these are products of ruminal microbial modification of dietary FA. The FA of ruminant milk has a dual origin. Those of chain length C4:0 to C14:0 are derived from de novo synthesis in the mammary gland and are considered as saturated fatty acids (SFA), whereas those of C18:0 and longer are derived from the diet and considered as unsaturated fatty acids (UFA). The FA of greatest proportion, C16: 0 arises from both sources; the relative amounts from each source can be influenced by the diet. Thus, it is not surprising that milk fat, containing about 70% of the fatty acids (FA) as saturated (SFA), 25% as monounsaturated (MUFA) and 5% as polyunsaturated (PUFA) (Grummer, 1991), has been perceived for long-time to be detrimental to humans' health (Lock *et al*. 2008). Nevertheless, it is worth noting that probably only some of these SFA such as C12: 0, C14: 0 and C16: 0 would be considered as cholesterol-raising saturates, while other SFA contained in milk such as C4: 0, C6: 0, C8: 0, C10: 0 and

C18:0 might suppose no risk of cardiovascular disease (Lock *et al.* 2008; Parodi, 2009). Roca-Fernández and González-Rodríguez (2012) reported that due to pasture-based milk production systems have high dependence on fresh grass for feeding dairy cattle is possible to increase the added value of the milk, with higher content of conjugated linoleic acid which helps farms to be more profitable and competitive.

Immunoglobulins

They are one of the calf's principal defenses against infectious organisms (viruses, bacteria and etc.). Concentrations of immunoglobulins are especially high in the colostrum, the milk produced immediately at the onset of lactation. Immunoglobulins are not produced in the mammary tissue but they are transferred directly from the blood serum into the milk. The calf can best adsorb the immunoglobulins after birth, with the ability to absorb decreasing to near zero by 36 h of age. This is because, in the first 12 h of life, the calf does not produce appreciable amounts of hydrochloric acid in its stomach so the immunoglobulins are not damaged. Colostrum should be given to the calf as soon after birth as possible. This wills at least double the young calf's chances of survival. Colostral immunoglobulins are stable in the calf's bloodstream for 60 days, providing protection until its own immune system is functional. Not only is colostrum of vital importance to the newborn calf, it also has no commercial value as it is not acceptable for commercial milk collection for human consumption. So, the milk from a cow that has freshly calved must not be included in the milk for sale for 3 to 4 days.

Lactose

The principal carbohydrate in milk is lactose (disaccharide composed of D-glucose and D-galactose). In addition to lactose there are a great variety of saccharides in milk (Jenness *et al.* 1964; Urashima *et al.* 2001). The concentration of lactose in milk is relatively constant and averages about 5% (4.8-5.2%) (Johnson, 1978). As opposed to the concentration of fat in milk, lactose concentration is similar in all breeds and cannot be altered easily by feeding practices. Lactose plays a major role in milk synthesis (Kuhn *et al.* 1980). Because of the close relationship between lactose synthesis and the amount of water drawn into milk, lactose is the least variable component of milk. In a significant portion of the human population, the deficiency of the enzyme lactase in the digestive tract results in the inability to digest lactose (Malagelada, 1995). Most individuals with low lactase activity develop symptoms of intolerance to large doses of lactose, but the majority can consume moderate amounts of milk without discomfort (Johnson *et al.* 1993). Not all dairy products contain similar proportions of lactose. The fermentation of lactose during processing lowers its concentration in yogurts and cheeses. Milk pretreated with lactase, which minimizes the problems associated with lactose intolerance, is now available.

Vitamins and minerals

Milk contains all the major vitamins. The fat soluble vitamins A, D, E and K are found mainly in the milk fat. The B vitamins are also found in the aqueous phase of milk. Milk in some countries such as Canada and USA is being fortified with vitamin D (Calvo Lacosata *et al.* 2004) and vitamin A is also added to fat reduced milk products. Milk is an excellent source of most minerals required for the growth of the young cattle. Furthermore, all minerals considered essential (22) to the human diet are present in milk. There are a great variety of minerals in milk present in a variety of chemical forms. The major cations are sodium (Na), potassium (K), calcium (Ca) and magnesium (Mg,) and the major anions are phosphorus (P) as phosphate, chloride (Cl) and citrate (Jenness, 1974; Peaker, 1977; Gaucheron, 2005). The digestibility of Ca and P are unusually high, in part because they are found in association with the casein of the milk (Holt, 1997).

As a result, milk is the best source of Ca for skeletal growth in the young (Black *et al.* 2002) and maintenance of bone integrity in adults. Another mineral of interest in the milk is iron (Fe). The low Fe concentration in milk cannot meet the needs of the young, but this low level turns out to have a positive aspect because it limits bacterial growth in milk-Fe and it is considered essential for the growth of many bacteria (Bullen *et al.* 1972).

Cells in Milk

The somatic cell count (SCC) in milk does not affect the nutritional quality per se. They are only significant as indicators of other processes that may be taking place in the mammary tissue including inflammation. When SC is present at rates of over half a million per milliliter, there is reason to suspect mastitis.

This is an inflammation of the mammary gland caused by mastitis-causing organisms and rarely physical or chemical trauma, characterized by pathological changes in the mammary tissue; an increased number of SC, physical, chemical and microbiological changes in milk. In cattle, both clinical and sub-clinical mastitis can affect the composition and manufacturing properties of milk (Auldist and Hubble, 1998; Pyorola, 2003).

The reductions in milk production probably are largely due to physical damage to the epithelial cells of the affected mammary gland and a reduction in the synthetic and secretory capacity of the gland as a whole. Shuster *et al.* (1991) hypothesized that part of the decrease of the milk production is due to increased demand for energy of the immune system against the infection, decreased appetite associated with any inflammation and lowered food intake due to pain and decreased movement. Holdaway (1990), Auldist *et al.* (1995) and Auldist and Hubble (1998) reported that there is an increase in proteins of blood serum origin during mastitis and this is due to a disruption to the integrity of the mammary epithelia by microbial toxins and opening of the tight junctions.

Auldist and Hubble (1998) also reported that the decrease in casein concentrations during mastitis is largely due to post-secretory degradation of casein by proteinases originating from mastitis causing organisms, leucocytes or the blood and in part to a reduction in the synthesis and secretion of casein as a result of physical damage to the mammary epithelial cells by microbial toxins during mastitis.

Pathogenic bacteria

During the milking process, presence of mastitis in the udder of dairy cows can cause the pathogenic bacteria in the milk. Bacteria present on the outside skin of the udders and unhygienic milking practices which allow the milk to come in contact with contaminants like feces may also cause pathogenic bacteria in the milk.

Studies on the pathogenesis and epidemiology of a number of these pathogens have been published by some authors (Barkema *et al.* 1998; Sears and Wilson, 2003) and they showed a difference between pathogens in pathogenesis, epidemiology and clinical presentation. Some of the most important pathogenic bacteria in milk are: *Escherichia coli* or *Coliform bacteria*, *Staphylococcus aureus*, *Salmonella*, *Listeria monocytogenes*, *Campylobacter jejuni* and *Yersinia enterocolitica.*

Milk prices in response to milk chemical composition

The need of producing high quality milk from sustainable pasture-based milk production systems is unquestionable, not only from the sanitary point of view but also from the economic aspect related to milk prices due to its direct relationship with the yield and quality of the final product. Table 1 shows that the bigger Galician milk delivers had the highest total milk price, with the highest base milk price and total milk premiums (Roca-Fernández, 2011).

Table 1 Milk price (€/100 kg) and premiums got by farmers for milk delivery at Galicia (NW Spain) in 2009

Milk quota (kg per year)	72001-240000	240001-500000	240001-500000
Total milk price	26.73	28.86	30.30
Base milk price	25.17	26.39	26.97
Total milk premiums	1.57	2.47	3.33
Milk fat	0.21	0.15	0.01
Milk protein	0.04	0.21	0.26
Bacteriology	0.06	0.07	0.05
Somatic cells	-0.04	-0.05	0.04
Other milk premiums	1.29	2.09	2.97

When milk fat and protein are higher than 3.7% and 3.1%, producers are paid a milk quality premium by the dairy industry. Bacteriology and SCC are other relevant parameters that influence milk quality premium when these are lower than 100000 bacteria and 400000 SC, respectively. Despite milk quality is an important factor that might be taken into account when milk price is established, its relevance on the total milk premium is lower than other milk premiums such a quantity of milk delivered by farmers to the dairy industry. In fact, according to the data presented in the table below milk quality premium only represents 0.34 € cents in the total milk price perceived by Galician milk delivers in 2009 while other milk premiums as quantity of milk delivered represented about 4-10 times this value, being three times higher for the biggest milk delivers than for the lowest milk producers.

Inhibitors

Other relevant parameter that nowadays is taking into consideration to improve milk quality is the absence of inhibitors. Milk should not contain antibiotic residues at all. Manufacturers buying milk from milk producers impose stringent financial penalties on farmers producing contaminated milk and have procedures to exclude this milk from the food chain.

Despite legislation and financial penalties, there is evidence to suggest that residues occasionally still cause problems. Antibiotics gain entry to milk because of mastitis treatment and the antibiotics that are commonly used in veterinary medicine to control mastitis belong to six major groups: aminoglycosides, penicillins and cephalosporins, macrolides, quinolones and fluroquinolones, sulphonamides and tetracyclines. Control the absence of these antibiotics in milk is an important issue for dairy producers.

CONCLUSION

Pasture dry matter intake should satisfy dairy cattle requirements for a determinate level of milk production, according to the lactation curve of cows. Animal characteristics (potential milk yield, body weight, body condition score, breed, parity, stage of lactation, fertility and etc. for each cow) and herd management (calving pattern, type and level of supplementation at pasture, grazing intensity, animal behaviour and etc. for each herd) condition cows' needs for milk production in the grazing system. Milk yield and milk quality (milk protein content, milk fat content, milk lactose content and milk fatty acids profile) vary according to cows' lactation stage and these are also dependent on the seasonality of grass production and the changes on sward quality across the grazing season. Adopting good grassland management practices at farm level in order to satisfy cows' needs is an essential tool for future develop ment of sustainable pasture-based milk production systems.

ACKNOWLEDGEMENT

To INIA and CIAM by supporting the research project RTA2005-00204-00.

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