IMPROVEMENT OF THE EFFICIENCY OF THE GRASS PROTEIN IN DAIRY CATTLE USING FOUR DIFFERENT FEED FORMULATIONS

MEJORAMIENTO DE LA EFICIENCIA DE LA PROTEINA DE LOS PASTOS EN BOVINOS DE LECHE UTILIZANDO CUATRO FORMULACIONES DE BALANCEADOS

Francisco Gutiérrez¹, Alex Estrella¹, Eduardo Irazábal¹, Víctor Quimiz¹, Arnulfo Portilla¹, Nancy Bonifaz²

¹Faculty of Agricultural Sciences, Agronomy Engineering, Universidad Central del Ecuador.
²Research group NUNKUI WAKAN, Universidad Politécnica Salesiana del Ecuador, Girón Campus: Av. Isabel la Católica N. 23-52 and Madrid.

*Corresponding authors: fgutierrez@uce.edu.ec, nbonifaz@ups.edu.ec

Abstract

Cattle raising is the most diffused activity in the world. The main source of feeding is the grass. In this research, the efficiency of the crude protein (CP) in grass was evaluated using four different feed formulations in the cattle feeding. When the grass had >18% of CP, the best results were obtained with cattle feed of 12% of CP and non-nitrogenous elements (NNE) in a 68%. It increased the milk production and the protein content, and the milk urea nitrogen (MUN) decreased. On the other hand, if the grass had between 14-18% of CP, the best results were obtained with cattle feed containing CP ≤ of 14% and the NNE ≥ of 61%. It also improved the milk production and the protein content, and the MUN level decreased. Whereas, if the CP in the grass was <14%, it required an additional quantity of CP coming from the cattle feed. The best results were obtained with cattle feed with CP>16% and NNE <57%, increasing the milk production and the protein content, improving the content of MUN in milk.

Keywords: bovine production, supplementation, fodder mix, milk urea nitrogen.
Resumen
La ganadería es la actividad agropecuaria más difundida en el mundo, siendo la principal fuente de alimento el pasto. En esta investigación se evaluó la eficiencia del uso de la proteína bruta (PB) en pastos, utilizando cuatro formulaciones diferentes de balanceados en la alimentación de bovinos. Cuando los pastos tuvieron >18% de PB, los mejores resultados se obtuvieron con un balanceado 12% de (PB), y los elementos no nitrogenados (ENN) en un 68% aumentó la producción de leche, disminuyendo el contenido de proteína (p<0,05) y el nivel de nitrógeno ureico en leche (MUN). Por otro lado, cuando los pastos se encuentran entre 14-18% de PB, se obtuvieron los mejores resultados con balanceados que contienen una (PB) ≤ 14% y los (ENN) ≥ 61%, mejoró la producción de leche y el contenido de proteína (p<0,05) y el nivel de MUN disminuye; mientras que si la (PB) en los pastos <14% requiere una cantidad adicional de (PB) proveniente del balanceado, se obtuvieron los mejores resultados con balanceados de (PB) >al 16% y un (ENN) <57%, aumentando la producción de leche y el contenido de proteína (p<0,05), mejorando el contenido de MUN en leche.

Palabras claves: producción bovina, suplementación, mezcla forrajera, nitrógeno ureico en leche.

1 Introduction

The demand for livestock products in the world has increased progressively according to (FAO, 2009), this is due to two factors: to the increase of the population worldwide, and to the purchasing power making that people can consume products with a more value; this presents a challenge as this activity has to be integrated into a context of finite natural resources, contribution to livelihoods, long-term food security and climate change response (FAO, 2017a). The livestock sector is the world’s largest consumer of agricultural land through grazing and the use of forage crops. Livestock is responsible for most of the world’s land use; pastures and farmlands dedicated to livestock feed production account for almost 80 percent of all agricultural land (Batallas, 2009).

Beef cattle contributes to 40% of the value of world agricultural production and sustains livelihoods and food security for nearly 1 300 million people. The livestock sector is one of the fastest growing sectors in the agricultural economy. The growth and transformation of the sector provides opportunities for agricultural development, poverty reduction and improves food security (FAO, 2017b).

The intensification of livestock systems evaluates the nitrogen losses produced by the gas emission into the atmosphere (ammonia, nitrous oxide, and nitric oxide), and the runoff of nitrates to the surface and underground water. Nitrogen losses can be reduced by improving the animal’s efficiency to use the food protein, reducing loss during storage and management of excreta (Díaz, 2016b). When a cow is only fed with pastures and forages its nitrogen losses to the atmosphere are lower than when it receives a diet composed of pastures and concentrates (Rua, 2016).

In Ecuador, the cattle ranches use a pastoral system for their production; it is important to analyze the systems as a whole, that is, the relation between soil-plant-animal, since the growth of the pasture obeys to the soil nutrients, and pasture nutrients have an impact on the animal production (Batallas, 2009).

This research relates the protein content of the pastures and the interaction they have with the protein content of the formula, and how they influence on the production of milk, protein content and urea in milk.

2 Materials and methods

This research was carried out in the Experimental teaching academic Campus “La Tola” of the Faculty of Agricultural Sciences, Universidad Central del Ecuador, located in Tumbaco parish, Quito, Pichincha, at 2 465 masl 00° 14’ 46” S, Longitude 78° 22’ 00” W, with an annual temperature of 16.3 °C, Annual precipitation 870.3 mm, and an annual relative humidity of 71.75%. For the study, 12 breed Holstein cows were chosen with more than one calving, which were distributed in three groups of 4 animals according to their third lactation. In the first third (0 to 100 days), second third of (100 to 200 days) and in the third third (>to 200 days)

<table>
<thead>
<tr>
<th>Treatments</th>
<th>(PB) %</th>
<th>(EE) %</th>
<th>(FB) %</th>
<th>Ashes</th>
<th>(ENN)%</th>
</tr>
</thead>
<tbody>
<tr>
<td>T1</td>
<td>12</td>
<td>4</td>
<td>8</td>
<td>8</td>
<td>68</td>
</tr>
<tr>
<td>T2</td>
<td>14</td>
<td>3</td>
<td>13</td>
<td>9</td>
<td>61</td>
</tr>
<tr>
<td>T3</td>
<td>16</td>
<td>4</td>
<td>13</td>
<td>10</td>
<td>57</td>
</tr>
<tr>
<td>T4</td>
<td>18</td>
<td>4</td>
<td>13</td>
<td>10</td>
<td>55</td>
</tr>
</tbody>
</table>

PB=Crude protein; FB=Crude fiber; EE=Ethereal extract; (ENN)=Non nitrogen elements

Four treatments were evaluated, which were balanced with different formulations in which the level of (PB) was considered, starting with 12% and increasing to 18%, also the amount of (FB) and (ENN) was considered (Table 1). The Latin square experimental design was used, using four experimental units with four treatments, the third of lactation was considered as a covariate for the variables.
Improvement of the efficiency of the grass protein in dairy cattle using four different feed formulations

wich and statistic differencie (p<0.05), a Tukey test was made. Animals were subjected to treatments with a transition period between diets of in two weeks. The two daily rations of the formulation were provided at 3:00 a.m. and 3:00 p.m. at the time of the milking routine. The amount of the daily ration was calculated according to the treatment and the milk production of animals, for every 5 liters of milk they received 1 Kg of the formula (Table 2).

Table 2. Treatment distribution of animals in the thirds of lactation

<table>
<thead>
<tr>
<th>Animals</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Animal 1</td>
<td>T1 T2 T3 T4</td>
</tr>
<tr>
<td>Animal 2</td>
<td>T4 T1 T2 T3</td>
</tr>
<tr>
<td>Animal 3</td>
<td>T3 T4 T1 T2</td>
</tr>
<tr>
<td>Animal 4</td>
<td>T2 T3 T4 T1</td>
</tr>
</tbody>
</table>

The milk production register was taken daily with the help of a decaliter. On days 7 and 21 of each of the treatments counted after the transition phase, a sample of 40 ml milk was taken in sterile vials, and taken in a refrigerated container to the Milk Quality Laboratory of Universidad Politécnica Salesiana, located in Cayambe. The samples were subjected to compositional analysis (protein, fat, lactose, total solids and non-fatty solids) and urea nitrogen in milk (MUN), the method used in the laboratory is infrared spectrophotometry, in the equipment MIL-KOSCAN FT 6200, PEE02 protocol.

The second analysis was to determine the crude protein of the pastures consumed by the animals at the time of the study. The sample of the forage mixture was taken with the aid of a quadrant of 50 cm long by 50 cm of width, which was made with a flush cut at a ground level; then it was weighed to determine the green matter. The pasture sample was taken to the Laboratory of Nutrition and Animal Health, of the Faculty of Agricultural Sciences of Universidad Central del Ecuador. To determine the dry matter content of the sample, 200 g of the specimen was analyzed in a range at 68 °C for 24 hours. The sample of dry matter obtained from each of the treatments and replications was grounded in a mesh 750 micron to be used in the analysis of protein ((PB)), ethereal extract (EE), crude fiber (FB) and ashes. The methods used were those determined by the AOAC (2010). For the protein analysis ((PB)), the Semimicro Kjeldahl method was used, which consists of a hot digestion with concentrated H2SO4 and catalyst and amino nitrogen; the sample becomes (NH4)2SO4, which subsequently by action of an alkali (NaOH) decomposes, releasing ammonia (NH3) which is distilled and collected in boric acid. Finally, the acid which is proportional to the amount of nitrogen is valued with a normalized acid and from the amount of acid that has reacted with the ammonia.

3 Results

3.1 Milk Production

The best milk productions were obtained with T1 and T2 (Figure 1), when the pastures had >18% (Pb) and between 14-18% (PB); while T3 and T4 recorded smaller productions; the best treatments were T3 and T4 with the (PB) in Pastures <14%. Gagliostro (2012) interprets the protein content in the pastures and concludes that a content higher than 20% is very high and causes excess of NH3 at a ruminal level, and its elimination is expensive and should be supplemented with grains of high ruminal degradability; content between 16 and 20% causes slight excess of NH3 at ruminal level and ensures adequate ruminal functioning, while a content of less than 12% is inadequate for the milk production and for good live weight gains except termination.

A protein-deficient diet causes body deposits to be emptied into the blood, liver, and muscles. As a consequence, the milk production and protein content reduce, and the body fat deposition increases (Velez, 2015). A deficient diet in protein causes body stores to empty into the blood, liver and muscles. As a consequence, milk production and protein content decrease and at the same time increases the deposition of body fat (Velez, 2015). The requirements of crude protein depends on the lactation sta-
ge, for an early stage is 18 - 17%, for an average stage is 17 and 16%, and late between 16 and 15% (Ishler, Heinrichs y Varga, 2013). For cows producing 20 to 25 l/day, 16% of CP in the diet appears as adequate, being feasible to provide it with most forages (INTA, 2014). There is a clear relationship between the increasing level of protein in the ration and milk production, with a disparity of criteria ranging from 14 to 18% of crude protein (Zaragoza, Seguí y Sanz, 1998). The supplementation of the cows allows to balance the meadow, the prairie protein must be corrected through the supply of concentrates with good contribution of fermentable non-structural carbohydrates in the rumen to increase the synthesis of microbial protein and decrease the high levels of ammonium and the excretion of urea in milk and urine, a cows could produce up to 26 liters of milk per day with only forages (Klein, N.d.). The pastures in the real production systems present physical and quality limitations that make that the consumptions achieved are lower than the potential values in milk production, which makes it necessary to introduce supplementation (Cangiano, 2011). The basis of the feeding of the cows should be the forage that must be complemented with a concentrate whose compositional characteristics vary to complete it (Shimada Miyasaka, 2003).

### 3.2 Protein content in the milk

For the protein variable in milk, the best behaved treatment was the T1 with pastures of >18%, reaching 3.6% (Figure 2); the other treatments with different pasture contents of (PB) had protein values in milk between 3.3% and 3.1%; except for treatment T2 and T1 with a content of (PB) in pastures <14%. This shows that at this level the animals had deficiencies of (PB) in their diet. The average protein in milk for the Holstein Friesian breed is 3.3% (Hazard, 2015), and the minimum protein content in raw milk to be marketed in Ecuador according to the standard (INEN, 2015) is 2.9%.
Pastoral systems of temperate zones have an imbalance between energy and protein, this is because pastures have high quality (high digestibility and high protein); the higher the quality of the pasture, the more effective the degradability of the starch (Santini, 2014). If the energy and protein ratio is adequate, the rumen bacteria can synthesize NH₃ and transform it to bacterial protein; the quality of the bacterial protein is high for ruminants, this is reflected in a greater synthesis of milk protein in the udder. Approximately 95% of the nitrogen in milk is in the form of protein; the rest is found in substances such as urea, creatine, glucosamine and ammonia, which pass from blood to milk (McDonald, 1999). The objective of the protein in ruminants is to provide the adequate amount of protein degraded in rumen to optimize its efficiency and obtain the desired animal production with the minimum amount of crude protein (Dairy Cattle Nutrition, 2001).

Currently, the efficiency of nitrogen use (EUN) is calculated as the proportion of nitrogen excreted in milk or meat on the nitrogen consumed; in ruminants (Eun) average is 25%, this can be improved by limiting the protein content in the diet and it has been shown that diets with a content of 16.5% are sufficient to obtain maximum yields in high production dairy cattle (Díaz, 2016a). The average efficiency of the use of nitrogen in dairy cattle is 26% (Calsamiglia, 2014). Gaining efficiency in the nutrients consumed by animals is a permanent challenge of dairy production and developing a technology that develops dynamic feeding systems (Martínez del Olmo, 2015). The inclusion of concentrated supplements modifies the composition of milk, generally with a decrease in fat concentration and an increase in milk protein content (Bargo, 2003).

### 3.3 Urea content in the milk (MUN)

In MUN, no difference was found (p<0.05) for values ranging from >18% to 14 to 18% of (PB); however, T4 registered an elevated value, 18 mg/dl of MUN (Figure ??); while the content of (PB) <14% had differences with the previous two levels, registering lower values than the accepted between 11 and 10 mg/dl of MUN. The most suitable values of urea in milk in works carried out in Ecuador are between 12 and 15 mg/dl, values higher than 18 mg/dl imply high risk in the productive and reproductive management of the bovines (Bonifaz y Gutiérrez, 2013). The values per treatment range from 9 to 18.3 mg / dL, the lowest MUN values were found in high production cows and were increased as lactation progressed and milk production decreased (Peña, 2002). Values of urea in blood or milk lower than 2.5 mmol / L (7.0 mg / dL MUN or PUN), indicate low content of degradable protein in the diet in relation to the ruminal availability of energy, while higher values at 7.0 mmol / L (19.6 mg / dL MUN or PUN) indicates a reverse situation (Scandolo, 2007). The MUN level can vary between breeds of dairy cows the MUN in Holstein cows is 14.18 mg / dL (Doska et al., 2012).

![Figure 3. Influence of crude protein content on pastures and formulations on the urea content in milk (MUN) mg= milligrams; dl=deciliter; (PB)=crude protein.](Image)
The excess of protein causes the animal to produce urea secretions synthesized from the rumen ammonia, the nitrogen excreted in milk and feces is positively and linearly related to the nitrogen consumed, while the nitrogen excreted in urine is exponentially related to nitrogen consumption; this demonstrates the importance of not supplying protein above the needs of the animal (Díaz, 2016b).

An excess of protein (more than 18% of PC) increases urea levels in the blood and milk, and especially if it is easily degradable, it affects the liver that has to transform the NH₃ into urea. This transformation is a gradual process; meanwhile, the excess of NH₃ circulating in the blood affects the reproduction by a change in the pH of the uterus after ovulation, and by the toxic effect of ammonia and its metabolites on the gametes and the embryo (Elrod y Butler, 1993).

The excess of urea may affect the reproductive processes through the toxic effects on the ovum, sperm, and the embryo (Melendez, 2011).

The transformation of ammonia into urea demands a considerable amount of energy, so it decreases the availability of energy for productive processes and also requires the amino acid arginine; if the amount of ammonia is high, arginine deficiency may occur, affecting the production (Zinn y Owens, 1993).

### 4 Conclusions and Recommendations

The (PB) of pastures have a direct influence on the production of milk, protein content in milk and MUN; depending on the (PB) in pastures the diet of the bovines must be completed with a specific formulation that would that potentiate the ruminal metabolism and the synthesis of final products such as milk production and nutrient content. This can be evidenced when pastures had a content of (PB) >18%, using formulations with 12% (Pb) and (ENN) above 60%, which increased the milk production and the protein content, and decreased the levels of MUN. When the content of (PB) in the pasture is between 14-18%, the (PB) in the formulation should be ≤ 14% for milk production and protein content to increase and decrease the MUN level. When the content of (PB) <14% in the pasture, it is deficient for milk production so a formulation with (PB) >16% should be used, to improve the production, the protein content and MUN levels.

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Improvement of the efficiency of the grass protein in dairy cattle using four different feed formulations


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