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Determination of Beta-Hydroxibutirate and Blood Glucose in Transition Cows to Prevent Clinical Ketosis in Tecamachalco, Puebla´S State

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Abstract

The term metabolic profile turns out to be an auxiliary method in the diagnosis of production disorders or diseases according to Contreras & Van Saun [1]; In this analysis, the metabolic profiles can contribute to the study of the nutritional balance of cattle, being nutritional disorders, which influence some metabolites in the blood. The metabolic profile is not considered a nutritional test, because metabolites are not indicators of nutritional status in animals, rather, they are indicating alterations in homeostasis, being then indicators of metabolic balance, as mentioned. When such homeostasis is broken, there is undoubtedly a decrease in productive performance [2] and depending on the degree of imbalance, the development of production diseases. The interpretation of blood components can therefore be useful for diagnosing imbalances derived from the animal's inability to maintain homeostasis. Considering the above, it is important to have representative diagnostic methods, which allow maintaining control of the animals through simple and low-cost exams, mainly during the transition period, considered this as the metabolically critical period, since it is the presence of so-called metabolic diseases is common, so that the nutritional, metabolic and general health status of the group of cows can be evaluated.

Recent studies mention prevalence of clinical ketosis of the order of 5 to 15% in herds in confinement, and in the case of the prevalence of subclinical ketosis in cows during early lactation, between 40 and 60% of the animals are affected, therefore the diagnosis of the subclinical form of the disease is extremely important [3]. The objective of this work is to compare and associate the presentation of subclinical ketosis and the negative energy balance (BEN), diagnosed by Beta hydroxybutyrate (BHB) plasma concentrations, blood glucose and the assessment of body condition, given the possibility of preventing the clinical manifestations of metabolic diseases such as ketosis or acetonemia, as well as the prevalence of the disease, in cows that begin lactation.

Keywords: Clinical Ketosis; Blood Glucose; Beta hydroxybutyrate (BHB) plasma

Introduction

It is currently considered that to obtain adequate profitability in intensive farms of specialized livestock in milk production, it is necessary to obtain adequate lactations and one calf per cow per year. This type of increasingly intensive farms and the selection of increasingly productive individuals has led to an increase in the called metabolic diseases, especially in the group of freshly born cows, which are in the transition period. In such a way that the demands caused by a higher productive demand, would favor the establishment of an imbalance between the entries of food into the body, the ability to metabolize these nutritional components and the levels of production achieved [4]. The specialized cows in milk production (Bos-Taurus), present from the preparatory period until the eighth postpartum week, the called negative energy balance (NEB) mainly due to the decrease in the consumption of dry matter in the delivery and the increase in energy demand for milk production in the immediate postpartum period, which leads to a lipid mobilization to meet its energy requirements [5].

Lipid mobilization increases the plasma concentration of non-esterified fatty acids (NEFA), which are transported to the liver for esterification or production of triacylglycerols. However, there is an important entry of NEFA into the hepatocyte mitochondria, producing ketone bodies, B-hydroxybutyrate (BHB), aceto-acetate (AcAc) and acetone (Ac), which are a source of energy in adult cattle. In turn, when its production exceeds the body's ability to use them, its concentrations increase producing ketosis [6,7]. Ketosis is a metabolic disease that mainly affects high-yielding dairy cows, between the 2nd. and 8th. postpartum week. It is characterized by a decrease in blood glucose and an increase in the concentration of ketone bodies in organic tissues and fluids [8]. Ketosis, in its clinical and subclinical forms, is one of the most frequent metabolic diseases in dairy herds, generating a negative impact due to the cost of treatment, reduction in milk production and higher incidence of diseases and reproductive problems [9].

In reference to subclinical ketosis, which is characterized by presenting plasma concentrations of βHB greater than 1.4 mmol / L, without evidence of clinical signs, it is commonly inadvertent in animals [10]. Considering that plasma concentrations ≥1.2 mmol / L achieve increases of 4.7 more in the presentation of clinical ketosis as mentioned [11], likewise, it mentions that concentrations ≥1.0 mmol / L during the first postpartum week (transition period), reductions of 25% in the pregnancy rate are observed during the first service [12]. On the other hand, plasma βHB concentrations at levels of 0.6 mmol / L are indicative of the negative energy balance in animals [13]. The statement by Carrier J [10]; in relation to considering that plasma BHB levels are considered a reliable test for the diagnosis of subclinical ketosis; remembering that there are different field tests for diagnosis such as the Rothera test, which attempts to identify the so-called ketone bodies by reacting Ac and AcAc with sodium nitroprusside. Comparing the rother test in milk samples, high specificity (98%) and low sensitivity (44%) are mentioned, following the same plasma BHB cut pattern ≥ 1.4 mmol / L as described [14]; however, in urine samples with the same cut-off point, this test now has a specificity of 96%, but an increase in sensitivity by 76% [10].

In relation to the mobilization of lipids, from the point of view of [15,16], if we monitor the plasma concentration of NEFA, together with the energy balance, this would be sufficient for its diagnosis, as well as to predict the risk of subclinical ketosis [7], It is known that NEFA concentrations greater than 300μmol / L, increase the presentation of ketosis by 3.6% according to data from [7]. Another important fact in the determination of glycemia is to continue monitoring the BEN, however, its sensitivity is low by presenting a strong hormonal homeostatic control, which maintains its concentrations without apparent changes [5]. In relation to the components of milk, both fat and proteins, they are considered important indicators in energy metabolism [9]. The optimal proportion of milk in terms of fat and protein varies from 1.0 to 1.25; and according to [3]; Cows with milk values G: P greater than 1.5 at the beginning of lactation, have a higher risk of ketosis.

Materials and Methods

During the months of March 2018 to February 2019, in the "El Salado" zootechnical post, belonging to the Faculty of Veterinary Medicine and Zootechnics of the "Benemérita Universidad Autónoma de Puebla", in the milk module, with Holstein-Friesian cows, 45 cows were sampled freshly born, that is, during the transition period, average 30 ± 5 postpartum days. Body condition was evaluated on a scale of 1 to 5. blood (0.5ml of the middle coccygeal artery) and urine samples were obtained from the group of cows, for analysis in relation to the presentation of BHB, using the Free Style equipment, test strips for ketone bodies, and test strips for blood glucose. The Rothera test was applied in urine samples to determine the presence of ketone bodies (acetone, acetoacetate and B-hydroxybutyrate). Considering cut-off points for ketosis> 1.4 mmol /L. The reaction intensity of the Rothera test was related to plasma BHB concentrations. The Rothera test was associated with the different cut-off points of ketosis, as well as glucose. The advantages of using the portable BHB meter for herd-based monitoring, rather than sending blood samples to the laboratory, are obvious. The cost of the test strips is less than the cost of laboratory tests, the results are known immediately, very small amounts of blood are required and no need to process and send serum or plasma samples to laboratories. The BHB blood field test also serves to sample individual cows for ketosis detection.

The information provided by the test then serves to make decisions about the individual treatment of each cow. Other field tests (that of BHB in milk and that of acetoacetate in urine) have been used for these same purposes. The test with the portable meter has much better sensitivity and specificity combined, than the other field tests. However, testing with the portable meter is more expensive, you need to have the ability to take the blood sample and training to handle the meter. The choice of the best protocol for the individual test of Ketosis in cows depends on the unique circumstances of each dairy. The blood glucose levels found with the portable meter with that reported by the laboratory have a correlation of 0.56 [17]. The portable meter with the glucose test strip had a sensitivity of 55% and a specificity of 99% to diagnose hyperglycemia (blood glucose> 79 mg / dL). It is suggested that this field test for glucose be used only in individual cows for treatment decisions but not as a herd-based monitoring system.

The determination of glucose is not useful at the level of herd monitoring because it is a highly regulated metabolite that is unlikely to change in response to the general management or nutrition of the herd. Problems at the herd level in terms of energy nutrition are diagnosed by monitoring BHB levels in fresh cows, milk production and body condition score in the rest of breastfeeding; and the Non-Esterified Fatty Acids (NEFA) blood in cows near delivery and freshly born. There are many situations that involve sick fresh cows in which the status of their glucose is uncertain and some information about the status of their glucose could serve to make some treatment decisions. It is particularly worrying that cows are treated with glucose when they already have hyperglycemia.

Situations in which a glucose test could be useful include cows with chronic ketosis (particularly after several treatments for ketosis) and sick cows that have less than about 4 days of calving (these cows are hyperglycemic because they have just given birth) and incompetent cows for no apparent reason. In these situations, knowing the status of your blood glucose could be valuable before treatment. Treating cows with intravenous glucose (or over-medicating with oral glucose precursors) can have negative consequences and should not be practiced in cows that are already

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hyperglycemic. For the determination of the statistical significance of the different cows of this group, the analysis of Variance will be applied to determine the correlation between the results obtained and the cut-off points.

Results

After the work is done, it is concluded that the mild and moderate reactions of the Rothera test in urine samples are indicators of BEN, while the intense reactions correspond to subclinical ketosis. In the case of BHB in blood, concentrations greater than 1.4 mmol / L indicate the presence of subclinical ketosis. The prevalence of ketosis according to the different types and the degree of disease involvement are presented in Table 1; Of the 45 animals evaluated at 30 ± 5 days after delivery, 21 had a BHB blood concentration lower than 1.4 mmol/ L (average 0.60), 19 from 1.4 to 1.9 mmol/ L BHB (average 1.52) and 5 more than 1.9 mmol.L BHB (average 2.38), which indicates that the prevalence of ketosis in its subclinical form in the herd evaluated was 42.2% and in the clinical form 11.1%; Similar data found in the Holstein breed, according to Oetzel [17]., when evaluating statistics on the incidence of ketosis in multiple scientific publications, reports that the average prevalence of this disease in its clinical form is around 15%.

Table 1: The prevalence of ketosis according to the different types and the degree of disease involvement.

Date	No. Cow	Birth and Number Date	BHB	Glucose	Rothera	C.C.
			Results	Result	Test	
26/05/2018	159	29/04/18, 6to	1,6 mmol/L	47mg/dL	++	2.5
26/05/2018	29	11/05/18, 2do	1.4 mmol/L	46mg/dL	+	3
26/05/2018	58	11/05/18, 1er	2,0 mmol/L	31mg/dL	++	2.5
26/05/2015	15	24/05/18, 3er	1,0 mmol/L	35mg/dL	+	2.5
2/6/2018	2	14/04/18, 4to	0,7mmol/L	62mg/dL	-	3.5
2/6/2018	8	29/05/18, 1er	0,6 mmol/L	40mg/dL	-	3
3/7/2018	8	14/06/18, 4to	0.9 mmol/L	55mg/dL	-	3
14/07/2018	685	06/06/18, 2do	1.4 mmol/L	44mg/dL	++	2
14/07/2018	29	10/06/18, 2do	0.4 mmol/L	60mg/dL	-	3
14/07/2018	780	25/06/18, 2do	0.6 mmol/L	42mg/dL	+	2.5
22/07/2018	749	24/05/18, 2do	1.4 mmol/L	54mg/dL	-	3
22/07/2018	28	25/06/18, 3er	2.5 mmol/L	44mg/dL	++	3
22/07/2018	400	23/05/18, 2do	0.6 mmol/L	54mg/dL	-	3.5
6/8/2018	54	08/06/18, 5to	1.4 mmol/L	71mg/dL	+	3
6/8/2018	572	27/05/18, 2do	0.6 mmol/L	50mg/dL	-	3.5
6/8/2018	292	23/05/18, 4to	1.5 mmol/L	47mg/dL	+	2.5
10/9/2018	727	29/05/18, 2do	0.6 mmol/L	44mg/dL	-	2.5
10/9/2018	864	22/07/18, 4to	1.6 mmol/L	57mg/dL	++	2
10/9/2018	774	22/04/18, 3er	0.2 mmol/L	80mg/dL	-	3
24/09/2018	913	19/07/18, 3er	1.7 mmol/L	27mg/dL	++	2
24/09/2018	133	21/07/18, 3er	0.7 mmol/L	61mg/dL	-	3
24/09/2018	739	21/07/18, 2do	1.0 mmol/L	40mg/dL	+	2.5
24/09/2018	801	06/07/18, 2do	1.5 mmol/L	230mg/dL	+	2
15/10/2018	11	29/06/18, 1er	2.6 mmol/L	40mg/dL	++	3.5
15/10/2018	14	29/06/18, 3er	1.4 mmol/L	34mg/dL	+	3
15/10/2018	16	05/06/18, 1er	0.8 mmol/L	49mg/dL	-	3
15/10/2018	62	04/08/18, 1er.	1.4 mmol/L	59mg/dL	+	3.5
22/10/2015	61	03/08/18, 1er.	0.6 mmol/L	58mg/dL	-	3
22/10/2018	59	04/07/18, 1er.	0.4 mmol/L	63mg/dL	-	3.5
22/10/2018	26	29/06/18, 2do.	0.4 mmol/L	72mg/dL	-	3
22/10/2018	167	22/09/18, 5to	1.9 mmol/L	39mg/dL	+	2.5
12/11/2018	9	08/09/18, 3er.	0.9 mmol/L	67mg/dL	-	3
12/11/2018	49	29/09/18, 2do.	0.4 mmol/L	55mg/dL	-	3.5

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12/11/2018	19	03/10/18, 3er.	1.5 mmol/L	41mg/dL	+	3
12/11/2018	24	02/10/18, 3er.	1.4 mmol/L	51mg/dL	-	3
10/12/2015	52	04/10/18, 2do.	2.4 mmol/L	31mg/dL	++	2
10/12/2018	90	23/10/18, 9no.	1.5 mmol/L	58mg/dL	-	3
21/01/2019	17	14/11/18, 4to.	1.7 mmol/L	41mg/dL	+	3
21/01/2019	48	06/01/19, 2do.	0.5 mmol/L	51mg/dL	-	3.5
21/01/2019	51	08/01/19, 2do.	2.4 mmol/L	33mg/dL	++	4
26/01/2019	172	20/02/19, 6to.	1.6 mmol/L	54mg/dL	+	3
29/01/2019	66	27/02/19, 1er.	0.3 mmol/L	61mg/dL	-	3
5/2/2019	198	09/03/19, 5to.	1.4 mmol/L	51mg/dL	-	3.5
5/2/2019	71	30/03/19, 1er.	0.4 mmol/L	67mg/dL	-	3
5/2/2019	99	30/03/19, 2do.	1.7 mmol/L	53mg/dL	+	2.5

The same author suggests using the 10% prevalence value of clinical ketosis as the value from which a comprehensive review of the management and feeding practices of the dairy herd should be made to make the corresponding corrections. In relation to the incidence of subclinical ketosis, Duffield [3] warns that the prevalence of this disease in stagnant herds is often greater than 40%, which increases the probability that animals suffer from different diseases and have a lower productive and reproductive performance than expected. The average concentrations of BHB during the period of 30 ± 5 days postpartum were 0.66 and 0.91 mmol/L, respectively. The values differ significantly ($p < 0.01$) from each other, suggesting that the animals in this herd are more affected by chronic hypoglycemia, because of an unmet demand for glucose precursors, rather than a high mobilization of body lipids due to a negative energy balance typical of the onset of breastfeeding (subclinical ketosis). The higher concentration of BHB in blood in the sampling performed at 30 ± 5 days postpartum indicates that animals mobilize more adipose tissue in this period to try to achieve an energy balance [18]. Only 46.6% of the animals had lower levels of BHB in the blood, as well as average blood glucose values.

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This situation could be due to a low mobilization of subcutaneous tissue during this period, caused by the effect of the breed and a moderate milk production at the beginning of lactation. French [20] indicates that animals of the Holstein breed decrease in a greater proportion the consumption of dry matter during the peripartum (35%) compared to cows of the Jersey breed (17%). The above suggests that Holstein cows may depend more on body reserves during the period of higher risk of ketosis than other breeds. In addition, Prendiville and Mackle indicate that Jersey cows have a greater capacity to consume dry matter per unit of live weight than those of the Holstein breed, which could explain a different propensity of breeds to the prevalence of this metabolic disease.

Discussion and Conclusion

Hormonal stimuli generate changes in the animal's tissue response, causing greater mobilization of NEFAs and reserves, from adipose tissue. Excessive transport of reserves raises blood ketone body concentrations (mostly BHB), causing ketosis. Some predisposing factors in the development of ketosis are race, dry matter consumption, body condition, hormonal concentrations, difficulty in childbirth, presence of other diseases and high milk production. There are several diagnostic tests for the detection of subclinical ketosis. Blood is the most recommended matrix for the sensitive and specific diagnosis of this disease, the main standard being blood detection of BHB. The presence and onset of subclinical ketosis may increase the likelihood of other diseases such as abomasum displacement and metritis, as well as decrease the productive and reproductive performance of the herd. There are nutritional and management practices to prevent the incidence of ketosis and if the disease is present, apply the most appropriate treatment to reverse the problem. Ketosis, although it does not show high heritability ($h2 = 0.09$), is related to the genetic selection of cows that reach a rapid increase in production and milk fat content at the beginning of lactation [21].

Several factors predispose to the presentation of ketosis, among them the high C.C. at birth and primary diseases that induce secondary ketosis, such as hypocalcemia, subacute ruminal acidosis, metritis, endometritis, placental retention, mastitis, abomasum displacement (especially on the left), peritonitis,

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laminitis, pyelonephritis, and muscle injuries associated with childbirth [19,22]. Cobalt deficiency is also considered a potential cause for ketosis presentation, since cobalt is required for the synthesis of B12 vitamin that is necessary for the conversion of propionate to glucose [18]. The use of bST (Synthetic Bovine Somatotropin) has been suggested as a factor associated with disease prevention; however, its effectiveness has not been proven [18]. Genetic and nutritional improvements have achieved an increase in milk production of 2% - 3% per cow and year. However, despite these great achievements, reproductive performance rates have worsened. In addition, it has been suggested that this increase in production implies a greater demand for the cow, which leads to an increase in the incidence of certain metabolic disorders and a higher rate of involuntary selective sacrifices. Epidemiological studies have also revealed an inverse relationship between milk production and reproduction in cattle for milk production [23,24].

Nevertheless, Lucy suggests that the effects on reproduction of an increase in milk production are relatively minor compared to the effects of other factors. For example, it has been described that diseases (eg retention of placenta, metritis and ovarian cysts) and the month of delivery (eg March to May) have a negative effect on the relative risk of conception, while It has been observed that the cumulative milk production of the first 60 days has no effect on the rate of conception. The reasons for poor reproductive performance in dairy cows are multiple. Anoestrus and nutrition of transitional cows have been identified as the most important variables in reproductive performance. The nutritional imbalance during the dry and early postpartum period results in a reduction in the concentration of IGF1 and a low frequency of LH (luteinizing hormone) pulses, followed by a delay in the resumption of the ovarian cycle. The concentrations of BHBA, NEFA and triacylglycerol increase, while cIGF1 decreases.

The literature mentions clinical ketosis prevalence from the order of 2 to 15%, in herds of countries, in which animals are managed in confinement. The prevalence of subclinical ketosis, in early lactation in these herds, affects between 40 and 60% of animals. Due to its high prevalence, the diagnosis of the subclinical form of the disease is considered very important [3].

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