INCIDENCE AND RISK FACTORS OF MILK FEVER AMONG CROSS-BRED DAIRY COWS IN KHARTOUM STATE, SUDAN

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Abstract:

The study was conducted on 206 cross-bred dairy cows in different dairy herds in Khartoum State, Sudan, during the period from March 2003 to June 2004 to determine the prevalence and incidence rate of milk fever (MF) based on clinical and laboratory diagnosis, and to recognize the risk factors associated with the disease in Khartoum State. The incidence rate was 5.3% among the examined dairy cows. Using Chi-square cross-tabulation statistics, herd and parity did not represent risk factors for the occurrence of MF. The incidence ranged between 2.2 and 8.0% among the examined herds. The incidence was 2.5%, 4.0% and 9.1% among dairy cows of 1-2, 3-4, and > 5 parities respectively. Milk yield, age and previous history represented high risk factors for the occurrence of MF in this study. Among age groups of 3-4, 5-6 and > 5 years the incidence rates were 3.3%, 2.0% and 10.5% respectively. Cows eler than 6 years constituted 72.7% of the positive cases. Among the milk yield groups, the incidence was significantly higher (19.5%) among dairy cows producing 12-25 liters per day, whereas no milk fever case was reported in dairy cows producing 10 liters or less per day. The disease was studied based on clinical signs and laboratory examinations of serum calcium, phosphorus and magnesium. In all positive cases, body temperature, heart rate and rumen motility were examined. Rectal temperature and rumen motility were significantly low (37.0± 0.5°C and 0.25± 0.2 per minute respectively). Heart rate was significantly increased (103.5± 6.9 beat per minute). Blood calcium and Phosphorus levels were significantly lower (4.6± 0.5 mg/dl and 2.3±0.4 mg/dl respectively) than the levels measured after treatment and recovery, but magnesium levels remained unaffected before and after treatment. The increased blood calcium levels were followed by immediate recovery of 81.8% of the diseased cows. The recovery delayed in 9.1% of cases. 9.1% of the affected cows died. This study is the first, of its kind, was based on laboratory diagnosis in Sudan with featured results indicating that the parity did not affect the incidence of the disease as the disease occurred in one primi-parous cow (9.1%), and magnesium has no role on the pathogenesis of the disease as the levels remained unchanged before and after treatment.

INCIDENCE ET FACTEURS DE RISQUE DE FIÈVRE VITOLAIRE CHEZ DES VACHES LAITIÈRES CROISÉES DANS L'ÉTAT DE KHARTOUM AU SOUDAN

Résumé

Cette étude a porté sur 206 vaches laitières croisées choisies dans différents troupeaux laitiers de l'État de Khartoum au Soudan, au cours de la période de mars 2003 à juin 2004. L'objectif de l'étude était de déterminer les taux de prévalence et d'incidence de la fièvre vitolaire (MF), sur la base du diagnostic clinique et de laboratoire, et de déterminer les facteurs de risque associés à la maladie dans l'État de Khartoum. Le taux d'incidence était de 5.3% chez les vaches laitières examinées. Les statistiques des tableaux croisés du Chi-carré utilisées ont montré que le troupeau et la parité ne représentaient pas des facteurs de risque d'apparition de la MF. Le taux d'incidence variait entre 2.2 et 8.0% chez les troupeaux examinés. Il était de 2.5%, 4.0% et 9.1% respectivement chez les vaches laitières de 1-2, 3-4, et > 5 parités. Dans cette étude, le rendement laitier, l'âge et les antécédents représentaient des facteurs à haut risque d'apparition de la MF. Parmi les groupes d'âge de 3-4, 5-6 et > 5 ans, les taux d'incidence étaient respectivement de 3.3%, 2.0% et 10.5%. Les vaches âgées de plus de 6 ans constituaient 72.7% des cas positifs. Parmi les groupes de vaches laitières, le taux d'incidence était significativement plus élevé (19.5%) chez les vaches produisant 12 à 25 litres par jour; alors qu'aucun cas de fièvre vitolaire n'a été signalé parmi celles produisant 10 litres ou moins par jour. La maladie a été étudiée sur la base des signes cliniques et des examens de laboratoire des taux sériques de calcium, de phosphore et de magnésium. Dans tous les cas positifs, la température du corps

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rythme cardiaque et la modilité du rumen ont été examinés. La température rectale et la modilité du rumen étaient très basses (respectivement 37,0± 0,5°C et 0,25± 0,2 par minute). Le rythme cardiaque a augmenté de manière significative (103,5± 6,9 battements par minute). Les taux de calcium et de phosphore dans le sang étaient significativement plus faibles (4,6± 0,5 mg/dl et 2,3±0,4 mg/dl respectivement) que les taux notés après le traitement et le rétablissement, mais les taux de magnésium étaient les mêmes avant et après le traitement. L'augmentation des taux de calcium dans le sang a été suivie par le rétablissement immédiat de 81,8% des vaches malades. Le rétablissement a tardé dans 9,1% des cas ; et 9,1% des vaches atteintes sont mortes. Cette étude inédite, menée sur la base du diagnostic de laboratoire au Soudan a noté que la parité n'avait pas affecté l'incidence de la maladie - car celle-ci est apparue chez une vache primipare (9,1%) - et que le magnésium n'avait pas de rôle dans la pathogenèse de la maladie puisque sa teneur est restée la même avant et après le traitement.

Introduction

Metabolic diseases (Production Diseases) are important group of diseases affecting mainly multi-parous dairy cows. The concentration on this group of diseases came from the improvement and advances in breeding programmes which focused on the genetic improvement of the production trait such as milk yield of the selected dairy cows. The increase in milk yield has been accompanied by increasing metabolic problems as milk is produced at the expense of many metabolites.

Milk fever (parturient paresis, parturient or peri-parturient hypocalcaemia,) is one of the most important metabolic disorders affecting mainly the lactating dairy cows and less frequently the pregnant cows, ewes and goats worldwide. The disease is characterized by muscular weakness and unconsciousness. Most cases of the disease occur during the first two days of calving because milk and clostrum production drain calcium from the blood and some cows are unable to compensate it fast enough, so that the depression of the levels of ionized calcium in tissue fluids is the main biochemical defect in the disease. Rare cases may occur as paresis in non-pregnant cows after oestrus (Hess, 1969) or more than 48 hours post-calving. The incidence of the clinical form of the disease varies greatly within countries and within areas in the same country, but it generally ranges from 5-10%, and may exceed 25%. The prevalence of the subclinical form of the disease is too difficult to be estimated. Age, milk yield, stage of lactation, previous history of the disease, parity, and breed were reported as risk factors associated with the disease. Older milking cows are more susceptible to the disease and age was reported to increase the risk of MF by approximately 9% per lactation. Heifers are rarely affected with milk fever. Among breeds, Jersey breed were reported as susceptible to the disease more than Friesian dairy cows.

The economical importance of the disease arises from its effect on milk yield and reproductive performance of the dairy cows. The occurrence of the disease around calving is a risk factor for many diseases like mastitis, metritis, lameness and abomasal displacement, the diseases which were encountered as major causes of disposal among dairy cows. The disease was reported as one of the causes of high culling rate among dairy cows in United State.

The clinical signs of the disease are highly specific and of great importance in diagnosing the disease. The objective of this study was to report the incidence of MF among cross-bred dairy cows in Khartoum State, as such study is lacking in the Sudan, and to correlate the reported risk factors with the incidence of the disease.

Material and Methods

Animals and Management:

A number of 206 per-parturient cross-bred (Friesian Zebu cattle) dairy cows in five different herds in Khartoum State were assigned for this study. The five herds (H-1, H-2, H-3, H-4 and H-5) were represented by 45, 46, 50, 40 and 25 cows respectively. H-1 was a governmental herd of the farm of University of Khartoum in Shambat, Khartoum North and the other four herds (H-1 to H-4) were in Khartoum North but privately owned. The animal's houses of H-1
to H-4 were constructed from iron poles and corrugated iron sheets with provided sheds. In H-5 the ranch was constructed from local materials with un-adequate shade.

Assessment of Risk Factors and Clinical Examination:
All dairy cows in this study were followed till the date of parturition and then for three weeks post-calving. Information for each cow was reported from the farm record and by direct questions to the animal's owners or attendants. The age, number of parity, milk yield and previous history of milk fever were reported. Cows which developed the clinical signs of milk fever were examined and data were reported. The period of the onset of the clinical signs related to the time of the parturition, response to treatment (immediate response, failure to respond and relapses) were reported. Clinical examination was carried out before the onset of treatment and 24 hours post-treatment and then weekly for three weeks. Rectal body temperature ruminal motility and heart rate were monitored.

Sampling:
Blood Samples were collected from all cows which developed the clinical signs of the disease pre-treatment, 24 hours post-treatment and then weekly for three weeks. Sera were separated and used for laboratory investigation. Blood serum levels of calcium (Ca), phosphorus (P) and magnesium (Mg) were determined using the colorimetric method and biochemical kits depending on the absorbance of the sample and standard against the blank.

Treatment:
All MF cases were treated using a solution containing per ml: 175.00 mg calcium gluconate, 100 mg glucose and 00.35 mg magnesium hypophosphite.

Statistical Analysis:
Data were presented as mean + standard deviation of the mean. The data were statistically analyzed using the descriptive Paired T-test to compare means before and after treatment. Data for correlation were analyzed using Chi-Square crosstabs descriptive statistics.

Results:
Incidence of Milk Fever: During the period of investigation, eleven cows out of 206 developed the conventional and non-conventional signs of MF. The diagnosis was confirmed by determination of pre-treatment blood calcium levels.

<table>
<thead>
<tr>
<th>Table 1: Percentage of milk fever in dairy cows in five dairy Herds in Khartoum State, 2003-2004.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herd</td>
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<tr>
<td>--------</td>
</tr>
<tr>
<td>H-1</td>
</tr>
<tr>
<td>H-2</td>
</tr>
<tr>
<td>H-3</td>
</tr>
<tr>
<td>H-4</td>
</tr>
<tr>
<td>H-5</td>
</tr>
<tr>
<td>Total</td>
</tr>
</tbody>
</table>

Chi-Square Value = 2.681; P = 0.6

<table>
<thead>
<tr>
<th>Table 2: Percentage of Milk fever in cows of different ages in Khartoum State, 2003-2004.</th>
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</thead>
<tbody>
<tr>
<td>Age/ year</td>
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<tr>
<td>-----------</td>
</tr>
<tr>
<td>3-4</td>
</tr>
<tr>
<td>5-6</td>
</tr>
<tr>
<td>&gt;6</td>
</tr>
<tr>
<td>Total</td>
</tr>
</tbody>
</table>

Chi-Square value = 10.322; P = 0.006
The incidence rate among the examined cows in the five herds during the period of the investigation (2003-2004) was 5.3% with no significance differences among different herds (Table-1).

Risk factors:

Herd: The incidence of MF among the examined different 5 herds in Khartoum State ranged between 2.2 and 8% with no significant different (P=0.6).

Age:

As shown in table-2, age has a significant effect (P=0.006) on the occurrence of milk fever. According to age groups, most cases of MF (72.7%) occurred in dairy cows more than 6 years old. One case (9.1%) occurred in age group of 3-4 years.

Parity:

The result of the effect of parity on the incidence of MF was shown in table-3. Most cases (54.5%) of the disease occurred in multiparous dairy cows of more than 5 parities. Four MF cases occurred in cows with 3-4 parities. One case (9.1%) occurred in uniparous cow. Statistics revealed that the Number of parity has no effect (P=0.243) on the incidence of MF reported in this study.

Milk Yield:

The effect of milk yield on the incidence of MF was reported in table-3. In this study, milk fever had not been reported in dairy cow yielding 10 liters of milk or less per day. Most cases of the disease occurred in milk yield group producing 21-25 liters per day. Milk yield has a significant effect (P<0.0001) on the incidence of MF and assumed as a great risk factor for the occurrence of the disease in this study.

Previous history of milk fever:

The previous history of milk fever as a risk factor for the disease was reported. Seven MF cases (63.6%) had previous history of the disease. The incidence of the disease is significantly higher in dairy cows with previous history of the disease (Chi-Square Value = 47.548a, P< 0.0001).

Clinical parameters:

Ten positive milk fever cases occurred during the first 24 hours post parturition. One case (9.1%) occurred 48 hours post-calving. The conventional signs of MF were not observed in pregnant cows in this study. The cows which developed the clinical signs of the disease were examined clinically before and after treatment. All cows were in fair body condition except one cow which was in poor body condition. Body temperature, heart rate and rumen motility were as shown in table-4. Rectal body temperature was significantly low (37.0±0.5°C) before treatment and 24 hours post-treatment and significantly increased at 8 hours post-treatment.
days P.T. and remained unchanged and within the normal range till 21 days. The heart rate was significantly higher before and even 24 hours P.T. The rate decreased significantly at 8 days P.T. and remained unchanged and within the normal limit till 21 days. Rumen motility was significantly reduced before treatment and did not improve till 15 days P.T. Hyperemia of the udders and involuntary letdown of bloody milk were observed in five MF cases and the edema of the udder was observed in one case. Partial uterine prolapsed was diagnosed in one MF case (2.2%).

**Table 5:** Clinical parameters of Positive Milk Fever Cases before and after Treatment in Khartoum State, 2003-2004.

<table>
<thead>
<tr>
<th>Clinical Parameter</th>
<th>A.T.</th>
<th>24h P.T.</th>
<th>8 days P.T.</th>
<th>15d P.T.</th>
<th>21d P.T.</th>
</tr>
</thead>
<tbody>
<tr>
<td>R.T. (°C)</td>
<td>37.0±0.5*</td>
<td>37.4±0.5*</td>
<td>38.6±0.5**</td>
<td>38.5±0.4*</td>
<td>38.9±0.6*</td>
</tr>
<tr>
<td>H.R (Beat/Min)</td>
<td>103.5±6.9**</td>
<td>91.0±14.9**</td>
<td>79.2±5.3**</td>
<td>80.9±7.4*</td>
<td>84.0±6.4*</td>
</tr>
<tr>
<td>R.M. (per min.)</td>
<td>0.25±0.2*</td>
<td>0.26±0.2*</td>
<td>0.8±0.2**</td>
<td>1.7±0.4**</td>
<td>2.4±0.5**</td>
</tr>
</tbody>
</table>

R.T. Rectal temperature, H.R. = Heart Rate, R. M. Rumen Motility, A.T. Before Treatment, P.T. Post-Treatment

* = Significant, P < 0.05,
* = Not significant, P > 0.05.

**Table 6:** Calcium, phosphorus and magnesium levels of MF cases before and after treatment in cross-bred dairy cows in Khartoum State, Sudan (2003-2004).

<table>
<thead>
<tr>
<th>Biochemical parameter</th>
<th>A.T.</th>
<th>24h P.T.</th>
<th>8 days P.T.</th>
<th>15d P.T.</th>
<th>21d P.T.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium (mg/dl)</td>
<td>04.6±0.5**</td>
<td>08.2±1.5**</td>
<td>07.4±0.5**</td>
<td>7.3±0.3*</td>
<td>07.3±0.4*</td>
</tr>
<tr>
<td>Phosphorus (mg/dl)</td>
<td>2.3±0.4**</td>
<td>2.9±0.7**</td>
<td>4.1±5**</td>
<td>4.0±0.3*</td>
<td>4.5±0.5*</td>
</tr>
<tr>
<td>Magnesium (mg/dl)</td>
<td>2.6±0.4*</td>
<td>2.7±0.5*</td>
<td>3.4±0.6*</td>
<td>3.0±0.6*</td>
<td>3.0±0.4*</td>
</tr>
</tbody>
</table>

* = Significant, P < 0.05,
* = Not significant, P > 0.05.

**Discussion**

Milk fever is a life threatening disease of dairy cows and predisposes them to other postpartum problems14. The study reported that the incidence of milk fever was 5.3% in a total number of 206 cross-bred dairy cows in Khartoum State, Sudan, during the period from January 2003 to June 2004. This study is the first one of its kind to report the incidence of MF based on clinical and laboratory findings. However a report based only on clinical observation reported an incidence of 3.5% among cross-bred dairy cows15.
Ten MF cases out of the 11 MF cases (90.9%) reported in this study occurred within the first 24 hours post-partum, one case (9.1%) occurred 48 hour post-partum. No MF case occurred before calving or more than 48 hours post-calving.

The incidence of MF was reported to be different within countries and within farms in the same areas. But generally the overall incidence of the disease is estimated at 0-10% in USA1,6, although higher incidence was reported10. In Australia, incidence ranges of 0-30% among different herds with an overall incidence of 3.0% were reported8. In the Sudan, prevention of MF is not a top priority as the disease is not highly prevalent. As proposed, specific control measurement is relevant when the incidence of the disease increases to above 10% among high risk cows17. Because there is a genetic predisposition of cows to milk fever18,19 and this is well defined to certain breeds like Jersey breed, in Sudan, the most dairy cows are cross-bred between Friesian and local breeds, this may decrease the risk for the disease.

Severe hypocalcaemia as a main cause of MF could be attributed due to several risk factors20,21. The risk factors recognized in this study included age, milk yield, parity and previous history of the disease. Variations in incidence of MF among different dairy herds were reported in many studies22,23. In this study the incidence of MF among 5 dairy herds was not significantly different (P=0.6). The same result was reported in Danish dairy herds 22. This may be due to the same management measures and practices adopted in different dairy farms in Sudan.

Milk yield, parity and breed were reported as important risk factors which contribute to the occurrence of MF4,9,10,22. Previous history and the age were also reported among the risk factors. Although the age of cows was reported to increase the risk of MF by approximately 9% per lactation11, some authors assigned no age effect on the incidence of MF24. As a result of this study, age represented an important risk factor as 8 out of 11 MF cases (72.7%) occurred in cows more than 6 years old. Increased prevalence rate is highly associated with increased milk yield as 81.8% of MF cases were among the cows producing 21-25 liters of milk per day. The previous occurrence of the disease was highly associated with the higher incidence of MF (P<0.0001) as 7 cows (63.6%) had milk fever at least once in their production lives. No association between the incidence of MF and the number of parities was found in this study (P=0.24), since 2.5% of MF cases occurred in cows of 1-2 parity. This result is not compatible with many authors who reported parity as a risk factor for the occurrence of MF10,22. Although MF in this study was prevalent in older cows which assumed to have more parities, this can be justified by the inappropriate management practices adopted in dairy herds in Khartoum State which results in prolonged calving intervals and thus less parities even in older cows.

The rectal body temperature, heart rate and rumen motility were examined. The pre-treatment rectal body temperature was 37.0±0.5°C and did not change after 24 hours PT, but elevated to reach the normal range when measured 8 days P.T. and remained steady till 3 weeks later. The relationships between rectal temperature and rumen motility and blood calcium levels were well established25. The heart rate was significantly high before the commencement of treatment followed by significant decrease 24 hours P.T. to settle at normal range thereafter. Rumen motility was significantly slow in affected cows and did not return to normal level even after 8 days P.T. The reported Low rectal temperature, increased heart rate and decreased rumen motility are all due to low level of blood calcium. Response to treatment and recovery from the first dose is related to several clinical characteristics such as the duration of recumbency, body temperature26 and biochemical parameters24. In this study 81.8% of MF cases responded rapidly from a single treatment. This is due to the immediate onset of treatment and shorter recumbency period. One case (9.1%) responded after the second treatment and one case (9.1%) died after 3 hours of administration of the drug. High recovery rates were reported among cows with phosphorus level > or = 0.9 mmol/l (2.7 mg/dl) and calcium level > or = 1.7 mmol/l (6.0 mg/dl) 24.

Blood Ca and Pi levels were significantly lower in pre-treatment samples of MF cases.
Magnesium was not affected before and after treatment. Ca level elevated significantly P.T and then significantly decreased to reach 7.4 ± 0.5 mg/dl and remained steady till 3 weeks later. The normal levels of blood metabolites in cross-bred dairy cows in Sudan are still not fully reported, but the levels of Ca, P, and Mg levels reported in this study 3 weeks P.T. were in accordance with those previously reported levels of these metabolites in healthy pregnant and non-pregnant cross-bred dairy cows in different seasons of the year in Khartoum State, Sudan by 27. The authors reported Ca levels ranging from 6.9 ± 1.4 to 7.9 ± 2.1 mg/dl, P levels from 4.1 ± 0.7 to 4.6 ± 0.9 mg/dl, Mg levels from 1.7 ± 0.6 to 1.8 ± 0.6 mg/dl. The role of P, and Mg in the pathogenesis of MF is still not fully understood. Hypomagnesaemia and metabolic alkalosis due to high potassium levels (Higher blood pH) was reported in MF cases 28. Other authors reported hypomagnesaemia as a risk factor for the occurrence of the disease. In this study, Mg was not affected in all diseased cows and had no role in the incidence of the disease. This finding is compatible with some reports 25. Magnesium levels were within the levels reported in cross-bred dairy cows in Khartoum State in different stage of pregnancies 27. Hypophosphataemia was accompanied with MF in this study as serum phosphorus levels were significantly lower in affected cows. This result confirmed that the pathophysiology of milk fever is still not fully understood as different authors reported different findings regarding the levels of Mg and P in affected dairy cows 28, 23. The disagreements on the effect of blood levels of electrolytes on the pathogenesis and prevalence of MF supported the hypothesis suggested that pathophysiology of the disease MF is still not completely understood as still more arguments are reported, moreover, these disagreements regarding the role of Mg, Ca and K on the occurrence of MF supported the theory of dietary cation anion differences (DCAD) reported by many authors 22, 29, 30, 31. The levels of serum calcium which identify the clinical and subclinical hypocalcaemia are not easy to be clearly cut identified. In this study, Serum Ca levels in all cows with clinical MF were less than 5.0 mg/dl (mean = 4.6 ± 0.5 mg/dl), became elevated after treatment and then fell to a level of 7.4 ± 0.5 mg/dl which did not change till three weeks after treatment.

In the literature, blood calcium levels in the adult cows are maintained between 8.5 and 10.0 mg/dl and blood calcium levels of 5.5-8.0 are reported as subclinical hypocalcaemia, and levels of <8.0 mg/dl but not <5.0 mg/dl was encountered for acute hypocalcaemia. In this study, during the acute hypocalcaemia, calcium levels were less than 5.0 mg/dl but more than 4.0 mg/dl. This difference is due to BREED difference since the normal levels of Ca in cross-bred dairy cows is less than those reported in dairy cows of pure-bred highly producing dairy cows. The lower calcium levels in cross-bred dairy cows in Sudan may be due to low milk yield capacity and low metabolic rate. In all recently calved cows, serum calcium falls to as low as 8.0 mg/dl and for this reason approximately all recently calved cows experience a degree of hypocalcaemia 32. In this study, Ca levels were raised after injection of calcium salts and fell to reach steady levels which did not exceed 8.0 mg/dl. This level though identified as hypocalcaemia in some countries, but it is the normal level of calcium among healthy dairy cows in Sudan 27.

Acknowledgements

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parameters on incidence risk, diagnosis, risk factors and biological effects as input for a decision system for disease control. Acta Veterinaria Scandinavica, 42(1): 1-29.


