

Study on blood gases, Acid-Base balance and the therapeutic evaluation of different protocol of treatment of mastitic cows.

Abdulkarim Jafar Karim

College of Vet. Medicine- University of Baghdad

Summary

This study was planned to reveal the changes in blood gases and acid-base values in mastitic cows and to state the intravenous fluid therapy regimen treatment. A herd with a population of 150 lactating cows was screened for clinical and subclinical mastitis for a period of 6 months. Based on inclusion and exclusion criteria, twenty-eight cows with clinical mastitis were divided in three groups (mastitic not treated (group M), mastitic treated with antibiotic only (group TA) and mastitic treated with antibiotic and fluid therapy (group TAF) and another five healthy cows (group C) served as controls. Blood gases, SCC, CMT scores, udder health status and cure rate were carefully investigated. There was significant differences between PO_2 , PCO_2 , BE, and HCO_3^- concentration in mastitic cows. Increased pH, BE, and HCO_3^- referred to metabolic alkalosis whereas increased PCO_2 indicate compensatory respiratory acidosis. The results of SCC in milk agreed to CMT scores, and to some extent to udder health status among the entire period of study. Cure rate was earlier in group TAF with less number of treatment than in group TA. It has been concluded from this study that administration of fluid therapy might be useful as an adjunct in case of clinical mastitis in cows to get quick clinical recovery with less number of treatments. However, the role of blood gases and acid – base status in mastitis should be investigated in details.

دراسة تقييم لطرق مختلفة في علاج التهاب الضرع

عبد الكريم جعفر

كلية الطب البيطري/جامعة بغداد

الخلاصة

تم تصميم هذه الدراسة لدراسة التغيرات في غازات الدم والقيم الحمضية القاعدية في أبقار مصابة بالتهاب الضرع، ومقارنة نظم علاج التهاب الضرع بإضافة نظام العلاج بالسوائل الوريدية. تم توزيع ثمان وعشرون بقرة مصابة بالتهاب الضرع السريري في ثلاث مجموعات (المجموعة الأولى وتضم سبعة أبقار مصابة بالتهاب الضرع ولم تُعالج، المجموعة الثانية وتضم سبعة أبقار مصابة بالتهاب الضرع عولجت بالمضاد الحيوي فقط، أما المجموعة الثالثة فتضم أربعة عشر بقرة مصابة بالتهاب الضرع عولجت بالمضاد الحيوي و السوائل الوريدية). وعولمت خمس أبقار سليمة أخرى كحيوانات سيطرة في المجموعة

الرابعة. تمت دراسة نسبة غازات الدم ، عد الخلايا البيضاء في الحليب ، اختبار كاليفورنيا لإلتهاب الضرع وحالة الضرع الصحية ونسبة الشفاء بعناية. كان هناك اختلافات هامة بين تركيز أيون الأوكسجين (PO_2) ، تركيز أيون ثاني أوكسيد الكربون (PCO_2) ، BE ، تركيز أيون البيكاربونات (HCO_3^-) في الأبقار المصابة بالتهاب الضرع. إنَّ الزيادة في قيم درجة حامضية الدم ، BE ، و تركيز أيون البيكاربونات يشير الى قاعدية الدم الأيضية metabolic alkalosis بينما يشير زيادة PCO_2 الى الحمّاض التنفسي التعويضي. تطابقت نتائج عد خلايا الحليب مع نتائج فحص كاليفورنيا لإلتهاب الضرع، وإلى حدّ ما مع حالة الضرع الصحية طيلة فترة الدراسة. وأظهرت نسبة الشفاء تماثلاً أسرع في المجموعة الثالثة مع عدد أقل من المعالجات عن المجموعة الثانية. إستنتج من هذه الدراسة أن اعطاء السوائل الوريدية أثبتت فائدة كعلاج ملحق مع المضادات الحيوية في حالة إلتهاب الضرع السريري في الأبقار للحصول على التحسّن السريري السريع مع فترة علاج أقل .

Introduction

Mastitis continues to be a problem of serious concern for dairy industry as well as public health throughout the world. The annual incidence of this disease in Iraq is as high as 70.6% for subclinical mastitis (1). Subclinical mastitis causes 18% and clinical form causes 50% milk loss in dairy cows (2). Moreover, economic importance is revealed by milk dumped for antibiotics, veterinary costs, labor costs, poor conditioned suckling calves, culling and death costs, and lost milk premiums through i.) Increased somatic cell counts, ii.) Decreased fat, and iii.) Decreased protein. It seems to be the most prevalent (35 to 40 cases/year per 100 cows) and the most costly disease in dairy cattle in the USA with losses exceeds \$ 2 billion/year (3,4).

Multidisciplinary approaches have been undertaken in recent years to find out a suitable remedy. In addition to that intramammary infusion of antibiotics has been reported to suppress the various activities of infiltrated polymorphnuclear leukocytes (PMNL) in mastitis udder (5), none of the available antibiotics have been proven to have more than 60% efficacy in field conditions against major mastitis pathogens (2) and the use of fluid therapy was often excluded.

Consideration the changes in blood gases and acid base values, which seem the more important point in stating the fluid therapy as a line of treatment in conjunction with antibiotics, has been usually missed, with the exception of the treatment of *E.coli* mastitis, in which the fluid therapy focused to treat the accompanied toxemia (2). Therefore the aim of this study is to reveal data on acid-base balance in mastitic cows and to insure a prompt treatment.

Materials and Methods

Animals: 150 healthy cows from small herds, in Abu-Ghraib village, 30 kilometers west to Baghdad, from early May to the end of October 1989, fed on balance ration, were put under clinical supervision. Hand milking was performed twice daily and there was no milking machine. Washing of udder with tape water before each milking was performed regularly. Teat dipping was not practiced regularly. General (including udder) health status and data of SSC, CMT, PO₂, PCO₂, pH, HCO₃⁻ and BE were recorded to fix the normal (pre-infection) values of all cows.

Selection of study animals was based on inclusion (Age: 4 to 7 years, Lactation: 2nd to 5th, Stage of lactation: 15th day to 4th month, Milk yield: > 10 liters, Breed: Crossbred of Friesian with indigenous breed) and exclusion (First 15 days or later stage of lactation, History of prior infection in any quarter or any major health problem in present lactation, Presence of skin lesions on teat, Chronic or Peracute form of mastitis) criteria.

Out of the 150 cows examined, the prevalence of mastitis was (57.3%) considerably high in this study, and in 6 months period 86 cases of mastitis (subclinical=58, clinical=28) were diagnosed. From the 86 mastitis cows twenty eight cows each with clinical mastitis, and five cows separately with normal udder and health were selected as study animals

Chemicals:

Ampiclox (Ampicillin plus Cloxacillin) (Beecham Pharmaceuticals, Brentford, England).

Glucose saline (from MIV: Mosul IntraVenous Project, Mosul, Iraq).

Experimental design:

Table 1 shows protocol for therapeutic evaluation of antibiotic therapy plus fluid therapy in mastitic cows. Five healthy animals with normal udder and milk served as healthy control (group C). Twenty-eight cows with clinical mastitis were divided into group M (n=7, mastitic cows not treated), group TA (n=7, mastitic cows treated with antibiotic only), and group TAF (n=14, mastitic cows treated with antibiotics and fluid therapy).

All cows were subjected to daily routine clinical examination including udder health status (swelling, pain reflex measured by pressing the udder with the help of fingers) and milk yield was estimated. Arterial blood samples were taken at healthy status (pre-infection), during infection, and 14 days after infection to determine the partial pressure of oxygen (PO₂) and carbon dioxide (PCO₂), pH, bicarbonate (HCO₃⁻) concentration and base excess (BE) using a blood gas analyzer (Acid-base calculator: Inst. Lab. Italy), in the Department of

Surgery- Collage of Vet. Medicine, Baghdad.

Sampling: 2-3 ml of blood from common carotid artery was taken 3-5 cm foreword the thoracic inlet in the jugular groove. Heparinised syringe was used. Needle fitted to the syringe was injected in vertical position to the artery. Moving the depth of the needle inside tissues without pulling the pistol of the syringe, blood will soon fill the syringe by the cardiac pumping force without any bubble appear in the sample (anaerobically). Samples were handled in an ice water bath and checked in the laboratory within 30 minutes (6).

Screening for mastitis and evaluation of treatment:

Diagnosis and classification of mastitis were based on physical examination of the udder; physical and chemical changes in the milk, California mastitis test (CMT) score of affected quarter. Milk Somatic Cell Count (SCC) was carried on in a healthy status, during infection, 5 and 14 days post infection. Subclinical mastitis was diagnosed on the basis of CMT score and SCC. The CMT was carried out at cow side at the time of milking by mixing 3 ml of fresh milk with equal volume of CMT reagent. The milk and CMT reagent were mixed together by swirling the paddle or by stirring with a glass rod and the results were read after 10-15 seconds. Interpretation of CMT results was carried according to (2). Standard procedures (7) were followed to collect the milk for SCC and CMT. Data were statistically analyzed using analysis of variance (ANOVA) and student's t-test ($P < 0.05$) (8).

Results

Udder health status, Milk SCC, CMT reaction, treatment duration and cure rate are shown in Table 2. Animals of group TAF showed faster recovery and required less number of antibiotic treatments than the group TA. The cure rate was 100% on day 6 in group TAF, and 85.7% on day 14 in group TA. Duration of antibiotic treatment was longer in group TA (5 days) than in group TAF (3 - 4 days). The effect of fluid therapy was apparent, the milk from quarters of group TAF showed moderate (++) and (-) while quarters of group TA showed severe (+++) and (+) reaction to CMT on the 2nd and 6th day of treatment respectively. Records of udder health status showed severe swelling, pain reflex and physical changes in milk in all quarters of all groups at the first day post treatment. Swelling of the quarters remained severe in group TA on the day 2 and tend to be moderate, mild, and normal on days 3, 4 and 14 respectively. The normal size of the quarters in group TAF was clear on day 5. There was moderate pain reflex in quarters of group TA on day 2 and became mild and nil on days 3 and 6 respectively, while it was mild on days 2,3 and nil on day 4 in group TAF.

Physical changes in milk became moderate, mild to nil on days 2,3,6 in group TA and mild to nil on days 2, 5 in group TAF respectively. Normal milk yield was achieved on day 14 in TAF group, while there was 7% milk loss in group TA. Somatic cell counts (SCC) of milk increased significantly ($P<0.05$) during infection in all groups. There was gradual decrease in SCC on day 5 post-infection in groups TA and TAF, and the data was still showing a significant high values ($P<0.05$) from the pre- infective stage in groups TA and M two weeks post-infection, while normal level was observed in group TAF.

Results of the blood gases and acid-base values are shown in Table 3. There were significant increases ($P<0.05$) in PO_2 , PCO_2 , and HCO_3 -concentration in all cows during mastitis compared with the pre-infective stage accompanied by slight increase in pH. These values showed a significant increase in group M two weeks post infection, with a tendency to slight increase and normal in group TA and TAF respectively.

Discussion

Duration of treatment was less frequent in group TAF than in group TA. The use of antibiotic therapy alone in bovine mastitis suppresses the functions of PMN Leukocytes of mammary gland (5, 9). Fluid therapy in conjunction with antibiotic may have the ability to treat toxemia and severe infection (2) accompanying mastitis, and so, general animal health including the udder health status could be improved faster. This was also manifested by earlier clinical cure rate in group TAF compared with group TA. The results of SCC in milk agreed to CMT scores among the entire period of study. The CMT reagent (Sodium arylalkyl sulphonate) causes the increased leukocytes (nucleic acids) in mastitic milk to form a gel (7).

The technique of sampling arterial blood in cattle could be applied from brachial, saphenous, coccygeal, and the auricular arteries, but the common carotid artery might be an easier and perfect method (10). Acid-base disorders of the blood (metabolic acidosis, metabolic alkalosis, respiratory acidosis, and respiratory alkalosis) depend primarily upon the ratio between bicarbonate (HCO_3) and PCO_2 in blood (HCO_3/H_2CO_3 system)(11). On applying the Henderson-Hasselbach equation (11), metabolic alkalosis is a prominent criterion in the mastitic cows in this study. It might be concluded that the more chloride ion secreted in the mastitic milk (12) resulted in more HCO_3 diffusion in the plasma. This result is agreed to that seen in mastitic ewes (13) with no any suggestion for the use of fluid therapy but only in cases of peracute coliform mastitis in cows (2). As per author's knowledge no study has been reported on the acid-base status of the blood in cows affected with mastitis in Iraq and very little worldwide (14), while many studies referred that acid-base status might be

affected by pathological conditions, e.g., carbohydrate intake, diarrhea, intestinal strangulation, abomasal displacement, and physiological conditions, e.g., stage of pregnancy, calving and grazing seasons, breeds with climate, sex, highest yielding cows (2).

The pathogenesis of mastitis starts from a damage to glandular tissue followed by invasion, colonization and infection. Infection results in further damage of mammary tissue, which should heal quickly by an effective therapy. Healing can take place only when sufficient amount of collagen is being synthesized by system(15). This process might be affected when any disturbance in the acid – base status referring to a disturbed metabolism. It was observed that SOD (Super Oxide Dismutase) is intracellular superoxide escaping from the neutrophil could damage surrounding tissue and cause inflammatory disease (16). Interstitial fluid, unlike the enzyme, is not limited to the cell and could serve as a proper media for metabolic processes. Antibiotics alone (in group TA) are less effective after the development of clinical signs as the multiplication of bacteria slowed down considerably (17,18). It has become further easy to accept that faster recovery in clinical mastitis and the normal return of PCO_2 , HCO_3 and BE in group TAF two weeks post infection could be attributed to administration of fluid therapy which maintain the metabolic processes.

The significant differences between SCC, PCO_2 , HCO_3 , BE in group TA and the normal return in group TAF, in addition to the udder health status and CMT scores two weeks post infection, give a conclusion, that the administration of fluid therapy might be useful as an adjunct in case of clinical mastitis in cows to get quick clinical recovery with less number of treatments. However, the role of blood gases and acid – base status in mastitis should be investigated in details.

Table 3: Arterial pH, PO₂, PCO₂, bicarbonate (HCO₃⁻) and base excess (BE) in mastitic cows (mean ± S.D.).

Parameters	Animal groups	Pre-infection	During infection	2 weeks post infection
PO₂ (mm Hg)	M	91.28±3.85	* 112.27±6.03	* 100.11±4.34
	TA	93.50±4.12	* 110.91±5.21	95.84±3.17
	TAF	92.50±4.32	* 113.26±6.27	94.24±3.04
	C	89.66±2.81	91.16±3.39	93.22±2.56
PCO₂ (mm Hg)	M	43.80±4.03	* 59.61±2.15	* 58.71±4.05
	TA	44.21±2.46	* 58.72±2.35	* 52.63±2.71
	TAF	41.32±6.22	* 58.10±4.98	43.17±3.48
	C	42.48±1.56	41.96±1.59	41.74±1.23
pH	M	7.408±0.009	* 7.577±0.010	* 7.528±0.013
	TA	7.410±0.008	* 7.572±0.017	7.431±0.021
	TAF	7.407±0.016	* 7.580±0.025	7.402±0.034
	C	7.408±0.007	7.410±0.006	7.410±0.006
HCO₃⁻ (mEq / L)	M	25.70±1.86	* 36.14±1.95	* 36.10±2.41
	TA	25.61±2.96	* 37.06±2.36	* 31.87±2.72
	TAF	27.66±3.42	* 35.60±0.66	25.98±0.68
	C	25.72±0.88	25.50±1.12	27.70±0.97
BE (mEq / L)	M	2.22±0.48	* 10.02±1.47	* 9.54±1.35
	TA	2.43±0.91	* 8.52±1.26	* 5.62±1.12
	TAF	2.71±0.64	* 8.24±1.44	2.02±0.80
	C	2.10±0.81	2.24±0.58	2.06±0.63

M (group one)(n=7) mastitic cows not treated, TA (group two) (n=7) mastitic cows treated with antibiotics only, TAF (group three) (n=14) mastitic cows treated with antibiotics and fluid therapy, C (group four) (n=5) controls.

* : Significantly different (P<0.05) from the corresponding pre-infection value

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