Effects of subclinical mastitis on biochemical and cellular alterations in goats

Deepak Lal Kumhar, PC Shukla, Brejesh Singh, Amir Amin Sheikh and Rakshanda Bhagat

Abstract
The present investigation was carried out in 30 goats with Subclinical mastitis (SCM) which were randomly divided into five groups with six animals each. Sample (milk) was collected aseptically from different livestock farm, Adhartal, Amanala farm and private goat keepers in the nearby areas of Jabalpur. A total of 260 lactating goats belonging to non descript, Jamnapari, Jamnapari crosses, Barbari and Sirohi breeds were screened. Milk samples from individual teat were subjected to various laboratory tests including pH, somatic cell count (SCC) and coliform count which were significantly increased compared to healthy control animals.

Keywords: biochemical, cellular, subclinical mastitis, goats

Introduction
The subclinical form of mastitis is more difficult to identify and can only be diagnosed bacteriologically or with some direct or indirect laboratory tests. It has been reported that infection changes the permeability of the vessels in udder tissue which causes an increase in Na and Cl levels and leucocytes number but a decrease in K level, lactose and fat rates that leads to an elevation of Somatic Cell Count (SCC). If the owner wishes to avoid the economic losses in their herd, early detection of SCM has always been a matter of great curiosity since subclinical mastitis evinces no gross signs of inflammation. Successful containment of SCM involves rapid diagnosis. The aim of this study was to find out the relationship between pH, somatic cell count, coliform count in the milk with subclinical mastitis in goats.

Material and Methods
The present investigation was conducted at livestock farm, Adhartal, Amanala farm and private goat keepers in the nearby areas of Jabalpur. A total of 260 lactating goats belonging to non descript, Jamnapari, Jamnapari crosses, Barbari and Sirohi breeds were screened. Equal quantity of milk and MCMT reagent was added in a mastitis paddle, giving gentle swirling motion in horizontal plane with minimum agitation. In negative cases mixture was liquefied. Grading of positive samples was done according to the intensity of viscous and gel formation, reflecting the degree of inflammation and leucocyte count. It was scored as trace, +1, +2 and +3 reaction depending on gel formation. The udder of each goat was thoroughly washed with potassium permanganate (0.01%) then wiped with clean cloth. 7ml milk was collected aseptically during mid stream from each half with sterilized vials and transported in ice to the department for further examination.

On the basis of the results of MCMT the infected does were subjected to therapeutic regimen. The results of the curative therapy were judged by retesting the milk samples after completion of the therapy on day-3, 7, 15th. On the basis of pH, somatic cell count, MCMT and reduction in coliform count. The infected Does were divided into 5 groups each comprised of 6 animals for this study.

1. pH
Milk pH was estimated by digital pH meter standardized by non buffer solution thereafter, the pH reading of the normal and mastitic milk sample was recorded on day 0 (pre-treatment) and on day 3, 7 and 15 (post-treatment) are presented in Table 1.
The rise in pH was due to the leakage of blood bicarbonate lowered the acidity as also reported by the milk) subsequently due to the treatment effect that chances of inflammation the pH got.

2. Somatic cell count

The leucocyte count in subclinical mastitis milk was performed to access the degree of infection is presented in Table 2. The SCC was performed on day 0 (pre-treatment) and on day 3, 7 and 15 (post-treatment) using the procedure adopted by [1]. The smear of milk for SCC was prepared within one hour of its collection to minimize disintegration of leukocyte. The sample was uniformly mixed by gentle shaking of vials and the milk (0.01%) and was spread with sterilized bacteriological loop, over one cm square rectangular area on a clean microslide. The milk smear from the test sample was stained by pyronin Y-methyl green stain. A total of 30 fields were counted under oil immersion lens and average number of the cells per field was worked out. The average number of cells was multiplied by the multiplication factor of the microscope i.e. 497512 to obtain the number of cells per ml of the milk.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Treatment</th>
<th>0 day</th>
<th>3 day</th>
<th>7 day</th>
<th>15 day</th>
<th>Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>T1</td>
<td>7.45±0.06^AB</td>
<td>7.26±0.04^A</td>
<td>6.60±0.10^B</td>
<td>6.36±0.04^B</td>
<td>6.92±0.09^B</td>
</tr>
<tr>
<td>2.</td>
<td>T2</td>
<td>7.50±0.08^A</td>
<td>7.25±0.04^AB</td>
<td>7.03±0.05^B</td>
<td>7.05±0.09^B</td>
<td>7.20±0.05</td>
</tr>
<tr>
<td>3.</td>
<td>T3</td>
<td>7.4±0.03^A</td>
<td>7.10±0.10^B</td>
<td>6.31±0.07^C</td>
<td>6.31±0.04^C</td>
<td>6.76±0.10^C</td>
</tr>
<tr>
<td>4.</td>
<td>T4</td>
<td>7.31±0.09^A</td>
<td>7.25±0.06^A</td>
<td>6.68±0.13^B</td>
<td>6.75±0.04^B</td>
<td>7.01±0.07^B</td>
</tr>
<tr>
<td>5.</td>
<td>T5</td>
<td>6.55±0.04^A</td>
<td>6.55±0.04^A</td>
<td>6.55±0.04^A</td>
<td>6.55±0.04^A</td>
<td>6.55±0.01^A</td>
</tr>
<tr>
<td>Overall</td>
<td></td>
<td>7.24^a</td>
<td>7.06^b</td>
<td>6.63^c</td>
<td>6.60^d</td>
<td>6.88</td>
</tr>
</tbody>
</table>

abcd – values with different superscript between treatment (lowercase) and between interval (Uppercase) different significantly (P < 0.05)

3. Coliform count

The MCMT positive sample was performed on agar plate count method using VRBA (violet red bile agar). In this method making tenfold serial dilution of sample in diluents and spreading in VRBA plate and incubated at 37°C for 24hrs. Present coliform colony was counted using colony counter is presented in Table 3.

Table 2: Comparison of Somatic Cell Count (SCC) at Pre and Post treatment groups at different intervals

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Treatment</th>
<th>0 day</th>
<th>3 day</th>
<th>7 day</th>
<th>15 day</th>
<th>Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>T1</td>
<td>14.00±3.45^A</td>
<td>4.00±0.45^B</td>
<td>4.50±0.85^B</td>
<td>4.00±0.52^B</td>
<td>6.62±1.23^NC</td>
</tr>
<tr>
<td>2.</td>
<td>T2</td>
<td>11.83±0.70^AB</td>
<td>8.83±0.87^AB</td>
<td>10.16±0.48^AB</td>
<td>7.16±0.49^B</td>
<td>9.50±0.46^C</td>
</tr>
<tr>
<td>3.</td>
<td>T3</td>
<td>11.50±1.50^aA</td>
<td>6.83±1.67^AB</td>
<td>5.33±0.95^B</td>
<td>5.20±0.43^B</td>
<td>6.54±1.15^NC</td>
</tr>
<tr>
<td>4.</td>
<td>T4</td>
<td>8.00±0.58^A</td>
<td>7.83±0.68^A</td>
<td>8.83±0.95^A</td>
<td>8.72±0.95^A</td>
<td>8.12±0.37^BC</td>
</tr>
<tr>
<td>5.</td>
<td>T5</td>
<td>5.50±0.77^A</td>
<td>5.50±0.77^A</td>
<td>5.50±0.77^A</td>
<td>5.50±0.77^A</td>
<td>5.50±0.36^A</td>
</tr>
<tr>
<td>Overall</td>
<td></td>
<td>10.10^a</td>
<td>6.60^b</td>
<td>6.60^c</td>
<td>5.40^d</td>
<td>7.25</td>
</tr>
</tbody>
</table>

abcd – values with different superscript between treatment (lowercase) and between interval (Uppercase) different significantly (P < 0.05)

Results and Discussion

During the present observation the mean value of milk pH in subclinical mastitis showed a significantly increase in all the groups as compared to their mean control value (Table 9). The mean value in T3 (6.3±0.04) groups was reported to be highly significant than on day 0 (7.4±0.03). The increased pH value is nearer to the pH 6.50 to 6.80 and 5.62, as reported by [2]. The decrease in pH due to reduction in alkalinity (resulted due to inflation by increased number of Na+, Cl ions in the milk) subsequently due to the treatment effect that lowered the acidity as also reported by [3]. The rise in pH was due to the leakage of blood bicarbonate into the milk following damage to the mammary epithelium. Whereas, [4] reported the viability in pH of milk in Sirohi, Jakharana and Barbari goats as between 6.532 to 6.34 and opined that this variation might be due to the effect of breed, stage of lactation, health of animal as well as bacterial contamination in raw milk. However, [5] reported the pH as 6.93±0.577. Similar observations have also been reported by [6] as 6.73±0.01.

In the present study the mean value of SCC got increased as per the stage of lactation due to more no. of handling for milking so, due to more chances of inflammation the pH got increased. The average milk SCC reported by [7] as 7.50x10^5 cells/ml and by [8] as 8.80x10^5 cells/ml. The increased in SCC can also be attributed to increase polymorph nuclear cells and neutrophils [9]. Whereas, on post treatment there was gradual reduction in SCC in all the treatment groups but significant reduction (2.50 to 0.42 cells x 10^5 cells/ml) was obtained in T3 groups indicated reduction of inflammation as a result of treatment drastically as also reported by [10]. The rise in SCC is higher
during mid lactation (July-Aug) may be due to the stress induced by high ambient temperature and the beginning of reproductive season [11].

In the present observation the mean value of coliform count in subclinical mastitis showed a significantly increase in all the groups as compared to their mean control value. The mean value in T3 (1.17±0.14) groups was reported to be highly significant than on day 0 (2.26±0.15). The increased coliform count value is nearer to the, 1.0 to 4.2 x 10^3 CFU/ml, as reported by [10]. The decrease in coliform count due to reduction in alkalinity (resulted due to inflammation by increased number of Na+, Cl- ions in the milk) subsequently due to the treatment effect that lowered the acidity as also reported by [12].

References