Effect of Clinical Mastitis on the Gross Morphometry and Histopathology of Mammary Glands of Sahel Goats

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Abstract
The effect of clinical mastitis (unilateral, n = 10 and bilateral, n = 90) on the morphometry of mammary glands of Sahel goats was evaluated in comparison with those of apparently normal glands (n = 20). The mammary glands were examined at slaughter and glandular measurements such as; glandular weight (GW), glandular mid circumference (GMC), glandular longitudinal length (GLL) and glandular teat lengths (GTL) were measured using a measuring tape. Tissue samples of the mammary gland were collected preserved in 10% buffered formalin, processed, embedded, section and stained with H&E and observed under light microscope at various magnifications for presence of pathological lesions. The mean GW was significantly higher (p < 0.05) in normal glands relative to the unilateral and bilateral mastitis. Mean GLL was significantly higher (p<0.05) in unilateral and bilateral mastitis than in normal glands. The mean GMC was not significantly different in all groups while, GTL was significantly higher (p<0.05) in normal mammary glands compared to the unilateral and bilateral mastitic goats. Histopathological evaluation of the mammary glandular tissue showed acute to sub-acute suppurative inflammatory response (85%) and non-suppurative inflammation (15%). Quantitative histopathology showed varying degrees of vascular responses (hyperemia and congestion), vacuolation of the alveolar cells, epithelial necrosis and fibroplasia in the glandular parenchyma. In conclusion, the results of this study showed that clinical mastitis in the Sahel goat causes a significant alteration in the glandular morphometry and histopathology

Keywords: Clinical Mastitis, Morphometry, Sahel Goat, Histopathology

Introduction
Mastitis is a common disease entity of cattle, buffaloes, dairy and non-dairy goats associated with the inflammation of mammary parenchyma, protracted production loss, risks of premature culling from the herd, the release of injurious toxins in the udder (Yousaf et al., 2010; Hussain et al., 2012), development of fibrosis or acute toxemia (Hussain et al., 2012) and the lesions might vary from increased milk leukocytes counts with no gross alterations in milk to amplified vascular permeability (Oviedo-Boyso et al., 2007; Ibrahim et al., 2011; Hussain et al., 2012) which is accompanied by physical, chemical, pathological and bacteriological changes in milk
and glandular tissues (Radostits et al., 2000; Ameh et al., 1993; Holland and Holland, 2005; Patel et al., 2007). The course of the disease in goats ranges from acute to chronic (Acland, 1995). The disease is caused by several microbial agents such as *Mycoplasma, Corynebacteria, Staphylococcus, Escherichia coli, Bacillus, Pasteurella*, yeast and other fungi (DaMassa, 1983; Ameh et al., 1993; Egwu et al., 1994; Ameh and Tsari, 1999; Alawa et al., 2000; Egwu et al, 2001; Pedersen et al., 2003; Ajuwape et al., 2004; Slaïma et al., 2009).

The distributions of mastitis among goat populations in Nigeria have been reported by several authors. A prevalence of 10% was reported from 900 does examined in Kaduna, Kano and Katsina States of Nigeria (Ameh et al., 1993), while in Maiduguri, a prevalence of 17% was reported from 300 does examined (Ameh and Tsari, 1999). In both studies, the occurrence of unilateral mastitis constituted 68-76% of all mastitis cases. The interactions between the resident and newly recruited leukocytes during the initial phases of mastitis play an imperative role in the establishment of intra-mammary infection (Hussain et al., 2012). However, there was no relationship between unilateral infection and the half (right or left) of the gland affected (Ameh et al., 1993; Ameh and Tsari, 1999). Similarly, Okoli et al (2006) reported a prevalence of 19% out of 1848 does presented for slaughter at an abattoir in Imo state, Nigeria. Necrosis of the mammary gland with gangrene formation was reported in a goat with coliform mastitis (Ameh et al., 1994).

Classification of mammary parenchyma specifies the influence of milk production on the proliferation and differentiation of secretory cell (Capuco and Akers, 1999; Hussain et al., 2012). Clinical mastitis in the Sahel goat is grossly characterized by an increase in size and distortion of shape of the mammary gland. Goss morphometric changes in Sahel goats with clinical mastitis have not been reported. This study was therefore carried out to compare the gross morphometry and histopathology of the mammary gland of Sahel goats with clinical mastitis (unilateral and bilateral) with that of normal goats.

**Materials and Methods**

The mammary glands of 100 adult Sahel goats with clinical mastitis (unilateral = 10, bilateral = 90) and normal goats (n = 20) were examined at slaughter at the Maiduguri Metropolitan abattoir. Clinical mastitis was identified based on physical examination of the gland by palpation...
and examination of expressed milk content for abnormal color and consistency (California mastitis test; Schalm et al., 1971). The goats were aged using dental examination as described by Chibuzo (2006) and body weight was measured using weighing balance. Gross measurements of the mammary glandular morphometry (total glandular weight, glandular longitudinal length, glandular mid-circumference and glandular teat length) were carried out using a weighing balance (kg) and a measuring tape (cm). Tissue sections of the mammary gland were obtained and fixed in 10% buffered formalin. For histological observations, 3 to 5 μm thick sections of the fixed tissues were processed, sectioned and stained with hematoxylin and eosin as described by Bancroft and Gamble, (2008). Microscopic examination of slides was carried out using different lens magnifications and photomicrographs were taken using a Canon PowerShot A470 digital camera. JMP 9 (SAS, QSAS Institute Inc, Cary, NC, USA) statistical software package was used to evaluate the data as mean ± standard deviation, which were then subjected to one way analysis of variance (ANOVA) for statistical variations amongst the means.

**Results**

The age range of the goats with unilateral, bilateral mastitis and the normal was between 3.2 to 3.6 years. Gross examination of the affected glands showed a considerable distortion in size and shape of the glands. The affected halves of the glands appeared irregular in size and were pendulous in nature. Expression of milk content produced milk that had clots, clumps or pus. Goats with bilateral mastitis had both halves affected while those with unilateral mastitis had only one half affected (left or right).

The mean age of the goats with unilateral, bilateral mastitis and the normal was not significantly different (p > 0.05) which was between 3.2 ± 1-3.6 ± 9 years. The mean body weight was significantly higher (p < 0.05) in the goats with unilateral mastitis compared to the bilateral mastitis and the normal. The mean total glandular weight (GW) was significantly higher (p < 0.05) in the goats with normal glands relative to those with unilateral and bilateral mastitis. The mean glandular longitudinal length (GLL) of the mastitic halve was significantly higher (p<0.05) in the goats with unilateral and bilateral mastitis than the normal goats. The mean GLL of the non-mastitic halve was significantly higher (p<0.05) in the goats with bilateral mastitis compared to the unilateral mastitis and the normal goats. The glandular mid circumference (GMC) was not
Table 1: Biometric and gross glandular morphometry of the mammary gland of normal Sahel goats and those with unilateral and bilateral clinical mastitis

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Normal (n = 20)</th>
<th>Unilateral mastitis (n = 90)</th>
<th>Bilateral mastitis (n = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>3.2±1.0a</td>
<td>3.6±0.9a</td>
<td>3.5±0.9a</td>
</tr>
<tr>
<td>Body weight (kg)</td>
<td>17.9±3.8a</td>
<td>21.0±5.1b</td>
<td>20.8±6.1ab</td>
</tr>
<tr>
<td>Glandular weight (kg)</td>
<td>3.6±1.4a</td>
<td>2.6±0.6b</td>
<td>2.4±0.6b</td>
</tr>
<tr>
<td>Glandular longitudinal length(cm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mastitic halve</td>
<td>10.6±3.7a</td>
<td>14.9±3.0b</td>
<td>14.2±1.7b</td>
</tr>
<tr>
<td>Non-mastitic halve</td>
<td>10.6±3.6a</td>
<td>10.2±1.8a</td>
<td>14.1±1.8b</td>
</tr>
<tr>
<td>Glandular mid-circumference</td>
<td>27.3±5.6a</td>
<td>30.7±6.4a</td>
<td>29.4±5.4a</td>
</tr>
<tr>
<td>Glandular teat length (cm)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mastitic halve</td>
<td>4.8±1.4a</td>
<td>6.1±2.0b</td>
<td>5.7±1.1ab</td>
</tr>
<tr>
<td>Non-mastitic halve</td>
<td>4.8±1.5a</td>
<td>4.7±1.2a</td>
<td>5.7±1.1a</td>
</tr>
</tbody>
</table>

All values are expressed as mean ± standard deviation. a, b, c. Means with different superscripts are significantly different (p < 0.05).

Table 2: Quantitative histopathological evaluation of the mammary gland of Sahel goats with clinical mastitis (n = 20).

<table>
<thead>
<tr>
<th>Histopathological changes</th>
<th>Number observed in population</th>
<th>Prevalence (%) in population</th>
<th>Lower 95% confidence interval</th>
<th>Upper 95% confidence interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inflammatory response Suppurative (Neutrophils)</td>
<td>17</td>
<td>85</td>
<td>0.6114</td>
<td>0.9604</td>
</tr>
<tr>
<td>Non-suppurative (Lymphocyte)</td>
<td>3</td>
<td>15</td>
<td>0.0396</td>
<td>0.3886</td>
</tr>
<tr>
<td>Mixed Vascular response</td>
<td>7</td>
<td>35</td>
<td>0.1631</td>
<td>0.5905</td>
</tr>
<tr>
<td>Hyperemia</td>
<td>6</td>
<td>30</td>
<td>0.1284</td>
<td>0.5433</td>
</tr>
<tr>
<td>Congestion Degenerative Changes</td>
<td>2</td>
<td>10</td>
<td>0.0175</td>
<td>0.3313</td>
</tr>
<tr>
<td>Vacuolation of epithelial cells</td>
<td>19</td>
<td>95</td>
<td>0.7306</td>
<td>0.9974</td>
</tr>
<tr>
<td>Glandular epithelial necrosis Fibroplasia</td>
<td>18</td>
<td>90</td>
<td>0.6687</td>
<td>0.9825</td>
</tr>
<tr>
<td>Lobular</td>
<td>2</td>
<td>10</td>
<td>0.0175</td>
<td>0.3313</td>
</tr>
</tbody>
</table>

significantly different (p > 0.05) in all the groups. The glandular teat length (GTL) of the mastitic halve was significantly higher (p < 0.05) in the unilateral mastitic halve compared to the bilateral mastitic halve and the normal. Furthermore, the GTL for non mastitic halve was not significantly different for both the unilateral, bilateral and the normal.
Histopathological evaluation of the mammary gland showed acute to sub-acute suppurative inflammatory response marked by neutrophil infiltration (85%) and non-suppurative inflammation marked by lymphocytic infiltration (15%) in the parenchyma (Table 2).

Quantitative estimates of twenty slides examined showed; vascular response ranging from hyperemia (30%) and congestion (10%), degenerative change marked by vacuolation of the alveolar epithelial cells (95%), epithelial necrosis (90%) and fibroplasia (10%) (Fig 1A, B, C and D).

**Discussion and Conclusion**

The alterations in the body weights of the goats in the various groups were not associated with the onset of mastitis. The initial body weights of the goats were as stated. However, there were
slight changes in the body weight of the mastitic goats due to their nutritional status. The total glandular weights in normal goats were significantly higher. The increase in the total glandular weight could be associated to the reservoir of milk in the lactiferous glands of the normal goats as they were actively lactating. The decrease in the total glandular weights in the mastitic goats was probably due to significant decreased in the number of alveoli, alveolar diameter and secretory alveolar cell population. This finding is similar to the study conducted by Hussain et al., 2012. The glandular longitudinal length was significantly higher in goats with clinical mastitis than in the normal goats, probably because the mammary gland of goats with clinical mastitis is less elastic, pendulous and stretched due to damage of the suspensory ligament by the inflammatory process, thus increasing the glandular length and distorting the glandular morphology. In the study carried out by (Hussain et al., 2012), they found the prevalence of mastitis was higher in quarters with small teat and streak canal length and large teat diameter. They postulated that the changes in the length and size of the teat could be due to the pathogens traversing less distance to establish infection in the mammary glands. The association of these factors with the incidence of mastitis has already been established globally (Klaas et al., 2004; Bhutto et al., 2010). In a frequency analysis the prevalence of mastitis in cattle was indicated to be higher in pendulous, round and bowl shaped udder (Hussain et al., 2012). Furthermore, higher prevalence of mastitis in compromised teat and long udder shape has been described (Klaas et al., 2004; Bhutto et al., 2010). Hussain et al. (2012) also confirmed that teat lesions, teat/udder shape and pendulous udder were significantly associated with mastitis and in a previous study conducted by (Shukla et al., 1997; Breen et al., 2009; Bhutto et al., 2010).

Furthermore, the main histopathological findings in the present study were the atrophy of alveoli, fragmented alveoli, acute to sub-acute suppurative inflammatory response marked by neutrophil infiltration, increased stromal tissue and presence of non-suppurative inflammation marked by lymphocytic infiltration in alveoli, hyperemia and congestion, degenerative change marked by vacuolation of the alveolar epithelial cells, epithelial necrosis and fibroplasia. These findings were in accord with the study conducted by (Hussain et al., 2012). The histopathological changes observed in this study were in line with that observed by Alawa et al (2000) in Nigerian goat breeds with chronic mastitis, where he observed mild to severe inflammation with involution of glandular parenchyma and replacement by fibrous connective tissue. Similarly, Singh et al
(1998) observed acute and chronic proliferative lesions in experimental Candidal mastitis in the goat.

In conclusion, the results of this study showed that clinical mastitis in the Sahel goat causes a significant alteration in the glandular morphometry (glandular weight, glandular longitudinal length and teat length) and histopathology of the glandular tissue resulting from acute to sub-acute suppurative and non-suppurative inflammatory responses.

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Conflict of Interest

The authors have no any potential conflict of interest.

References