Original Article

Buffalo Bulletin (January-March 2026) Vol.45 No.1
https://doi.org/10.56825/bufbu.2026.4515747

ELIMINATING SUBCLINICAL URINARY TRACT INFECTION AMELIORATES PRODUCTION AND REPRODUCTION PERFORMANCE IN BUFFALOES

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Received: 01 February 2024
Accepted: 27 March 2026

ABSTRACT

An organized Murrah buffalo farm with a herd strength of 180 was considered for the study. Forty-six apparently healthy buffaloes were included in the study excluding the pregnant animals and animal in estrus and screening was done to detect subclinical infection. Urine was collected in sterile container by catheterization. Bacteriological examination of urine was done along with routine urine analysis. Subclinical UTI was detected in 12 buffaloes (26.08%). Affected animals were divided into two groups in first group enrofloxacin 5 mg/kg b.wt was given intramuscularly for 5 days and in second group amoxicillin was given 10 mg/kgb.wt was given intramuscularly for 5 days. Animals were given treatment on the basis of pre-determined sensitivity. Bacteriological cure rate was 83.33% in Group 1 and 100% in Group 2. Retrospective analysis of production and reproduction data was compared with post treatment data. Statistical analysis revealed significant increase in milk yield by a mean of 0.725 kg per head per day after the treatment. Five out of 12 treated animals who were repeated breeder, conceived with first AI post treatment. Data of affected animals revealed conception rate of 2.60±0.38 in normal animal as compared to 4.67±0.84 in affected animal group. Average service period was 117 days in normal animal group while it was 129 days in affected animal group. On the basis of present study, it can be concluded that subclinical UTI can be a cause of impaired production and reproduction performance in buffaloes.

Keywords: *Bubalus bubalis*, buffaloes, subclinical urinary tract infection, production and reproduction traits, Murrah buffaloes, treatment

INTRODUCTION

Production and Reproduction play crucial role in economics of a dairy herd. It is mainly affected by poor nutrition, reproductive tract problems and urinary tract infections (UTI). Urinary system regulates the body fluid components and is responsible for removal of toxic waste from the body (Divers and Peek, 2018). In cattle, urinary tract affections are commonly associated with cystic calculus, difficult parturition, contaminated catheterization, late pregnancy and sequel to paralysis of the bladder

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(Constable *et al*., 2017). Females are generally more effected as compared to males owing to the reasons related to the anatomical features (short urethra), hormonal status (high levels of estrogen may affect the functional integrity of the epithelium in the urethra and urinary bladder), risks associated with pregnancy or iatrogenic procedures like catheterization (Stevens *et al*., 2007).

Bacteria are the most common reason for UTI and mainly caused by *Escherichia coli*, followed by *Corynebacterium renale* group (*C. renale, cystidis* and *pilosum*), *Trueperella* (formerly *Arcanobacterium*) *pyogenes*. (Yeruham *et al*., 2006).

UTI affects the herd badly by its indirect effect on reproductive system. Its morbidity rate in lactating cattle has been recorded to be 4% and involuntary culling rate of 47% by Yeruham *et al.* (2004) in four-year survey in Israel (1997 to 2000). Cases of UTI remains underdiagnosed because signs of urinary tract infections are subtle in cattle. Most urinary bladder and renal lesions are subclinical, and they might have remarkably higher frequencies than expected; such lesions could result in the poor production of the involved animals (Divers *et al*., 1982; Yeruham *et al*., 2006). UTI may also elicit vascular damage to the urinary bladder and decrease kidney function competence, with subsequent disturbances in protein, acid-base, water and solute homeostasis and in the excretion of final metabolic products (El-Deeb and Elmoslemany, 2016).

For therapy, antimicrobial agents are indicated to control the infection and determination of the antimicrobial susceptibility of causative bacteria are essential. Treatment is continued for a minimum of 7 days and preferably for 14 days. Repeated bacterial culture of urine at least once and again within 7 to 10 days after completion of treatment should be used to assess the success of therapy (Constable *et al*., 2017).

Keeping the above point under consideration, the present study was conducted to evaluate the impact of treatment of subclinical urinary tract infection in buffaloes and its impact on production and reproduction traits.

MATERIALS AND METHODS

Sample area, sample size and sample collection

The present investigation was carried out at Murrah Buffalo Farm, LUVAS, Hisar. A total of 46 animals were sampled from a herd of strength 180, purposively excluding the pregnant animals and animal in estrus. Urinary catheterization using a sterile two-way foley 18F catheter was done to collect the urine samples. Initial stream of urine through catheter was discarded as it contains contaminants from urethra. Then a sterile syringe was applied to the catheter and approximately 30 ml of urine was collected. Then this urine was transferred to sterile urine containers in duplicate for further examination.

General clinical examination and anamnesis

Complete general clinical examination of all the animals was made which included recording of vital parameters, signalmentand clinical signslike pyrexia, hematuria, pyuria, stranguria,hemoglobinuria, dysuria, decreased milk yield and decreased conception rate were recorded.

Physical, chemical and microscopic examination of urine samples

Physical examination

Color of urine sample was noted to be

normal, which is light yellow, yellow or amber color and abnormal colors (red, brown, green, orange or pale). Other physical parameters were examined like odour and turbidity.

Chemical examination

Chemical examination of urine was done using dipstick urinalysis method.

Microscopic examination

Urine samples were shaken and then a centrifuge tube filled with 15 ml of urine and it was centrifuged for 3 minutes at 1000 to 1500 rpm. The supernatant was poured off leaving the sediment and one or two drops of urine. The sediment completely mixed with the urine. A drop of this was put on a clean glass slide and covered with a cover glass. It was examined under the microscope with the low power objective with subdued light and then examined with the high-power objective to identify the smaller objects. Casts were recorded as type and number of casts seen per low power field (LPF) (l0 X objective). RBCs, WBC and epithelial cells were recorded as the no. of cells per high power field (40X objective). Crystals were recorded as few, many or abundant.

Bacteriological culture examination of urine samples

Streaking of urine samples were done on 5% sheep blood agar (BA), Nutrient agar (NA) and MacConkey‘s lactose agar (MLA) to isolate the bacteria involved in infection. Incubation was done at 37^{o}C for 24 to 48 h and subsequent purification and identification of bacteria was done on the basis of Gram‘s reaction, morphology and colony characteristics. The Gram-positive cocci were subjected to catalase test to differentiate between *Staphylococci* spp. and *Streptococci* spp. Other organisms like *Escherichia coli* and *Klebsiella* spp. were differentiated on the basis of growth on eosin methylene blue agar (EMB) and MLA.

Antibiogram pattern

Different strains of various organisms isolated from urine samples of the infected animals were subjected to *in vitro* drug sensitivity testing, using 26 antimicrobials belonging to seven groups of antibiotics by disc-diffusion method (Bauer *et al*., 1966).

Treatment of subclinical urinary tract infections

A total of 12 animals identified to be suffering from urinary tract infection were selected for the treatment based on sensitivity pattern. Buffaloes were divided into two groups and were given antibiotic, in first group enrofloxacin 5 mg/kg b.wt was given intramuscularly for 5 days and in second group amoxicillin was given 10 mg/kgb. wt was given intramuscularly for 5 days along with the supportive therapy. Evaluation of therapeutic efficacy was done on the basis of clinical cure and bacteriological cure rate. For this urine samples were collected 3 days after administration of last dose of antibiotic. Cultural examination was done to compare day 0 (start of therapy) sample result with that of day three post therapy

Impact on production and reproduction traits

For determining the impact on production and reproduction traits, retrospective data was collected regarding milk yield, service period, number of AI per conception and about repeat breeding syndrome. The data was compared between affected and normal animals, before and after treatment. Data was analyzed using appropriate statistical tools.

RESULTS

The overall prevalence rate on the basis of cultural examination was 26.08% (12/46). Affected buffaloes manifested non-specific signs of UTI. Major observations were inappetence, decreased milk production and repeat breeding without any signs of uterine infection involvement. Temperature was found to be within normal range with normal mucus membrane on examination. Comparison of urine parameters between normal and infected animals is shown in Table 1 which showed significant difference in specific gravity of urine.

In the UTI positive animals (n=12), maximum animals belong to Age group 4 to 6 years (50%) followed by 1 to 3 years Age group (33.33%) and >6 years age (16.67%). Out of 12 samples positive for UTI, 5 (41.66%) samples yielded pure culture whereas mixed growth was observed in 7 (58.33%) samples. The percentage efficacy of treatment has been recorded on the basis of bacteriological cure of urine sample collected 3 days after administration of last dose of antibiotic. It was seen that percentage efficacy of first group was 83.33% after treatment for 7 days which increased to 100% after treatment for 14 days. The percentage efficacy of second group was 100% even when treatment was given for 7 days.

On evaluating the impact of subclinical urinary tract infection on the reproductive status of buffaloes data was evaluated retrospectively as mentioned in Table 2, service period of infected animals was found to be more (129 days) in comparison to that of normal animals (117 days) but there was no significant difference between the two groups. There was significant difference between number of AI required to get animal conceived between the normal animals (2.60±0.38) and that for the infected animals (4.67±0.84). It was found that although there was no gross abnormality in reproductive tract and animals were coming in estrus regularly with clear vaginal discharge, but they were not able to conceive. Five out of 12 animals were treated with first AI post treatment. Animals showed improvement in conception rate after treatment.

Production wise mean milk yield of affected Animals group was 9.45±0.60 kg per day before treatment which increased to 10.17±0.48 kg per day after treatment (Table 3). There was significant difference ($P<0.05$) in milk yield before and after the treatment. Milk yield increased by a mean of 0.725 kg per head per day after the treatment of infected animals. Following treatment, significant increase ($P<0.05$) in milk yield per head per day was reported.

DISCUSSION

Non typical subtle clinical signs were shown by effected animals. Yeruham *et al.* (2004) which reported almost similar signs in affected cattle such as reductions in feed intake, reduction in milk production, and weight loss. In contrast to the present study, Braun *et al.* (2008) however reported colic and stranguria and Floeck (2007) recorded arched back and tensed abdominal wall in 16 out of 19 UTI affected cows.

Presence of bacteria was detected at 26.08% (12/46). Although different studies have been conducted to evaluate the percentage of UTI in cattle, but all were done postmortem whereas in present study live animals were considered. Various study reported variable results *viz.* Hajikolaei *et al.* (2015) reported the percent infection as 17.75% in slaughtered female cattle

Table 1. Comparison of urine parameters between normal and infected animals.

Parameter	Normal animals	Infected animals
pH	7.58±0.73	7.75±0.75
Sp. gravity	1.03±0.01[b]	1.01±0.01[a]

*Mean (±SE) bearing different superscripts (a, b) differ significantly (P<0.05) between normal and infected animals.

Table 2. Comparison of reproductive parameters between normal and infected animals.

Parameter	Normal animals	Infected animals
Service period (in days)	117	129
Number of AI	2.60±0.38[a]	4.67±0.84[b]

*Mean (±SE) bearing different superscripts (a, b) differ significantly (P<0.05) between normal and infected animals.

Table 3. Effect of treatment on production parameters.

Parameter	Before treatment	After treatment
Mean of milk yield (in kg)	9.45±0.60[a]	10.17±0.48[b]

*Mean (±SE) bearing different superscripts (a, b) differ significantly (P<0.05) before and after treatment for milk yield.

and Nikvand *et al.* (2014) to be 6.30% at slaughter in female buffaloes (353 animals) on bacterial culture while on histopathological examination the chronic cystitis rate was found to be 14.4%. Herenda *et al.* (1990) also reported the prevalence of (5.3%) in slaughtered cattle when examination was carried out by cultural examination of urine collected from urinary bladder.

In present study animals in age group of 4 to 6 years were mostly affected. Similar findings have been reported by Markusfeld *et al.* (1989) who found that cows in the second, fourth or higher lactation had the maximum incidence rate while the cows at third lactation were somehow protected. In contrast to the present study, Hajikolaei *et al.* (2015) reported the highest percentage of UTI in animals of age 2 years (53.84%) and Solomon *et al.* (2020) reported that the risk of contracting pyelonephritis increases with parity.

Successful treatment of UTI with enrofloxacin and amoxicillin have been reported in present study which is in conjunction with the findings of Braun *et al.* (2008); Reddy *et al.* (2017). Although treatment in enrofloxacin group resulted in 83.33% bacteriological treatment after 7 days which reached 100% after 14 days treatment.

In the present study it was found that the animals suffering from UTI have decreased reproduction potential. There may be either direct or indirect relationship between UTI and

reproduction system. Menin *et al.* (2008) reported that 93.98% of sows with clinical signs of some reproductive disorders were diagnosed as positive for UTI. Similarly, there is decrease in production of milk yield in UTI affected animals. Nyabinwa *et al*. (2020) reported that a case of endometritis had milk yield reduced in 30-day by 40.7±5.0 litres/ cow representing a reduction of 1.4±0.2 litres/ cow/day (15.3%) when compared to milk yield of healthy cows. In present study, milk yield has been found to increase after the therapy which is similar to the findings of Yeruham *et al.* (2006). Hence, it was found that UTI both affects reproduction and production. The effect of treatment may be due to improvement in feed intake and decreased inflammation in the various body systems leading to overall good health of animals.

In conclusion subclinical UTI should be ruled out in farm with problem of decreased milk yield, repeaters and decreased conception rate along with other factors. Treatment of subclinical urinary tract infection should be done for at least 14 days and bacteriological evaluation to be made post therapy.

ACKNOWLEDGMENT

The authors acknowledge in-charge, Buffalo Farm, LUVAS, for providing the facilities.

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