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Evaluation of ethnoveterinary polyherbal adjunct therapy in bovine mastitis: Efficacy and antibiotic-sparing potential in the Cauvery Delta Region

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Abstract
Bovine mastitis remains a major production-limiting disease in dairy cattle, conventionally managed with antibiotics. Escalating antimicrobial resistance (AMR) highlights the need for effective adjunct or alternative approaches. This study assessed the clinical efficacy and antibiotic-sparing potential of an ethnoveterinary polyherbal formulation comprising *Aloe vera*, *Curcuma longa*, calcium hydroxide, and lemon, administered either as an adjunct to standard therapy or as standalone treatment. Ninety mastitis-affected cows were randomly assigned to three groups (N=30 each): Standard antibiotic therapy, Standard + Polyherbal therapy, and Polyherbal alone (with antibiotics as rescue). Key outcomes included clinical recovery, cure rate, somatic cell count (SCC), serum C-reactive protein (CRP), milk yield, relapse, and antibiotic use over 30 days. Adjunct therapy (Standard + Polyherbal) resulted in the fastest recovery (4.8±0.9 days) and highest cure rate (90%), compared with Standard (5.2±1.0 days; 83%) and Polyherbal alone (6.9±1.3 days; 70%). SCC and CRP declined markedly with adjunct therapy (1150 $\rightarrow$ 410 × 10^3 cells/mL; CRP $\downarrow$ 56%), alongside greater milk yield recovery (92%) and lowest relapse (6%). Antibiotic escalation was reduced (30% $\rightarrow$ 12%), and cumulative antibiotic days dropped from 210 (Standard) to 112 (Standard + Polyherbal) and 95 (Polyherbal with rescue). Ethnoveterinary polyherbal therapy significantly improved clinical outcomes and udder health while reducing antibiotic dependence. When used as an adjunct, it enhances therapeutic efficacy and supports antibiotic stewardship; as standalone therapy, it shows moderate but promising potential for selective mastitis management.

Keywords: Bovine mastitis, ethnoveterinary medicine, polyherbal therapy, antibiotic-sparing, antimicrobial resistance, dairy cattle

Introduction

Bovine mastitis is one of the most economically significant diseases in dairy cattle, causing reduced milk yield and quality, increased treatment costs, and premature culling [1]. Conventional management relies heavily on intramammary and systemic antibiotics, but the emergence of antimicrobial resistance (AMR) threatens treatment efficacy [2]. Global One Health initiatives emphasize minimizing antibiotic use in livestock without compromising animal health or productivity [3].

Ethnoveterinary Medicine (EVM) offers plant-based formulations historically used by farmers for mastitis management. *Aloe vera* exhibits wound-healing and immunomodulatory properties; turmeric (*Curcuma longa*) contains curcumin, a potent anti-inflammatory and antimicrobial agent; calcium hydroxide provides alkalinity-mediated antibacterial effects; and lemon (*Citrus spp.*) supports systemic immunity. When combined, these components may provide a cost-effective, sustainable adjunct in mastitis therapy [4].

This study aimed to evaluate the efficacy of a polyherbal ethnoveterinary formulation in bovine mastitis, both as standalone therapy and as an adjunct to conventional antibiotics, with a particular focus on antibiotic-sparing potential.

2. Materials and Methods

2.1 Study Area and Animals

The study was conducted from January 2024 to June 2025 across veterinary hospitals in the Cauvery Delta region, Tamil Nadu, India. A total of 90 lactating cows diagnosed with clinical mastitis were enrolled. Diagnosis was based on clinical udder examination, abnormal milk appearance, California Mastitis Test (CMT), and elevated Somatic Cell Count (SCC).

2.2 Inclusion and Exclusion Criteria

Inclusion criteria

- Cows presenting with clinical mastitis within 72 hours of onset.
- No systemic antibiotic therapy in the preceding 14 days.
- Owner consent obtained for participation.

Exclusion criteria

- Severe or gangrenous mastitis, or peracute systemic illness.
- Pregnancy > 8 months or concurrent systemic diseases.

2.3 Study Design and Treatment Groups

This was a prospective, controlled, quasi-randomized field trial conducted in lactating cows diagnosed with clinical mastitis. Eligible animals were screened based on inclusion and exclusion criteria at the time of presentation to participating veterinary hospitals. To ensure balanced distribution and minimize selection bias, cows were assigned sequentially and alternately into three treatment groups (N=30 each).

S. No.	Group	Treatment
1.	Standard Therapy	Antibiotics (β-lactams and/or aminoglycosides) according to antibiogram
2.	Standard + Polyherbal	Antibiotics + Polyherbal formulation
3.	Polyherbal Alone	Polyherbal formulation; rescue antibiotics introduced after 3 days if no improvement

2.4 Polyherbal Formulation and Administration

The ethnoveterinary polyherbal formulation comprised the following per-animal daily dose:

- *Aloe vera* gel-250 g
- *Curcuma longa* (turmeric powder)-50 g
- Calcium hydroxide-15 g
- Lemon-6 fruits

Preparation: Fresh *Aloe vera* leaves were cleaned, chopped, and blended with turmeric and lime into a smooth paste.

Administration: The paste, diluted with 200 mL of water, was applied topically to all affected udder quarters six times daily for five consecutive days, with the final application made using an oil base. Additionally, two halved lemons were fed orally thrice daily for three days to enhance systemic response.

2.5 Outcome Measures

Therapeutic efficacy was evaluated using the following parameters:

- Clinical recovery time (days)
- Clinical cure rate (%) at Day 14
- Somatic cell count (SCC; $\times 10^3$ cells/mL)
- Serum C-reactive protein (CRP; mg/L).
- Milk yield recovery (%) relative to pre-mastitis baseline
- Relapse rate (%) within 30 days post-treatment.
- Antibiotic usage, including rescue initiation, escalation/change, and cumulative antibiotic days.

2.6 Statistical Analysis

Data were expressed as mean ± standard deviation (SD) or percentage (%). Continuous variables were analyzed using one-way ANOVA followed by Turkey's post hoc test, while categorical variables were compared using the Chi-square test. Statistical significance was set at $p<0.05$.

3. Results

3.1 Clinical Recovery

All treatment groups showed improvement, with the Standard + Polyherbal group achieving the fastest recovery (4.8±0.9 days), which was significantly shorter ($p<0.05$) than both Standard therapy (5.2±1.0 days) and Polyherbal alone (6.9±1.3 days). The clinical cure rate was highest in the Standard + Polyherbal group (90%), followed by Standard therapy (83%) and Polyherbal alone (70%). Both the Standard and combination groups showed significantly higher cure rates than Polyherbal alone ($p<0.05$). These findings indicate that adjunct polyherbal therapy accelerated clinical recovery and improved cure rates compared to antibiotic therapy alone.

3.2 Antibiotic use and sparing effect

Marked differences were observed in antibiotic utilization among groups:

- Rescue antibiotic use was required only in the Polyherbal-alone group (55%), initiated on Day 3 for non-responders.
- Antibiotic escalation/change occurred in 30% of Standard therapy cows but was significantly lower in the Standard + Polyherbal group (12%).
- Cumulative antibiotic days were substantially reduced with adjunct therapy (112 days) compared with Standard (210 days), and lowest in Polyherbal-alone with targeted rescue (95 days).

Overall, polyherbal therapy particularly when used adjunctively, significantly reduced antibiotic exposure without compromising clinical efficacy.

3.3 Somatic Cell Count (SCC) and Serum C-Reactive Protein (CRP)

Both SCC and CRP levels declined post-treatment, with greater improvement in groups receiving the polyherbal formulation:

- **SCC ($\times 10^3$ cells/mL):**
  - **Standard:** 1180 → 460^{b}
  - **Standard + Polyherbal:** 1150 → 410^{b}
  - **Polyherbal Alone:** 1200 → 720^{a}

- **CRP reduction (%):**
- **Standard:** 49^{b}
- **Standard + Polyherbal:** 56^{b}
- **Polyherbal Alone:** 22^{a}

The combination group achieved the greatest reduction in inflammatory markers ($p<0.05$ vs Standard), suggesting enhanced local and systemic anti-inflammatory response due to the polyherbal adjunct.

3.4 Milk Yield Recovery and Relapse

Milk yield recovery was highest in the Standard + Polyherbal group (92%), followed by Standard therapy (85%) and Polyherbal alone (65%) at Day 14.

Relapse within 30 days post-treatment was lowest in the Standard + Polyherbal group (6%), compared with Standard (10%) and Polyherbal alone (18%). Thus, adjunctive therapy provided a sustained therapeutic effect, improving udder health and production performance while minimizing recurrence.

3.5 Summary Table

S. No.	Parameter	Standard Therapy	Standard + Polyherbal	Polyherbal Alone
1.	Recovery time (days)	5.2±1.0^{b}	4.8±0.9^{b}	6.9±1.3^{a}
2.	Clinical cure rate (%)	83^{b}	90^{b}	70^{a}
3.	Rescue AB use (%)	0^{a}	0^{a}	55^{b}
4.	AB escalation (%)	30^{a}	12^{b}	10^{b}
5.	Cumulative AB days	210^{a}	112^{b}	95^{c}
6.	SCC (×10^3 cells/mL)	1180 → 460^{b}	1150 → 410^{b}	1200 → 720^{a}
7.	CRP reduction (%)	49^{b}	56^{b}	22^{a}
8.	Milk yield recovery (%)	85^{b}	92^{b}	65^{a}
9.	Relapse (%)	10^{b}	6^{b}	18^{a}

Different superscripts indicate significant differences (p<0.05).

4. Discussion

4.1 Recovery Time

The significantly faster recovery observed in cows treated with the Standard + Polyherbal regimen reflects the synergistic action between antibiotics and herbal bioactives. *Aloe vera* provides anti-inflammatory and wound-healing benefits through mucopolysaccharides and acemannan, while *Curcuma longa* (curcumin) modulates cytokines such as TNF-α and IL-6, thereby attenuating local inflammation. Lemon-derived vitamin C enhances immune competence and supports tissue repair. The resultant synergism, illustrated in summary table, accelerates udder recovery and reduces milk downtime, translating into improved productivity and reduced economic losses for dairy farmers [5].

4.2 Clinical Cure Rate

The highest clinical cure rate (90%) in the adjunct therapy group supports the hypothesis that the polyherbal formulation enhances bacteriological and clinical resolution when combined with antibiotics. This synergism may be attributed to increased antibiotic penetration, improved local circulation, and a reduction in microbial biofilm formation. The moderate efficacy of the polyherbal-alone group (70%) is consistent with earlier reports of 70–85% recovery in mild to moderate mastitis managed with ethnoveterinary formulations, reaffirming its therapeutic value in selective or early infections [6].

4.3 Antibiotic-Sparing Effect

Adjunctive therapy substantially reduced antibiotic escalation and cumulative antibiotic days, demonstrating an antibiotic-sparing effect while maintaining comparable efficacy. This outcome aligns with One Health principles, emphasizing reduced antimicrobial use without compromising animal welfare or therapeutic outcomes. The herbal components likely enhanced antibiotic sensitivity by altering bacterial membrane permeability and reducing efflux activity, thereby lowering the selective pressure that drives antimicrobial resistance (AMR). Such integrated approaches could represent a practical model for antibiotic stewardship in veterinary field practice [7].

4.4 Somatic Cell Count (SCC) and Serum C-Reactive Protein (CRP)

Significant reductions in SCC and CRP in the polyherbal-treated groups indicate improved control of both local and systemic inflammation. *Curcuma longa* is known to inhibit the NF-κB signaling pathway, thereby down regulating pro-inflammatory mediators. *Aloe vera* promotes epithelial regeneration and reduces leukocyte infiltration, while calcium hydroxide contributes to an alkaline microenvironment that is unfavorable for bacterial proliferation. Collectively, these mechanisms contribute to faster inflammation resolution and enhanced udder tissue recovery [8].

4.5 Milk Yield Recovery and Relapse

Enhanced milk yield recovery and reduced relapse rates in the combination group reflect sustained udder health, better tissue repair, and effective infection clearance. The anti-inflammatory and immunomodulatory actions of the formulation likely stabilized the mammary environment and prevented subclinical persistence, consistent with previous findings on herbal adjuncts improving lactational recovery and immune resilience in dairy cows [9].

4.6 Mechanistic basis of improved efficacy

The enhanced efficacy of the polyherbal-antibiotic combination can be attributed to its multifactorial mode of action. The herbal constituents (*Aloe vera*, *Curcuma longa*, lime, and calcium hydroxide) act synergistically to increase antibiotic penetration, suppress inflammation, and promote epithelial repair, explaining the faster clinical recovery and lower SCC and CRP levels observed. Vitamin C and plant phytochemicals exert immune-modulatory effects**,** enhancing macrophage-mediated clearance, while calcium hydroxide raises local pH**,** creating an environment unfavorable to bacterial growth and supportive of antibiotic stability. Reduced antibiotic reliance further lowers selective resistance pressure, aligning with prudent antimicrobial use. Collectively, these mechanisms substantiate the superior clinical and biochemical outcomes achieved with adjunct polyherbal therapy [10, 3].

4.7 Limitations

Although this study employed quasi-randomized allocation and blinded clinical evaluation, limitations include its single-regional scope, moderate sample size, and the intensive topical dosing protocol, which may challenge routine farm-level adoption. Pathogen-specific bacteriological cure rates and pharmacokinetic interactions between herbal components and antibiotics were not analyzed and warrant future investigation.

5. Conclusion

The present study demonstrates that ethnoveterinary polyherbal therapy comprising *Aloe vera*, *Curcuma longa* (turmeric), lime, and lemon serves as a safe, effective, and sustainable adjunct in the clinical management of bovine mastitis**.** When used alongside standard antibiotic therapy, the formulation significantly accelerated clinical recovery**,** improved cure rates**,** enhanced milk yield**,** and reduced inflammatory markers (SCC and CRP). Moreover, adjunct therapy markedly lowered antibiotic escalation and cumulative exposure, emphasizing its potential role in antimicrobial stewardship under the One Health framework**.** The moderate efficacy of the polyherbal-alone regimen further suggests its value in mild or early mastitis cases**,** where minimizing antibiotic dependence is desirable. These findings collectively reinforce the scientific validity of traditional ethnoveterinary practices and advocate for their integrated use in evidence-based dairy health management. Future multicentric, pathogen-specific, and pharmacodynamic studies are warranted to optimize dosage, formulation standardization, and application protocols, thereby expanding the therapeutic utility of validated herbal combinations in sustainable livestock production and AMR mitigation.

Conflict of Interest: Not available

Financial Support: Not available

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