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Clinical Evaluation of Reproductive Disorders in Buffaloes under Field Conditions Using Ultrasonography and Its Diagnostic and Practical Implications

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Abstract

Reproductive inefficiency is a major constraint in buffalo-based smallholder dairy systems, where delayed diagnosis of pregnancy and uterine–ovarian disorders adversely affects calving interval, milk yield, and farm income. This study evaluated portable ultrasonography (USG) as a field-level diagnostic and extension intervention under rural conditions of western Uttar Pradesh, India. A three-year field investigation was conducted across 21 reproductive health camps involving 368 buffaloes, each examined by both per-rectal (PR) examination and transrectal ultrasonography during the same visit. Diagnostic agreement was assessed using McNemar's chi-square test, and farmer perception was evaluated using a structured Likert-scale questionnaire. Pregnancy diagnosis was recorded in 258/368 (70.1%) animals by PR and 345/368 (93.8%) by USG ($\chi^2 = 62.1$, $p < 0.0001$). Early pregnancy diagnosis was 212/368 (57.6%) by PR and 331/368 (89.9%) by USG ($\chi^2 = 95.4$, $p < 0.0001$). Ultrasonography showed significantly higher diagnostic performance for anoestrus/silent heat (22.8% vs 14.1%), repeat breeding (14.7% vs 11.1%), pyometra/metritis/endometritis (11.1% vs 4.9%), ovarian cysts (8.7% vs 3.8%), hydrometra/mucometra (4.1% vs 1.1%), early embryonic death (5.7% vs 1.9%), retained fetal membranes (7.1% vs 4.1%), abortion (8.1% vs 5.7%), stillbirth/early neonatal loss (6.0% vs 3.3%), and mummified fetus (2.2% vs 0.3%) ($p < 0.05$). Animals categorized as normal/no disorder decreased from 206/368 (56.0%) by PR to 110/368 (29.9%) following USG evaluation. Farmer perception analysis demonstrated significantly higher trust, preference, and overall satisfaction with ultrasonography ($p < 0.01$), while willingness to spend on treatment did not differ significantly. The study was limited to field conditions within a defined geographic region and did not include long-term reproductive performance follow-up. Under rural field conditions, ultrasonography demonstrated superior diagnostic performance compared with per-rectal examination and positively influenced farmer perception, supporting its role as a diagnostic and extension intervention in buffalo herds.

Keywords: Anestrus, buffalo, early pregnancy diagnosis, field extension, ultrasonography

Introduction

Buffaloes (*Bubalus bubalis*) constitute a critical component of dairy-based livelihood systems in South Asia and contribute substantially to India's national milk production, thereby functioning as a primary income buffer for smallholder farmers (FAO, 2023). In such production systems, reproductive efficiency is not merely a biological trait but a central economic determinant governing calving interval, lactation continuity, replacement rate, and lifetime productivity. Even marginal impairment in fertility extends days open, increases insemination frequency, and amplifies cumulative economic losses at the household level (Noakes *et al.*, 2018) [8]. Field-level reproductive inefficiency in

buffaloes commonly presents as anoestrus, repeat breeding, uterine infections (metritis and endometritis), ovarian cystic conditions, hydrometra, retained fetal membranes, abortion, and early embryonic death (EED). Among these, early embryonic mortality represents a biologically silent yet economically significant cause of reproductive wastage, frequently occurring before clinical recognition and often misinterpreted as repeat breeding (Sahu *et al.*, 2025) [9]. Such diagnostic ambiguity leads to repeated inseminations, irrational hormonal use, delayed treatment, and erosion of farmer confidence in reproductive interventions. Additionally, postpartum disorders including genital prolapse and fetal mummification, though relatively less

frequent, reflect underlying reproductive management deficiencies and further compromise herd productivity (Sahu *et al.*, 2024a; Sahu *et al.*, 2024b) ^[12, 13]. Per-rectal (PR) palpation remains the most widely practiced reproductive examination method under field conditions because of its low cost and minimal equipment requirement. However, PR examination has intrinsic limitations in accurately differentiating uterine pathologies, detecting early pregnancy, assessing embryonic viability, and characterizing ovarian structures with precision (Noakes *et al.*, 2018) ^[8]. Subtle intrauterine fluid accumulation, early conceptus development, and luteal morphology are particularly difficult to evaluate reliably through palpation alone. These diagnostic constraints often translate into delayed therapeutic decision-making and avoidable economic loss.

Real-time transrectal ultrasonography (USG) has transformed bovine reproductive diagnostics by enabling direct visualization of ovarian follicular dynamics, corpus luteum morphology, uterine lumen characteristics, embryonic vesicles, and fetal heartbeat (Ginther, 2014) ^[4]. Early pregnancy detection is feasible as early as 25–30 days post-insemination, substantially improving management precision compared with palpation-based diagnosis (Noakes *et al.*, 2018) ^[8]. Furthermore, Doppler ultrasonography has expanded diagnostic capacity beyond structural imaging by allowing assessment of uterine artery hemodynamics and tissue perfusion, thereby offering functional insight into estrous cyclicity, uterine inflammation, and recovery dynamics (Sahu *et al.*, 2026a; Sahu *et al.*, 2026b) ^[10, 11]. This integration of structural and vascular assessment strengthens evidence-based reproductive management. Despite its established diagnostic superiority in organized dairy systems, the systematic deployment of portable ultrasonography as a structured field extension intervention in buffalo-dominated smallholder systems remains inadequately documented. Importantly, ultrasonography functions not only as a diagnostic modality but also as a visual communication tool. Real-time visualization of fetal heartbeat, uterine pathology, or ovarian activity enhances farmer understanding, builds trust in veterinary recommendations, and improves compliance with reproductive management protocols. Thus, its value extends beyond diagnostic accuracy into the domains of extension effectiveness and income protection.

Therefore, the present study was undertaken to evaluate ultrasonography-led field extension interventions in buffalo herds under rural conditions. Specifically, the objectives were to (i) compare the diagnostic performance of ultrasonography with conventional per-rectal examination for common reproductive disorders, and (ii) assess changes in farmers' perception following ultrasonographic demonstration. By integrating diagnostic precision with participatory extension outcomes, this work positions portable ultrasonography not merely as a reproductive imaging tool, but as a scalable intervention capable of strengthening reproductive efficiency and enhancing income resilience in buffalo-based dairy systems.

Materials and Methods

Study Area and Duration

The study was conducted in Bareilly and adjoining districts

of western Uttar Pradesh, India, under field conditions typical of buffalo-dominated smallholder dairy systems (Fig. 1). The intervention was implemented over a period of three years through organized reproductive health camps conducted in collaboration with field extension units (Fig. 2).



Fig 1: Study region depicting Bareilly and adjoining districts of western Uttar Pradesh, India, where reproductive health camps were conducted during the three-year field investigation.

Study Population

A total of 21 field reproductive health camps were organized during the study period. Across these camps, 368 buffaloes presented with reproductive complaints were examined. Animals included both cyclic and non-cyclic females of varying parity brought by farmers for infertility, suspected pregnancy, repeat breeding, anoestrus, postpartum complications, or other reproductive concerns.



Fig 2: Field-based extension activities conducted various health camps, including farmer interaction and advisory discussions, conventional per-rectal examination, transrectal ultrasonographic evaluation, on-site data recording, and administration of structured questionnaires for assessment of farmer perception.

Diagnostic Protocol

Each animal underwent a two-step diagnostic procedure during the same visit. Initially, conventional per-rectal (PR) examination was performed following standard large-animal reproductive evaluation procedures as described in established veterinary texts (Noakes *et al.*, 2018) ^[8]. Uterine size, tone, horn symmetry, cervical status, ovarian structures, and presumptive pregnancy indicators were assessed manually. Following PR examination, transrectal ultrasonography (USG) was performed using a portable ultrasound unit equipped with a linear-array transducer appropriate for bovine reproductive imaging. Ultrasonography was conducted according to standard reproductive imaging criteria, allowing visualization of

ovarian follicles, corpus luteum morphology, intrauterine fluid accumulation, embryonic vesicle presence, and fetal heartbeat where applicable (Ginther, 2014) [4]. Diagnostic categorization was based on ultrasonographic visualization of reproductive structures and pathological changes. Diagnostic outcomes recorded through PR and USG were documented separately for comparative evaluation.

Farmer Perception Assessment

To evaluate the extension impact of ultrasonography-based reproductive examination, farmers attending the camps were assessed using a structured Likert-scale questionnaire administered before and after ultrasonographic demonstration. The perception survey evaluated farmer understanding of reproductive status, confidence in diagnosis, satisfaction with examination, willingness to use ultrasonography in future, and likelihood of recommending the method to other farmers. Responses were recorded on a standardized five-point Likert scale. Pre- and post-demonstration perception scores were compared to determine changes attributable to visual diagnostic confirmation.

Statistical Analysis

Diagnostic agreement between PR and USG findings was assessed using McNemar's chi-square test for paired nominal data. Differences in detection rates between the two diagnostic modalities were expressed as frequency and percentage. Changes in farmer perception scores before and after ultrasonographic demonstration were analyzed using paired t-test. Data were analyzed using GraphPad Prism 10.1.2. Statistical significance was considered at $p < 0.05$.

Results

A total of $n = 368$ buffaloes examined across 21 field camps were evaluated using both per-rectal (PR) examination and transrectal ultrasonography (USG). The comparative diagnostic findings are summarized in Table 1 and representative images are shown in Fig. 3.

Comparative Diagnostic Performance

A total of 368 buffaloes examined across 21 field reproductive health camps underwent both per-rectal (PR) examination and transrectal ultrasonography (USG). Comparative diagnostic findings are presented in Table 1. Pregnancy Diagnosis was recorded in 258/368 (70.1%) animals by PR and 345/368 (93.8%) by USG, with 87 (23.6%) PR- / USG+ cases (McNemar $\chi^2 = 62.1$, $p < 0.0001$). Early Pregnancy Diagnosis (EPD) was 212/368 (57.6%) by PR and 331/368 (89.9%) by USG, with 119 (32.3%) PR- / USG+ cases ($\chi^2 = 95.4$, $p < 0.0001$). Anoestrus / Silent Heat was identified in 52/368 (14.1%) by PR and 84/368 (22.8%) by USG, with 32 (8.7%) PR- / USG+ cases ($\chi^2 = 20.9$, $p < 0.0001$). Repeat Breeding was recorded in 41/368 (11.1%) by PR and 54/368 (14.7%) by USG (12; 3.3% PR- / USG+, $\chi^2 = 7.5$, $p = 0.006$). Confirmative PD in Repeat Breeders increased from 18/368 (4.9%) by PR to 41/368 (11.1%) by USG (23; 6.3% PR- / USG+, $\chi^2 = 16.4$, $p < 0.0001$). Abortion was recorded in 21/368 (5.7%) by PR and 30/368 (8.1%) by USG (10; 2.7% PR- / USG+, $\chi^2 = 5.4$, $p = 0.020$). Retained Fetal Membranes (RFM) was 15/368 (4.1%) by PR and 26/368 (7.1%) by USG (11; 3.0% PR- / USG+, $\chi^2 = 7.4$, $p = 0.006$). Pyometra / Metritis / Endometritis increased from 18/368 (4.9%) by PR to 41/368 (11.1%) by USG (23; 6.3% PR- / USG+, $\chi^2 = 16.4$, $p < 0.0001$). Ovarian Cysts (COD / FC / LC) were detected in 14/368 (3.8%) by PR and 32/368 (8.7%) by USG (18; 4.9% PR- / USG+, $\chi^2 = 12.8$, $p = 0.0003$). Hydrometra / Mucometra increased from 4/368 (1.1%) to 15/368 (4.1%) (11; 3.0% PR- / USG+, $\chi^2 = 6.9$, $p = 0.008$). Stillbirth / Early Neonatal Loss was 12/368 (3.3%) by PR and 22/368 (6.0%) by USG (10; 2.7% PR- / USG+, $\chi^2 = 6.4$, $p = 0.011$). Mummified Fetus increased from 1/368 (0.3%) to 8/368 (2.2%) (7; 1.9% PR- / USG+, $\chi^2 = 4.3$, $p = 0.038$). Early Embryonic Death (EED) increased from 7/368 (1.9%) by PR to 21/368 (5.7%) by USG (14; 3.8% PR- / USG+, $\chi^2 = 8.7$, $p = 0.003$). Normal / No Disorder was 206/368 (56.0%) by PR and 110/368 (29.9%) by USG.

Table 1: Comparative diagnostic performance of per-rectal palpation and transrectal ultrasonography for reproductive disorders in buffaloes under field conditions

Reproductive Disorder	Observed field occurrence (%)	Detected by PR (n/368, %)	Detected by USG (n/368, %)	PR- / USG+ (n, %)	McNemar χ^2	p-value
Pregnancy Diagnosis	35–50	258/368 (70.1%)	345/368 (93.8%)	87 (23.6%)	62.1	<0.0001
Early Pregnancy Diagnosis (EPD)	-	212/368 (57.6%)	331/368 (89.9%)	119 (32.3%)	95.4	<0.0001
Anoestrus / Silent Heat	10–30	52/368 (14.1%)	84/368 (22.8%)	32 (8.7%)	20.9	<0.0001
Repeat Breeding	8–16	41/368 (11.1%)	54/368 (14.7%)	12 (3.3%)	7.5	0.006
Confirmative PD in Repeat Breeders	-	18/368 (4.9%)	41/368 (11.1%)	23 (6.3%)	16.4	<0.0001
Abortion	5–9	21/368 (5.7%)	30/368 (8.1%)	10 (2.7%)	5.4	0.020
Retained Fetal Membranes (RFM)	5–10	15/368 (4.1%)	26/368 (7.1%)	11 (3.0%)	7.4	0.006
Pyometra / Metritis / Endometritis	6–15	18/368 (4.9%)	41/368 (11.1%)	23 (6.3%)	16.4	<0.0001
Ovarian Cysts (COD / FC / LC)	4–10	14/368 (3.8%)	32/368 (8.7%)	18 (4.9%)	12.8	0.0003
Hydrometra / Mucometra	1–4	4/368 (1.1%)	15/368 (4.1%)	11 (3.0%)	6.9	0.008
Dystocia	4–6	-	-	-	-	-
Uterine / Vaginal Prolapse	3–8	-	-	-	-	-
Stillbirth / Early Neonatal Loss	3–6	12/368 (3.3%)	22/368 (6.0%)	10 (2.7%)	6.4	0.011
Mummified Fetus	<1	1/368 (0.3%)	8/368 (2.2%)	7 (1.9%)	4.3	0.038
Early Embryonic Death (EED)	3–8	7/368 (1.9%)	21/368 (5.7%)	14 (3.8%)	8.7	0.003
Normal / No Disorder	25–50	206/368 (56.0%)	110/368 (29.9%)	-	-	-

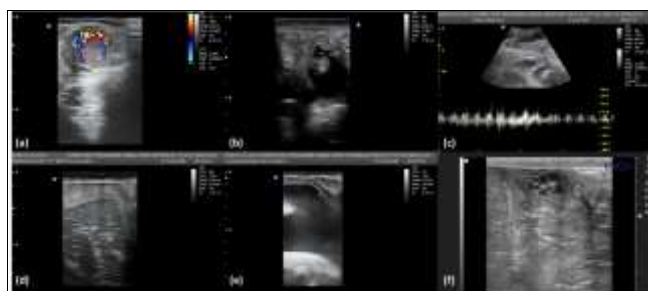


Fig 3: Representative ultrasonographic images recorded during field examination: (a) real-time visualization of functional corpus luteum; (b) early pregnancy confirmation showing embryonic vesicle; (c) early pregnancy confirmation showing fetal heartbeat; (d) pyometra/metritis with echogenic intrauterine fluid accumulation; (e) mucometra presenting as anechoic uterine fluid distension; and (f) anestrus/silent heat characterized by absence of active follicular or luteal structures;

Farmer Perception Analysis

Farmer perception outcomes comparing per-rectal (PR) examination and ultrasonography (USG) are presented (Fig. 4, Fig. 5). Ultrasonography significantly enhanced farmer understanding of reproductive status, trust in diagnosis, and confidence in reproductive decision-making ($p < 0.01$) (Fig. 4). Perception regarding differentiation between pregnancy and reproductive disorders did not differ significantly, likely reflecting moderate baseline awareness among farmers. Preference patterns clearly favored ultrasonography over conventional palpation ($p < 0.01$) (Fig. 5). Farmers reported greater perceived economic benefit, improved clarity of demonstration, and a higher likelihood of recommending USG-based examination to others ($p < 0.01$). However, willingness to spend on treatment did not differ significantly between methods, indicating that increased diagnostic clarity strengthened confidence but did not immediately alter cost-related decisions. Overall satisfaction levels were substantially higher following ultrasonographic evaluation compared with PR examination (Fig. 6), underscoring the extension value of visual diagnostic confirmation under field conditions.

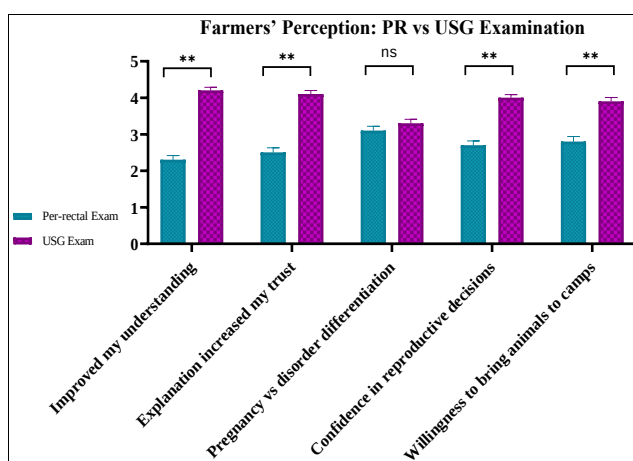


Fig 4: Comparative farmer perception scores for per-rectal examination and ultrasonography-based reproductive diagnosis under field conditions.

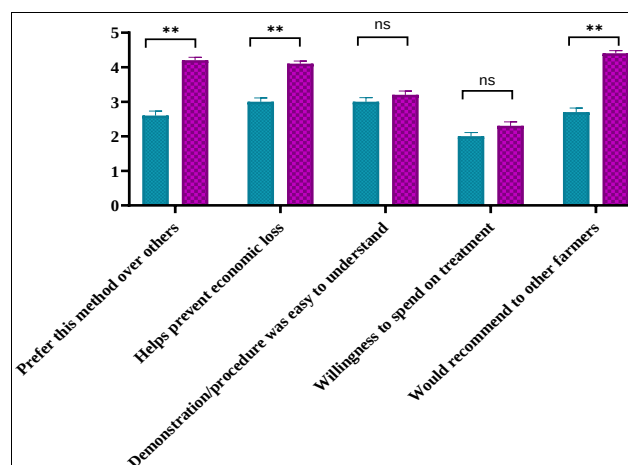


Fig 5: Farmer preference and adoption willingness toward ultrasonography versus conventional per-rectal examination during reproductive health camps.

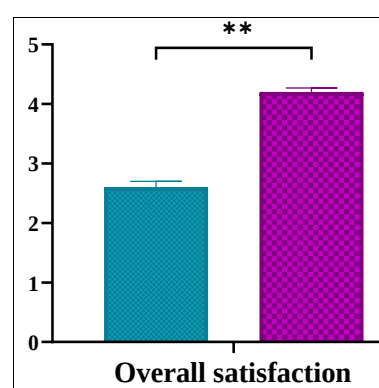


Fig 6: Overall satisfaction levels of farmers following ultrasonographic demonstration compared with per-rectal examination.

The overall findings indicate that ultrasonography significantly enhanced detection rates of early pregnancy, uterine inflammatory conditions, ovarian cysts, hydrometra, and early embryonic death under field conditions. In parallel, ultrasonographic demonstration positively influenced farmer perception, diagnostic trust, and willingness to adopt reproductive management interventions.

Discussion

Accurate reproductive diagnosis under field conditions remains one of the most decisive determinants of fertility management efficiency in buffalo-based smallholder systems. Conventional per-rectal palpation, while economical and widely practiced, is inherently limited by its dependence on tactile interpretation and operator experience. Subtle intrauterine fluid accumulation, early embryonic structures, and differentiation between follicular and luteal ovarian states often fall below the resolution threshold of manual assessment (Noakes *et al.*, 2018) [8]. In contrast, real-time ultrasonography provides direct visualization of reproductive structures, thereby reducing diagnostic ambiguity and enabling physiologically informed

decision-making. The enhanced diagnostic capacity of ultrasonography observed in this study is biologically consistent with established reproductive physiology. Early gestation detection through visualization of embryonic vesicles and cardiac activity allows confirmation of pregnancy at stages where uterine asymmetry may not yet be appreciable by palpation (Ginther, 2014) [4]. Similarly, differentiation of uterine inflammatory conditions depends on identification of echogenic versus anechoic intrauterine contents, endometrial changes, and structural integrity parameters that cannot be reliably evaluated through tactile examination alone. This capacity for structural differentiation has long been recognized as a defining advantage of ultrasonographic imaging in bovine reproduction (Noakes *et al.*, 2018) [8].

Beyond structural imaging, contemporary research has expanded the role of ultrasonography into functional assessment. Doppler-based evaluation of uterine hemodynamics has demonstrated measurable variations in blood flow associated with estrous cyclicity, inflammation, and luteal activity, offering insights into reproductive physiology that extend beyond gross morphology (Sahu *et al.*, 2026a; Sahu *et al.*, 2026b) [10, 11]. Such developments reinforce the conceptual shift from purely anatomical diagnosis toward integrated structural–functional reproductive assessment. Importantly, the significance of ultrasonography in this study extends beyond diagnostic sensitivity. The observed improvement in farmer perception and trust following ultrasonographic demonstration highlights the role of visualization in extension science. In smallholder systems, where reproductive disorders are often interpreted through experiential heuristics, the ability to visually demonstrate fetal heartbeat, ovarian dynamics, or uterine pathology transforms abstract explanations into observable evidence. Behavioral adoption theory consistently emphasizes that visibility, perceived reliability, and experiential validation strongly influence technology acceptance (Neethirajan, 2021) [7]. Thus, ultrasonography functions not merely as a diagnostic tool but as an extension catalyst that strengthens veterinarian–farmer interaction and informed reproductive decision-making.

Early embryonic mortality and subclinical uterine pathology represent major hidden contributors to infertility in bovines (Sahu *et al.*, 2025; Andrade *et al.*, 2024) [9, 1]. Without imaging support, such conditions are frequently misclassified as repeat breeding, leading to repeated inseminations and inappropriate hormonal interventions. By enabling earlier and more accurate differentiation between embryonic loss, luteal insufficiency, and uterine pathology, ultrasonography supports targeted therapeutic strategies and rational reproductive management. Despite these advantages, several practical considerations merit attention. Ultrasonographic accuracy remains operator-dependent, requiring structured training and calibration to ensure reproducibility. Equipment cost, maintenance, and portability may also influence scalability in resource-limited settings. Furthermore, while visual confirmation enhances confidence, economic constraints may still limit treatment uptake, underscoring the need for integrated financial and advisory support mechanisms within extension programs. Collectively, the present findings align with the growing body of literature supporting ultrasonography as a

cornerstone of modern bovine reproductive management (Fricke, 2022) [3]. When deployed through structured field extension camps, portable ultrasonography bridges the gap between advanced reproductive diagnostics and rural livestock production systems. Its dual role as a diagnostic and educational instrument positions it as a scalable intervention capable of strengthening reproductive efficiency and, ultimately, income resilience in buffalo-based dairy enterprises.

Statements and Declarations

Ethics Approval and Consent to Participate

The clinical examinations performed in this study involved non-invasive reproductive evaluation procedures routinely conducted under veterinary field practice. No experimental interventions were performed beyond standard diagnostic procedures. All animal handling complied with accepted animal welfare guidelines for large-animal clinical examination. Verbal informed consent was obtained from livestock owners prior to examination and inclusion of their animals in the study.

Consent for Publication

All participating farmers provided consent for the use of anonymized clinical data for research and publication purposes.

Availability of Data and Materials

The datasets generated and analyzed during the current study are available from the corresponding author upon reasonable request.

Competing Interests

The authors declare that they have no competing financial or non-financial interests that could have influenced the work reported in this manuscript.

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