



Pregnancy Diagnosis in Cattle: The Way Forward

*Rameez Ali, Anees A Shah, Syed M Shah, Manzoor A Yattoo, Gowhar G Sheikh,
Arif A Pandit and Syed Ansar Ul Haq

Mountain Livestock Research Institute, Manasbal, Safapora, SKUAST-Kashmir, India

*Corresponding Author's email: dr.rameezali@gmail.com

Efficient reproductive management is fundamental to profitable cattle production, and pregnancy diagnosis remains one of its most critical components. Early and accurate identification of pregnant and non-pregnant animals facilitates timely reproductive interventions, reduces calving intervals, improves herd fertility and enhances economic returns. Traditional methods such as rectal palpation and transrectal ultrasonography continue to be widely employed; however, advances in reproductive biotechnology have expanded the range of available diagnostic tools. Pregnancy-associated glycoprotein assays, Doppler ultrasonography, molecular biomarkers and precision livestock farming technologies are increasingly being investigated and adopted for earlier and more reliable pregnancy detection. The integration of artificial intelligence, sensor-based monitoring system and omics technologies is expected to revolutionize reproductive management in cattle. This article discusses current pregnancy diagnosis methods, recent advances, existing challenges and future prospects for improving reproductive efficiency in cattle.

Keywords: cattle, pregnancy diagnosis, ultrasonography, pregnancy-associated glycoproteins, biomarkers, precision livestock farming, reproductive management

Introduction

Reproductive efficiency is a key determinant of profitability in both dairy and beef cattle enterprises. Failure to identify non-pregnant animals promptly results in extended calving intervals, increased insemination costs, reduced milk production efficiency and substantial economic losses. Consequently, accurate and timely pregnancy diagnosis is essential for effective reproductive management (Fricke, 2002). Conventional methods such as rectal palpation and ultrasonography have served as the cornerstone of bovine pregnancy diagnosis for decades. However, the increasing demand for earlier detection, improved accuracy and reduced labor dependency has stimulated the development of novel diagnostic approaches. Recent advances in reproductive physiology, molecular biology and precision livestock farming offer promising opportunities to transform pregnancy diagnosis from a periodic clinical procedure into a continuous monitoring process.

Conventional Pregnancy Diagnosis Methods

1. Rectal Palpation

Rectal palpation remains one of the most commonly used methods for pregnancy diagnosis in cattle because of its low cost, simplicity and practicality under field conditions. Pregnancy can generally be confirmed from approximately 35–45 days after insemination through detection of uterine asymmetry, amniotic vesicles, membrane slip, placentomes or fetal structures (Youngquist and Threlfall, 2007). Despite its widespread use, rectal palpation requires considerable expertise and carries a small risk of embryonic loss if performed improperly. Furthermore, the method provides limited information regarding embryonic viability during early gestation.

2. Transrectal Ultrasonography

The introduction of ultrasonography significantly improved the accuracy and scope of pregnancy diagnosis. Embryonic structures and heartbeat can be visualized as early as 26–30 days after conception, allowing confirmation of fetal viability and estimation of gestational age (Fricke, 2002). Ultrasonography also facilitates diagnosis of reproductive disorders, twin pregnancies, embryonic mortality and fetal abnormalities. Nevertheless, high equipment costs and the requirement for trained personnel limit its routine use in some production systems.

Biomarker-Based Pregnancy Diagnosis

1. Progesterone Assays

Progesterone measurement in milk or blood was among the earliest biochemical approaches to pregnancy diagnosis. Low progesterone concentrations approximately 21 days after insemination indicate luteolysis and non-pregnancy. However, elevated progesterone concentrations may occur in conditions such as persistent corpus luteum or pyometra, reducing diagnostic specificity (Sasser *et al.*, 1986).

2. Pregnancy-Associated Glycoproteins

Pregnancy-associated glycoproteins (PAGs) are placental proteins secreted by trophoblastic cells into maternal circulation. Their detection in serum, plasma, or milk has become an increasingly reliable method of pregnancy diagnosis. PAG concentrations can be measured from approximately 28 days after breeding and exhibit high sensitivity and specificity (Green *et al.*, 2005). Commercial PAG-based enzyme-linked immunosorbent assay (ELISA) kits are increasingly adopted due to their convenience, scalability, and suitability for large herd screening. In addition to pregnancy detection, PAG concentrations may provide information regarding fetal viability and risk of pregnancy loss (Pohler *et al.*, 2016).

Advanced Imaging Technologies

1. Doppler Ultrasonography

Color Doppler ultrasonography represents a significant advancement over conventional B-mode imaging. Assessment of blood flow to the corpus luteum and uterine tissues provides valuable information regarding luteal function and embryonic development. Research suggests that Doppler-based evaluation of luteal vascularization may enable earlier differentiation between pregnant and non-pregnant animals and facilitate prediction of embryonic survival (Pugliesi *et al.*, 2018).

2. Three-Dimensional Ultrasonography

Three-dimensional ultrasonography is emerging as a valuable research tool for evaluating embryonic development and placental structures. Although its routine field application remains limited, improvements in imaging technology may eventually enhance its practical utility.

Molecular Approaches and Omics Technologies

- 1. Interferon-Stimulated Genes:** Maternal recognition of pregnancy in cattle is mediated by interferon tau (IFN- τ), secreted by the conceptus during early gestation. IFN- τ stimulates the expression of interferon-stimulated genes (ISGs) in maternal blood leukocytes. Increased ISG expression has been proposed as an early marker of pregnancy before conventional diagnostic methods become effective (Han *et al.*, 2006).
- 2. MicroRNAs:** MicroRNAs (miRNAs) are small non-coding RNA molecules involved in gene regulation. Pregnancy-specific changes in circulating miRNA profiles have attracted considerable attention as potential non-invasive biomarkers for early pregnancy diagnosis. Several candidate miRNAs have demonstrated promising diagnostic accuracy; however, further validation is required before commercial application.
- 3. Metabolomics and Proteomics:** Advances in metabolomic and proteomic technologies have facilitated the identification of pregnancy-specific molecular signatures in biological fluids. These approaches may enable the discovery of novel biomarkers capable of detecting pregnancy during the earliest stages of embryonic development.

Precision Livestock Farming and Artificial Intelligence

Precision livestock farming (PLF) technologies are transforming cattle reproduction management. Automated monitoring systems continuously collect information related to animal activity, rumination, feed intake, body temperature, and physiological parameters. Artificial intelligence (AI) and machine learning algorithms can integrate these datasets with reproductive records to predict pregnancy status and reproductive outcomes. Such systems offer several advantages such as, continuous monitoring of individual animals, reduced dependence on manual examinations, early identification of reproductive failures, improved decision-making and herd management and enhanced labor efficiency. The integration of wearable sensors, cloud computing, and predictive analytics is expected to become increasingly important in modern cattle production systems.

Challenges and Limitations

Despite significant progress, several challenges continue to hinder widespread adoption of advanced pregnancy diagnostic technologies:

- High implementation costs.
- Limited accessibility in developing regions.
- Need for specialized training.
- Variability in biomarker expression among animals.
- Requirement for large-scale validation studies.
- Integration challenges between diagnostic platforms and farm management systems.

Addressing these constraints is essential for maximizing the practical benefits of emerging technologies.

Future Perspectives

The future of pregnancy diagnosis in cattle is likely to involve a multimodal approach combining imaging technologies, biomarker assays, and precision farming systems. Development of rapid cow-side diagnostic tests capable of detecting pregnancy within the first three weeks after insemination remains a major research objective. Furthermore, integration of genomic information, reproductive biomarkers, and AI-driven predictive models may facilitate individualized reproductive management strategies. Such advances could shift the focus from merely confirming pregnancy to predicting pregnancy success and preventing embryonic loss.

Conclusion

Pregnancy diagnosis in cattle is undergoing a significant transformation driven by advances in biotechnology, molecular diagnostics, and precision livestock farming. While rectal palpation and ultrasonography remain indispensable tools, emerging technologies such as PAG assays, Doppler ultrasonography, interferon-stimulated gene analysis and artificial intelligence-based monitoring systems offer unprecedented opportunities for earlier and more accurate pregnancy detection. Future developments will likely focus on integrating multiple diagnostic modalities to improve reproductive efficiency, animal welfare, and profitability in cattle production systems.

References

1. Fricke, P. M. (2002). Scanning the future—Ultrasonography as a reproductive management tool for dairy cattle. *Journal of Dairy Science*, 85(8), 1918–1926.
2. Green, J. A., Parks, T. E., Avalle, M. P., Telugu, B. P. V. L., McLain, A. L., Peterson, A. J., McMillan, W., Mathialagan, N., Hook, R. R., Xie, S., & Roberts, R. M. (2005). The establishment of an ELISA for the detection of pregnancy-associated glycoproteins in the serum of pregnant cows and heifers. *Theriogenology*, 63(5), 1481–1503.
3. Han, H., Austin, K. J., Rempel, L. A., & Hansen, T. R. (2006). Low blood ISG15 mRNA and progesterone levels are predictive of non-pregnancy in cattle. *Journal of Endocrinology*, 191(2), 505–512.

4. Pohler, K. G., Geary, T. W., Atkins, J. A., Perry, G. A., Jinks, E. M., & Smith, M. F. (2016). Pregnancy-associated glycoproteins as indicators of pregnancy status and fetal viability in cattle. *Animal Reproduction Science*, 174, 4–11.
5. Pugliesi, G., de Melo, G. D., Ataíde, G. A., Pellegrino, C. A. G., Silva, L. A., & Binelli, M. (2018). Use of Doppler ultrasonography in embryo transfer programs and pregnancy diagnosis in cattle. *Animal Reproduction*, 15(Suppl. 1), 875–886.
6. Sasser, R. G., Ruder, C. A., Ivani, K. A., Butler, J. E., & Hamilton, W. C. (1986). Detection of pregnancy by radioimmunoassay of a novel pregnancy-specific protein in serum of cows and a profile of serum concentrations during gestation. *Biology of Reproduction*, 35(4), 936–942.
7. Youngquist, R. S., & Threlfall, W. R. (2007). *Current Therapy in Large Animal Theriogenology* (2nd ed.). Saunders Elsevier.