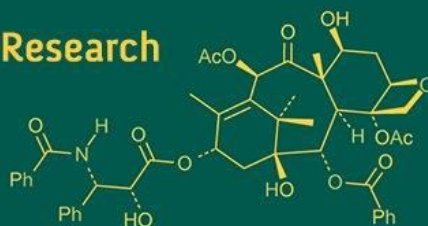


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Determinants of pregnancy success in bovine embryo transfer

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Abstract

Embryo transfer (ET) is a widely used reproductive technology in cattle breeding; however, its efficiency is constrained by inconsistent conception rates and high costs associated with recipient management. Achieving successful pregnancy at the first transfer is therefore critical for improving both biological and economic outcomes. Conception success in ET programs is influenced by multiple interrelated factors, including recipient physiology, corpus luteum function, embryo characteristics, technical proficiency during transfer, and environmental conditions. Recipient-related factors such as body condition score, parity, and health status play a crucial role in determining uterine receptivity, while optimal luteal function ensures an adequate progesterone environment necessary for embryo survival. Embryo origin and handling conditions further influence developmental competence with *in vivo* derived and fresh embryos generally yielding higher pregnancy rates. In addition, technical precision during embryo transfer and minimization of uterine stress are essential to prevent luteolysis and embryo loss. Environmental stressors, particularly heat stress, further compromise reproductive efficiency by altering endocrine and follicular dynamics. Despite advancements in ET technology, challenges related to management, infrastructure, and standardization continue to limit consistent outcomes. Emerging approaches integrating phenomics and transcriptomics with practical herd management strategies offer promising opportunities to enhance prediction of pregnancy success and improve the overall efficiency of ET programs.

Keywords: Embryo transfer, conception, recipient, corpus luteum, embryo quality, heat stress

1. Introduction

Embryo transfer (ET) is an important reproductive technology in cattle breeding, but its efficiency is limited by inconsistent conception rates. The major economic constraint in ET programs is the cost associated with recipient management, making successful conception at the first transfer highly critical (Looney *et al.*, 2006) [13]. Pregnancy success is influenced by multiple interrelated factors, including recipient physiology, luteal function, embryo quality, technical precision, and environmental conditions (Schmidt, 2010) [22]. Therefore, understanding these determinants is essential to improve reproductive efficiency and overall program success.

Variability in these factors often leads to suboptimal uterine receptivity, impaired embryo-maternal signaling, and increased embryonic loss, thereby reducing overall program efficiency (Hansen, 2020) [6]. In particular, suboptimal progesterone exposure, and compromised embryo handling can disrupt early developmental events critical for implantation (Jaśkowski *et al.*, 2021; Estrada-Cortés *et al.*, 2019) [8, 4]. Furthermore, technical inconsistencies during embryo transfer may induce uterine stress responses that negatively affect luteal stability and embryo survival (Upreti *et al.*, 2025) [27]. Environmental stressors, especially heat stress, further exacerbate these challenges by altering endocrine function and follicular dynamics (Puicón *et al.*, 2018) [18]. Collectively, these constraints highlight the need for a precise and integrated understanding of the biological and management factors governing ET success.

2. Key Drivers of Conception Success in Embryo Transfer

The major economic cost in embryo transfer (ET) programs is associated with the procurement and upkeep of recipient animals (Looney *et al.*, 2006) ^[13]. Consequently, efficient recipient herd management depends on a thorough understanding of physiological status, nutritional management, estrous synchronization, disease control, and overall herd planning (Schmidt, 2010) ^[22]. Achieving conception at the first transfer is significantly more economical than performing repeated transfers and is more efficient than replacing recipients that fail to conceive (Looney *et al.*, 2006) ^[13]. Pregnancy success is influenced by multiple factors that act in a coordinated manner to affect implantation and the maintenance of pregnancy (Fig. 1).

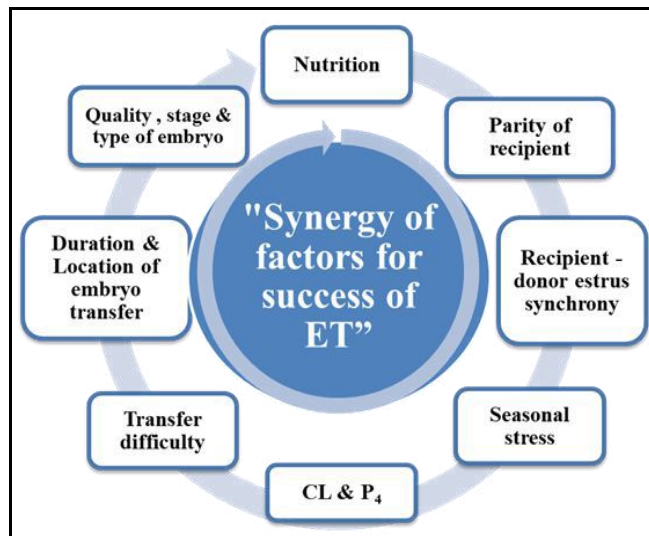


Fig 1: Comprehensive and synergistic management practices for successful embryo transfer.

2.1 Recipient Physiology and Management

The physiological status of the recipient is a cornerstone of ET success. Key factors include body condition score (BCS), parity, lactation status, temperament, and freedom from periparturient or postpartum disease (Hansen, 2020) ^[6]. Body condition is one of the most consistent predictors of fertility, as both low and excessive BCS impair reproductive efficiency, whereas moderate scores are associated with improved conception rates (Lalman *et al.*, 1997; Whitman, 1975) ^[11, 28]. The postpartum period is particularly critical, requiring animals to balance lactation demands, regain body reserves, and restore reproductive function within an optimal timeframe (Lamb, n.d.).

Parity also significantly influences outcomes. Heifers are generally more suitable recipients than multiparous cows due to the absence of postpartum uterine disease and metabolic stress. Reported pregnancy rates in heifers (47.05%) exceed those in multiparous cows (34.61%), indicating a substantially higher likelihood of conception in younger animals (Sahu, 2022) ^[20].

2.2 Corpus Luteum Function and Hormonal Environment

The corpus luteum (CL) is the key determinant of uterine receptivity in ET recipients, with its size, morphology, and secretory capacity defining the progesterone environment required for embryo survival. Both homogeneous and cavitary CLs are observed in cattle, and evidence indicates

that cavitary CLs do not impair luteal function. In fact, higher pregnancy rates have been reported in recipients with cavitary CLs (46.15%) compared to homogeneous CLs (32.55%), along with higher progesterone concentrations (Jaśkowski *et al.*, 2021; Sahu, 2022) ^[8, 20].

Progesterone concentrations ranging from 0.58 to 16.0 ng per mL have been associated with comparable pregnancy outcomes, suggesting that luteal functionality and timing of progesterone exposure are more critical than absolute circulating levels (Spell *et al.*, 2001) ^[25]. Inadequate uterine preparation during early stages remains a major cause of embryonic loss. Hormonal interventions such as GnRH administration can enhance follicular maturation, increase LH secretion, and improve ovulation and conception rates (Sahu, 2022) ^[20].

2.3 Embryo Characteristics and Handling

Embryo characteristics significantly influence conception rates. *in vivo*-derived embryos consistently outperform *in vitro*-produced (IVP) embryos due to reduced exposure to suboptimal culture conditions (Jones & Lamb, 2008) ^[9]. Fresh embryos also yield higher pregnancy rates than frozen-thawed embryos, although cryopreservation provides logistical advantages (Estrada-Cortés *et al.*, 2019) ^[4].

The frequency of embryo transfer attempts further affects outcomes. Pregnancy rates remain relatively stable between the first and second attempts but decline sharply with subsequent transfers, falling below 20% by the third attempt (Looney *et al.*, 2006) ^[13]. These findings emphasize the importance of careful recipient selection and turnover in ET programs. Collectively, these observations indicate that embryo competence is influenced not only by intrinsic quality but also by production and handling conditions.

2.4 Technical Skill and Embryo Transfer Procedure

Technical proficiency during embryo transfer is a critical determinant of success. Proper cervical passage, minimal uterine trauma, and accurate embryo deposition are essential (Sreenan & Diskin, 1987) ^[26]. Excessive manipulation during nonsurgical transfer can increase prostaglandin F_{2α} release, leading to luteolysis and embryo loss (Wann & Randel, 1990) ^[29]. The use of flunixinmeoglumine to inhibit prostaglandin synthesis has been shown to improve pregnancy outcomes (Kasimanickam *et al.*, 2019) ^[10].

Embryo deposition site also influences success rates. While placement in the middle third of the uterine horn is traditionally recommended, higher pregnancy rates have been reported with deeper deposition toward the cranial region (Alkan *et al.*, 2020) ^[1]. However, excessive manipulation associated with deeper placement may counteract this benefit, emphasizing that atraumatic and precise deposition is more critical than location alone (Santos, 2019) ^[21].

2.5 Environmental and Seasonal Factors

Environmental stressors significantly affect reproductive performance. Pregnancy rates are typically highest during cooler seasons and decline during periods of heat stress. Elevated temperatures impair embryonic interferon production, disrupt maternal recognition of pregnancy, and alter endocrine function (Puicón *et al.*, 2018) ^[18]. Heat stress also reduces feed intake and compromises follicular dynamics, ultimately impairing fertility. Mitigation strategies such as cooling systems, shade provision, and

optimized timing of ET procedures can improve outcomes. Additionally, comprehensive herd health programs, including vaccination and parasite control, are essential to maintain recipient readiness and reproductive efficiency (Looney *et al.*, 2006; Schmidt, 2010) ^[13, 22].

3. Challenges in ET Programs and Practical Solutions

The widespread application of ET, particularly in developing regions, is constrained by structural, economic, and technical challenges. Limited access to resources, lack of organized livestock databases, and variability in indigenous breeds hinder the standardization of protocols. These limitations are compounded by high costs, inadequate biosafety measures, and insufficient policy support (Seidel & Seidel, 1992) ^[23]. A major constraint in ET programs remains inconsistency in recipient selection and management, which often has a greater impact on success rates than technological limitations. Additional technical factors including embryo quality, synchronization accuracy, and progesterone status, further influence pregnancy outcomes (Montiel *et al.*, 2006) ^[13].

Even minor procedural aspects, such as performing ET while the recipient is in motion or prolonged manipulation of the transfer device, can reduce pregnancy success (Rowe *et al.*, 1980) ^[19]. Strategies such as optimizing fixed-time embryo transfer protocols and minimizing handling stress have been shown to improve efficiency (Baruselli *et al.*, 2010) ^[2]. Emerging approaches such as immune modulation, including intrauterine administration of peripheral blood mononuclear cells, have demonstrated potential to enhance pregnancy rates, highlighting the importance of immune-uterine interactions (Ideta *et al.*, 2010) ^[7]. Addressing these challenges requires an integrated approach involving veterinarians, producers, and researchers to ensure both biological and economic sustainability.

4. Future Directions: Omics and Precision Reproduction

The future of ET lies in improving embryo competence and maternal receptivity through advanced biotechnologies. While MOET remains relevant, increasing emphasis is placed on IVP systems due to their expanding global use and potential for optimization (Ealy *et al.*, 2019) ^[9]. Advances such as stem cell-derived gametes may further transform embryo production systems (Goszczynski *et al.*, 2019) ^[5].

Phenomics offers a promising approach by enabling systematic large-scale phenotypic data collection to improve prediction of pregnancy outcomes (Koltes *et al.*, 2019) ^[14]. At the molecular level, transcriptomic studies have identified differential gene expression patterns associated with uterine receptivity, particularly during critical stages of the estrous cycle (Hansen, 2020; Moran *et al.*, 2017) ^[6, 17]. For instance, genes involved in extracellular matrix remodeling, immune regulation, and growth factor signaling have been shown to differ between pregnant and non-pregnant recipients (Mazzoni *et al.*, 2020) ^[15]. The integration of phenomics, transcriptomics, and other omics technologies with practical herd management strategies has the potential to establish a precision reproduction framework. Such an approach could transform ET from an empirical practice into a predictive and highly reliable reproductive technology central to modern cattle breeding systems (Hansen, 2020) ^[6].

5. Conclusion

Conception success in embryo transfer programs is determined by a complex interplay of recipient-related, embryonic, technical, and environmental factors that collectively influence uterine receptivity and embryo survival. Among these, recipient physiology and corpus luteum function play central roles in establishing an optimal hormonal milieu, while embryo characteristics and handling conditions directly affect developmental competence. In addition, technical proficiency during transfer and minimization of procedural stress are critical to prevent luteal disruption and enhance implantation success. Despite existing challenges in management and standardization, integration of advanced approaches such as phenomics and transcriptomics with practical herd strategies holds promise for enhancing the efficiency and reliability of ET programs.

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