Animal Reproduction Science 212 (2020) 106251

Contents lists available at ScienceDirect

Animal Reproduction Science

journal homepage: www.elsevier.com/locate/anireprosci

Pregnancy loss in beef cattle: A meta-analysis

S.T. Reese, G.A. Franco, R.K. Poole, R. Hood, L. Fernadez Montero, R.V. Oliveira Filho, R.F. Cooke, K.G. Pohler*

Department of Animal Science, Texas A&M University, College Station, TX, United States

A R T I C L E I N F O

Keywords:
Pregnancy
Beef cattle
Embryo mortality

A B S T R A C T

Pregnancy loss in beef cattle causes both management and economic challenges to a producer. A meta-analysis was conducted to quantify reproductive failures that occur during fertilization, early embryonic development, and late embryonic/early fetal development periods of gestation in beef cattle. The meta-analysis included more than 56,000 diagnostic records in 159 studies from 48 papers with 12 studies included in fertilization and pre- blastocyst loss analysis (FERT; days 1–7 of gestation), 107 in early embryo (EEM; days 7–32), and 40 in late embryo/early fetal period (LEF; days 32–100) analysis. Although fertilization rates are reportedly high in beef cattle, significant developmental failure occurs within the first 7 days of gestation. Approximately 28.4 % of embryos will not develop past day 7 of gestation with most embryonic losses occurring before day 4. By the conclusion of the first month of gestation, 47.9 % of cows submitted to a single insemination at day 0 will not be pregnant. Overall, LEF between days 32–60 and 100 was 5.8 %. *Bos indicus* animals had greater ($P = 0.001$) EEM compared to *Bos taurus,* but there was no difference ($P = 0.39$) for the LEF period between subspecies. Primiparous cows had greater EEM ($P = 0.002$) compared to nulliparous heifers and multiparous cows; and nulliparous heifers had a greater LEF compared to primiparous and multiparous cows ($P = 0.048$). Collectively, these cumulative findings provide a baseline assessment of pregnancy loss specific to beef cattle.

1. Introduction

A main principle for most profitable cowherd models is to maximize the number of cows that produce a marketable calf yearly; however, calf crop percentage often fall below the level of expectation due to reproductive failures. Many cow calf operations are less intensively managed than dairy herds resulting in minimal awareness of reproductive failure within a herd. Understanding the timing of reproductive failure such as pregnancy loss can assist scientists and producers in making important management decisions; however, results from conducting studies aimed at quantifying pregnancy loss during specific periods of gestation in beef cattle have been somewhat inconsistent. It is generally accepted that fertilization rates in beef cattle are considerably greater than pregnancy rates due to embryonic mortality occurring within the first 30 days of gestation which accounts for the largest percentage of pregnancy loss. The amount of embryonic loss reported after day 30 until the early fetal period, however, is variable (Diskin and Morris, 2008; Diskin et al., 2016). Causes of embryonic and fetal mortality are wide ranging from genetic lethal mutations and uterine asynchrony to failure in maternal recognition of pregnancy, placental insufficiency and disease (Pope, 1988; Farin et al., 2006; Diskin and Morris, 2008; Cheng et al., 2016; Pohler et al., 2016b; Abdalla et al., 2017).

Within beef cattle production, type of cattle and management strategies can significantly affect the extent of reproductive failures.

* Corresponding author at: 2471 TAMU, College Station, TX, United States.
E-mail address: kpohler@tamu.edu (K.G. Pohler).

https://doi.org/10.1016/j.anireprosci.2019.106251
Received 2 September 2019; Received in revised form 25 October 2019; Accepted 22 November 2019
Available online 23 November 2019
0378-4320/

Ayalon (1978) provided one of the earliest reviews of embryonic loss in cattle which is still commonly cited in recent publications. During the last 40 years, there have been few publications in which there has been a specific review or in which there has been a summary of pregnancy loss throughout gestation in beef cattle. Furthermore, there has been no systematic review or meta-analysis of pregnancy loss in beef cattle. This gap in knowledge has a fundamental impact in measuring reproductive success and obtaining an accurate estimate of when there are reproductive failures during the various reproductive processes that result in production of calves. The primary objective of this meta-analysis is to conduct a review of studies and data to predict accurate values for reproductive failures during multiple periods of gestation including fertilization, early embryonic, late embryonic/early fetal development in beef cattle using quantitative analyses procedures. Secondarily, there was use of moderator analyses procedures of subspecies and parity to evaluate the effect of these characteristics on reproductive failures during critical periods of gestation in beef cattle. While many factors, including disease, environmental condition and management strategy, can increase or decrease reproductive success, the aim with this meta-analysis is to identify an updated baseline value for critical periods of loss throughout gestation in beef cattle.

2. Materials and methods

2.1. Data collection

Relevant literature was identified through comprehensive searches of Web of Science, PubMed, Google Scholar, pertinent scientific journals and meeting proceedings. In addition to articles accessed as a result of original searches, reference lists from these articles were used to identify additional articles in which there was relevant research reported. Search terms included “pregnancy loss”, “embryo mortality”, “embryo loss”, “fertilization”, “conception rate”, “pregnancy rate”, “early embryo”, “late embryo”, “beef cattle”, “beef cow”, and “beef heifer.” More than 1000 articles were identified and were further examined to determine suitability for inclusion utilizing PRISMA guidelines for systematic reviews. Primary screening of every article was undertaken by S.T. Reese with secondary reviews by G.A. Franco and K.G. Pohler. Each reviewer recommended or excluded articles based on a series of criteria to avoid bias. Primary screening was based on title and abstract information to establish whether in the article there was reporting on original research, determination of pregnancy rates in beef cattle and in the study(ies) conducted that there were not treatments that were intended to be detrimental to pregnancy. Articles meeting these criteria were further evaluated for data extraction and, subsequently, appraisal by G.A. Franco and K.G. Pohler. Mandatory inclusion criteria included i) cows or heifers of beef breeds ii) published after adoption of ultrasonic technology for early pregnancy diagnosis to allow for accurate pregnancy determination between days 28 and 32 of gestation and iii) day of gestation of pregnancy diagnosis, subspecies, location, parity, and/or breeding method was listed. Studies with first pregnancy diagnosis after day 32 of pregnancy or that included dairy animals and trials with treatments that could bias pregnancy success, such as induced twinning, were excluded from the meta-analysis. Articles were sourced from countries with modern beef production systems, including North America, Europe, Brazil and Australia. In papers where there was reporting on results from multiple treatment and/or control groups, and/or where there were detrimental losses as a result of treatment (induced disease states, severe nutrient restriction, etc.), there was exclusion of these data from the average analysis.

Each study was assigned a pregnancy loss time period (fertilization, early embryo, late embryo/early fetal) based on when pregnancy diagnoses occurred. Unfortunately, the physiological periods of pregnancy development do not coincide with common time points of pregnancy diagnosis in herd management protocols. Time of pregnancy diagnosis in many studies does not correspond to a single development period, therefore, some periods of this meta-analysis were extended beyond the usual physiological developmental period to include a greater number of studies (Fig. 1). Results from studies in which the pregnancy rate or embryo recovery and survival was determined before day 7 of gestation (approximately a blastocyst stage embryo) were included in the

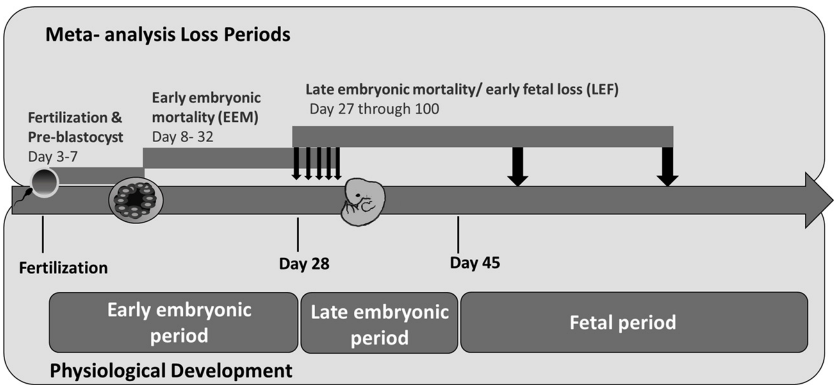

Fig. 1. Loss periods and physiological development of pregnancy. Divisions between periods used to classify studies in the meta-analysis did not align with physiological development periods. Effort was made to best utilize the most possible trials within a logical distance from the true periods. The black arrows represent the most common times for pregnancy diagnosis in beef cattle: an initial diagnosis between days 27 and 32 of gestation, and a second diagnosis around day 60 or day 100 of gestation.

initial period subsequently referred to in this manuscript as the period of fertilization and pre-blastocyst loss (FERT; days 1–7 of gestation). This allowed for results of a more substantial number of studies to be included in the meta-analysis because actual fertilization data are difficult to collect in *in vivo* studies. Pregnancy status was most commonly diagnosed before day 7 by flushing of the uterus after uterine tissues were collected. With many studies there was reporting of individual stages of embryo development but with this particular meta-analysis there was utilization of data from the most advanced stage of embryo present on the day of collection (i.e., cleaved on day 4; blastocyst on day 7 of gestation). Studies in which there was flushing of the uterus strictly for evaluation of embryo transfer factors were not included due to large variability and discrepancies on how data were reported and the potential basis for the techniques used. Data collected from days 27–32 of gestation were classified as being collected during a period when there is early embryonic mortality (EEM) for this meta-analysis, although the physiological period of early embryonic development is considered to have concluded by day 28 of gestation. Transrectal ultrasound was the primary method of pregnancy diagnosis; however, reproductive tract collections and pregnancy associated glycoprotein blood testing were utilized in some studies. Importantly, collected EEM data will be confounded by loss that occurs during FERT because the data related to losses cannot be separated in the studies reported in the original publications and are cumulative as the pregnancy progresses. While embryo developmental stage shifts to stage of fetal development between days 42 and 45 of gestation, there is most commonly reporting in these studies of a secondary pregnancy diagnosis between days 60 and 100 of gestation evaluated using transrectal ultrasonography. Data from all studies in which there was diagnosis of pregnancy between days 60–100 are combined to assess reproductive failures occurring from days 60–100 of gestation which is termed late embryo/early fetal loss (LEF) for purposes of this analysis. This meta-analysis included more than 56,000 diagnostic records in 159 studies reported in 48 papers with 12 FERT studies, 107 EEM studies, and 40 LEF studies. Classification of studies is reported in Table 1.

2.2. Effect size and moderator variables

A meta-analysis of reproductive failures during various reproductive processes was conducted to determine percentage of pregnancy losses and periods when there were significant reproductive failures. Although meta-analyses are generally conducted to examine a relationship between two groups or treatments, pregnancy loss was the single group effect size for this analysis. Effect sizes were calculated from data provided within the publications as percent pregnancy loss during each developmental period. Fertilization and EEM classification effect sizes are reported as percent of cows diagnosed as being non-pregnant when there were uterine flushing or ultrasonic diagnosis and percentage determinations of reproductive failures relative to total cows inseminated. In studies where both conception rate based on ovulation or estrus expression and pregnancy rate based on total cows inseminated were reported, pregnancy rate was utilized for effect size calculations to maintain consistency across all studies. Effect sizes for LEF are reported as a percent of cows that were diagnosed pregnant between days 28 and 32 but not pregnant at a secondary diagnosis, not as a percent of total cows inseminated.

Variables that may have contributed to variation among pregnancy losses were collected to be used as moderators. Described as third variables, moderators are variables which may have an effect on the extent or direction of change in the dependent variable and is generally a subset of the independent variable (Baron and Kenny, 1986). Moderators that were subjected to analysis included country of study, subspecies, parity and breeding method. Other moderator variables collected, if available, included state/region, service sire, synchronization protocol, body condition score and objective of original paper. Availability of all moderators did not affect eligibility for inclusion in the analysis; however, all papers did include descriptions of parity, subspecies (or breed) and country of study. Acquisition of these data allowed for an adequate number of studies to be included in each group for moderator analysis of parity and subspecies. Country of study was closely aligned with subspecies moderator analysis; therefore, it was not reported separately. In all studies, there was utilization of only cows with adequate body condition scores.

2.3. Meta-analysis

When conducting the meta-analysis, the methodology established by Borenstein et al. (2009) was utilized. Summary effects and associated statistics were computed using Comprehensive Meta-Analysis Version 3 (CMA) software (Biostat, Englewood, NJ, USA; 2014). Due to the high probability that true effects vary among studies, the random-effects model was used. A nonparametric variance was calculated using the following to weight studies within the meta-analysis as standard errors and standard deviations were not reported in a majority of papers

$$V = \frac{P \times (P - 1)}{n} \times m^{0.5}$$

where V is the variance, P is the point estimate, n is the sample size for the specific period and m is the number of studies extracted from the individual paper. For some papers, there were results from numerous studies from a single cow herd reported; the m correction was used to decrease weight that may be given when there were multiple studies with one herd so as to decrease the bias. Heterogeneity was calculated to evaluate the variation of random true effects that exist in pregnancy loss populations across multiple studies. Heterogeneity was assessed using the Q test for which the formula is subsequently described. This is a chi-square statistic that can be used to evaluate total weighted variability by accounting for both true heterogeneity (variation among studies) and expected sampling error (within study variation). The formula for this determination is as follows.

$$Q_t = Q_b + Q_w$$

Table 1
Period and moderator classification of studies.

Reference	Country	Loss Period[1]	Subspecies[2]	Parity[3]	Breeding Method[4]	No. of Animals
Aono et al., 2013	Brazil	E, L	I	P, M	FTAI	12,357
Beal et al., 1992	USA	L	T	M	AI	205
Breuel et al., 1993	USA	F	T	M	N, AI	50
Burns et al., 2008	USA	E, L	T	P, M	FTAI	676
Carter et al., 2008	Ireland	F, E	T	N	AI	125
Colazo et al., 2004	Canada	E	T	N, M	FTAI	363
Cooke et al., 2017	Brazil	E, L	I	M	FTAI,	1,209
Cordeiro et al., 2015	Brazil	E	I	N, M	FTAI, ET	350
Diskin and Sreenan, 1980	Ireland	F, E	T	N	AI	145
Dobbins et al., 2009	USA	E, L	T	P, M	FTAI	605
Dunne et al., 2000	USA	E	T	N	AI	158
Ferreira et al., 2016	Brazil	E, L	I	M	FTAI	604
Franco et al., 2018	Brazil	E, L	I	M	FTAI	1,228
Garrett et al., 1988	USA	F, E	T	M	N	31
Jinks et al., 2013	USA	E, L	T	M	ET	350
Kill et al., 2013	USA	E, L	T	N	FTAI	679
Lamb et al., 2001	USA	E	T	P, M	AI, FTAI	365
Lamb et al., 2006	USA	E	T	N	AI, FTAI	1,019
Larson et al., 2006	USA	E, L	T	P, M	AI, FTAI	2,417
Lopes et al., 2009	Brazil	E	I	P, M	FTAI, ET	2,667
Martinez et al., 2002a	Canada	E	T	N	FTAI	503
Martinez et al., 2002b	Canada	E	T	N, M	FTAI	622
Meneghetti et al., 2009	Brazil	E	I	P, M	FTAI	3,260
Mercadante et al., 2015	USA	E	T, X	N, M	FTAI	2,370
Mialon et al., 1993	France	L	T	N	AI	1,102
O'hara et al., 2014	Ireland	E	T	N	AI	33
Parr et al., 2017	Ireland	E	T	N	AI	83
Peres et al., 2009	Brazil	E	I	N, M	FTAI	1,855
Perry et al., 2003	USA	E, L	T	P, M	FTAI	174
Perry et al., 2007	USA	E, L	T	N	AI, FTAI	208
Pessoa et al., 2012	Brazil	E, L	I	N, P, M	FTAI	658
Pfeifer et al., 2017	Brazil	E	I	P, M	FTAI	253
Pohler et al., 2013	USA	E, L	T	M	FTAI, ET	354
Pohler et al. (2016a, 2016b)	Brazil	E, L	I	P, M	FTAI	2,205
Pontes et al., 2009	Brazil	E, L	I	N	ET	1,199
Pontes et al., 2011	Brazil	E, L	X	N	ET	5,938
Pradebon et al., 2017	Brazil	E	T	N	FTAI	414
Radigonda et al., 2017	Brazil	E	X	M	FTAI	150
Roche et al., 1981	England	F	T	N	AI	131
Sá Filho et al., 2010	Brazil	E	I, X	M	AI, FTAI	2,388
Sá Filho et al. (2009)	Brazil	E	I	M	FTAI	2,491
Sa Filho et al., 2014	Brazil	E, L	I	P, M	FTAI	1,538
Sales et al., 2011	Brazil	E, L	X	N	ET	495
Smith et al., 1982	USA	F, E	I	N	AI	101
Spitzer et al., 1978	USA	F	T	N	AI	30
Starbuck et al., 2006	USA	E, L	T	M	AI, FTAI	267
Stevenson et al., 2003	USA	E, L	T	M	FTAI	1,048
Unpublished Pohler Lab	USA	E, L	T	P, M	FTAI	229

[1] F = Fertilization (diagnosed before day 7 of gestation), E = early embryo mortality (loss prior to day 32 of gestation), L = late embryo/ early fetal mortality (pregnancy loss between initial pregnancy diagnosis at days 28–32 of gestation and second pregnancy diagnosis between day 60 and 100).
[2] Subspecies evaluated: T = *Bos taurus*, I = *Bos indicus,* X = cross breed of *Bos taurus* x *Bos indicus*.
[3] N = nulliparous, P = primiparous, M = multiparous.
[4] AI = artificial insemination based on estrus expression, FTAI = fixed time artificial insemination based on protocol specifications, N = natural service, ET = embryo transfer 7 days post predicted ovulation.

Heterogeneity was quantified using the formula for calculation of I^2 as an index that provides the proportion of variation due to true effects if sampling error was removed:

$$I^2 = \frac{Q_t - df}{Q_t} \times 100$$

where *df* (degrees of freedom; number of trials – 1 for each period of loss) represents expected variation (Q_w) and $Q_t - df$ represents the excess variation (Q_b). Lesser I^2 values close to 0 % indicate most variation is due to sampling error or no heterogeneity; whereas, I^2 values closer to 100 % denote variation in true effect sizes and indicate there is heterogeneity with the data (Higgins et al., 2003). For heterogeneity analysis, the prediction intervals (PI) were reported. Prediction intervals are dispersion indexes based on standard deviation that indicates how the effect sizes vary among all populations (95 % confidence that an individual study will fit), whereas, confidence intervals (CI) are more specific as it relies on the standard error and is dependent on the number of studies (essentially there is 95 % confidence that the mean will fall in this range) (Borenstein et al., 2017). Heterogeneity *P* values are reported among moderator subgroups and denote the probability that all groups share a common effect size.

Although this meta-analysis was conducted to examine a single effect size rather than a treatment effect, publication bias analysis was conducted to ensure balance between the results of large and small studies for each of the periods when reproductive failures were assessed. Two separate tests were used to detect potential bias. Funnel plot analysis can be used to provide a visual assessment to determine whether sample size affects the distribution of data around the mean (Borenstein et al., 2009). A symmetrical funnel plot can be used to indicate large and small studies are equally represented on either side of the mean. Secondly, Duval and Tweedie's (2000) trim and fill test can be used to adjust the effect size by removing data from small studies with extreme effect sizes and imposing studies to make the funnel plot symmetrical on both sides of the found effect size (Duval and Tweedie, 2000). Once the potentially missing studies are filled the possibility of exaggerated effect size can be assessed.

3. Results

3.1. Fertilization and pre-blastocyst failures

Due to the difficulty and cost associated with conducting fertility studies, a limited number of studies ($n = 12$) that examined pregnancy loss during the earliest periods of gestation were identified. Studies that determined outcomes through day 7 of gestation (approximately blastocyst developmental stage) were included in FERT analysis. It is recognized this does not accurately represent the actual percentage of zygote production but includes all loss during the initial stages of embryo development and cell division. Across 12 studies, the average pregnancy loss was 28.4 % (CI, 19.4 %–37.4 %) by day 7 after fertilization. Interestingly, in studies with data collected before day 4 ($n = 6$), reproductive failures were 23 %, indicating that most losses during this time period are due to fertilization or initial cell division failures. Heterogeneity was low ($I^2 = 18.5$ %). The prediction interval indicated that 95 % of pregnancy failures by day 7 of gestation will be in the range of 9.3%–47.5%. For the limited number of studies of fertilization analysis, publication bias did not affect the analysis based on funnel plot and trim and fill analysis publication bias tests. There were inadequate numbers of *Bos indicus* studies to provide a subspecies comparison and all but one study was conducted using heifers, thus, there was not moderator analysis for the FERT period.

3.2. Early embryo loss

In most studies the end of the early embryonic period was defined as ending on day 28 of gestation from a physiological perspective, therefore, the initial pregnancy diagnosis in beef cattle usually occurs after this timepoint, around day 30–32 of gestation. To utilize data from the maximum number of studies possible, EEM analysis included studies of data collected using pregnancy diagnosis occurring between days 27 and 32 of gestation ($n = 107$). Pregnancy loss during the EEM period was 47.9 % (CI, 45.8 %–50.0 %) for more than 53,000 individual cows. Additionally, 11 separate studies were identified in which there was diagnosis of pregnancy between days 12 and 16 of gestation using data collected at the time of detection of an embryo following collection of uterine tissues (slaughter) and reported a pregnancy loss point estimate of 32.3 % (CI, 24.9 %–37.8 %). The 47.9 % reproductive failure rate that occurs during the first month of gestation in beef cattle as detected using the meta-analysis can be refined: 28.4 % by day 7 of gestation, 3.9 % between days 7 and 16, and 15.6 % between days 16 and 32.

Reproductive failures during the EEM period was highly variable and moderator factors were more easily evaluated than fertilization data (Fig. 2). Moderator analysis of subspecies indicated a point estimate of 50.4 % reproductive failure during the first month of gestation for cattle of *Bos indicus* breeds while *Bos taurus* counterparts had a lesser reproductive failure (44 %; $P = 0.001$). Fewer data were available for crossbred cattle with both *Bos indicus* and *Bos taurus* genetic influence ($n = 9$) and data were highly variable (52.3 %, CI, 44.1 %–60.4 %). Parity also affected early embryonic mortality ($P = 0.002$). For parity moderator analysis, average early embryonic mortality for nulliparous heifers ($n = 39$) was 44.3 %, and for primiparous cows ($n = 17$) was 54.7 % and multiparous cows ($n = 49$) was 48.0 %. Breeding method affected early embryonic mortality ($P = 0.001$), with pregnancy loss in cows bred using AI after natural estrous expression being 32.2 % ($n = 10$), fixed time AI (FTAI) 49.5 % ($n = 83$) and embryo transfer (ET) 54.6 % ($n = 13$).

Heterogeneity of the EMM data set and by moderator sub level analysis was low as indicated by overall I^2 value equaling 13.1 %. Based on PI calculations, about 95 % of populations will have an overall EEM effect size in the range of 40.9%–54.9%. There was no indication of publication bias contributing to the effect size of early embryo loss.

3.3. Late embryo and early fetal losses

The late embryonic period has been defined as day 29 to approximately day 45 of gestation (Hubbert et al., 1972). Due to limited number of trials in which there was diagnosis of pregnancy at day 45, day 60 was considered as the last day of the late embryonic period. Additionally, there were a significant number of studies in which pregnancy diagnosis was conducted at day 30 and again around day 100 of gestation. Other than when there are infectious causes, there is little late fetal mortality in beef cattle and data for losses after day 100 were not included in the meta-analysis. After including data from studies in which there was a final pregnancy diagnosis between days 60 and 100, there was identification of 40 studies including 30,500 individual animals that were classified as

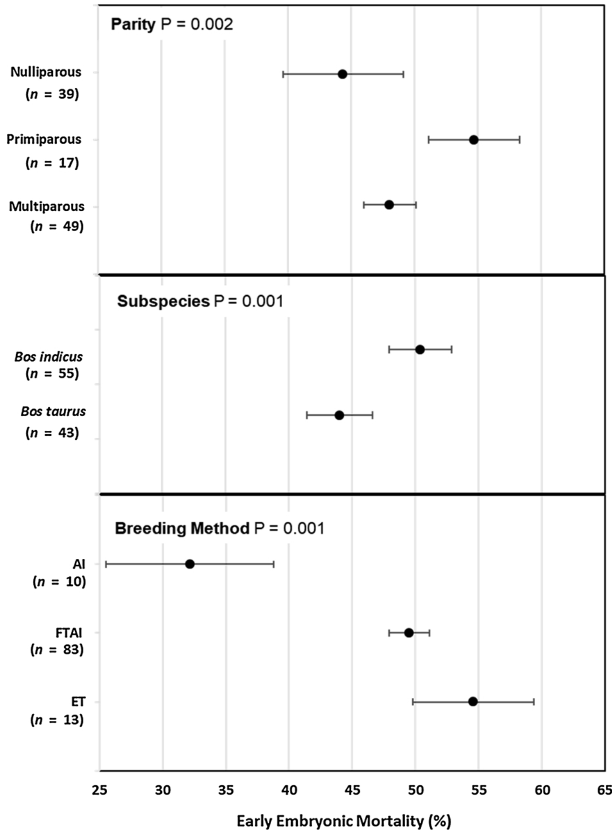

Fig. 2. Point estimates and 95 % CI for moderators explaining early embryonic mortality during the first month of gestation. n = number of trials ; heterogeneity P denotes the probability all trials share a common point estimate. Some publications that utilized multiple moderators in a single trial and could not be separated were excluded from moderator analysis.

LEF. Reproductive failures during the LEF period averaged 5.8 % (CI, 4.8 %–6.9 %). There was no subspecies did not effect on the frequency of pregnancy loss during this period (*Bos indicus* 5.0 % and *Bos taurus* 5.9 %, $P =$ 0.389, Fig. 3). Moderator analysis of parity indicated there were differences ($P =$ 0.048) between nulliparous heifers (n = 10; 8.1 %), primiparous cows (n = 4; 5.4 %), and multiparous cows (n = 14; 5.1 %) (Fig. 3). When there were pregnancies resulting from ET, there was a greater ($P =$ 0.001) LEF (n = 7; 10.2 %) compared with pregnancies resulting from FTAI (n = 26; 4.9 %). Consistent with other periods, results from heterogeneity analysis indicated there was a significant sampling variation compared to actual variation with an I^2 value of 8.7 %. Late embryonic/early fetal loss data were not affected by publication bias.

3.4. Pregnancy losses through gestation

Reproductive failures during the various developmental periods can be combined to determine the overall losses from the time of fertilization to the end of gestation (Fig. 4). In beef cattle, more than 50 % of the total reproductive failures occur prior to day 16 after insemination. Between day 16 and 32, there will be reproductive failures (pregnancy losses) in an additional 15.5 % of cows. Reproductive failures after the first month of gestation, on average, occurs in less than 6 % of beef cows; however, this is primarily affected by moderators and environmental factors.

4. Discussion

Meta-analysis results indicate that incidence of reproductive failure in beef cattle has not drastically changed since the first scientific reports (Ayalon, 1978); although a detailed description of periods during which pregnancy losses occur has potential impacts for research advancements and modified industry recommendations. Collecting large quantities of accurate reproductive data from beef cattle is more difficult when compared with dairy cattle, as less intensive management routines limit collection of large quantities of field data. This has led to limited information regarding timing of pregnancy loss in beef cattle which have different patterns of fertility and reproductive failure compared to dairy cattle.

Pregnancy loss periods as reported in this meta-analysis differ in terms of days of gestation compared to developmental period definitions based on physiological events. Although overlap may occur between physiological periods, the main objective of the

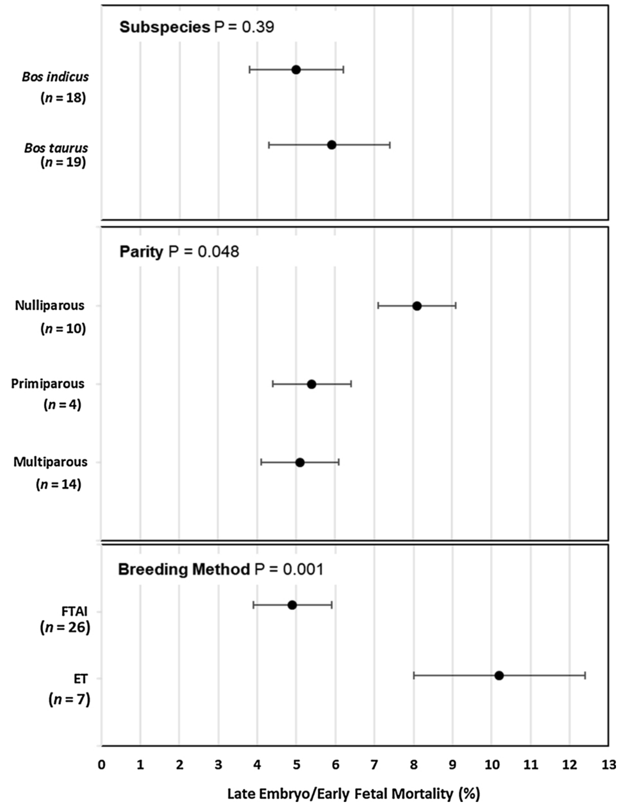

Fig. 3. Point estimates and 95 % CI for moderators explaining late embryonic/ early fetal mortality. n = number of studies; heterogeneity P denotes the probability all studies share a common point estimate. Some publications that utilized multiple moderators in a single trial and could not be separated were excluded from moderator analysis.

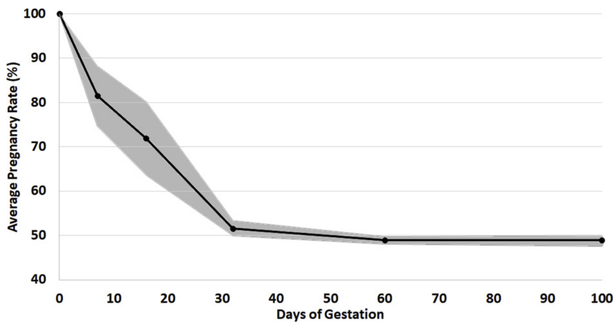

Fig. 4. Average predicted pregnancy rate by day of gestation in beef cattle. Grey area indicates area of 95 % confidence interval.

present meta-analysis was to identify and report a summary of the pregnancy loss based on available reports in research articles. While fertilization is generally thought of as a singular event at the initiation of pregnancy, results from all studies were included in which there was identification of pregnancies before day 7 accounting for fertilization and initial embryo development failure. Embryonic period, when strictly classified according to the physiological events during gestation, should refer to the period from conception to the end of embryonic differentiation stage, which is around day 42–45 of gestation (Hubbert, et al., 1972). It is commonly subdivided into early embryonic period (conception to day 28) and late embryonic period (days 28–42) marked by placental attachment and delineation of the fetal shape; however, pregnancy diagnoses are often reported at days 30–32 and later at days 60–100. The timeline used for the present meta-analysis maximizes the number of trials included in the analysis to obtain a more accurate prediction of reproductive efficiency data.

Fertilization and blastocyst formation are the initial processes for any pregnancy to occur. In early reviews of reproductive failure, there is reports indicating fertilization rates in beef cattle are approximately 90 % which is consistent with findings of structures collected at day 7 of gestation with embryo transfer (Maurer and Chenault, 1983; Santos et al., 2004). Unfortunately, significant embryo failure occurs between fertilization and day 7. Furthermore, collecting fertilization data is difficult and often requires uterine flushing after collection of uterine tissues. In the current meta-analysis, fertility and pre-blastocyst development failures during the FERT period averaged 28.4 %, with a range from 2.9%–44.4%. In comparison, embryo mortality during the first week of gestation in lactating dairy cattle can average 50 % when there is no evidence of excess stressors (Wiltbank et al., 2016). Although beef cattle have limited production stress compared to dairy cattle, there are physiological factors that may have important functions in pregnancy success during the first week of gestation. Data suggest beef cows with large (> 15.7 mm) or persistent dominant follicles are less fertile, likely due to decreased concentrations of progesterone and estradiol during follicular development (Ahmad et al., 1995; Perry et al., 2007; Abreu et al., 2014). Body condition score (BCS) and effects of nutrient restriction also impact initial embryo development. Cows and heifers with decreasing BCS or body weight post-AI not only have greater pregnancy losses but specifically have embryos with lesser quality grades and a greater percentage of immature staged embryos when collected at day 7 of gestation (Bridges et al., 2012; Perry et al., 2013; Kruse et al., 2017). Results from studies support that these failures are not due to fertilization failure or less than optimal progesterone concentration, but some other developmental incompetency related to the maternal environment (Spitzer et al., 1978; Kruse et al., 2017). Animals in studies included in this meta-analysis were bred after observation of estrus using semen of acceptable fertility or by natural service. Sire effects could not be assessed but paternal genetics can contribute significantly to early embryonic mortality (Ledoux et al., 2015). Samples sizes in studies were small and that may contribute to the variation in pregnancy loss. Furthermore, the absence of studies in which there was comparison of different factors such as parities and subspecies, indicate that there is a gap in current knowledge of pregnancy development during the first week of gestation in beef cattle. More research could result in enhanced knowledge about how factors, including parity and breeding method, contribute to pregnancy loss in the first week of gestation in beef cattle. The current meta-analysis is one of the more homogeneous; however, limitations of sample population diversity may mask differences between subspecies or parities, as only *Bos taurus* animals were represented and most studies conducted with heifers.

A significant amount of pregnancy loss in cattle occurs during the first month of gestation in beef cattle. There is, however, some debate on when this loss is most significant: during initial embryo elongation (days 7–14) or during maternal recognition of pregnancy and beyond (days 15–28). There are reports indicating the greatest single period of pregnancy loss is the second week of gestation when there is hatching of the blastocyst and initiation of elongation of the embryo (Diskin and Sreenan, 1980; Carter et al., 2008; Lonergan et al., 2016). Alternatively, other recent evaluations of available data, including this meta-analysis, may indicate otherwise (Burns et al., 2015; Wiltbank et al., 2016). It is important to note that reports of increased pregnancy loss during the second week of gestation may be heavily influenced by data collected from lactating dairy cattle. Only 11 studies in beef cattle were identified in which there was measurement of pregnancy loss before day 16, likely due to inconsistencies in identifying pregnancies at this early stage of development. Of the 11 studies, in one there was reporting of data collected from *Bos indicus* cattle which warrants further research to establish potential subspecies differences. The results from this meta-analysis suggest increased pregnancy loss after the second week of gestation whereas the traditional assumption is there are greater pregnancy losses during the second week of gestation prior to maternal recognition of pregnancy. During the second half embryonic development between days 15 and 28 of gestation, for successful pregnancy maintenance there is reliance on proper maternal recognition of pregnancy and important processes protecting the embryo from the maternal immune system (Roberts and Schalue-Francis, 1990; Bauersachs and Wolf, 2013; Yang et al., 2014). Losses during this period provide significant challenges to the adoption of early pregnancy diagnosis methods including the use of information related to interferon stimulated genes (Green et al., 2010; Pugliesi et al., 2014).

This meta-analysis provides a baseline value based on large quantities of data in current research that model beef production systems utilizing assisted reproductive technologies. Based on the consistency in results from this meta-analysis, there is approximately a 50 % pregnancy rate at day 30 of gestation when utilizing estrous synchronization, regardless of moderator combinations. Although there are a limited number of trials available, results from the current meta-analysis indicate there is a 15 % increase in pregnancy rate in cattle bred following estrus expression compared to those bred using a FTAI protocol following synchronization of estrus. This may be confounded by results from studies where there is the requirement for controlled data collection and use of FTAI protocols, especially in large *Bos indicus* trials conducted in South America. In a meta-analysis of expression of estrus in FTAI protocols, heifers exhibiting estrus before AI had a 27 % greater conception rate compared to heifers that did not express estrus (Richardson et al., 2016). Estrous expression with use of FTAI protocols is highly variable with there being reports of between 20%–80% of animals not exhibiting estrus prior to AI in both *Bos indicus* and *Bos taurus* subspecies (Richardson et al., 2016; Thomas et al., 2017; Rodrigues et al., 2018). Additionally, *Bos indicus* cattle are generally located in regions where the severe climatic conditions result in greater physiological stress, particularly as a result of nutritional factors, as compared with *Bos taurus* beef cows which contribute to trends of decreased fertility (Chenoweth, 1994; Bó et al., 2003). Cows with a lesser BCS or that are anestrous will have decreased estrous expression which is a variable that is correlated with pregnancy rates (Richardson et al., 2016). A less than optimal BCS is a critical factor in reduced fertility of primiparous cows (DeRouen et al., 1994; Ciccioli et al., 2003); however, data were not variable enough to utilize BCS as a moderator in the current meta-analysis. Additionally, results from studies in which there was examination of the combination of growth, lactation and reproduction stressors indicate there is an increased pregnancy loss in primiparous cows compared to heifers and multiparous cows (Werth et al., 1996; Lalman et al., 1997; Freetly et al., 2006).

Hormone manipulation, nutritional management, health protocols and other factors that may increase day 30 pregnancy rates have been studied extensively. Late embryonic and fetal mortality is the focus of less research and, thus, how these factors affect fertility failures is less understood than other areas of pregnancy loss. Late embryonic/early fetal mortality has significant negative impacts on reproductive efficiency and economic consequences because cows may be retained in the herd for an entire season without producing a marketable product. Based on the current study, overall LEF in beef cattle is 5.8 % which is significantly less compared to what occurs in dairy cattle. In most reports, there is an estimation of late embryonic mortality of lactating dairy cows between 10 % and 20 % (Grimard et al., 2006; Pohler et al., 2016a; Wiltbank et al., 2016), although in some studies results indicate there is about a 7 % late embryonic loss (Silke et al., 2002). With beef cattle herd management, there is more crossbreeding utilized than occurs in dairy cattle, thus, there is less inbreeding and expression of recessive genetically lethal traits which are known contributors to increased late embryonic mortality in dairy cattle (Cassell et al., 2003; Diskin and Morris, 2008). Additionally, use of advanced reproductive technologies, such as *in vitro* produced embryos, result in increased LEF; however, these technologies are not widely used in beef production (Farin et al., 2006). With the current meta-analysis, the studies included were where there was a confirmed pregnancy on either day 60 or 100 of gestation. Interestingly, results from studies with pregnancy diagnosis on day 60 indicated there was no difference ($P = 0.39$) in pregnancy loss compared to studies where there was pregnancy diagnosis at day 100 when initial diagnosis occurred around day 30. This indicates that fetal loss during the third month of gestation between days 60 and 100 is limited. Most studies in which there is pregnancy diagnosis on day 100 are conducted with *Bos indicus* cows and most day 60 studies were conducted in *Bos taurus* cows. It, therefore, may be interesting to analyze data from *Bos indicus* cows at day 60 and *Bos taurus* cows at day 100 to ascertain if a subspecies effect exists. Heifers had a greater late embryonic/early fetal mortality rate (8.1 %) than cows (5.1 %) but there were inadequate numbers of trials included in the meta-analysis for detection of a difference between *Bos indicus* heifers and *Bos taurus* heifers. From a management perspective, it is unknown if there is a physiologic factor affecting parity differences or if animals more susceptible to LEF are culled as heifers before being retained for placement in the mature cow population.

While the results from available studies only provide enough data for moderator analysis of subspecies, breeding method and parity, other factors may have important effects when quantifying embryo loss. Using results from available studies, there was no identification of other moderators or additional variables that significantly affected the results from the meta-analysis, therefore, estimates for pregnancy loss during multiple periods are both statistically and biologically sound. Optimal reproductive management strategies are dependent on numerous factors and with future analyses there should be comparisons of the impact of estrous synchronization protocols, sire effects, and nutritional status on overall reproductive performance to make recommendations for field use.

5. Conclusion

Gestational loss during the early stages of pregnancy can be detrimental to calving rates in beef cattle. The results from the current meta-analysis and further heterogeneity analysis indicates early fertilization failures are variable among cattle types and ages providing opportunity for both research and improved production strategies. Fertilization rates may be as high as 95 % in some scenarios; however, current research is limited to a few studies in beef cattle. Loss during the early embryonic period is dependent on many factors, the most impactful being parity with primiparous heifers where there are large amounts of reproductive failure early in gestation. Approximately 48 % of cows will not be pregnant at day 30 of gestation following a single insemination. Late embryonic mortality is variable among beef cattle and significantly less than what is reported in dairy cattle. Further reporting of pregnancy loss data is of great interest to identify other factors that may positively or negatively affect pregnancy loss at different points in gestation.

Funding

The authors would like to acknowledge that projects within were supported by Agriculture and Food Research Initiative Competitive Grant no. 2017-67015-26457 from the USDA National Institute of Food and Agriculture as well as USDA-NIFA Hatch/ Multistate Project W3112-TEX07702 – Reproductive performance in domestic animals.

Declaration of Competing Interest

The authors declare that there is no conflict of interest that could affect our objectivity in this study.

References

Abdalla, H., Elghafghuf, A., Elsohaby, I., Nasr, M.A., 2017. Maternal and non-maternal factors associated with late embryonic and early fetal losses in dairy cows. Theriogenology 100, 16–23.
Abreu, F., Geary, T., Cruppe, L., Madsen, C., Jinks, E., Pohler, K., Vasconcelos, J., Day, M., 2014. The effect of follicle age on pregnancy rate in beef cows. J. Anim. Sci. 92, 1015–1021.
Ahmad, N., Schrick, F., Butcher, R., Inskeep, E., 1995. Effect of persistent follicles on early embryonic losses in beef cows. Biol. Reprod. 52, 1129–1135.
Aono, F., Cooke, R.F., Alfieri, A., Vasconcelos, J.L.M., 2013. Effects of vaccination against reproductive diseases on reproductive performance of beef cows submitted to fixed-timed AI in Brazilian cow-calf operations. Theriogenology 79, 242–248.
Ayalon, N., 1978. A review of embryonic mortality in cattle. J. Reprod. Fertil. 54, 483–493.
Baron, R.M., Kenny, D.A., 1986. The moderator–mediator variable distinction in social psychological research: conceptual, strategic, and statistical considerations. J. Pers. Soc. Psychol. 51, 1173.
Bauersachs, S., Wolf, E., 2013. Immune aspects of embryo-maternal cross-talk in the bovine uterus. J. Reprod. Immunol. 97, 20–26.
Beal, W.E., Perry, R.C., Corah, L.R., 1992. The use of ultrasound in monitoring reproductive physiology of beef cattle. J. Anim. Sci. 70, 924–929.
Bó, G., Baruselli, P., Martınez, M., 2003. Pattern and manipulation of follicular development in Bos indicus cattle. Anim. Reprod. Sci. 78, 307–326.
Borenstein, M., Hedges, L.V., Higgins, J., Rothstein, H.R., 2009. Introduction to Meta-Analysis. Wiley Online Library.
Borenstein, M., Higgins, J., Hedges, L.V., Rothstein, H.R., 2017. Basics of meta-analysis: I2 is not an absolute measure of heterogeneity. Res. Synth. Methods 8, 5–18.
Breuel, K., Lewis, P., Schrick, F., Lishman, A., Inskeep, E., Butcher, R., 1993. Factors affecting fertility in the postpartum cow: role of the oocyte and follicle in conception rate. Biol. Reprod. 48, 655–661.
Bridges, G., Kruse, S., Funnell, B., Perry, G., Gunn, P., Arias, R., Lake, S., 2012. Changes in body condition on oocyte quality and embryo survival. Proc. Appl. Reprod. Strategies Beef Cattle 23, 269–283.

Burns, G., Wehrman, M., Geary, T., Moss, J., Denicol, A., Dobbs, K., Ortega, M., Hansen, P., Wojtowicz, A., Neibergs, H., 2015. 147- Systems biology approach to understanding uterine receptivity and pregnancy loss. Society for the Study of Reproduction.
Burns, M., Buttrey, B., Dobbins, C., Martel, C., Olson, K., Lamb, G., Stevenson, J., 2008. Evaluation of human chorionic gonadotropin as a replacement for gonadotropin-releasing hormone in ovulation-synchronization protocols before fixed timed artificial insemination in beef cattle. J. Anim. Sci. 86, 2539–2548.
Carter, F., Forde, N., Duffy, P., Wade, M., Fair, T., Crowe, M., Evans, A., Kenny, D., Roche, J., Lonergan, P., 2008. Effect of increasing progesterone concentration from Day 3 of pregnancy on subsequent embryo survival and development in beef heifers. Reprod. Fertil. Dev. 20, 368–375.
Cassell, B., Adamec, V., Pearson, R., 2003. Maternal and fetal inbreeding depression for 70-day nonreturn and calving rate in Holsteins and Jerseys. J. Dairy Sci. 86, 2977–2983.
Cheng, Z., Abudureyimu, A., Oguejiofor, C.F., Ellis, R., Barry, A.T., Chen, X., Anstaett, O.L., Brownlie, J., Wathes, D.C., 2016. BVDV alters uterine prostaglandin production during pregnancy recognition in cows. Reproduction 151, 605–614.
Chenoweth, P., 1994. Aspects of reproduction in female Bos indicus cattle: a review. Aust. Vet. J. 71, 422–426.
Ciccioli, N., Wettemann, R., Spicer, L., Lents, C., White, F., Keisler, D., 2003. Influence of body condition at calving and postpartum nutrition on endocrine function and reproductive performance of primiparous beef cows. J. Anim. Sci. 81, 3107–3120.
Colazo, M., Kastelic, J., Whittaker, P., Gavaga, Q., Wilde, R., Mapletoft, R., 2004. Fertility in beef cattle given a new or previously used CIDR insert and estradiol, with or without progesterone. Anim. Reprod. Sci. 81, 25–34.
Cooke, R., Schubach, K., Marques, R., Peres, R., Silva, L., Carvalho, R., Cipriano, R., Bohnert, D., Pires, A., Vasconcelos, J., 2017. Effects of temperament on physiological, productive, and reproductive responses in beef cows. J. Anim. Sci. 95, 1–8.
Cordeiro, M.B., Peres, M.S., de Souza, J.M., Gaspar, P., Barbiere, F., Sá Filho, M.F., Maturana Filho, M., Dinardi, R.N., Nogueira, G.P., Mesquita, F.S., 2015. Supplementation with sunflower seed increases circulating cholesterol concentrations and potentially impacts on the pregnancy rates in Bos indicus beef cattle. Theriogenology 83, 1461–1468.
DeRouen, S., Franke, D., Morrison, D., Wyatt, W., Coombs, D., White, T., Humes, P., Greene, B., 1994. Prepartum body condition and weight influences on reproductive performance of first-calf beef cows. J. Anim. Sci. 72, 1119–1125.
Diskin, M., Sreenan, J., 1980. Fertilization and embryonic mortality rates in beef heifers after artificial insemination. J. Reprod. Fertil. 59, 463–468.
Diskin, M., Waters, S., Parr, M., Kenny, D., 2016. Pregnancy losses in cattle: potential for improvement. Reprod. Fertil. Dev. 28, 83–93.
Diskin, M.G., Morris, D.G., 2008. Embryonic and early foetal losses in cattle and other ruminants. Reprod. Domest. Anim. 43 (Suppl 2), 260–267. https://doi.org/10.1111/j.1439-0531.2008.01171.x.
Dobbins, C., Eborn, D., Tenhouse, D., Breiner, R., Johnson, S., Marston, T., Stevenson, J., 2009. Insemination timing affects pregnancy rates in beef cows treated with CO-Synch protocol including an intravaginal progesterone insert. Theriogenology 72, 1009–1016.
Dunne, L., Diskin, M., Sreenan, J., 2000. Embryo and foetal loss in beef heifers between day 14 of gestation and full term. Anim. Reprod. Sci. 58, 39–44.
Duval, S., Tweedie, R., 2000. Trim and fill: a simple funnel-plot–based method of testing and adjusting for publication bias in meta-analysis. Biometrics 56, 455–463.
Farin, P.W., Piedrahita, J.A., Farin, C.E., 2006. Errors in development of fetuses and placentas from in vitro-produced bovine embryos. Theriogenology 65, 178–191.
Ferreira, L.C.L., Cooke, R.F., Marques, R.S., Fernandes, H.J., Fernandes, C.E., Stelato, R., Franco, G.L., Lemos, R.A.A., 2016. Effects of vaccination against foot-and-mouth disease virus on reproductive performance of Bos indicus beef cows. J. Anim. Sci. 94, 401–405.
Franco, G.A., Peres, R.F.G., Martins, C.F.G., Reese, S.T., Vasconcelos, J.L.M., Pohler, K.G., 2018. Sire contribution to pregnancy loss and pregnancy-associated glycoprotein production in Nelore cows. J. Anim. Sci. 96, 632–640.
Freetly, H., Nienaber, J., Brown-Brandl, T., 2006. Partitioning of energy during lactation of primiparous beef cows. J. Anim. Sci. 84, 2157–2162.
Garrett, J., Geisert, R., Zavy, M., Morgan, G., 1988. Evidence for maternal regulation of early conceptus growth and development in beef cattle. J. Reprod. Fertil. 84, 437–446.
Green, J., Okamura, C., Poock, S., Lucy, M., 2010. Measurement of interferon-tau (IFN-τ) stimulated gene expression in blood leukocytes for pregnancy diagnosis within 18–20d after insemination in dairy cattle. Anim. Reprod. Sci. 121, 24–33.
Grimard, B., Freret, S., Chevallier, A., Pinto, A., Ponsart, C., Humblot, P., 2006. Genetic and environmental factors influencing first service conception rate and late embryonic/foetal mortality in low fertility dairy herds. Anim. Reprod. Sci. 91, 31–44.
Higgins, J.P., Thompson, S.G., Deeks, J.J., Altman, D.G., 2003. Measuring inconsistency in meta-analyses. BMJ 327, 557–560.
Hubbert, W., Dennis, S., Adams, W., Bierschwal, C., Biggers, J., Carroll, E., Dunne, H., Hutton, N., Kendrick, J., Kenney, R., 1972. Recommendations for standardizing bovine reproductive terms. Cornell Vet. 62, 216–237.
Jinks, E., Smith, M., Atkins, J., Pohler, K., Perry, G., MacNeil, M., Roberts, A., Waterman, R., Alexander, L., Geary, T., 2013. Preovulatory estradiol and the establishment and maintenance of pregnancy in suckled beef cows. J. Anim. Sci. 91, 1176–1185.
Kill, L.K., Pohler, K.G., Perry, G.A., Smith, M.F., 2013. Serum Ovine Pregnancy Associated Glycoproteins and Progesterone in Beef Heifers That Experienced Late embryonic/fetal Mortality ASAS Midwestern Section. p 2, Des Moines, IA. .
Kruse, S., Bridges, G., Funnell, B., Bird, S., Lake, S., Arias, R., Amundson, O., Larimore, E., Keisler, D., Perry, G., 2017. Influence of post-insemination nutrition on embryonic development in beef heifers. Theriogenology 90, 185–190.
Lalman, D., Keisler, D., Williams, J., Scholljegerdes, E., Mallett, D., 1997. Influence of postpartum weight and body condition change on duration of anestrus by undernourished suckled beef heifers. J. Anim. Sci. 75, 2003–2008.
Lamb, G., Larson, J., Stevenson, J., Johnson, S., Day, M., Geary, T., Kesler, D., DeJarnette, J., Schrick, F., DiCostanzo, A., 2006. Synchronization of estrus in suckled beef cows for detected estrus and artificial insemination and timed artificial insemination using gonadotropin-releasing hormone, prostaglandin F, and progesterone. J. Anim. Sci. 84, 332–342.
Lamb, G., Stevenson, J., Kesler, D., Garverick, H., Brown, D., Salfen, B., 2001. Inclusion of an intravaginal progesterone insert plus GnRH and prostaglandin F2alpha for ovulation control in postpartum suckled beef cows. J. Anim. Sci. 79, 2253–2259.
Larson, J., Lamb, G., Stevenson, J., Johnson, S., Day, M., Geary, T., Kesler, D., DeJarnette, J., Schrick, F., DiCostanzo, A., 2006. Synchronization of estrus in suckled beef cows for detected estrus and artificial insemination and timed artificial insemination using gonadotropin-releasing hormone, prostaglandin F, and progesterone. J. Anim. Sci. 84, 332–342.
Ledoux, D., Ponsart, C., Grimard, B., Gatien, J., Deloche, M., Fritz, S., Lefebvre, R., Humblot, P., 2015. Sire effect on early and late embryonic death in French Holstein cattle. Animal 9, 766–774.
Lonergan, P., Fair, T., Forde, N., Rizos, D., 2016. Embryo development in dairy cattle. Theriogenology 86, 270–277.
Lopes, C., Scarpa, A., Cappellozza, B., Cooke, R.F., Vasconcelos, J.L.M., 2009. Effects of rumen-protected polyunsaturated fatty acid supplementation on reproductive performance of beef cows. J. Anim. Sci. 87, 3935–3943.
Martinez, M., Kastelic, J., Adams, G., Mapletoft, R., 2002. The use of a progesterone-releasing device (CIDR-B) or melengestrol acetate with GnRH, LH, or estradiol benzoate for fixed-time AI in beef heifers. J. Anim. Sci. 80, 1746–1751.
Maurer, R., Chenault, J., 1983. Fertilization failure and embryonic mortality in parous and nonparous beef cattle. J. Anim. Sci. 56, 1186–1189.
Meneghetti, M., Sá Filho, O., Peres, R., Lamb, G., Vasconcelos, J., 2009. Fixed-time artificial insemination with estradiol and progesterone for Bos indicus cows I: basis for development of protocols. Theriogenology 72, 179–189.
Mercadante, V., Kozicki, L., Ciriaco, F., Henry, D., Dahlen, C., Crosswhite, M., Larson, J., Voelz, B., Patterson, D., Perry, G., 2015. Effects of administration of prostaglandin F at initiation of the seven-day CO-Synch + controlled internal drug release ovulation synchronization protocol for suckled beef cows and replacement beef heifers. J. Anim. Sci. 93, 5204–5213.
Mialon, M., Camous, S., Renand, G., Martal, J., Menissier, F., 1993. Peripheral concentrations of a 60-kDa pregnancy serum protein during gestation and after calving and in relationship to embryonic mortality in cattle. Reprod. Nutr. Dev. 33, 269–282.
O'Hara, L., Forde, N., Carter, F., Rizos, D., Maillo, V., Ealy, A., Kelly, A., Rodriguez, P., Isaka, N., Evans, A., 2014. Paradoxical effect of supplementary progesterone between Day 3 and Day 7 on corpus luteum function and conceptus development in cattle. Reprod. Fertil. Dev. 26, 328–336.
Parr, M., Scully, S., Lonergan, P., Evans, A., Crowe, M., Diskin, M., 2017. Establishment of critical timing of progesterone supplementation on corpus luteum and

embryo development in beef heifers. Anim. Reprod. Sci. 180, 1–9.
Peres, R., Júnior, I.C., Sá Filho, O., Nogueira, Gd.P., Vasconcelos, J.L.M., 2009. Strategies to improve fertility in Bos indicus postpubertal heifers and nonlactating cows submitted to fixed-time artificial insemination. Theriogenology 72, 681–689.
Perry, G., Perry, B., Walker, J., Wright, C., Salverson, R., Patterson, H., 2013. Evaluation of prior grazing experience on reproductive performance in beef heifers. Prof. Anim. Sci. 29, 595–600.
Perry, G.A., Smith, M.F., Lucy, M.C., Roberts, A.J., MacNeil, M.D., Geary, T.W., 2003. Effect of ovulatory follicle size at time of GnRH injection or standing estrus on pregnancy rates and embryonic/fetal mortality in beef cattle. In: Western Section. Proceedings-American Society of Animal Science 54. pp. 281–284.
Perry, G., Smith, M., Roberts, A., MacNeil, M., Geary, T., 2007. Relationship between size of the ovulatory follicle and pregnancy success in beef heifers. J. Anim. Sci. 85, 684–689.
Pessoa, Jd.S., Rolin Filho, S.T., Ribeiro, H.F.L., Garcia, O.S., Nunes, Kd.B., Amorim, B.Sd., 2012. Embryonic loss in cows submitted to timed AI in the northeast of Para. Ciencia Animal 22, 239–241.
Pfeifer, L., Castro, N., Neves, P., Cestaro, J., Schneider, A., 2017. Comparison between two estradiol-progesterone based protocols for timed artificial insemination in blocks in lactating Nelore cows. Anim. Reprod. Sci. 181, 125–129.
Pohler, K., Pereira, M., Lopes, F., Lawrence, J., Keisler, D., Smith, M., Vasconcelos, J., Green, J., 2016a. Circulating concentrations of bovine pregnancy-associated glycoproteins and late embryonic mortality in lactating dairy herds. J. Dairy Sci. 99, 1584–1594.
Pohler, K., Peres, R., Green, J., Graff, H., Martins, T., Vasconcelos, J., Smith, M., 2016b. Use of bovine pregnancy-associated glycoproteins to predict late embryonic mortality in postpartum Nelore beef cows. Theriogenology 85, 1652–1659.
Pohler, K.G., Geary, T.W., Johnson, C.L., Atkins, J.A., Jinks, E.M., Busch, D.C., Green, J.A., MacNeil, M.D., Smith, M.F., 2013. Circulating bovine pregnancy associated glycoproteins are associated with late embryonic/fetal survival but not ovulatory follicle size in suckled beef cows. J. Anim. Sci. 91, 4158–4167. https://doi.org/10.2527/jas.2013-6348.
Pontes, J., Nonato-Junior, I., Sanches, B., Ereno-Junior, J., Uvo, S., Barreiros, T., Oliveira, J., Hasler, J., Seneda, M., 2009. Comparison of embryo yield and pregnancy rate between in vivo and in vitro methods in the same Nelore (Bos indicus) donor cows. Theriogenology 71, 690–697.
Pontes, J., Sterza, F.M., Basso, A., Ferreira, C., Sanches, B., Rubin, K., Seneda, M., 2011. Ovum pick up, in vitro embryo production, and pregnancy rates from a large-scale commercial program using Nelore cattle (Bos indicus) donors. Theriogenology 75, 1640–1646.
Pope, W., 1988. Uterine asynchrony: a cause of embryonic loss. Biol. Reprod. 39, 999–1003.
Pradebon, E., Machado, A., Gambin, L., Gonsioroski, A., da Silva, M., Bernardi, M., Dias, M., da Cunha Chaiben, M., Borges, J., 2017. Ovarian structures, estrus expression, and pregnancy rate in beef heifers using estradiol cypionate or GnRH as ovulation inductors in timed AI protocols. Reprod. Fertil. Dev. 29, 113–114.
Pugliesi, G., Miagawa, B.T., Paiva, Y.N., França, M.R., Silva, L.A., Binelli, M., 2014. Conceptus-induced changes in the gene expression of blood immune cells and the ultrasound-accessed luteal function in beef cattle: how early can we detect pregnancy? Biol. Reprod. 91 (95) 91-12.
Radigonda, V.L., Pereira, G.R., da Cruz Favaro, P., Júnior, F.A.B., Borges, M.H.F., Galdioli, V.H.G., Júnior, C.K., 2017. Infrared thermography relationship between the temperature of the vulvar skin, ovarian activity, and pregnancy rates in Braford cows. Trop. Anim. Health Prod. 49, 1787–1791.
Richardson, B.N., Hill, S.L., Stevenson, J.S., Djira, G.D., Perry, G.A., 2016. Expression of estrus before fixed-time AI affects conception rates and factors that impact expression of estrus and the repeatability of expression of estrus in sequential breeding seasons. Anim. Reprod. Sci. 166, 133–140.
Roberts, R.M., Schalue-Francis, T., 1990. Maternal recognition of pregnancy and embryonic loss. Theriogenology 33, 175–183.
Roche, J., Bolandl, M., McGeady, T., 1981. Reproductive wastage following artificial insemination of heifers. Vet. Rec. 109, 401–404.
Rodrigues, A., Cooke, R., Cipriano, R., Silva, L., Cerri, R., Cruppe, L., Meneghetti, M., Pohler, K., Vasconcelos, J., 2018. Impacts of estrus expression and intensity during a timed-AI protocol on variables associated with fertility and pregnancy success in Bos indicus-influenced beef cows. J. Anim. Sci. 96, 236–249.
Sa Filho, M.F., Marques, M.O., Girotto, R., Santos, F.A., Sala, R.V., Barbuio, J.P., Baruselli, P.S., 2014. Resynchronization with unknown pregnancy status using progestin-based timed artificial insemination protocol in beef cattle. Theriogenology 81, 284–290. https://doi.org/10.1016/j.theriogenology.2013.09.027.
Sá Filho, O., Dias, C., Lamb, G., Vasconcelos, J., 2010. Progesterone-based estrous synchronization protocols in non-suckled and suckled primiparous Bos indicus beef cows. Anim. Reprod. Sci. 119, 9–16.
Sá Filho, O., Meneghetti, M., Peres, R., Lamb, G., Vasconcelos, J., 2009. Fixed-time artificial insemination with estradiol and progesterone for Bos indicus cows II: strategies and factors affecting fertility. Theriogenology 72, 210–218.
Sales, J.N.S., Pereira, R.V.V., Bicalho, R.C., Baruselli, P.S., 2011. Effect of injectable copper, selenium, zinc and manganese on the pregnancy rate of crossbred heifers (Bos indicus x Bos taurus) synchronized for timed embryo transfer. Livest. Sci. 142, 59–62. https://doi.org/10.1016/j.livsci.2011.06.014.
Santos, J., Thatcher, W., Chebel, R., Cerri, R., Galvao, K., 2004. The effect of embryonic death rates in cattle on the efficacy of estrus synchronization programs. Anim. Reprod. Sci. 82, 513–535.
Silke, V., Diskin, M., Kenny, D., Boland, M., Dillon, P., Mee, J., Sreenan, J., 2002. Extent, pattern and factors associated with late embryonic loss in dairy cows. Anim. Reprod. Sci. 71, 1–12.
Smith, M., Nix, K., Kraemer, D., Amoss, M., Herron, M., Wiltbank, J., 1982. Fertilization rate and early embryonic loss in Brahman crossbred heifers. J. Anim. Sci. 54, 1005–1011.
Spitzer, J., Niswender, G., Seidel, G., Wiltbank, J., 1978. Fertilization and blood levels of progesterone and LH in beef heifers on a restricted energy diet. J. Anim. Sci. 46, 1071–1077.
Starbuck, M.J., Inskeep, E.K., Dailey, R.A., 2006. Effect of a single growth hormone (rbST) treatment at breeding on conception rates and pregnancy retention in dairy and beef cattle. Anim. Reprod. Sci. 93, 349–359.
Stevenson, J., Johnson, S., Medina-Britos, M., Richardson-Adams, A., Lamb, G., 2003. Resynchronization of estrus in cattle of unknown pregnancy status using estrogen, progesterone, or both. J. Anim. Sci. 81, 1681–1692.
Thomas, J., Locke, J., Bishop, B., Abel, J., Ellersieck, M., Yelich, J., Poock, S., Smith, M., Patterson, D., 2017. Evaluation of the 14-d CIDR-PG and 9-d CIDR-PG protocols for synchronization of estrus in Bos indicus-influenced and Bos taurus beef heifers. Theriogenology 92, 190–196.
Werth, L., Whittier, J., Azzam, S., Deutscher, G., Kinder, J., 1996. Relationship between circulating progesterone and conception at the first postpartum estrus in young primiparous beef cows. J. Anim. Sci. 74, 616–619.
Wiltbank, M.C., Baez, G.M., Garcia-Guerra, A., Toledo, M.Z., Monteiro, P.L., Melo, L.F., Ochoa, J.C., Santos, J.E., Sartori, R., 2016. Pivotal periods for pregnancy loss during the first trimester of gestation in lactating dairy cows. Theriogenology 86, 239–253.
Yang, L., Zhang, L., Qiao, H., Liu, N., Wang, Y., Li, S., 2014. Maternal immune regulation by conceptus during early pregnancy in the bovine. Asian J. Anim. Vet. Adv. 9, 610–620.