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# Efficacy of progesterone supplementation during early pregnancy in cows: A meta-analysis

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## ABSTRACT

Progesterone is a critical hormone during early pregnancy in the cow. As a result, a number of studies have investigated the effects of progesterone supplementation on pregnancy rates. In this study, a meta-analysis using a univariate binary random effects model was carried out on 84 specific treatments reported in 53 publications involving control ( $n = 9905$ ) and progesterone-treated ( $n = 9135$ ) cows. Although the results of individual studies showed wide variations ( $-40\%$  to  $+50\%$  point changes), progesterone treatment resulted in an overall increase in pregnancy rate odds ratio ( $OR = 1.12$ ;  $P < 0.01$ ). Improvements in pregnancy rate were only observed in cows treated at natural estrus ( $OR = 1.41$ ,  $P < 0.01$ ) and not following synchronization of estrus or ovulation. Although treatment between Days 3 to 7 postinsemination was beneficial ( $OR = 1.15$ ;  $P < 0.01$ ), treatment earlier or later than this was not. Progesterone supplementation was beneficial in cows of lower fertility ( $<45\%$  control pregnancy rate) but not in cows with higher fertility. These results indicated that the benefit of progesterone supplementation on fertility of cows required exogenous progesterone supplementation to start between Day 3 to 7 and the appropriate reproductive status (i.e., lower fertility, natural estrus) of the treated cows.

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## 1. Introduction

Over the past few decades reproductive efficiency of dairy cows has continually declined. E.g., Royal et al. [1] reported a fall in pregnancy rates to first service of approximately 1% per year in the UK, coupled with a dramatic increase in the incidence of reproductive problems. Over a similar period, the rate of decline in the USA was around 0.5% per annum [2]. This trend has continued with Hudson et al. [3] reporting declining pregnancy rates between 2000 and 2006 across 250 UK herds.

An important factor contributing to early pregnancy failure is embryo mortality; early embryo loss because of the failure of maternal recognition of pregnancy is thought to account for up to 25% to 30% of pregnancy failures in dairy cows [4,5]. The establishment of a successful pregnancy in the cow requires the embryo developing sufficiently to produce adequate amounts of the antiluteolytic protein, interferon tau to prevent luteolytic  $PGF_{2\alpha}$  secretion from the endometrium. One of the principal hormones in controlling pregnancy establishment is progesterone, which stimulates the production of endometrial secretions that are beneficial for the successful development of the embryo and subsequently survival [6,7]. Insufficient circulating progesterone concentration has been clearly linked to poor embryo development and failure to prevent the development of the luteolytic signal in dairy cows [8]. Problems associated with the physiological mechanisms

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underpinning the establishment of pregnancy continue to compromise the reproductive performance of dairy cows.

In the past 60 years, the importance of progesterone to embryonic development has resulted in a plethora of studies that have attempted to improve pregnancy rate through postinsemination progesterone treatment. However, the results of published studies have failed to deliver any clear consensus on the therapeutic benefits of such treatments. Although a number of studies have reported progesterone supplementation as beneficial, other studies have shown no benefit or even a marked reduction in pregnancy rate (e.g., [4,5]). Overall, despite the fact that progesterone has a major influence on the outcome of pregnancy in the dairy cow and its widespread use, the reason for these different outcomes is still not clear and requires further study. The objective of this study was to assess the efficacy of progesterone supplementation in improving pregnancy rates of cows, by conducting a meta-analysis of data from all available progesterone administration studies.

## 2. Materials and methods

### 2.1. Identification of studies and inclusion criteria

All English-language papers and abstracts that published the results of studies investigating the efficacy of direct progesterone supplementation on pregnancy rates in cattle were identified by literature search (Web of Science, PubMed, Science Direct and Google Scholar) using the combination of keywords “progesterone” and “cattle” or “cow”. Studies included in the analysis needed to meet the criteria of being carried out in dairy or beef cows or heifers, reporting reproduction outcomes in sufficient detail for analysis and containing an appropriate control group. Published data in journal papers were crosschecked with conference papers to avoid repetition of data.

Extensive literature searches revealed a total of 55 papers and abstracts reporting studies where direct progesterone supplementation had been undertaken during early pregnancy (treatment started before Day 21) in cattle. Two studies were excluded as one had no control group [9] and another because treatment was started after pregnancy had been diagnosed [10]. The remaining 53 papers (Supplementary Table 1), reporting 84 specific progesterone therapies applied to a total of 9135 treated cows, were then analyzed.

### 2.2. Summary of experimental animals and approaches represented in the analysis

Papers were published between 1953 and 2014 and reported studies carried out in 16 different countries in Asia, Europe, Oceania, North and South America. Data extracted from studies included number of treated and control cows, conception or pregnancy rates, treatment protocol (start and end day of progesterone treatment, administration method), premating, and mating protocol (mated at natural estrus or synchronized estrus or synchronized ovulation). Progesterone was administered as range of natural and synthetic progesterone and assorted

progestagens by a variety of routes of administration (intravaginal, oral or injection). Treatment was started between Days 0 (estrus) and 14 with a range of single, repeated, and continuous treatment approaches. Across the various studies, treatment was ended on as early as Day 0 and as late as Day 34. Although a number of reported studies did not specify the types of animals treated, in those that did treatment was administered to both beef and dairy cows and/or heifers and to both lactating and nonlactating animals. Once again, Although some studies made no reference to the reproductive status of the experimental animals, in those that did, animals were classified as “fertile” or “sub fertile” or “repeat breeders” though the definitions used were not consistent, and for the purpose of the present analysis, conception rate in the control group was used to define reproductive status of the experimental animals in a particular trial. Although many of the studies were carried out before the discovery of cycle synchronization technologies with mating at natural estrus, in more recent studies, a number of trials used either cycle synchronization protocols, or in some more recent studies, full synchronization of ovulation coupled with fixed time AI. For the purpose of the present analysis, the term pregnancy rate was used to define the successful establishment of a pregnancy. However, it should be noted that, where specifically stated, pregnancy diagnosis took place at a range of times between Days 25 and 100 after insemination.

### 2.3. Classification of studies for analysis

Three separate analyses were undertaken. In the first analysis, studies were classified according to the mating protocol:

- natural estrus,
- synchronized estrus,
- synchronized ovulation.

In the second analysis, studies were classified according to the time that treatment was initiated in relation to anticipated physiological endogenous progesterone phases. These were:

- before the postovulatory progesterone rise (<Day 3),
- during the postovulatory progesterone rise (Day 3–7),
- following the postovulatory progesterone rise (>Day 7).

In the third analysis, studies were classified according to the conception rate of the experimental animals in the control group giving categories of:

- very poor (<30%),
- poor (30%–45%),
- good (45.1%–60%),
- very good (>60%).

Furthermore, in the classification groups in which positive results were identified and where the necessary information was provided, analysis of the influence of both type of progesterone used (progesterone or progestogen)

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