



Concentrations of steroid hormones, estrous, ovarian and reproductive responses in sheep estrous synchronized with different prostaglandin-based protocols



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ABSTRACT

To determine estrous, ovarian and reproductive responses after different prostaglandin (PG)-based protocols, ewes were assigned to groups PG10, PG12, PG14 or PG16 (two PG injections administered 10, 12, 14 or 16 days apart; respectively). Experiment I ($n = 132$) was conducted to evaluate the estrous response, ovulation rate (OR), conception and fertility. Experiment II ($n = 24$) was conducted to evaluate ovarian follicle growth, steroid concentrations and the interval from the second PG injection to estrus (PG-estrus) and ovulation (PG-ovulation). Estrous response was less with the PG16 ($P < 0.05$) treatment, and the extent of estrous synchrony was greater with the PG10 and PG12 treatments. Ovarian follicle growth and the intervals for the variables PG-estrus, PG-ovulation and OR were similar among groups ($P > 0.05$). From 8 to 4 days before estrus, progesterone (P4) concentrations were greater for the PG14 and PG16 than for the PG10 and PG12 ($P < 0.05$) groups. There were more days where concentrations of P4 were above 3.18 nmol/L with the PG14 and PG16 than PG10 and PG12 ($P < 0.05$) treatments. Use of the PG14 and PG16 treatments resulted in greater estradiol (E2) at estrus and 12 h later than use of the PG10 and PG12 treatments. A positive correlation was observed between the duration of the luteal phase and maximum E2 concentrations, and between duration of the luteal phase and days with E2 concentrations above 10 pmol/L. Conception and fertility were greater with use of the PG14 compared with PG10 and PG12 ($P < 0.05$) treatments. The administration of two PG injections 10, 12, 14 or 16 days apart resulted in different durations of the luteal phase that were positively associated with E2 concentrations and the reproductive outcome. The shorter luteal phases were associated with greater synchrony in time of estrus. The intervals for the variables PG-estrus, PG-ovulation and OR were similar among groups.

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1. Introduction

Timed artificial insemination (TAI) represents a practical tool in genetic programs, but requires hormonal treatments that ensure a synchronized time of ovulation and acceptable pregnancy rates (Menchaca and Rubianes, 2004). Progestagen-based protocols are the

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preferred option by technicians and farmers to manage flock reproduction (Gordon, 1999) even though there are potentially environmental and tissue contamination risks due to residues of progestagen devices, as well as risks with use of eCG, or the addition of antibiotics to avoid vaginitis (Gonzalez-Bulnes et al., 2005; Viñoles et al., 2011). Also, progestagen based estrous synchronization protocols have been associated with alterations in oocyte quality that can result in lesser fertilization rates and impaired embryo development (Gonzalez-Bulnes et al., 2005; Berlinguer et al., 2007). Because consumers demand foods produced by “clean, green and ethical” guidelines (Martin et al., 2004), prostaglandins became a desirable alternative because lungs rapidly metabolize the drug hence it does not accumulate in tissues of treated animals (Piper et al., 1970; Davis et al., 1980). Furthermore, prostaglandin-based protocols are easily applied by intramuscular injection, thus improving animal management and welfare (Abecia et al., 2012), and are more economically feasible compared with intravaginal devices plus eCG in sheep production enterprises.

Prostaglandin $F_{2\alpha}$ and its synthetic analogues (PG) have been widely studied since its discovery in 1970 as a powerful luteolytic agent (McCracken et al., 1970). Different alternatives of PG-based protocols have been used to synchronize time of estrus in sheep for TAI (rev: Fierro et al., 2013). However, use of most of these treatments is associated with lesser pregnancy rates compared with use of progesterone-eCG based protocols (Boland et al., 1978; Olivera-Muzante et al., 2011a; Viñoles et al., 2011).

Traditional PG-based protocols consist of two PG injections administered 9–14 days apart (Fierro et al., 2013), however, there is considerable variability in timing of estrous onset and ovulation (Acritopoulou et al., 1978; Loubser and van Niekerk, 1981; Houghton et al., 1995; Viñoles and Rubianes, 1998), that limit the practicality of use of these protocols for TAI programs (Menchaca and Rubianes, 2004). When a PG-based protocol of two injections given 7 days apart is used (Rubianes et al., 2004), a highly synchronized time of estrus and timing of ovulation are observed (Rubianes et al., 2003; Menchaca et al., 2004), but undesirable pregnancy rates are often achieved that are related to an altered profile and lesser progesterone (P4) concentrations that in turn result in a lesser ovulation rate (OR), fertility and prolificacy compared with what occurs when inseminations occur as a result of spontaneous estrus (Fierro et al., 2011). Attempts to develop alternatives to improve this protocol (two injections given 7 days apart) have not been successful (Olivera-Muzante et al., 2011b, 2013; Fierro et al., 2013, 2014).

Lesser fertilization rates were reported when the interval between PG injections was reduced from 14 to 8 days (Fairnie et al., 1977). Furthermore, treatment with a supplemental P4 source provided via an intra-vaginal impregnated device that is inserted 8 days before the PG injection increased the number of ewes in estrus (93.4% compared with 82.0%), and pregnancy rate (84.9% compared with 75.3%) than occurred with untreated ewes (Loubser and van Niekerk, 1981). The interval between PG-injections has been defined as important by Fairnie and Wales (1980), and other reports indicated that the inter-

val between PG injections should not be reduced to less than 11 days (Greyling and van der Westhuysen, 1980) or 13 and 14 days (Fairnie et al., 1978). Similarly, Fierro et al. (2013) suggested that the extension of the period between PG injections (with the second PG applied during the mid-to-late luteal phase) may contribute to developing a protocol to prolong the exposure of follicles from which ovulation occurs to adequate P4 concentrations during the growth phase of these developing follicles. The evaluation of longer periods between PG injections is necessary to understand the physiologic processes (estrous response, hormonal profile, time of ovulation) when these protocols are applied.

To the best of our knowledge, there are no previous studies that compared the concentrations of steroid hormones, as well as estrous, ovarian and reproductive responses in sheep where time of estrus was synchronized with use of different PG-based protocols under the same conditions (breed, photoperiod, nutrition, and health management). In the present study, the working hypothesis was that the administration of two PG injections 10, 12, 14 or 16 days apart would allow for development of an estrous synchronization protocol that resulted in desirable pregnancy rates in ewes. The aim of these experiments was to study ovarian follicular growth, concentrations of steroid hormones, estrous response, ovulation time, OR, conception and fertility rates after estrous detection when different PG-based protocols were used for sheep estrous synchronization. This information is necessary to identify which of these protocols are most effective if used in TAI programs.

2. Materials and methods

Experiment I was conducted at Escuela Agraria “La Carolina” (33° S–57° W; Flores, Uruguay), and Experiment II at Estación Experimental “Dr. Mario A. Cassinoni” (32° S–58° W; Paysandú, Uruguay) during the breeding season for ewes at these locations (March to April). The experimental procedures were approved by the Universidad de la República’s Animal Ethics Committee (CUEA-Universidad de la República, Facultad de Veterinaria, Exp: 111400-000079-12).

2.1. Animals and management

2.1.1. Experiment I

Multiparous Corriedale ewes (older than 2.5 y old, $n = 132$), in a moderate body condition (3.2 ± 0.4 , scale 0–5; Russel et al., 1969) and weighing 51.5 ± 6.2 kg were used. The flock used had an OR, based on previous records for this flock, that ranged from 1.20 to 1.61 (Fierro et al., 2011; 2014). Ewes grazed pastures that are typically used for sheep production in the region where the studies were conducted with more than 600 kg of dry matter (DM) available per hectare (8% CP and 8.5 MJ ME/kg DM), and water was available *ad libitum*.

2.1.2. Experiment II

Multiparous (older than 2.5 y old, $n = 21$) and nuliparous (1.5 y old, $n = 3$) Corriedale ewes, in a moderate body condition (3.1 ± 0.4 and 2.6 ± 0.1), and weighing 51.7 ± 5.3 kg

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