



Effect of location and stage of development of dominant follicle on ovulation and embryo survival rate in alpacas

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ABSTRACT

This study was designed to determine the effect of location of the preovulatory dominant follicle and stage of ovarian follicle development on ovulation rate and embryo survival in alpacas. In Experiment 1, mature lactating alpacas were randomly assigned to one of two groups according to the location of the dominant follicle detected by ultrasonography: (a) Right ovary (RO, $n = 96$) or (b) Left ovary (LO, $n = 108$). All females were mated once by an intact adult male. Ovulation rate, CL diameter and embryo survival rate (heart-beat) were assessed by ultrasonography on Days 2 (Day 0 = mating), 8 and 30, respectively. Ovulation rate (96.5 and 96.3% for RO and LO group, respectively), corpus luteum (CL) diameter (10.2 and 10.6 mm for RO and LO group, respectively) and pregnancy rate (60.2 and 56.7% for RO and LO group, respectively) did not differ among groups. In Experiment 2, lactating alpacas ($n = 116$) were submitted to ultrasonic-guided follicle ablation to synchronize follicular wave emergence. Afterwards, daily ultrasonography examinations were performed and females were randomly assigned to the following groups according to the growth phase and diameter of the dominant follicle: (a) early growing (5–6 mm, $n = 27$), (b) growing (7–12 mm, $n = 30$); (c) static (7–12 mm, $n = 30$), or (d) regressing phase (12–7 mm, $n = 29$). All alpacas were mated with a proven intact male, except five alpacas from early growing group that rejected the male. Females were examined by ultrasonography on Day 2 (ovulation rate), Day 8 (CL diameter), and Days 15, 20, 25, 30 and 35 (embryo survival by the presence of embryo proper and heartbeat). No differences were detected in ovulation rate among groups (96%, 97%, 100%, and 97%) or in CL size (10.3, 11.7, 11.1, and 11.1 mm, for early growing, growing, early static and regressing, respectively). Although, embryo survival rate at Day 35 after mating was numerically greatest in growing (65.5%), intermediate in early growing (52.4%) and static (53.3%), and least in regressing phase (42.9%), there were no differences among groups. Results suggest that neither location nor stage of development of the dominant follicle has an influence on ovulation and embryo survival rate in alpacas.

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1. Introduction

Reproductive efficiency in alpacas inhabiting the Peruvian High Andes is affected by a rate of embryo mortality (Bravo et al., 1987, 2010; Fernandez-Baca, 1970). Previous alpaca studies (Bravo et al., 1987; Fernandez-Baca,

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1970) have reported that embryo loss in these species ranges from 50% to 80% by Day 30 and 80 of gestation, respectively. Nutritional constraints, hormonal imbalance, inadequate mating systems, improper day of mating and chromosomal aberrations could be responsible for such embryo loss (Sumar and Adams, 2006). Either in llamas (Sumar and Leiva, 1979) or alpacas (Fernandez-Baca et al., 1973) ovulation takes place at similar rates in the right or left ovary. However, more than 98% of fetuses from both species occupy the left uterine horn (Bravo and Varela, 1993; Fernandez-Baca et al., 1973), indicating that embryos conceived in the right uterine horn must migrate to the left to maintain the gestation (Sumar and Adams, 2006). Considering that luteal phase is short in these species (Adams et al., 1989, 1990; Sumar and Bravo, 1991; Sumar et al., 1988) embryos originating in the right horn have a narrow period of time to achieve migration to the left side and send a signal to the uterus to avoid the maternal luteolytic mechanism. As found for specimens obtained from abattoirs, embryos derived from right ovulations do not compromise pregnancy rate in alpacas and llamas (Fernandez-Baca et al., 1973). However, due to the nature of this previous study, the viability of the embryos that had migrated could not be determined.

Location of preovulatory follicle could have an effect on embryo survival, because embryos originating from the right or left sides have obviously different challenges to overcome during early pre implantation stages. The use of heartbeat as a sign of early embryo viability using transrectal ultrasonography may be a powerful diagnosing tool to elucidate whether embryos derived from right ovulations affect pregnancy rate in alpacas.

Alpacas are considered reflex or induced ovulators and, therefore, do not show regular estrous cycles (Fernandez-Baca, 1970). In alpacas, ovarian follicle development follows a wave-like pattern (Bravo and Sumar, 1989; Vaughan et al., 2004). Once one of the follicles has become dominant it continues to grow (growing phase) until it reaches the pre-ovulatory size (7–12 mm), where it will remain (static phase) 4–5 days until eventually decreasing in diameter (regressing phase) if ovulation is not induced (Vaughan et al., 2004). Females with follicles ≥ 7 mm during the growing phase and with mature static follicles (8–12 mm) ovulate after mating, whereas females with small follicles (4–5 mm) or a regressing dominant follicle do not (Bravo et al., 1991). Mating management in the High Andes depends on female sexual receptivity; however, sexual receptivity was not related to the stage of development of the dominant follicle, which results in the breeding of females with an unknown follicular stage that could compromise ovulation and subsequently gestation (Sumar et al., 1993; Vaughan et al., 2003). Accordingly, in a previous stage of follicular synchronization study in llamas (Ratto et al., 2003), ovulation was not affected between follicular stage non-synchronized and synchronized females, but pregnancy rate was greater in the latter group, suggesting that the stage of the dominant follicle may play an important role in oocyte competence.

The purposes of the present study were to determine the effect of the dominant follicle location (right or

left ovary) and the stage of development of the dominant follicle (early growing, growing, static and regressing phase) at mating on ovulation and embryo survival rate in alpacas.

2. Materials and methods

Two experiments were conducted during the breeding season (January–March) at the Quimsachata Research Station, in the Department of Puno, Peru ($15^{\circ}04'S$, $70^{\circ}18'W$, and 4500 m above sea level). Alpacas were kept on natural pastures and housed in outdoor pens during nights. All females were subjected to the same management and handling conditions.

2.1. Experiment 1

Mature lactating female alpacas ($n=204$), ≥ 3 years of age, weighing between 57 and 62 kg (59 ± 3 kg) without any history of recent dystocia and with a resting period ≥ 25 days after birthing, were examined daily by transrectal ultrasonography using a B-mode scanner with a 7.5 MHz linear array-transducer (Aloka SSD 500; International Clinics, Chile) as previously described (Adams et al., 1989, 1990). Females with a follicle ≥ 7 mm in diameter that had grown for 3 consecutive days were classified according to the site of location of pre-ovulatory follicle in two groups: (a) Right ovary (RO, $n=96$) and (b) Left ovary (LO, $n=108$). Afterwards, each female was mated (Day 0 = day of mating) once with an intact male, under continuous observation. Different males ($n=70$) were used for the breeding of both groups of alpacas. All females were examined by transrectal ultrasonography on Days 2, 8 and 30 after mating to determine the occurrence of ovulation, presence and size of the CL and embryo survival rate, respectively. Ovulation was defined as the disappearance of the dominant follicle detected on Day 0 and was confirmed by the detection of a corpus luteum on Day 8 (Adams, 1991). Only females with single ovulation were used in this experiment. An embryo was considered viable when heartbeats were detected by ultrasonography.

2.2. Experiment 2

Mature lactating female alpacas ($n=116$), ≥ 3 years of age and weighing 58–64 kg (58 ± 4 kg) without any history of recent dystocia and with a resting period ≥ 30 days after birthing, were submitted to ultrasonic-guided follicle ablation from all follicles ≥ 5 mm to synchronize stage of follicular wave emergence similar to that described previously (Ratto et al., 2003). After follicle ablation, alpacas were examined daily by transrectal ultrasonography to detect follicular wave emergence and to follow the growing pattern of the new dominant follicle. Based on a previous study (Ratto et al., 2003), follicular wave emergence was expected to occur 1.5 days after follicle ablation. Females were assigned according to the stage of follicle development, based on previous alpaca studies (Bravo et al., 1990; Bravo et al., 1991; Vaughan and Tibary, 2006), to one of the following groups: (a) early growing phase ($n=27$): alpacas with a growing follicle of 5–6 mm in diameter, (b)

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