

Ovarian function in pony mares undergoing porcine zona pellucida immunocontraception

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An advantage of the porcine zona pellucida (pZP) vaccine over other immunocontraceptives is the preservation of reproductive cyclicity and associated behaviors. Few studies have investigated ovarian function following pZP vaccination in the mare despite reported ovarian dysfunction in other species. The objectives of this study were to investigate ovarian function and estrous cyclicity in pony mares during immunocontraception with the conventional pZP vaccine. Fourteen mares were randomized into two groups of seven. Group I received 100 µg of pZP with Freund's complete modified adjuvant (FCMA; V1), followed after five weeks by booster vaccination with 100 µg of pZP with Freund's incomplete adjuvant (FIA; V2). Group II (controls) received two treatments five weeks apart of saline with FCMA (V1) and saline with FIA (V2) respectively. Treatments were administered via intramuscular injection into the gluteal muscles. Data were collected by an investigator blinded to treatment group over a period of 24 weeks during the physiological breeding season. All mares underwent estrus monitoring via trans-rectal palpation and ultrasound examination of the internal reproductive tract, on D0 (day of ovulation), D7, and D14 of consecutive estrous cycles, with daily monitoring between D14 and D0 of the following cycle. Artificial insemination was performed using fresh semen for up to two consecutive estrous cycles, commencing five weeks post-V2. Serum samples were collected weekly for the analysis of antibody titres and ovarian steroid (progesterone and estradiol) levels. Data were compared using Mann-Whitney U tests using commercially available software (IBM SPSS Statistics Version 22, International Business Machines Corp., Armonk, NY). Statistical significance was set as $P < 0.05$. All Group II mares showed normal estrous cyclicity throughout the study. Four Group I mares showed signs of anestrus within seven weeks of V2, characterised by small, inactive ovaries and baseline progesterone and estradiol levels. One Group I mare entered anestrus within 11 weeks of V2 and a second showed estrus with ovulation between variable periods of anestrus. Ovarian volumes, follicle counts and maximal follicle diameters in Group I were significantly lower than Group II. Per-cycle pregnancy proportions in Groups I and II were 0% and 78% respectively. This study demonstrated suppression of ovarian function in six of seven (86%) mares following pZP immunocontraception. Further research into the mechanism of action of zona pellucida-based vaccines is warranted.

Keywords: *Equus caballus*, mare, immunocontraception, porcine zona pellucida, ovarian function