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Seminal Plasma Induced Ovulation in the One Humped Arabian Camel (*Camelus Dromedrius*)

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ABSTRACT. Fifty females and six males one-humped Arabian camels were used in the breeding season to determine factors inducing ovulation. After insemination ovaries were checked for ovulation by rectal palpation. The results indicated that ovulation was induced by seminal plasma but not by the spermatozoa, and the incidence of ovulation after insemination was 80%. Sixty percent of the females ovulated by 36 h after insemination and the rest by 48 h. The least amount of semen required to elicit ovulation was about 1.0 ml. Intramuscular injection of LH, hcG and LHRH also caused ovulation, even in females that did not ovulate in response to insemination.

Introduction

Reproduction is often the key to improved livestock performance. Reproduction in the camel is not as well understood as in more common species of domestic animal (Merkt *et al.,* 1990). Puberty in females occurs at 3 to 4 years of age and the first calf is born when the mother is 5 to 6 years old. Females remain sexually active for 20 to 30 years (Yagil, 1985). It is a common practice to withhold female camels from breeding until they are 4 to 6 years and the age at first calving would be 5 to 7 years. Because camels can live up to 40 years, it is possible to produce a number of calves similar as are possible for cow (Musa and Merkt, 1990).

Puberty in males occurs at 6 years of age and good service ability is maintained until 18 to 20 years (Novoa, 1970 and Wilson, 1984). The female camel

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is considered to be a seasonal breeder with a marked peak in sexual activity (Wilson, 1984). Factors that affect the beginning of the breeding season, its duration and intensity of sexual activity include: local climatic condition, nutrition and management. It may be pointed out that, the male camels are sexually active for a few months each year, the so called rutting. The time of rutting season differs from region to region (Yagil, 1985).

In the Libyan dromedary the breeding season start in October (El Tamour) and terminates in March (El Rabia), (Zaid *et al.,* 1991) .

Novoa (1970) showed that sexual activity in camelids appeared to be acyclic; females did not have estrus cycles comparable to those of spontaneous ovulators. Chen *et al.* (1980) indicated that female bacterian camels exhibited follicular cycles, the follicles developing and regressing one by one. It took 19.10 ± 4.25 days from the start of development of follicle (~ 0.5 cm in diameter) to begin its regression. One mature or developing follicle was usually present in an ovary. When not allowed to mate, the camel manifested prolonged period of estrus however; if mating occurred, ovulation would take place in 30-48 h later. Shalash and Nawito (1964), Shalash, (1980) suggested that copulation, mechanical or electrical stimulation of the cervix and other afferent stimuli would be necessary for ovulation in the dromedary. San Martin *et al.* (1968) reported that ovulation in alpaca was induced by coitus, and that follicles ovulated 26 h after coital stimulation or 24 h after intravenous injection of hcG. Fernandez-Baca *et al.* (1970) indicated that mounting accompanied by intermission were necessary to provide adequate stimulation for LH release and subsequent ovulation in alpaca. Musa and Abusineina (1978) reported that ovulation in the dromedary camel was not spontaneous and required coitus stimulation, and that manual stimulation of the cervix did not induce ovulation. Chen *et al.* (1983) reported that mounting and stimulation of the cervix did not elicit ovulation in the bacterian camel however, vaginal insemination did induce ovulation, but the numbers of animals were very small. Chen *et al.* (1985) reported that semen-induced ovulation in the bactrian camel. Agarwal and Rai (1995) reported that camel is an induced ovulators and needs mating stimulus for ovulation.

The present study was therefore undertaken to investigate the ovulation-inducing effect of semen in the one-humped Arabian camels .

Materials and Methods

Animals and Methods

Fifty breeding camels, 8-14 years of age, free from any detectable genital abnormalities and 6 high fertile male camels, 5-10 years of age, were kept in separate areas. The range was good and the camels were grazed or browsed freely

all the year round were given supplementary food with 2 kg grain every evening in the winter months. The average body weight of female camels was about 450 kg and that the males was about 580 kg .

From the beginning of the breeding season (started from October and terminated in March) development of follicles was followed daily by rectal palpation. When the follicles had reached a diameter of $\geq$ 12 m (Skidmor *et al.,* 1996), the females were treated with materials deposited intravaginally or by intramuscular injection of hormones. The ovaries were checked for ovulation by rectal palpation at 24, 36, 48 h after treatment.

Intravaginal Administration

A rubber inseminating tube of the type used for horse AI, was inserted gently and as deeply as possible into the vagina. The materials to be tested were injected through the tube by means of syringe .

Whole Semen

Semen of male camels was collected by means of artificial vagina as used for cattle. The semen was used immediately or stored at 0 to –10^{o}C for different lengths of time. The semen was thawed and warmed to 37^{o}C before use. The insemination volume was 0.5 to 7.0 ml. Bull, goat semen samples (2-3 ml) stored in the refrigerator (0 to –2^{o}C) was also tested.

Seminal Plasma

After collection, the semen was centrifuged (500 g), the supernatant was examined microscopically and when sperm count was < 10000 / ml, the seminal plasma was used on a volume of 3 ml (Chen *et al.,* 1985).

Washed Spermatozoa of High Concentration

After centrifugation of semen and removal of the supernatant, saline (9 g NaCl / 1) was added to the sediment and the suspention was recentrifuged. The procedure was repeated 3 times and the final pellet was examined. Washed spermatozoa were used at a concentration 4-7$\times$ 10^{8} ml and an insemination volume of 3 ml (Chen *et al.,* 1985).

Accessory Sex Gland Secretions

Male 6 was vasectomized, collection of semen started 6 days after operation and was repeated every 2 days until for a period of 18 days (Chen *et. al.,* 1985).

Skim Milk

As control, skim milk from a cow was deposited into the vagina in a volume of 6 ml.

Natural Mating

Male camels were allowed to copulate and 4-10 min after the end of coitus, the duration of which was about 3 min, semen was flushed out of vagina with saline.

Exogenous Hormones

When ovulation did not occur in response to intravaginal treatment (except for the control) hormones were injected i.m. to determine whether the materials were ineffective or the follicle were unable to ovulate. The hormones used were LH (Wuhan Biochem Pharmaceutical Co., Hankow, China), 300 i.u in 4 ml saline: hcG (Tong-Feng Pharmaceutical Co., Beijing), 1000-2000 i.u. in 4 ml saline and LHRH analogue (Biochemical Institute – Shanghai), 250-500 μg in 2-4 ml saline.

Results

Ovulation-Inducing Effect of Components of Camel Semen

Whole semen: Eight females from ten inseminated ovulated (80%). The least amount of semen required for inducing ovulation was about 1.0 ml. All females inseminated with 1.0 ml semen or less (0.1-0.2 ml semen was left in the inseminating tube) ovulated. One semen sample stored at about –10°C for two months was effective in inducing ovulation. The ovulation time was similar to that after natural service, with 6 (60%) females ovulated after 36 hr and 3 (30%) having ovulated by 48 hr. While only one female (10%) did not ovulate. There was no differences in the effect on ovulation when the semen samples were fresh and contained live spermatozoa (9/10 ovulating) or had been frozen at –10°C for several days and contained dead spermatozoa (8/10 ovulating).

Seminal plasma: Among 10 females inseminated 8 ovulate.

Washed spermatozoa: None of 10 females inseminated with high concentration washed spermatozoa ovulated.

Accessory sex gland secretion: All females (10) inseminated (including 4 natural services by the vasectomized male), ovulated.

Control substance: None of the 10 females tested with skim milk ovulated.

Natural service: Among five females given natural service followed by flushing of vagina, four did not ovulate.

Effect of bull and goat semen: Among five females inseminated with bull semen, 2 had ovulated after 36 hr and 2 had ovulated after 48 h. Five females inseminated with goat semen did not ovulate.

Ovulation-inducing effect of semen of individual male camels in this part of the present study 6 male camels were used and the effect of their semen on

ovulation is summarized in Table 1. Camel 3 appeared different from the others (fewer females ovulated in response to its semen). Cystic follicle were detected in the ovaries of 2 of the females treated with semen of this male, and also 4 females that had not ovulated in response to male 3 did ovulate in response to hcG or LHRH injection, indicating that the follicles were able to ovulate .

TABLE 1. Ovulation inducing effect of individual male camel.

Male	Insemination		Mating	
	No. of ♀ tested	No. of ovulation	No. of ♀ tested	No. of ovulation
1	3	3	1	1
2	14	13	6	6
3	15	9	5	3
4	11	8	3	3
5	4	3	2	2
6	3	3	–	–

Reaction of individual female camel to semen: Out of fifty female camels investigated, 5 showed abnormal ovulatory response. One female suffered from chronic cervicies, and another had cystic follicles after insemination with semen of fertile male camels in two successive seasons. The other 3 females did not ovulate in response to insemination, but did ovulate after injection of the hcG; other female camels reacted to semen of the same male and ovulated.

Ovulation-inducing effect of some reproductive hormones: The results of intramuscular injection of hormones are summarized in Table 2. The interval between injection and ovulation in all cases was 36 h.

TABLE 2. Ovulation-inducing effect of reproductive hormones.

Hormone	Dose	No. of female tested	No. of ovulation
LH	300 i.u.	5	5
LHRH	250 µg	4	4
LHRH	500 µg	5	4
hcG	1000 i.u.	2	2
hcG	1500 i.u.	3	2
hcG	2000 i.u.	3	3

Discussion

Results indicated that ovulation in Arabian one-humped camels is not spontaneous and is induced by seminal plasma. Spermatozoa were not effective in inducing ovulation. This conclusion agrees with those obtained by Agarwal and Rai (1995) and Chen *et al.* (1985), who reported that, insemination of seminal plasma can cause ovulation indicating the presence of some ovulation inducing factor in it. Bull semen also seemed to contain the ovulation inducing factors in our study however, goat semen did not. The nature of the ovulation inducing factor was found similar to GnRH (Agarwal and Rai, 1995), The relation of ovulation with reproductive hormones was described by previous authors, who found that the preovulatory peak of LH and the postovulatory peak of progesterone seem to be important indicators of ovulation .

Ovulation can be induced by luteinising hormone (LH), human chrionic gonadotrophin (hcG) and gonadotrophin releasing hormone (GnRH) (Musa *et al.,* 1993). However, the nature of the inducing factor and the mechanism of stimulation of LH release after its absorption remains to be clarified. The vagina or the uterus may be the place of absorption, since intrauterine insemination of females also led to ovulation. Absorption may be very rapid because 1 of 5 females ovulated after mating though the semen was flushed from the vagina 4-10 min later.

In the present study ovulation occurred in 23/31 (74.19%) camels after insemination (Chen *et al.,* 1985 reported 41/47 = 87%) although in the previous work (Chen *et al.,* 1980) reported that, ovulation took place in all of 26 females mated naturally. The mating behavior might therefore have some augmentative effect on the semen-induced ovulation. There are also variations in the response of individual females and the ability of particular males to induce ovulation (Chen *et al.,* 1985).

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التبـويض يستحث ببـلازمـا السائـل المنـوي في الجمال العربية وحيدة السنام

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قسم الإنتاج الحيواني ، كلية الزراعة ، جامعة عين شمس ، حدائق شبرا القـــاهـرة – مصـــر

المستخلص . لتحديد العوامل التي تسبب حدوث التبويض في الجمال العربية ذات السنام الواحد استخدام في هذه الدراسة ٥٠ أنثى و ٦ ذكور في موسم التلقيح . وبعد التلقيح بعينات من السائل المنوي في المهبل ، تم فحص التبـويض عن طريق الجس المستقيمي . ولقد أظهرت النتائج أن التبويض في إناث الجمال (النوق) يستحث ببلازما السائل المنوي ، وليس بالحيـوانات المنوية . وأظهرت النتـائج أن نسبـة حدوث التـبويض بعد التلقيح كانت ٨٠٪ . ولقد اتضح أن معظم الإناث (٦٠٪) حدث لها تبويض بعد ٣٦ ساعة من التلقيح ، وبقية الإناث حدث لها تبويض بعد ٤٨ ساعـة . واتضح أن أقل كـميـة من السـائل المنوي يمكن أن تحـدث التبـويض هي ١ ملل. وأظهرت النتائج أيضًا أن الحقن بهرمونات LH ، hcG ، LHRH يسـبب حـدوث التبـويض حـتى في تلك الإناث التي لم يحدث لها تبويض نتيجة حدوث التلقيح .

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