ULTRASONOGRAPHIC ASSESSMENT OF FOLLICULAR SIZE TO REDUCE THE AGE OF FIRST SERVICE IN THE DROMEDARY HEIFERS

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ABSTRACT

In the present study we attempted to breed dromedary camel heifers (n= 68) at a younger age compared to the common practice of breeding camel heifers at 4 years of age. In the first experiment, heifers (n=15) were maintained on different nutritional groups viz. intensive system with fodder (ISF; n=5), intensive system with extra protein (ISP; n=5) and semi-intensive system with fodder (SIF; n=5) starting from 2.5 years of age. In experiment II, heifers (n=18) were maintained under SIF (n=12) and ISF (n=6) and in experiment III, heifers (n=35) were maintained under SIF. The heifers in all experiments were evaluated every 10th day during the breeding season by transrectal ultrasonography and those evidencing a follicle (diameter ≥ 9 mm) were mated with virile stud camels. Ultrasound examination revealed follicular growth diameter ≥ 9 mm in 14/15 (93.3%), 18/18 (100%) and 31/35 (88.5%) of the camel heifers in Experiments I, II, III, respectively, at 3 yr ± 2 months of age and weighed at least 360 kg; these were successfully mated and 28 out of 68 heifers conceived at approximately 3 yr and delivered calf at around 4 yr age. The age of first service was positively correlated with body weight, heart girth and body girth. It was concluded that supplementary feeding of camel heifers had positive effects on the body weight and reduced the age at first service. Camel heifers attained puberty at 3 yr of age and >360 kg body weight and were successfully bred through ultrasonographic monitoring of follicle.

Key words: Age of first service, body measurements, Camelus dromedarius, heifers, ultrasound for ovarian status

The reproductive performance of Camelus dromedarius is termed poor because of limited breeding season, lengthy gestation period and delayed puberty (Purohit and Pareek, 2000; Vyas et al, 2004a; Purohit et al, 2020). Improved reproductive performance is required in camel to provide an opportunity for selection, genetic improvement and profitable production. The reported age at puberty in the female camel varies widely; 3-4 yr (Musa et al, 1993), 5 yr (Rathore, 1986) of age. It is reported to vary from 4-6 yr for camels maintained at farm (Khanna et al, 1990) and at field level from 5-7 yr (Arthur, 1985). The age of first service at an organised farm is reported to vary between 1679.5±49.1 to 1863±44.1 days (NRCC, 2012). In India, dromedaries are not bred until 4 yr of age, resulting in an age at first calving of 5 yr or more (Khanna et al, 1990; Agarwal et al, 1996). In Saudi Arabia, majority of camel herds breed females at 4 yr (77/115 herds, 67%) and fewer at 5 yr (30/115 herds, 26%) and less

than 7% at 3 yr (8/115 herds) (Ali *et al*, 2018). Tura *et al* (2010) observed that the age at first calving was 60 months in camels in Kenya implying that 47-48 months was the age of first service. Reducing the age of first service can improve the reproductive performance and quality of camel herds. The prospect of increasing the reproductive performance of camels is impeded by the persistent use of traditional systems of reproductive management in most breeding herds (Al Eknah, 2000; Almutairi *et al*, 2010; Skidmore *et al*, 2010). The lack of strategy to improve age at first calving affects reproductive efficiency and productivity of camel herds (Gherissi *et al*, 2020).

Detecting oestrus efficiently and accurately is one of the key factors for reproductive success in camels (Manjunath *et al*, 2015). The oestrus behaviour in female camel is not characteristic and is rather vague because it does not correlate with the ovarian status or presence of follicle over ovaries (Vyas *et al*, 2004b). The male can be observed following

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females, sniffing their vulva exhibiting flehmen and these females also exhibit signs like frequent urinating, straddling hind legs and even sitting beside a copulating male, but upon ovarian examination, follicular activity was not observed in many of females exhibiting such behaviour (Skidmore et al, 1996; Vyas et al, 2008). At many times, male suddenly select a female which is not receptive, running away from male and chase her and force her to sit and engage in copulation. These females, on examination, were reported to possess mature follicle. The information about ovarian status, bodyweight and body size in relation to age at puberty is lacking in camel (Mahla et al, 2015). Therefore, in the present study trans-rectal ultrasonography was used to detect follicular growth with the following objectives -

(i) To know the effect of intensive and semiintensive feeding systems on body measurements and follicular growth at 3 yr of age in dromedary camel heifers.

(ii) To ascertain the presence of follicle at 3 yr of age and its relation with body weight and other body measurements.

Materials and Methods

Experimental animals

A total of 68 dromedary camel heifers belonging to ICAR- National Research Centre of Camel, Bikaner situated at 28° 01′ N and 73° 22′ E and 228 m above sea level were utilised for the experiments.

Experiment 1 (first year)

Fifteen camel heifers (2.5 yr) were divided into three groups. Group I (ISF, intensive stall-fed fodder, n=5) camel heifers were stall fed 6 kg *Cymopsis tetragonaloba* crop residue (Guar phalgati*; CP- 8%; TDN- 48%) and not allowed to graze outside the stall.

Group II (ISP, intensive stall-fed pellets, n=5) camel heifers were fed 6 kg diet containing 20% protein and energy guar phalgati 50%, moong fali chara** (*Arachis hypogea*)- 20%, khejri leaves*** (*Prosopis cineraria*)- 5%, moong churi- 2%, crushed maize- 17%, mineral mixture- 2%, common salt-1% and molasses- 3% in pellet form and were not allowed to graze out side the stall.

Group III (SIF, semi-intensive fodder n=5) camel heifers were fed 6 kg *Cymopsis tetragonaloba* crop residue and were kept under semi-intensive

Ultrasound examination

The ultrasound examination was performed using linear probe (6.5 MHz, V-5 portable ultrasound machine, Med-India) with camels restrained in sitting posture as described previously (Vyas and Sahani, 2000). The heifers were examined when they attained the age of 3 yr ± 2 months at fortnightly intervals till a follicle was observed.

Body measurements

Body weight and body measurements like heart girth (HG), height at withers (HE), body length (BL) and tail length (TL) were measured (using a measurement tape) at the age of 3 yr as per standard protocol at the farm. HG was measured as the circumference of anterior base of hump and behind the sternum. HE was the measurement between wither or front base of hump and foot pad of fore limb. BL was the measurement between shoulder joint to pin bone (pelvic). BG was the circumference at posterior base of hump and anterior to udder. TL was the length of tail from the base. The measurements were done by same technician. The heifers were mated with virile studs when a follicle of ≥ 9 mm diameter was observed (Skidmore et al, 1996) and copulation time was recorded.

Experiment II (second year)

The camel heifers (age 2.5 yr) were either maintained under ISF (n=6, Group I) or SIF (n=12, Group II), examined with ultrasound at similar intervals and mated as in experiment I once they attained the age 3 yr.

Experiment III (third year)

The experiment III was planned in subsequent year for validation of findings of experiment II and camel heifers (n=35) were maintained under SIF condition. They were examined for ovarian status at fortnightly interval from the age of 3 yr±2 months. The heifers were mated with virile studs when a follicle of \geq 9 mm diameter was observed.

The heifers in all experiments were kept separated from male camels during the period of experiment including grazing period except at mating.

All animal experiments were conducted adhering strictly to institutional ethical management practices.

^{*} Vernacular name of Cymopsis tetragonoloba

^{**} Vernacular name of Arachis hypogea

^{***} Vernacular name of Prosopis cineraria

Statistical analysis

The ANOVA was used to analyse the results of experiment I and unpaired t test was used to compare the results of experiment II.

Results

Experiment I

The overall mean age at first service in camel heifers was 1147±5.93 days and body weight were 422.4±12.5 kg. The body weights attained in the three different groups were significantly different (P<0.05; Table 1). The heifers in ISF group were the heaviest. The age at first service (days) i.e., when follicle was observed for the first time was lowest in heifers under SIF, but difference was not statistically significant. Ultrasound examination revealed that 13 out of 15 females had follicle at 3 yr ± 2 months. The successful calving was higher in Group III. Camel heifers maintained under ISF and ISP attained higher body weights compared to SIF but there was no difference in age of the first service. The overall means of different body measurements like body length (BL), heart girth (HG), height at wither (HE), tail length (TL), body girth (BG) were 143.87±2.0, 203.2±1.4, 188±1.18, 55.47±0.78 and 146.1±1.39 cm, respectively. The body measurements did not differ among different groups.

The age of the first service was positively correlated with body weight, heart girth and body girth and the heart girth was positively correlated with body length (Table 2).

Experiment II

A high proportion (91.66%; 11/12) of camel heifers in SIF group revealed follicles at 3 yr±2 months and were mated with virile stud camels. Six out of these 11 heifers (54.54 %) delivered the calf. All camel heifers (100%) in ISF group revealed ovarian follicles, were mated with virile stud and two out of six heifers (33.3%) delivered the calf. The age at first service in SIF camel heifers was 1108±5.0 days and it was significantly higher (P<0.05) compared to ISF camel heifers (1062±13.6 days) (Table 3).

Birth wt of Age of first Feeding B. wt. (kg) CT (s) BL (cm) HG (cm) HE (cm) TL (cm) BG (cm) calves born service group (days) (kg) (n) 189.2±0.7 ISF 1162 ± 11.2 403.6±31.8 206±2.7 56.4±0.7 $500.6 \pm 21.4^{\circ}$ 146.6±4.0 146.6±2.8 36(1) (ISP 1143.6±10.3 471.8±16.3^b 424.6±119.6 143.2±3.6 202.6±2.5 188.8±3.0 54.8±2.2 148 ± 2.8 40 (2) SIF 1134.8±5.9 422.4±27.0^a 353±99.7 141.8±3.4 201±1.9 186±1.2 55.2±0.8 143.8±1.4 32.5 (1) Overall 1147±5.93 464.9±12.5 393.73±49.6 143.8±2.0 203.2±1.4 188±1.2 55.5±0.8 146.1±1.39 35 f 2.25 5.357 0.160 0.44 1.139 0.69 0.344 0.76 0.52 P value 0.147 0.021 0.85 0.65 0.35 0.72 0.48 F crit 3.88 3.88 3.88 3.88 3.88 3.88 3.88 3.88 2 df between within 14 MS 44.1

Table 1. Mean ± SEM of growth parameters and age at first service in camel heifers.

P<0.05; s- seconds; B.wt- Body weight; CT- Copulation time; BL-Body length; HG- Heart girth; HE- Height at wither; TL- Tail length; BG- Body Girth; ISF- heifers under intensive management with fodder; ISP- Heifers under intensive management with concentrate pellets; SIF- Heifers under semi-intensive management with fodder; n- number.

Table 2. Correlation of body measurements and age at first service in camel heifers.

| Correlation coefficient | | | | | | | | | | | | |
|-------------------------|----------------------|-------------|---------|---------|---------|---------|--------|--------|--|--|--|--|
| | Age of first service | Body weight | СТ | BL | HG | HE | TL | BG | | | | |
| age of first service | 1 | 0.3058 | -0.0182 | 0.1092 | 0.4595 | 0.262 | 0.3161 | 0.3062 | | | | |
| Body weight | | 1 | 0.1417 | 0.1649 | 0.562 | 0.1906 | -0.028 | 0.3605 | | | | |
| СТ | | | 1 | -0.0045 | -0.2088 | -0.0432 | 0.2242 | 0.0535 | | | | |
| BL | | | | 1 | 0.5649 | 0.1099 | 0.4195 | 0.7453 | | | | |
| HG | | | | | 1 | 0.4358 | 0.3704 | 0.6648 | | | | |
| HE | | | | | | 1 | 0.4696 | 0.4025 | | | | |
| TL | | | | | | | 1 | 0.2331 | | | | |
| BG | | | | | | | | 1 | | | | |

CT- Copulation time; BL-Body length; HG- Heart girth; HE- Height at wither; TL- Tail length; BG- Body Girth.

The difference in mean bodyweight at first service in SIF (402.17±8.7 kg) and ISF camel heifers (421.1±9.5 kg) was not significant.

Experiment III

Ovarian follicle (>0.9 cm) was observed in a high proportion of camel heifers (88.57%; 31/35) managed under SIF at the mean age of 1089±6.51 days. The mean body weight at the time of first service was 407.48±8.58 kg (range- 360-506 kg). They were mated with virile stud. Eighteen heifers (58.06%; 18/31) became pregnant and sixteen heifers delivered healthy calf at around four years of age.

Discussion

In the present study positive effects of intensive feeding on the body weight and body dimensions of camel heifers were recorded. ISF camel heifers had significantly (P<0.05) higher body weight and nonsignificantly higher body dimensions compared to ISP and SIF camel heifers, yet SIF camel heifers had the lowest age at first service. However, in subsequent trials (Expt. II) ISF camel heifers had non-significantly higher body weight and significantly (P<0.05) lower age at first service compared to SIF camel heifers. This reflects that supplementary feeding has positive effects on the body weight of camel heifers, yet the appearance of an ovulatory sized follicle was less closely synchronised with feeding in camel heifers. A previous study (Saini et al, 2014) found that supplementary stall feeding of young camels (330 kg, n=20) resulted in significantly higher body weight compared to camels allowed grazing only. Similarly, Bakheit et al (2017) noticed significant higher growth of male and female camel calves maintained under semi-intensive management from 3 to 18 months of age compared to those maintained under traditional management system. Another study observed no appreciable difference in weight gain in male camels receiving supplementary diet (13% crude protein) although supplemented camels had higher testosterone concentrations (Al-Saiady et al, 2013). Under pastoral management conditions in Algeria a recent study recorded that the age at first oestrus in camels was 31.07±8.97 months and the age at first mating was 35.52±8.55 months (Gherissi et al, 2020). The body dimensions at puberty recorded in the present study are similar to previous report in camel (Kamoun and Wilson, 1994). Ultrasound examination of camel heifers (n= 68) at the age of 3 yr±2 months with a body weight of at least 360 kg revealed presence of follicle in 63 out of 68 heifers. They were mated and 28 out of 63 heifers got conceived at 3 yr±2 months of age and delivered healthy calf at around 4 yr of age. This finding is like a previous study (Abdel-Rahim, 1997) that recorded puberty in stall fed two camel breeds at 336 to 360 kg body weight attained at 173 weeks of age.

In most management systems dromedaries are not bred until the female has almost reached her mature physical size at 4 years of age, resulting in an age at first calving of 5 yr or more (Khanna *et al*, 1990). Growth and weight of young females also seem to be important factors in the onset of ovarian activity and influence the chances of conceiving. Low reproductive performance in camels is mainly ascribed to higher age at first calving, long calving interval and limited breeding season (ElWishy, 1987). The age of puberty and first service is reported to be influenced by complex interactions among genetic, nutritional, environmental and economic factors (Perry, 2016).

Studies on puberty in the female dromedary are based on a few clinical and field observations (Agarwal *et al*, 1996). Inadequate body weight caused by lack of sufficient food appears to be the cause of delayed puberty in the camel (Chatty, 1972). Nutrition seems to play a vital role in the various physiological

 Table 3. Comparison of semi-intensive and intensive management systems on age at first service in camel in camel heifers (Experiment II).

| | Age at first service | | Body weight | | Calves born; mean Calf birth weight (kg) | | |
|------------------|--------------------------|--------------------------|-------------|-----------|--|-------------|--|
| | SIF | ISF | SIF | ISF | SIF | ISF | |
| Mean±SE | 1108.1±5.04 ^a | 1062.5±33.5 ^b | 402.2±8.7 | 421.2±9.5 | 6; 31.5±0.8 | 2; 32.5±1.5 | |
| Ν | 12 | 6 | 12 | 6 | 12 | 6 | |
| P value | 0.0014 | | 0.1963 | | 0.56 | | |
| t | 3.8494 | | 0.3485 | | 0.6124 | | |
| df | 16 | | 16 | | 16 | | |
| SE of difference | 11.84 | | 14.09 | | 1.633 | | |

ISF- heifers under intensive management with fodder; SIF- Heifers under semi-intensive management with fodder Values with different superscripted letters in a row are significantly (P<0.05) different.

events for the attainment of sexual maturity and in the reproductive process (Maynard, 1979). The overall pregnancy rate did not differ significantly between the herds where camel heifers were bred by the age of 3 yr or age > 3 yr (Ali *et al*, 2018).

Another report suggests that first service is allowed when the female reaches about 70 % of their adult body weight (Tibary and Anouassi, 1997). The average adult body weight in females of different Indian camel breed is reported to be 577.95±0.79, 537.0±11.66, 563.74±14.73 kg for Bikaneri, Jaisalmeri and Kachchi breeds, respectively (Khanna et al, 2004). The bodyweight of heifers considered for mating in the present study is 380 kg which comes to be 65.8, 70.7 and 67.5 per cent of adult body weight for Bikaneri, Jaisalmeri and Kachchi breeds, respectively. Breeding in camels based on visual observations appears to be difficult and sonographic presence of a follicle can be a better option for mating. In one previous study parturient camels were monitored ultrasonographically and when a follicle was observed these were mated resulting in pregnancies in 52.7% camels (Vyas et al, 2000). A recent study (Faraz et al, 2022) reported that the age at first service was 1091, 1109 and 1162 days in camel heifers that were forcefully mated with male camels. This reflects the complexity of camel breeding and probably the forcefully mated camel heifers had an ovulatory sized follicle yet did not evidence overt signs of oestrus or receptivity.

Conclusion

It was concluded that supplementary feeding of camel heifers has positive effects on the body weight and age at first service. Camel heifers attain puberty at 3 yr of age and >360 kg .and can be successfully bred if the follicle is monitored by ultrasonography.

Declaration of competing interest

Authors of the manuscript declare no conflict of interest for this research work.

Contributions of authors

SV - conceptualisation, methodology, investigation, execution, writing-original draft preparation; RKS- investigation, execution; MMAinvestigation, execution; KN- investigation, execution; BDK- investigation, execution, SV- Statistical analysis, GNP-Writing-reviewing and editing.

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