DOI : 10.5958/2277-8934.2023.00003.6 *Vol 30 No 1, p 19-24*

SUPEROVULATION IN CAMEL: STATE OF THE ART

Amir Niasari-Naslaji[1] and Darab Nikjou[2]
[1]Department of Theriogenology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran
[2]NiaNik Animal Technology, Tehran, Iran

ABSTRACT

Superovulation (multiple ovulation) and embryo transfer is relatively an old, cheap and efficient reproductive technique for taking advantage of an elite she-camel's genome. It starts with the selection and preparation of donors followed by the stimulation of ovaries (superovulation) to produce more ovulatory follicles. The rest of the procedure including mating and/or AI, fertilisation, and construction of zygote and embryo follow the same natural sequence of embryo production *in vivo*. The quality of embryo produced by this technique has nearly similar competence to the one produced normally during reproductive cycle in camel resulting in the birth of healthy and genetically selected calf. This review article summarises the majority of references used for superovulation in camel in order to sum up the approaches for synchronisation of elite donors, varying types of gonadotropins and the way of their administration in camel.

Key words: Follicular wave synchronisation, gondadotropins, multiple ovulation, superovulatory response

Camel has low reproductive efficiency resulting in low genetic progress (Skidmore, 2003). She-camel could deliver the first calf around 5 to 6 years of age (Merkt *et al,* 1990). Considering the calving rates of 40% (Wilson, 1984), she-camel may produce 7 to 8 calves throughout the life span of 15 years (Merkt *et al,* 1990). Therefore, embryo production technology could boost reproductive efficiency of she-camel.

Three main approaches were used to produce camel embryos. The first approach relied on *in-vivo* production of embryos and introduced since early nineties in camel (Anouassi and Ali, 1990; Cooper *et al,* 1990; McKinnon and Tinnson, 1992; Skidmore *et al,* 1992). This is the most accepted approach to produce camel embryos well known as MOET (Multiple Ovulation and Embryo Transfer). In this particular method, the elite donors receive FSH and/or FSH like factors during the procedure named superovulation to induce more follicles to grow. Then, the donor is mated and following fertilisation and construction of embryo *in vivo*, the uterus is flushed and the embryos are recovered, graded and transferred into synchronised recipients (Niasari-Naslaji *et al,* 2009, 2014; Ararooti *et al,* 2018a,b). Due to the relatively simple, unexpensive and good embryo quantality and quality, MOET is well established in several labs and became the main source of embryo production in camel (Anouassi and Tibary, 2013).

The second approach, introduced in 2006 in camel (Khatir and Anouassi, 2006), relied on the aspiration of oocyte from ovary either collected from abattoir or through ultrasound-guided transvaginal ovum pickup (OPU). Good quality oocytes go through maturation and fertilisation procedures to produce presumptive zygotes from which after several cleavage cycles, the embryos are produced *in vitro* (IVEP; Wani, 2021). Unfortunately, due to the special configuration of follicle in camel ovary (El-wishy, 1992) and high incidence of bleeding following OPU in camel, the *in vitro* production of embryos in camel did not become a popular approach for embryo production like in other species. Moreover, as a result of several abnormalities of embryos and health issues in newborns following *in vitro* production of embryos in cattle (Ealy *et al,* 2019), there could be a chance of such abnormalities in camel as well which requires further investigation.

The third approach, introduced in 2010 in camel (Wani *et al,* 2010), relied on somatic cell nuclear transfer (SCNT/cloning). In this particular technique, the somatic cell was transferred into enucleated oocyte. Following reprogramming of the somatic cell, the totipotent cells started to grow toward embryo production. Apart from low success rates and being relatively expensive technique in camel (100000 USD per calf), there are great concerns for the quality of embryos and the health of offspring produced by SCNT (Malin *et al,* 2022). Recently, there are great investment on producing an expensive cloned embryo in camel (Olsson *et al,* 2021); however, like other

SEND REPRINT REQUEST TO AMIR NIASARI NASLAJI email: niasarinaslajiamir@gmail.com

species, there might be several future health problems in cloned camels as well, mainly due to epigenetic modifications (Malin *et al,* 2022), which is neglected in camel at the present time.

In this review, superovulation was described in detail as the most important part of MOET programme in camel. Synchronisation of donors prior to superovulation, different types of gonadotropins, their advantages and disadvantages and the way of gonadotropin administration were elaborated.

Follicular growth in camel

Camel follicular growth occurs in a wave like pattern so called as follicular wave cycle (Nawito *et al,* 1967; Skidmore *et al,* 1995; Nikjou *et al,* 2009). There is no luteal phase in camel, therefore using the term "oestrous cycle" may not be appropriate in this species. Camel is an induced ovulator (Marie and Anouassi, 1987), seasonal breeder, displaying complete follicular wave cycles from November to March in most countries (Al Eknah, 2000). Each follicular wave cycle includes recruitment, growing, mature and regressing phases (Skidmore *et al,* 1995, 1996; Nikjou *et al,* 2009). During non-breeding season, the follicle may not reach to the diameter suitable for mating or AI (Nawito *et al,* 1967). It is possible to enhance follicle growth to the final stage of development or superovulate during non-breeding season using gonadotropins (Nowshari and Ali, 2005). However, due to the lack of suitable male with proper libido and sperm production during non-breeding season (Ali *et al,* 2021) breeding of female during non-breeding season could be a challenging subject until the availability of fertile frozen semen to achieve successful fertilisation and pregnancy.

Preparing donors for superovulation

The main concept for successful superovulation is to initiate superovulation between the emergence of follicular wave (Ararooti *et al,* 2018a, b) and before follicle deviation (Skidmore *et al,* 2005; Nikjou *et al,* 2008; Anouassi and Tibary, 2013; Manjunatha *et al,* 2019). Using this strategy, the follicles that initially emerged by endogenous FSH surge grow simultaneously with the support of exogenous FSH. Moreover, there is no mature follicle to suppress the growth of subordinate follicles. To achieve the emergence of new follicular wave, nearly all synchronising treatments relied on eliminating the existing follicle either by inducing ovulation in mature follicle using GnRH (Moghiseh *et al,* 2008), or by regressing existing follicles using progestogens (McKinnon and Tinson, 1992; McKinnon *et al,* 1994). Another non-practical way to eliminate mature follicles in camel is to ablate mature follicle by ultrasound-guided transvaginal follicle aspiration (Skidmore *et al,* 2009).

Camel is induced ovulator with follicular wave cycles and corpus luteum is present only in camels that have previously had sterile mating or have been ovulated by ovulating agents (Nawito *et al,* 1967; Skidmore *et al,* 1996; Moghiseh *et al,* 2008). In non-mated camel, the interval between formations of two consecutive mature follicles (inter-wave interval) is about 18 days for dromedary (Skidmore *et al,* 1996) and 19 days for Bactrian camels (Nikjou *et al,* 2009). Induction of ovulation in the mature follicle reduces the inter-wave interval to about 14 days. When GnRH is injected at a random stage of follicular wave cycle, emergence of new follicular emergence occurs variably within 2 to 6 days after GnRH injection, depending on the size of follicle at the time of treatment (Nikjou *et al,* 2008). However, the tightness of synchrony for follicular wave emergence is increased when the second GnRH is administered 14 days later (Nikjou *et al,* 2008; Skidmore *et al,* 2009). Similar results were found using the combination of GnRH and prostaglandin $F_2\alpha$ (Manjunatha *et al,* 2014, 2015). Accordingly, she-camel receives three GnRH injections on day -22, -12 and 0 (Day of the last GnRH injection) and two prostaglandin $F_2\alpha$ on Days -15 and -5. Superovulation could be started at any time between emergence of new follicular wave (2 days after the last GnRH injection; Ararooti *et al,* 2018a, b), and prior to the follicle deviation (4 days after the last GnRH injection; Skidmore *et al,* 2005; Nikjou *et al,* 2008; Anouassi and Tibary, 2013; Manjunatha *et al,* 2019).

The second approach for synchronising donors is to use follicle regressing agents such as progesterone (McKinnon and Tinson, 1992; McKinnon *et al,* 1994). In this particular approach used in dromedary camel, superovulation is initiated when there is limited follicular activity in the ovaries (i.e. no follicle > 7 mm in diameter) which can be achieved by daily intramuscular injection of progesterone (100 mg) for 8–15 days (McKinnon and Tinson, 1992, 1994). However, we could not find any benefit of using progestogens in controlling follicular dynamics in Bactrian camel (Nikjou *et al,* 2008).

Evaluation of superovulatory response

Superovulatory response could be evaluated by the number and diameter of follicles induced

to grow prior to ovulation, the number of corpora lutea (CL), the number of total ova/embryos, the number of transferable embryos, and the number of un-ovulated follicles at the time of embryo recovery. The best superovulatory response occurs when there is high number of CL and transferable embryos (≥ 5; Table 1) with low number of un-ovulated follicles and no unfertilised ova. This could be achieved when good type, right amount and right procedure for gonadotropin injections is used. Having good and safe gonadotropin such as highly purifies FSH (Folltropin-V) and/or hMG, still it is necessary to use right total amount and correct procedure of daily gonadotropin administration (Ararooti *et al,* 2017, 2018a). Small proportion of camels (10–20%) do not respond at all; whereas, some donors (15–20%) could be over-stimulated and produce ≥ 30 follicles in each ovary (McKinnon *et al,* 1994; Tibary and Anouassi, 1997; Skidmore *et al,* 2002; Anouassi and Tibary, 2013). The great number of un-ovulated follicles at the time of embryo recovery, in association with the low number of CLs and recovered embryos, could imply that the type, the amount and/or the way of gonadotropin administration may not be suitable. The presence of great number of un-fertilised ova could imply over-response to gonadotropin and/or using immature bull or using the bull at the beginning of breeding season.

Prediction of superovulatory response could be an essential part of superovulation program in order to estimate the right number of recipients necessary to receive embryo. We have demonstrated positive correlations between the ratio of follicles >6 mm/follicles ≤6 mm, detected on Day 4 of 5.5 days program, and the number of CLs and embryos in FSH treated camel (r=0.9; P<0.05; Ararooti *et al,* 2018a). As a result, a great percentage of follicles >6 mm, detected on Day 4 of superovulation, ovulated and established corpora lutea (90%), and embryos (70 %). This protocol allows the estimation of superovulatory response and the number of recipients that has to be prepared prior to embryo recovery in dromedary camel.

Table 1. Summary of the number of transferable embryos found following different superovulation protocols in dromedary camel since 1990.

Investigators	Year	No. Donors	No. Responders (%)	Superovulation	Transferable Embryo (mean)
Anouassi & Ali	1990	9	3 (33)	eCG (1500 IU)	2.1
Anouassi & Ali	1990	21	7 (33)	eCG (2000 IU)	4.9
Skidmore *et al*	1992	30	5 (17)	eCG (2000-4000 IU)	1.3
Skidmore *et al*	1992	11	3 (27)	Ovagen (20—30 IU)	2.5
Mckinnon and Tinson	1992	14	9 (62)	eCG (4500 IU)	2.7
Mckinnon and Tinson	1992	13	9 (69)	Ovine FSH (?)	4.5
Mckinnon *et al*	1994	68	N/A	Folltropin-V (?)	3.8
Mckinnon *et al*	1994	84	N/A	eCG (3000-6000IU)	2.3
Skidmore *et al*	2002	42	36 (86)	eCG (2500IU) + Folltropin-V (400 mg)	6.7
Skidmore *et al*	2005	12	10 (83)	eCG (2500IU) + Folltropin-V (400 mg)	4.0
Nowshari *et al*	2005	15	15 (100)	eCG (2000IU) + Folltropin-V (400 mg)	5.1
Nowshari *et al*	2005	11	10 (91)	Folltropin-V (400 mg)	1.8
Anouassi & Tibary	2013	153	106 (69)	eCG (3000 IU)	7.1
Anouassi & Tibary	2013	176	123 (70)	Folltropin-V (400 mg)	8.2
Ararooti *et al*	2017	6	6 (100)	hMG (16.5 ampules)	5.8
Ararooti *et al*	2018	5	5 (100)	Folltropin-V (390 mg)	16.2
Ararooti *et al*	2018	5	5 (100)	eCG (1000IU) + Folltropin-V (330 mg)	7.2
Manjunatha *et al*	2019	13	N/A	Pluset (2000 IU)	6.1
Manjunatha *et al*	2019	45	N/A	Folltropin-V (200 mg) dissolved in hyaluronan	5
Manjunatha *et al*	2019	42	N/A	Pluset (1000 mg) dissolved in hyaluronan	5.2
Manjunatha *et al*	2020	11	N/A	Folltropin-V (400 mg)	5.6
Manjunatha *et al*	2020	14	12 (86)	Recombinant eCG (3000 IU)	4.6
Average number of transferable embryos					5.1

Superovulation in camel

Three main gonadotropins used for superovulation in camel are equine Chorionic Gonadotropin (eCG), Follicle Stimulating Hormone (FSH) and human Menopausal Gonadotropin (hMG). Early efforts to superovulate camel in the early 1990s used eCG at various doses (Anouassi and Ali, 1990; McKinnon Tinson, 1992; Skidmore *et al,* 1992; Table 1). Anouassi and Tibary (2013) summarised the result of single injection of eCG (3000 IU) on 153 dromedary camels. They found that 31% of donors did not respond to eCG and the number of transferable embryos were 7.1±4.3 with great variation (0-19; Table 1). More recently, Manjunatha *et al* (2020) used newly released recombinant eCG (reCG) to superovulate camel. Accordingly, single injection of 3000 IU reCG could produce between 5-25 ovulation and 4.6±1.3 (0-7) transferable embryos. Most studies using eCG suffers the variation in response due to the inherent impact of eCG of any kind on superovulatory response. Two main advantages of using eCG for superovulation are easy administration (single injection) and low cost compared to other gonadotropins. However, several negative impacts of eCG on superovulatory response need to be considered including long half-life, the possibility of producing different generation of follicles, having both FSH and LH like effects, the possibility of luteinisation of follicles prior to ovulation which delays or entirely inhibits ovulation, and antigenic properties resulting in ovarian refractoriness following repeated administration of eCG, which ultimately results in low or lack of desired superovulatory response (Skidmore, 2003; Anouassi and Tibary, 2013).

The main negative impact of gonadotropins with prolonged half-life or at the doses beyond the requirement for suitable superovulation is continuous daily recruitment of different generation of follicles resulting in the formation of follicles with different diameters that could interfere in the process of ovulation followed by the production of un-ovulated follicles at the time of embryo recovery, which in turn, interrupt with the process of uterine flushing and embryo recovery (Ararooti *et al,* 2018a).

Ovine FSH (20 or 30 IU; Ovagen), twice daily, over 3 days were used in camel (Cooper *et al,* 1992; Skidmore *et al,* 1992; Table 1). However, follicular response and embryo recovery using these protocols were poor. From more than two decades ago, highly purified porcine FSH (Folltropin-V; LH to FSH ratio:0.12; Mikkola and Taponen, 2017) was used for superovulation in camel. Because of very short half-life of FSH, about 5 hours, typically, donor animals received FSH twice daily in decreasing doses manner, over 4 days (Anouassi and Tibary, 2013; Ararooti *et al,* 2017, 2018a). Anouassi and Tibary (2013) summarised the result of Folltropin-V injections (400 mg, twice daily over 4 days) on 176 dromedary camels. They found that 30% of donors did not respond to FSH and the number of transferable embryos were 8.2±6.1 with great variation (0-36; Table 1).

Three gonadotropins for superovulation in dromedary camel have been investigated (Ararooti *et al,* 2018a). Donors received porcine FSH alone (Folltropin-V; 390 mg), or combination of eCG (1000 IU im) and FSH (330 mg), and hMG (17.5 ampules. Donors in the first group, received twice daily FSH in decreasing doses (80, 50, 30, 20, 10 mg) for 5 days and single dose of FSH on Day 6 (10 mg). Donors in eCG-FSH group, received a single dose of eCG (1000 IU) on Day 0, followed by twice daily FSH in decreasing doses (60, 40, 30, 20, 10 mg), beginning on Day 0, followed by a single dose of 10 mg FSH on Day 5. In hMG group, donors received hMG (3, 2, 1.5, 1, 1 ampules) twice daily between Days 0 to 4, and 0.5 ampule on Day 5. The number of transferable embryos were 16.2, 7,2 and 1.6 in FSH alone, eCG-FSH and hMG, respectively (Table 1). We hypothesised that low superovulatory response in hMG group could be due to high total dose of hMG and/or the way of hMG administration. In another study, we have reduced the total dose of hMG to 16.5 ampules and we changed the injection protocol to twice daily injections over 5 days of 4, 2, 1, 0.5, 0.5 ampules and 0.5 ampule on Day 6. These modifications resulted in 5.8 transferable embryos which was comparable to FSH treatment (Ararooti *et al,* 2017; Table 1).

In earlier study porcine FSH (Folltropin-V; 400 mg) in decreasing doses (80, 60, 40, 20 mg) over 4 days was associated with low response (1.8 transferable embryos; Nowshari and Ali, 2005). However, in more recent study using the same amount of porcine FSH, twice daily, in decreasing dosages (60, 50, 40, 30, 20 mg), over 5 days was associated with good superovulatory response (5.6 transferable embryos; Manjunatha *et al,* 2020).The later authors used 2000 IU Pluset (LH to FSH ratio: 1; Mikkola and Taponen, 2017) in twice daily, in decreasing dosages (300, 250, 200, 150, 150 mg), over 5 days, which was associated with good superovulatory response (6.1 transferable embryos; Manjunatha *et al,* 2019). In order to simplify the administration of FSH, two intramuscular injections of FSH dissolved into 5 mg/

ml hyaluronan solution, 48 hrs apart, were examined to superovulate dromedary camel (Manjunatha *et al,* 2019). Using 200 mg Folltropin-V (120 and 80 mg at 48 hrs interval) or 1000 IU Pluset (600 and 400 IU at 48 hrs interval) were associated with 5 and 5.2 transferable embryos, respectively (Manjunatha *et al,* 2019; Table 1). Although this method of FSH delivery seems to be very simple, it needs to be repeated in large scale and by different groups to become routine procedure for superovulation in camel. The combination of eCG and FSH were also used in camel. Donors received a single injection of eCG (2000-2500 IU), in association with porcine FSH (400 mg) administered in decreasing doses (80, 60, 40, 20 mg twice daily) over 4 d. The average numbers of 4, 5.1 and 6.7 embryos were recovered using the latter protocol (Skidmore *et al,* 2002, 2005; Nowshari and Ali, 2005).

In conclusion, different gonadotropins could be used for superovulation in camel with great variation in superovulatory response. Although eCG seems to be a simple and cheap gonadotropin for superovulation, great variation and several drawbacks of eCG, might remove this gonadotropin as the first choice for superovulation in camel. Highly purified FSH seems to be the first choice for superovulation in camel. Although it is more expensive than eCG, it provides better response. The number of FSH injections could be reduced by dissolving FSH in hyaluronan. It could be possible to predict superovulatory response by using the right amount and the way of gonadotropin administration.

References

Al Eknah MM. Reproduction in old world camels. Animal Reproduction Science. 2000; 60:583-592.

Ali A, Derar DR, and Almundarij TI. Aetiological analysis and diagnosis of reproductive disorders in male dromedary camels. Reproduction in Domestic Animals. 2021; 56(10):1267-73.

Anouassi A and Ali A. Embryo transfer in camel (*Camelus dromedarius*). In: Saint-Martin (Ed). Proceedings of the UCDEC workshop 'Is it Possible to Improve the Reproductive Performance of the Camel'. Paris. 1990; pp. 327-332.

Anouassi A and Tibary A. Development of a large commercial camel embryo transfer program: 20 years of scientific research. Animal Reproduction Science. 2013; 136(3):211-21.

Ararooti T, Niasari-Naslaji A, Asadi-Moghaddam B, Razavi K and Panahi F. Superovulatory response following FSH, eCG-FSH and hMG and pregnancy rates following transfer of hatched blastocyst embryos with different diameter and shape in dromedary camel. Theriogenology. 2018a; 106:149-56.

Ararooti T, Niasari-Naslaji A, Razavi K and Panahi F. Comparing three superovulation protocols in dromedary camels: FSH, eCG-FSH and hMG. Iranian Journal of Veterinary Research. 2017;18(4):249.

Ararooti T, Panahi F and Niasari-Naslaji A. Superovulation protocols before ovum pickup in camel. Journal of Camel Practice and Research. 2018b;25(3):261-5.

Cooper MJ, Skidmore J, Ali M, Billah T, Wensvoort S, Billah AA and Allen WR. An attempt to induce and synchronise ovulation and superovulation in dromedary camels for embryo transfer. In: Saint-Martin (Ed). Proceedings of the UCDEC workshop 'Is it Possible to Improve the Reproductive Performance of the Camel'. Paris. 1990; pp. 313-326.

Ealy AD, Wooldridge LK and McCoski SR. Post-transfer consequences of *in vitro* produced embryos in cattle. Journal of Animal Science. 2019; 97(6):2555-2568.

El-Wishy AB. Functional morphology of the ovaries of the dromedary camel. In: Allen, WR, Higgins AJ, Mayhew IG, Snow DH, Wade JF (Eds). Proceedings of the First International Camel Conference. Dubai: R and W Publications (Newmarket) Ltd. 1992; pp.149-154.

Khatir H and Anouassi A. The first dromedary (*Camelus dromedarius*) offspring obtained from *in vitro* matured, *in vitro* fertilized and *in vitro* cultured abattoir-derived oocytes. Theriogenology. 2006; 65(9):1727-1736.

Marie M and Anouassi A. Induction of luteal activity and progesterone secretion in the nonpregnant one-humped camel (*Camelus dromedarius*). Reproduction. 1987; 80(1):183-192.

Manjunatha BM, Al-Bulushi S and Pratap N. Synchronisation of the follicular wave with GnRH and PGF2α analogue for a timed breeding program in dromedary camels (*Camelus dromedarius*). Animal Reproduction Science. 2015; 160:23-29.

Manjunatha BM, Al-Bulushi S and Pratap N. Ultrasonographic Characterization of Follicle Deviation in Follicular Waves with Single Dominant and Codominant Follicles in Dromedary Camels (*Camelus dromedarius*). Reproduction in Domestic Animals. 2014; 49(2):239-242.

Manjunatha BM, Al-Hosni A and Al-Bulushi S. Simplified superovulation protocols in dromedary camels (*Camelus dromedarius*). Theriogenology. 2019; 126:214-221.

Manjunatha BM, Al-Hosni A and Al-Bulushi S. Superovulation of dromedary camels with eCG: an effective and practical technique. Theriogenology. 2020; 151:112-118.

Malin K, Witkowska-Piłaszewicz O and Papis K. The many problems of somatic cell nuclear transfer in reproductive cloning of mammals. Theriogenology. 2022; 189:246-254.

McKinnon AO and Tinson AH. Embryo transfer in Dromedary camels. In: Allen, WR, Higgins AJ, Mayhew IG, Snow DH, Wade JF (Eds). Proceedings of the First International Camel Conference. Dubai: R and W Publications (Newmarket) Ltd. 1992; pp. 203-208.

McKinnon AO, Tinson AH and Nation G. Embryo transfer in dromedary camels. Theriogenology. 1994; 41(1):145-150.

Merkt H, Rath D and Musa B. Reproduction in camels. FAO Animal Production and Health. Bulletin No. 1990; No, 82.

Mikkola M and Taponen J. Embryo yield in dairy cattle after superovulation with Folltropin or Pluset. Theriogenology. 2017; 88:84-8.

Moghiseh A, Niasari-Naslaji A, Nikjou D, Gerami AL, Razavi K, Mostafaey M. The effect of LH and GnRH analogues on induction of ovulation in Bactrian camel (*Camelus bactrianus*). Iranian Journal of veterinary research. 2008 Dec 30;9(4):324-9.

Nawito MF, Shalash MR, Hoppre R and Rakha AM. Reproduction in female camel. Bulletin Animal Science Research Institute. 1967.

Nowshari MA and Ali SA. Effect of season and gonadotropins on the superovulatory response in camel (*Camelus dromedarius*). Theriogenology. 2005; 64(7):1526-1535.

Niasari-Naslaji A, Nikjou D, Skidmore JA, Moghiseh A, Mostafaey M, Razavi K and Moosavi-Movahedi AA. Interspecies embryo transfer in camelids: the birth of the first Bactrian camel calves (*Camelus bactrianus*) from dromedary camels (*Camelus dromedarius*). Reproduction, Fertility and Development. 2009; 21(2):333-337.

Niasari-Naslaji A, Barani M, Adel HR, Ghanbari A and Moosavi-Movahedi AA. Adaptation of bactrian camel to hot desert climate using interspecies embryo transfer. Journal of Camel Practice and Research. 2014;21(2):161-4.

Nikjou D, Niasari-Naslaji A, Skidmore JA, Mogheiseh A, Razavi K, Gerami A and Ghanbari A. Synchronisation of follicular wave emergence prior to superovulation in Bactrian camel (*Camelus bactrianus*). Theriogenology. 2008; 69(4):491-500.

Nikjou D, Niasari-Naslaji A, Skidmore JA, Mogheiseh A, Germai A, Razavi K and Ghanbari A. Ovarian follicle dynamics in bactrian camel (*Camelus bactrianus*). Journal of Camel Practice and Research. 2009; 16(1):97-105.

Olsson PO, Tinson AH, Al Shamsi N, Kuhad KS, Singh R, Son YB, Jeong Y, Jeong YW, Cai L, Sakaguchi K and Kim S. Blastocyst formation, embryo transfer and breed comparison in the first reported large scale cloning of camels. Scientific Reports. 2021; 11(1):14288.

Skidmore JA, Adam GP and Billah M. Synchronisation of ovarian follicular waves in the dromedary camel (*Camelus dromedarius*). Animal Reproduction Science. 2009; 114: 249-255.

Skidmore JA, Allen WR, Cooper MJ, Chaudhry MA, Billah M and Billah MA. The recovery and transfer of embryo in the dromedary camel: results of preliminary experiments. In: Allen, WR, Higgins AJ, Mayhew IG, Snow DH, Wade JF (Eds). Proceedings of the First International Camel Conference. Dubai: R and W Publications (Newmarket) Ltd. 1992; pp 137-1 42.

Skidmore J, Billah M and Allen WR. The ovarian follicular wave pattern in the mated and non-mated dromedary camel (*Camelus dromedarius*). Journal of reproduction and fertility. Supplement. 1995; 49:545-548.

Skidmore JA, Billah M and Allen WR. The ovarian follicular wave pattern and induction of ovulation in the mated and non-mated one-humped camel (*Camelus dromedarius*). Reproduction. 1996; 106(2):185-192.

Skidmore JA, Billah M and Allen WR. Investigation of factors affecting pregnancy rate after embryo transfer in the dromedary camel. Reproduction, Fertility and Development. 2002; 14(2):109-116.

Skidmore JA. The main challenges facing camel reproduction research in the 21st Century. Reproduction-Cambridge-Supplement. 2003; 1:37-47.

Skidmore JA and Billah M. Embryo transfer in the dromedary camel (*Camelus dromedarius*) using asynchronous, meclofenamic acid-treated recipients. Reproduction, Fertility and development. 2005;17(4):417-421.

Skidmore JA, Billah M and Loskutoff NM. Comparison of two different methods for the vitrification of hatched blastocysts from the dromedary camel (*Camelus dromedarius*). Reproduction, Fertility and Development. 2005; 17(5):523-527.

Tibary A and Anouassi A. Artificial breeding and manipulation of reproduction in camelidae. In: Institut Agronomique et Vétérinarie Hassan II, Rabat, Maroc. (Ed.), Theriogenology in Camelidae. Anatomy, Physiology, Pathology and Artificial Breeding. Abu Dhabi Printing and Publishing Company. UAE. 1997; pp. 413-52.

Wani NA, Wernery U, Hassan FA, Wernery R and Skidmore JA. Production of the first cloned camel by somatic cell nuclear transfer. Biology of reproduction. 2010; 82(2):373-379.

Wani NA. *In vitro* embryo production (IVEP) in camelids: Present status and future perspectives. Reproductive Biology. 2021; 21(1):100471.

Wilson RT. The Camel. Longman: Harlow Essex. 1984; pp. 97-98.