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Use of assisted reproduction for the improvement of milk production in dairy camels (*Camelus dromedarius*) $\stackrel{\text{\tiny{}}}{\approx}$

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ABSTRACT

Despite their production potential and ability to survive on marginal resources in extreme conditions, dromedaries have not been exploited as an important food source. Camels have not been specifically selected for milk production, and genetic improvement has been negligible. High individual variation in milk production both within the population and within breeds provides a good base for selection and genetic progress. In this paper, we discuss the possibilities and constraints of selective breeding for milk production in camels, and include a summary of the use of embryo transfer at the world's first camel dairy farm. Embryo transfer is an integral part of the breeding strategy at the camel dairy farm because it increases selection intensity and decreases the generation interval. Using high milk-producing camels as donors and low producing camels as recipients, 146 embryos were recovered $(6.1 \pm 1.0 \text{ embryos/donor; range: 0–18})$. Embryo transfer is an of 28 twin embryo transfers). Pregnancy rate at 21 days and 5 months was 55% (61/111) and 45% (50/111), respectively. Finally, a total of 46 recipients delivered a live calf. These results document the utility of embryo transfer using high milk producing dromedaries as donors.

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1. Introduction

Globally, the consumption of non-cattle milk has doubled over the last 50 years reaching approximately 16.9% of the total 702 billion tons of milk produced in the world by 2009 (Faye and Konuspayeva, 2012). Amongst non-cattle species, dromedary and Bactrian camels play an important role as a milk source in many arid and semi-arid countries, as they can produce more milk over a longer period of

time than any other species under these harsh conditions (Farah and Younan, 2005). In the Horn of Africa, where 60% of the world camel population lives, approximately 10% of the total milk produced is of camel origin (Faye and Konuspayeva, 2012), but despite their potential to survive on marginal resources in extreme conditions, camels have not been exploited as an important food source. Camel milk also seems to have medicinal properties such as antimicrobial, hypoallergenic, hypoglycemic, anti-hypertensive effects and it has been used in different parts of the world (India, Russia, Sudan) for the treatment of a number of diseases (Al Haj and Al Kanhal, 2010).

Dromedaries have not been specifically selected for milk production and no systematic methods have been applied for genetic improvement. In addition, there is little differentiation among breeds and the distinction is not based on sound quantitative parameters (Abdallah and Faye, 2012; Almathen et al., 2012; Hermas, 1998). Milk production has



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not been recorded in a systematic fashion in camels, and the inconsistency of information gained to date has not provided a solid base for genetic improvement programs. Similarly, breeding records are not usually kept and studbooks have not been established, so the origin of individual animals is uncertain. As a result, genetic improvement has been negligible for specific production traits. Only one study was found in the literature that reported an annual genetic progress; 0.05 kg milk/camel improvement in 305 day lactation over a 23-year period (Almutairi et al., 2010).

The camel dairy industry has progressed in the last decade with the development of the first large-scale camel dairy farm (Emirates Industries for Camel Milk and Products, EICMP) in Dubai. The farm has over 1500 adult dromedaries that are machine-milked twice daily, and the milk is processed on-site before being distributed. A genetics improvement program was implemented which includes identification and phenotypic characterization, consistent and systematic milk data collection and analysis, reproductive data recording, and establishment of an inhouse studbook, and the use of synchronization protocols and assisted reproductive technologies (Juhasz and Nagy, 2012; Nagy et al., 2012). This large pool of animals, supported by a solid database has provided an unprecedented opportunity for genetic improvement for dairy production.

The aims of the present paper are (i) to review the production potential, reasons for slow genetic progress and the use of embryo transfer and related techniques in dromedary camels and (ii) to provide new data on the use of embryo transfer as part of a breeding strategy to improve milk production in this species.

2. Milk production potential of dromedary camels and factors affecting milk production

Data on the production potential of dromedaries is scarce, and reports vary widely and are difficult to compare (Fave, 2008). Authors have used different measurement procedures for the estimation of milk production (e.g., the calf suckling method, hand milking of two or four quarters, the oxytocin technique; Simpkin and Rowlinson, 1994), and some include an estimated quantity of milk consumed by the calf. Moreover, milk production has been expressed in different ways, such as daily average, daily maximum, total lactation, 305-day production, or herd average. Some suggest that daily maximum production could reach 35-401 and that total production varies between 1000 and 12,0001 during an 8-18 month lactation period (Faye, 2008). Based on 388 complete lactation cycles, the length of lactation, total production per lactation, and average daily milk production in the EICMP herd were 585 ± 11.0 days, 3152 ± 73.5 kg and 5.8 ± 0.08 kg, respectively (mean ± SEM). Daily maximum production exceeded 25 kg only rarely. The mean daily milk production for 400 days is a good overall indicator of production potential in dromedaries and was 6.9 ± 0.10 kg (mean \pm SEM) with a range of 2.4 kg/day to 17.4 kg/day in the EICMP herd.

As in other species, the most important factors affecting milk production are breed, age, parity, season, nutrition,

milking frequency and the presence of the calf. In dromedaries, considerable phenotypic variation can be observed between the different breeds, but breeds are not well defined and are not genetically characterized (Abdallah and Faye, 2012; Almathen et al., 2012; Simpkin, 1994). The breeds or types are differentiated on the basis of origin and distribution of the camels, on the ethnic groups they belong to, or on their color. Some report differences between breeds based on color (Faye, 2008; Ismail and Al Mutairi, 1994), and others reported 20-50% less milk in the first lactation of the Arvana breed (Cherzekov and Saparov, 2005) with maximum milk production in the 2nd or 3rd lactation (Ismail and Al Mutairi, 1994). In pastoral systems, milk production corresponds to the wet season when feed and water is plentiful. Simpkin (1994) described a two-peak lactation curve in dromedaries; the first peak is marked and occurs during the first few weeks of lactation, the second corresponds to the following wet season. In traditional systems, the presence of the calf is essential for milking, as the milk let-down reflex is initiated by suckling. Some report that increasing milking frequency from twice to three or four times a day resulted in a 10–15% increase in milk yield (Simpkin, 1994). We did not observe an increase when we compared the daily yield after two or three times a day milking following a 2-week adaptation period (Nagy et al., 2012).

3. Reasons for slow genetic improvement for milk production

Genetic progress in dairy camels has been limited by low accuracy and intensity of selection, unknown heritability of specific production traits, and long generation intervals (Hermas, 1998). The accuracy of estimated breeding value could be increased by combining individual and progeny performance; however, progeny testing takes a long time in camels because of the long production cycle, late age at first parturition and present reproductive management systems. For example it may take 8–12 years for a female offspring of a high-producing dromedary to start lactating so that her potential can be evaluated. Even for adult males, it takes 6–7 years before their female offspring reach production age (Tibary and Anouassi, 1997b).

Selection intensity is influenced by reproductive efficiency, and may be described by the number of offspring from an individual dromedary within a defined period of time. Any factor decreasing the number of calves slows selection intensity. Reproductive efficiency and fertility are generally regarded to be low in dromedary camels. Although end-of-season pregnancy rates of 50–80% can be achieved with improved nutrition and reproductive management, birthing rates barely exceed 40% in traditional management systems (Tibary et al., 2005). Fertility after an ovarian synchronization protocol and natural mating was found to be similar to that in other domestic species (Nagy and Juhasz, 2012).

The generation interval and the age at first birthing are high in dromedaries; first parturition varies from 3 to 7 years of age (Tibary and Anouassi, 1997b). The long generation interval for camels is due in large part to long production cycles and management methods. Both lactation (\approx 585 days) and gestation (\approx 390 days) periods are long, resulting in a calving interval of 2–2.5 years (Tibary et al., 2005). Unlike dairy cows, lactation and pregnancy cannot overlap in camels. Milk yield drops significantly by 60 days of gestation in camels; hence, shortening the calving interval by early breeding would result in a significant decrease in milk production (Nagy and Juhasz, 2010). Embryo transfer offers a way overcoming the above-mentioned constraints of camel physiology.

4. Overview of embryo transfer and related techniques in dromedaries in the last two decades

Due to the growing demand from the camel racing industry in the Arabic peninsula, embryo transfer has been developed in dromedary camels over the last 20 years. Non-surgical transfer of fresh Day 7 embryos from superior racing male and female pairs has gained widespread acceptance and is practiced routinely (McKinnon et al., 1994; Skidmore et al. 2002; Tibary and Anouassi, 1997a). However, this technique has not been applied to dairy animals. The method of embryo transfer has been described in detail by Tibary and Anouassi (1997a), and the development of reliable superovulation protocols, the management and synchronization of donors and recipients and other factors that affect success rate have been reported by McKinnon et al. (1994), Nowshari and Ali (2005), Skidmore (2003), Skidmore et al. (2002). For example, in the study by Skidmore et al. (2002) the pregnancy rate increased to a maximum of 67% when ovulation in the recipient occurred 1 day later than that of the donor, but fell dramatically when the level of asynchrony between recipient and donor increased to +1(9%) or -3(10%) days. Recently, new methods have been tested using progesterone, nonsteroidal anti-inflammatory drugs or a combination of progesterone and eCG for the management of recipients, so that such tight synchrony between donors and recipients is not required (Skidmore and Billah, 2005, 2011).

Until recently there has been very limited data on the cryopreservation of camel embryos. Skidmore and Loskutoff (1999) conducted preliminary studies to determine the minimum exposure time required for camel embryo in the most appropriate cryoprotective agent. Using two different methods for vitrifying Day 6 and Day 7 camel embryos, no pregnancies resulted from the open pulled-straw method but 8 pregnancies from the 21 transferred embryos (38%) resulted after use of the French straw method with the smaller Day 6 embryos (Skidmore et al., 2005). Nowshari et al. (2005) reported the birth of the first camel calf from cryopreserved embryos but the efficiency was very low.

Improvements in culture conditions resulted in the first camel offspring obtained from in vitro matured, in vitro fertilized and in vitro cultured abattoir-derived oocytes (Khatir and Anouassi, 2006). Optimization of somatic cell nuclear transfer techniques has led to the production of the first cloned camel by Wani et al. (2010), but the efficiency for nuclear transfer in camels is between 0 and 10%.

5. Breeding strategies to improve milk production on a large-scale camel dairy farm

At EICMP, the aim is to establish a good milk producing (400-day average daily yield >10–12 kg), mastitis resistant and fertile camel population whilst keeping the original breeds/types. The current breeding program is based on phenotypic selection of qualitative and quantitative traits and on the assumption that heritability of the milk yield is similar to that of dairy cattle (Hermas, 1998). The breeding strategy is based on (i) accurate data recording and analysis, (ii) categorizing and selecting female camels according to their type and milk production, (iii) selecting bulls for natural mating from high-producing dams, (iv) an embryo transfer program for high-producing female camels using males from high-producing dams, (v) natural mating of average producing camels with selected bulls and (vi) culling of camels with low production and/or low resistance to diseases.

Categorizing and selecting female camels: Camels at EICMP originated from different regions of the Middle-East, Africa and Asia. Currently there are 8 different types/breeds based on phenotype, origin and color: Emirati, Emirati-mix, Cross-breed, Saudi, Saudi-mix, Sudani, Black, and Pak-istani. Remarkable individual variation in milk yield both within the population and within breeds has been observed (Fig. 1). Such a high individual variation within the same breed/type provides a good opportunity for selection and genetic progress.

Bull selection: During the first five years, males with unknown genetic background and potential had to be used for breeding. Recently, however, young bulls from high-producing mothers have been introduced into the breeding program. Although, the parents of these young males were known, only the performance of the dam could be evaluated. Progeny testing of these young bulls will take another 6–8 years.



Fig. 1. Distribution of average daily milk yield for 400 days of lactation (n = 388 domedary camels; mean \pm SEM = 6.9 ± 0.10 kg/day). Average daily production was >1SD above the mean (8.95 kg/day) in 48 dromedaries (12.4%), and >2SD above the mean (11.0 kg/day) in 11 dromedaries (2.8%).

Table 1

Results of superovulation and embryo transfer during two consecutive breeding seasons (2010 and 2011) using high- and low-milk producing dromedaries as embryo donors and recipients, respectively.

	Responded donors	Number of embryos	Number of transfers ^a	Pregnant recipients ^b	Embryo mortality	Mean pregnancy per donor ^c
Year 1 Ratio or mean (±SEM)	9/10 90%	$\begin{array}{c} 56 \\ 6.2 \pm 1.5 \end{array}$	46 (36+10)	20 43.5%	5 25%	1.7
Year 2 Ratio or mean (±SEM)	15/18 83.3%	$\begin{array}{c} 90 \\ 6.0 \pm 1.4 \end{array}$	65 (47+18)	41 63.1%	6 16.6%	2.3
Total Ratio or mean (±SEM)	24/28 85.7%	$\begin{array}{c} 146 \\ 6.1 \pm 1.0 \end{array}$	111 (83+28)	61 55.0%	11 18.0%	2.1

^a Numbers in brackets indicate single and twin embryo transfers.

^b Pregnancy at 21 days of gestation.

^c Pregnancy at 5 months of gestation.

Embryo transfer: Embryo transfer is a useful tool for increasing selection intensity and decreasing the generation interval. The selected donors are the best producing female camels from each type/breed, whose 400-day average daily milk production is >1 SD above the mean. Donor camels are mated with bulls that are the offspring of the best producing dams, whereas the selected recipients are low-producing camels, whose milk production is >1SD below the mean. These recipient camels represent a substantial proportion of the herd and they have a long life span, hence they are invaluable in the embryo transfer program. In addition, they will not add to the genetic pool of the next generation of the herd. The low-producing recipients may be milked for 4–5 months after delivery, and then return to the embryo transfer program.

Natural mating with selected bulls and culling: The rest of the female camels are mated with bulls from highproducing mothers. Female calves from low-producing mothers are culled yearly at the time of weaning and only the yearlings that will be in the future recipient herd are kept.

6. Superovulation and embryo transfer in high producing dromedaries during lactation

Twenty eight (8-15 years), high-producing, multiparous, lactating dromedaries were selected as donors at the end of 2 consecutive breeding seasons (April-May 2010 and 2011) at EICMP in Dubai, UAE. All camels were milked by machine twice daily during the study, as described (Nagy et al., 2012). During the first season, donors (n = 10) were in late lactation (560 ± 45 days post-partum; mean \pm SEM), whereas during the second season donors (n=18) were at the beginning of their lactation $(132 \pm 21 \text{ days post-}$ partum). Average daily milk production of camels in the first and second season was 8.1 ± 0.4 kg and 9.4 ± 0.7 kg, respectively. Recipients were selected from the breeding herd of the farm. They had been recently weaned from their calves, were healthy, had normal genital organs and their 400-day average daily milk yield during the previous lactation was $\leq 5 \text{ kg/day}$. All camels were in moderate to good body condition with body weight ranging from 550 to 650 kg.

In general, superovulation, embryo recovery and transfer were performed as described (Skidmore et al., 2002).

In brief, follicular activity was monitored by regular rectal examination and ultrasonography (Aloka 500, 5 MHz, Japan). Donors were induced to ovulate at random with 20 µg Buserelin i.v. (Receptal, Intervet, Boxmeer, The Netherlands). Starting on Day 4 (GnRH treatment = Day 0), each donor was treated with a combination of 2000 IU eCG im (Folligon, Intervet, Boxmeer, The Netherlands) administered as a single injection, and a total of 400 mg pFSH (Folltropin, Bioniche Animal Health, Ireland) given im twice daily in declining doses on Days 4-7 inclusive. Donors were mated twice with a fertile bull, 24 h apart, when follicles reached 10-15 mm in diameter, and embryo recovery was carried out 7 days after ovulation. Recovered blastocysts were transferred non-surgically into recipients that had been induced to ovulate one day after the donors and pregnancy was diagnosed by ultrasonography.

The results are summarized in Table 1. Superovulation was successful in 24 of 28 camels (85.7%), resulting in the development of 19.1 ± 1.9 (mean \pm SEM, range: 6–45) follicles \geq 10 mm and 12.0 \pm 1.3 (range: 2–25) corpora lutea after mating per successful donor. Multiple ovulation failed to occur in four camels. A total of 146 embryos were recovered $(6.1 \pm 1.0 \text{ embryos/donor; range: } 0-18)$; the recovery rate (number of embryos recovered relative to the number of ovulations detected) was $43.1 \pm 6.0\%$ (range: 0–90%). Embryos were transferred non-surgically into 111 recipients (83 single and 28 twin embryo transfers) and the pregnancy rate at 14, 21, 35, 60 days and 5 months was 57.7% (64/111), 55.0% (61/111), 53.2% (59/111), 47.7% (53/111) and 45.0% (50/111), respectively. There was no difference in pregnancy rate between single and double embryo transfers. Pregnancy loss between 21 days and 5 months of pregnancy was 18.0% (11/61) which led to an average of 2.1 pregnancies (50/24) per responded donor. After 5 months of gestation, 1 animal died and 3 aborted (2 single and 1 twin conceptuses) so a total of 46 recipients delivered a live calf from selected donor dromedaries with high genetic potential.

In this study, embryo transfer was carried out during lactation of the donors when camels would traditionally not be bred. The results demonstrated that multiple offspring can be obtained from high-producing donors while the animals continue milking and the pregnancy rates were similar to those reported earlier in commercial embryo transfer programs (McKinnon et al., 1994; Tibary and Anouassi, 1997a). Embryonic mortality was also in the same range of previous studies following embryo transfer or natural mating (Nagy and Juhasz, 2012; Skidmore and Billah, 2005). This finding is surprising as embryo transfer has not been recommended at the end of the breeding season due to increased embryonic death during the hot summer months (Tibary and Anouassi, 1997a). In the first and second seasons 3.5% (15/422) and 9% (35/390) of the pregnant camels in the herd were embryo transfer recipients, respectively.

7. Conclusion

Due to several physiological and management reasons, genetic improvement for milk production has been negligible in dromedary camels. The large pool of various types of animals at the EICMP, supported by the solid production database, provides a unique opportunity for selective breeding to accelerate genetic progress. Embryo transfer has great potential this regard, and has been an indispensable tool for increasing selection intensity and decreasing the generation interval by obtaining multiple offspring from females of high genetic potential. In addition, embryo transfer can be performed during lactation when camels would not traditionally be bred. This is the first report on embryo transfer using high-producing lactating dromedary camels as donors and low-producing animals as recipients. The model has been effective and resulted in acceptable pregnancy rates.

Conflict of interest statement

None.

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