Reproduction in the endangered African wild dog: basic physiology, reproductive suppression and possible benefits of artificial insemination.

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Abstract

The African wild dog (*Lycaon pictus*) is an endangered exotic canid with less than 5500 animals remaining in the wild. Despite numerous strategies to conserve this species, numbers of free-living animals are in decline. It is a highly social species with a complex pack structure: separate male and female dominant hierarchies with, typically, participation of subdominant adults in the rearing of the dominant breeding pairs' pups. Basic reproductive knowledge is largely missing in this species, with only limited information available on the profile of reproductive hormones, based on non-invasive endocrine monitoring. The dominant or alpha male and female are reproductively active and the subdominants are generally reproductively suppressed. However, the occasional production of litters by subdominant females and evidence of multiple paternity cases suggests fertility of subordinates is not completely inhibited. Despite some general knowledge on the reproductive biology of this species there are considerable gaps in our knowledge about the mechanisms governing reproduction and reproductive suppression in African wild dogs, particularly the influence of dominance and pack structure on both male and female fertility. Given concerns over the long-term survival of this species, further research in this area is essential to provide valuable information for their captive breeding and conservation. Reproductive information can also be applied in the development of Assisted Reproductive Techniques in this species, of which the utility in African wild dog conservation is hereby discussed.

Key words

African wild dog; Reproduction; Dominance; Artificial Insemination; Seasonality; Oestrous Cycle; Pregnancy; Paternity

1. Introduction

The African wild dog (*Lycaon pictus*), like the domestic dog (*Canis familiaris)* and wolf (*Canis lupus*), belongs to the Canidae (order Carnivora). Formerly occurring throughout sub-Saharan Africa, African wild dogs have disappeared from most of their original range with less than 5500 animals left in the wild, and are now one of the most endangered canids in the world (McNutt *et al.*, 2008). The key threats affecting free-living African wild dogs are habitat fragmentation and loss, infectious diseases, intra- and interspecies competition (mostly with lions *Panthera leo* and hyaenas *Crocuta crocuta*) and anthropogenic mortality (e.g. persecution and road accidents) (Creel & Creel, 1998; Woodroffe *et al.*, 2007). Numerous strategies have been undertaken to preserve the species, including re-introduction (Graf *et al.*, 2006), community awareness and education (Gusset *et al.*, 2008), and captive breeding programs (Frantzen, Ferguson & de Villiers, 2001), but still their numbers are in decline (McNutt *et al.,* 2008). African wild dogs have large home ranges from 1500 - 2000 km^2 and low population densities (Creel & Creel, 2002; Woodroffe *et al.,* 2007), making them relatively vulnerable to habitat fragmentation (Buettner *et al.*, 2006) and to contact with humans or human activities. In South Africa, the Kruger National Park is the only protected habitat considered large enough to contain a viable self-sustainable population (Gusset, Slotow & Somers, 2006). Re-introductions and translocations have been performed in order to supplement the original population (Gusset, Slotow & Somers, 2006). These translocations are performed to sustain a single population composed of different isolated subpopulations. This human intervention is called metapopulation management (Davies-Mostert, Mills & MacDonals, 2009).

African wild dogs commonly live in packs of five to 15 adults and yearlings, and show a complex social structure consisting of separate male and female dominance hierarchies (Creel & Creel, 2002). The alpha male and female have almost exclusive reproductive privileges, while subdominants rarely breed but help to rear pups (Girman *et al.,* 1997). This cooperative breeding system is also seen in several other carnivores including meerkats (*Suricata suricatta*), gray wolves (*Canis lupus*) and dwarf mongooses (*Helogale parvula*) (Creel, 2005; Young *et al.,* 2006). In the wild, there is a positive relationship between pack size and successful breeding, hunting and survival (Courchamp & Macdonald, 2001), with a critical threshold of at least five animals in a pack (Buettner *et al.,* 2006; Graf *et al.*, 2006). Thus, the failure of some reintroduction attempts might be explained by the inability to form or maintain a pack of five or more animals, coupled with a lack of sufficient numbers of separate packs to establish a genetically self-sustaining population (Gusset *et al.,* 2009). Dispersal of adult animals typically involves single-sex groups and occurs at an older age in males (median age 28.1 months) than in females (median age 21.8 months) (McNutt, 1996). A new pack is mostly formed when two opposite-sex groups join together and, after a 'trial period', a stable reproductive unit is formed (McCreery & Robbins, 2001; Creel & Creel, 2002). During the 'trial period', different factors like group size, mate choice and competition are responsible for the formation of a stable social structure. Annulment of a pack can occur within several months of initial association (McCreery & Robbins, 2001).

In this article we provide an overview of current reproductive knowledge and the possible mechanisms of reproductive suppression in the African wild dog. In addition, we identify areas requiring further research, and discuss the merits of using Assisted Reproductive Techniques (ART) towards the conservation of this species.

2. Reproduction in African wild dogs

2.1. Oestrous cycle and mating

Most Canidae studied to date show similar reproductive features: a mono-oestrous cycle with a long pro-oestrous and oestrus, a pregnant or non-pregnant (pseudopregnancy) period of dioestrus and an obligatory period of anoestrus.

As with most endangered wild-living species, there is a dearth of knowledge about female reproductive physiology; research into which is difficult to perform. Limited data has been collected using blood serum in African wild dog bitches (van Heerden & Kuhn, 1985). However, improved techniques using non-invasive endocrine monitoring, now permits basic reproductive information to be obtained on a more regular basis (Goodrowe *et al.,* 2000). Faecal samples have been used to assess steroid metabolites by radio and enzyme immunoassays in African wild dogs (Creel *et al.*, 1997; Monfort *et al.*, 1997; Johnston *et al.*, 2007; Santymire & Armstrong, 2009).

Behaviourally the approach (approximately 1½ months prior to the onset of pro-oestrus) of the breeding season in captive dogs in South Africa is marked by increased intra-pack aggression, which mostly involves females (Boutelle & Bertschinger, 2010). Fighting as a result of aggression may become so severe that deaths occur and is indeed the most common cause mortality in adult and subadult dogs in captivity in South Africa (van Heerden, 1986; van Heerden *et al.*, 1996). Mortality resulting from intra-pack aggression is presumably less common in free-ranging dogs whereas avoidance amongst a pack held in captivity is difficult (Boutelle and Bertschinger, 2010).

Studies in captive females show that the period of pro-oestrus and oestrus takes 14-20 days (van Heerden & Kuhn, 1985). During pro-oestrus female receptivity slowly increases before mating, during which time the bond between the dominant male and female strengthens (Creel *et al.,* 1997). During pro-oestrus the vulva is obviously tumescent but serosanguineous vaginal discharge, although it may occur, has not been described in the African wild dog (H.J. Bertschinger, personal communication). Oestrus, which is accompanied by a reduction in vulval tumescence, lasts six to nine days and is associated with an increase in oestrogen metabolites (Monfort *et al.,* 1997). Mating occurs over a period of three to seven days at the time of peak or declining oestrogen and increasing progesterone metabolite concentrations (Creel *et al.,* 1997; Monfort *et al.,* 1997). The copulatory tie observed in all canids (Asa & Valdespino, 1998), is very short in African wild dogs and can easily go undetected in captivity (H. Verberkmoes, personal communication). A copulatory tie was observed in Madikwe Game Reserve and lasted of 15 minutes and, contrary to domestic dogs, the male did not turn around once the tie had been achieved. The female first sat and then lay down on her side during the tie with the male obviously following suit. On the other hand a tie was observed at the Ann van Dyk Cheetah Centre where the male did turn and was of short duration (H.J. Bertschinger, personal communication). In addition, the measurement of faecal progesterone metabolites collected individually from group-housed individuals, has also shown that females appear to cycle in the absence of males (Paris *et al*., 2008). Research is currently ongoing to investigate this in more detail (L. Van der Weyde, personal communication).

2.2. Pregnancy, parturition, litters and the bias in sex-ratio of offspring

Pregnancy lasts approximately 72 days (van Heerden & Kuhn, 1985; Creel *et al.,* 1997; Monfort *et al.,* 1997), and similar to the domestic dog, where pregnancy dioestrus takes 57 days (Gudermuth *et al.*, 1998; Verstegen-Onclin & Verstegen, 2008), parturition coincides with a drop in progesterone, as evidenced by faecal progesterone metabolites (Monfort *et al.,* 1997). The number of nipples on an African wild dog bitch can vary between 6 and 8 pairs (van Heerden & Kuhn, 1985). Litter sizes are large, consisting of approximately 10-12 pups

(Comizzoli *et al.,* 2009). Some populations of African wild dogs show a bias in the sex-ratio of litters with primiparous bitches producing more male pups than multiparous ones (Creel, Creel & Monfort, 1998; McNutt & Silk, 2008). The exact mechanism underlying this phenomenon is yet to be determined, but elevated oestrogen levels in primiparous bitches has been proposed as a potential cause by either affecting uterine implantation of zygotes or by sperm selection in the female reproductive tract (Creel, Creel & Monfort, 1998). While lactation in subdominant females is common in wolves (Asa & Valdespino, 1998), it is rare in African wild dogs (Creel *et al.,* 1997). Weaning takes place at about 10 weeks (reference) although they start taking in regurgitated food already at 14 days of age (Smithers, 1983).

2.3. Seasonality of reproduction

Most Canids seasonally reproduce (Asa & Valdespino, 1998). In the case of the African wild dog, most pups are born in the southern hemisphere between May and July (McNutt, 1996; Buettner *et al.,* 2006), however it shifts by up to 6 months in animals living in the northern hemisphere (Monfort *et al.,* 1997; Asa & Valdespino, 1998; Verberkmoes, 2008; Table 1).

Table 1: Major reproductive season of the African wild dog in the Southern and the Northern hemisphere.

	Southern hemisphere	***Northern hemisphere***
Oestrus	February - May	Late August - early October
Birth	May - July	October - December

Based on European regional studbook information, the major breeding season for captive African wild dogs in Europe is around August/September resulting in a peak of births in November (Verberkmoes, 2008) (Fig. 1a). When births were grouped by the latitude at which animals were housed, no obvious differences in the pattern of peak births was observed (Fig. 1b). Births however, do occur at lower levels year-round, with the least occurring in July and August (1.1 and 0.5% respectively). Although considered strictly mono-oestrus, some captive African wild dog bitches in Europe also show a second minor breeding season between February and March (with a corresponding increase in births during April), if they fail to become pregnant during the main breeding season or if they lose their pups (H. Verberkmoes, personal communication; Fig. 1b). Similarly, a second minor breeding season in African wild dogs has been observed in South Africa (Boutelle and Bertschinger, 2010).

The collective evidence suggests that reproduction of the African wild dog is generally seasonal, yet births can occur at every month of the year, supporting the idea that the window of fertility of the African wild dog is broader than described for temperate and arctic zone canids. Valdespino (2007) showed a negative relationship between latitude and the duration of the reproductive season in canid species, with longer reproductive seasons occurring at lower latitudes. Seasonal reproduction is mainly influenced by photoperiod, but also by other factors like temperature, body condition and nutritional intake as described for example in mares (Nagy et al., 2000).

In most seasonally breeding Canidae including maned wolves (*Chrysocyon brachyurus*; Velloso et al., 1998 and Maia et al., 2008), red and blue foxes (*Vulpes vulpes* and *Alopex lagopus*; Farstad, 1998 and Andersenberg et al., 2001) and coyotes (*Canis latrans*; Minter and DeLiberto, 2008), testosterone levels, testis size and semen production increase in

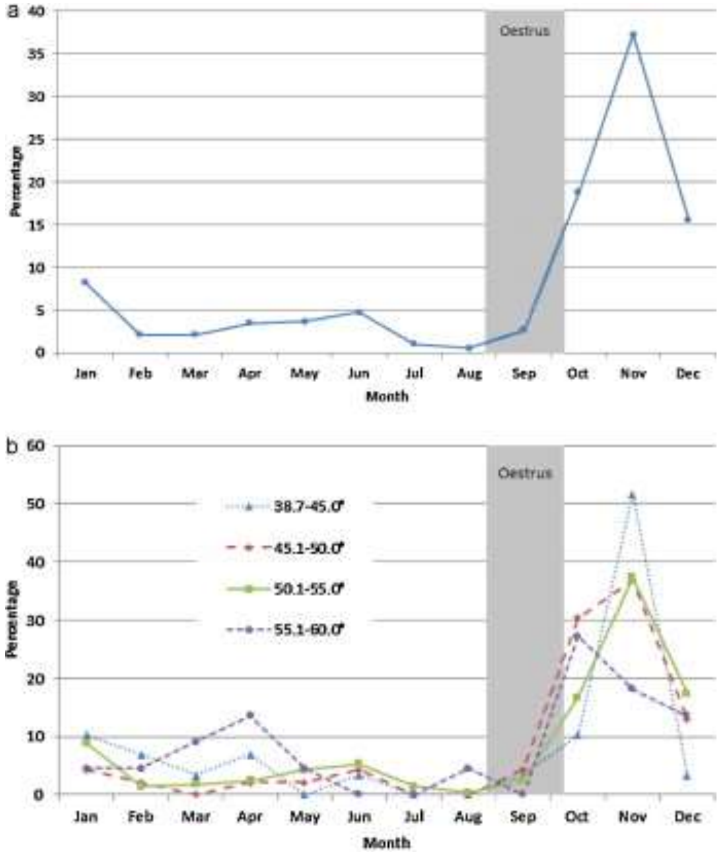

Fig. 1. Monthly percentage of African wild dog litters born in captivity in Europe from 1938 to 2008 presented as (a) combined data and (b) grouped by latitude. The period of oestrus is indicated in grey. Data based on the European regional studbook (Verberkmoes, 2008).

males during the breeding season. In the African wild dog, the measurement of testosterone levels has given conflicting results (Creel et al., 1997, Monfort et al., 1997 and Johnston et al., 2007), but testis size appears to increase in the breeding season and semen could not always be collected at other times of the year (Johnston et al., 2007). However, failure to collect semen by electroejaculation does not conclusively prove spermatogenic arrest in this species, because seasonal reduction in male accessory glands may also reduce the surface contact of the probe and effectiveness of the electrostimulation (D. Paris, personal observation; H.J. Bertschinger, personal observation). Moreover, the European regional studbook data (Verberkmoes, 2008) presented in Fig. 1 suggest that males are reproductively fertile throughout most of the year. This could indicate that female fertility is generally photoperiod-dependent while male fertility is opportunistically primed by female pheromonal cues. A similar phenomenon has been observed in the tammar wallaby (*Macropus eugenii*) in which female seasonality is strictly controlled by photoperiod while males are fertile year-round; but the quality of their semen decreases outside the main breeding season (Paris et al., 2005). Similarly, in African wild dogs, semen quality (percentage motile sperm, motility rating and sperm morphology) was poorer in out-of-season samples than in-season samples (Nöthling et al., 2002). In male red wolves (*Canis rufus*), it has been shown that faecal androgen concentrations begin to rise four months prior to the onset of oestrus in females, with peak concentrations coinciding with maximal sperm production (Walker et al., 2002). Thus, although spermatogenesis does not appear to be completely arrested in male African wild dogs, they would need to detect and respond to female pheromonal cues sufficiently early to ensure optimal reproductive synchronicity between the sexes.

In conclusion, even though there were a few births of captive African wild dogs in Europe outside the main breeding season, the chances of fertilization and successful pregnancy are likely to be much lower due to (i) a lack of female cyclicity, (ii) poor semen quality influenced by lower testosterone levels, (iii) a lack of reproductive synchronicity between males and females, and (iv) a myriad of environmental effects that could include latitude, photoperiod, temperature, changes in housing and diet during winter, etc.

3. Suppression of reproduction

3.1. Stress-induced suppression of reproduction?

The mechanism underlying reproductive suppression that inhibits or greatly reduces the fertility of subordinates is not well understood. The relationship between social dominance and circulating steroid hormones may be one possible mechanism involved in reproductive suppression. In many species with a complex social structure, hierarchy and dominance are related to the level of circulating glucocorticoids (GC) (Creel, 2001 and Creel, 2005). In rats and primates, dominant animals show lower cortisol concentrations than subdominant animals (de Villiers et al., 1997 and Creel, 2001). ‘Social stress’ experienced by subordinates, can cause a chronic increase of GC secretion (Creel, 2001). It is known that chronic augmentation of these hormones costs/requires energy and can suppress other physiological systems not immediately necessary for survival and, in the case of the reproductive system, can cause ‘psychological castration’ (Creel et al., 1997 and Barja et al., 2008). One cooperative breeder in which stress-related suppression of reproduction is an important strategy is the meerkat (*Suricata suricatta*), where pregnant females chase subordinate females from the group resulting in an increase of GC levels in subordinates and a down-regulation of their reproduction (Young et al., 2006). However, in many other cooperative breeders such as wolves (*Canis lupus*), female dwarf mongooses (*Helogale parvula*) and male and female African wild dogs, it is dominant animals that exhibit higher GC levels than subordinates without any adverse effect on their fertility (Creel et al., 1997, Creel, 2005 and Barja et al., 2008). These conflicting observations across species exemplify the Wingfield's challenge hypothesis stating it is sometimes more stressful to be dominant and it is sometimes more stressful to be subordinate (Wingfield et al., 1990).

3.2. Multiple maternity in packs and suppression of female reproduction

Stress-induced reproductive suppression may also be an unlikely mechanism in African wild dogs because both subdominant males and females remain fertile to some extent. In the wild, the alpha female produces 75–81% of all litters (Creel, 2005). Multiple maternity in packs, where subdominant females also reproduce, occurs in 40% of the pack years, but only eight percent of the subdominant's pups survive beyond their first year (Girman et al., 1997). This high pup mortality is probably partly due to infanticide by the alpha female observed both in the wild and captivity (Girman et al., 1997 and Robbins and McCreery, 2000). In cases where subdominant females copulate, it is generally the beta female that does so (Creel et al., 1997). Subdominant females do cycle (van Heerden and Kuhn, 1985 and Paris et al., 2008), but the frequency and extent to which beta or lower ranked females can reproduce is unknown, leaving crucial gaps in our understanding of reproductive suppression in this species. Bertschinger et al. (2002) observed oestrus at two to three week intervals in three captive females housed together in the same enclosure. Hofmeyr (1997) observed breeding by two sub-ranking females after three captive-born females were co-housed in a pre-release enclosure for five months with three wild males. However, none of the pups survived. One year after release, the alpha female was mated in February by all three males, gave birth in May but emerged from the den without pups which were presumed dead. Subsequently, both the second and third-ranking females exhibited oestrus and were mated at different times in April, and gave birth in a shared den at the end of June and middle of July respectively. The third-ranked female emerged with her seven pups after fighting displaced the second-ranked female, whose litter presumably had been killed. However, the following year, the original second-ranking female became alpha female and produced 12 pups. Thus, it appears that subdominant females of any rank are reproductively fertile, but the opportunity to successfully raise their pups appears to be status/hierarchy-dependent.

In gray wolves, all subdominant females ovulate and mating is suppressed by dominant female behaviour (Asa and Valdespino, 1998), while in some primates, reproduction is physiologically suppressed by arresting pubertal development (Abbott et al., 1981). Behavioural inhibition of copulation among subdominant animals has often been observed and is recognized as a mechanism of reproductive suppression in African wild dogs (van Heerden and Kuhn, 1985). It is possible that behavioural suppression allows all African wild dog females to ovulate but prevents copulation in subordinate females, resulting in a period of pseudopregnancy that makes them more receptive as maternal carers. This may reflect the high energy demands required to successfully raise a single litter within the pack.

3.3. Suppression of male reproduction

In males, subordinates occasionally copulate but to a lesser extent than the alpha male (Creel et al., 1997). This raises questions about the fertility of these subdominant matings. Testosterone levels, testis size, and semen production are positively correlated in mammals (Preston et al., 2001 and Gomendio et al., 2007). During the breeding season in African wild dogs, the dominant male shows higher testosterone levels than subdominants (Creel et al., 1997, Monfort et al., 1997 and Johnston et al., 2007). High testosterone levels can positively influence both spermatogenesis and the size and secretory activity of accessory sex glands (Paris et al., 2005 and Gomendio et al., 2007). Although spermatogenesis does not improve further once a certain threshold of testosterone has been reached (Walker, 2009), dominant males with higher testosterone could have higher quality semen than subordinates. The fact that testis size in subdominant male African wild dogs also increases during the breeding season (Johnston et al., 2007), support the idea that spermatogenesis is not arrested as a result of dominance. However, when subjected to electroejaculation during the breeding season, most males in the captive pack produced spermatozoa, but mean ejaculate quality was reduced once the dominance hierarchy was established (Johnston et al., 2007). This suggests that dominance may affect subordinate male fertility. Unfortunately, samples were pooled for analysis in this study, making it unclear whether the overall decrease in semen quality was specifically caused by poor semen from subdominant males. Thus, the extent to which dominance and pack structure may positively or negatively affect fertility requires further investigation.

Moreover, dominance and optimal timing of or higher rates of copulation do not always result in higher reproductive success. In the tammar wallaby (*Macropus eugenii*), despite dominant males guarding and being the first to mate with oestrous females at the optimal time of copulation, they sire only half of the offspring born compared to second, third and fourth ranking males (Hynes et al., 2005). In Soay rams (*Ovis aries*), larger dominant males show a very high rate of mating, but this eventually leads to sperm depletion as the mating season progresses, making them less fertile than subordinate males (Preston et al., 2001). In African wild dogs, Girman et al. (1997) showed multiple paternity in at least 10% of litters and, in one case, paternity was also assigned to the brother of the dominant male. This suggests that intra-pack mating does exist in the wild, and demonstrates that at least some subdominant males are fertile irrespective of whether they are related or unrelated to the dominant male. Multiple paternity was reported in five litters sampled at 9–12 months old, from three free-ranging packs in South Africa (Mouiex, 2006). In one litter of eight, four pups were sired by the alpha male, three by the second and one by the third ranking male. In the four remaining litters there were two sires each and the numbers of pups sired by the alpha and second ranking male were 11 and one, three and one, five and one and seven and one, respectively. This supports observations in Madikwe Game Reserve of one female being mated by three different males in order of ranking (M. Hofmeyr, personal communication). Subdominant male fertility was also recently demonstrated by Spiering et al. (2010) in which approximately half of the pups were sired by the alpha male, while remaining pups were sired by the second and third ranking males.

Although subdominant males appear to be able to sire offspring, the extent of this fertility/sub-fertility is not definitively clear. Indeed evidence of reduced semen quality and limited paternity success support the idea that there is at least some degree of reproductive suppression that limits mating access and fertilization success of subdominant males. Besides behavioural suppression, at this stage we cannot exclude other hormonal or pheromonal cues that may act on the hypothalamic–pituitary–gonadal axis to induce sub-fertility in male African wild dogs.

4. Further research in African wild dog reproduction

Infertility or declining reproduction is not the cause of endangerment in African wild dogs (Comizzoli et al., 2009). When pack size is sufficiently large and resources plentiful, the dominant pair will produce large litters once per year in the wild (Courchamp and Macdonald, 2001). In captivity, sufficient numbers of pups are also produced to maintain an adequate population size. However, it is critically important for the long-term captive propagation of this species, to carefully regulate breeding partners and maintain a population size that will effectively maximize and maintain current levels of genetic diversity (Frantzen et al., 2001). In order to achieve this and to avoid inbreeding depression, current strategies involve the regular translocation of live animals. In captivity, male and female single-sex groups are often combined to constitute a new social unit in an attempt to imitate dispersal patterns that occur naturally in the wild (H. Verberkmoes, personal communication). Such introductions are made difficult because of the complex social structure of these animals coupled with the unnatural space-limited environment often present in zoos; that can cause stress to animals and result in aggression and sometimes morbidity and mortality. Thus research directed at overcoming problems of aggression is essential. One such approach has been undertaken by Vlamings et al. (2009) who investigated whether Dog Appeasing Pheromone (DAP) can be used to minimise aggression during introductions, with the hope of reducing cases of morbidity and mortality. A second approach, could involve the use of artificial insemination to infuse new genes into existing groups without disrupting their social hierarchy by the introduction of new individuals (see the following section).

In addition to modifying behaviour, studies directed at further understanding and controlling the female reproductive cycle of the species could help improve animal welfare and captive conservation management. For example, there is an urgent need to improve current methods of contraception for genetically over-represented captive individuals. Previous contraceptive administration of progestins, have been shown to greatly increase the risk of developing pyometra, to which the African wild dog bitch is highly susceptible (Hermes et al., 2001 and Boutelle and Bertschinger, 2010). Deslorelin, a GnRH agonist, is currently the safest method for inducing reversible contraception in all carnivores, including the African wild dog, but further research is needed to gain more information on dosage and reversibility (Bertschinger et al., 2001, Bertschinger et al., 2002 and Boutelle and Bertschinger, 2010).

Basic knowledge is still missing on reproductive hormone profiles in both female and male African wild dogs, as well as the effect of season, dominance and pack structure on fertility. Endocrine monitoring of faecal samples can be used non-invasively to answer many of these questions. Such endocrine data coupled with behavioural observations collected in the northern hemisphere during the 2009 season from group-housed individuals in the presence or absence of males, is currently being analysed (L. Van der Weyde, unpublished data). These studies also incorporate endocrine data collected from free-ranging animals in Hluhluwe–Umfolozi Game Reserve, South Africa.

5. Potential role for assisted reproductive techniques in the African wild dog

Artificial Insemination (AI) coupled with semen cryopreservation has long been considered one of the most powerful and least invasive forms of Assisted Reproductive Techniques (ART) for the preservation, distribution and improvement of animal genetics

(Durrant, 2009). Its value and success in overcoming infertility in humans and animals as well as improving livestock production (e.g. increased milk production or meat quality) is illustrated by its widespread application (Mastromonaco et al., 2011). These techniques are being increasingly incorporated into the captive breeding programs of a wide range of wildlife species. In this regard, perhaps one of the greatest AI success stories has been the birth and wild re-introduction of over 139 endangered black-footed ferret (*Mustela nigripes*) kits using AI and cryopreserved semen (Howard and Wildt, 2009). Multiple births have resulted from AI in other mammals including a variety of non-domestic felids, cervids, non-domestic bovids, camelids, marsupials, primates, ursids and pachyderms (Paris and Mastromonaco, 2009). Moreover, in at least one case, AI has already been conducted successfully in captive North-American cheetahs (*Acinonyx jubatus*) using frozen–thawed semen from wild-caught males without the need to remove these males from their natural habitat (Howard and Wildt, 2009). Despite these successes, the lack of a strong working relationship between conservation biologists/animal managers and reproductive specialists, as well as general distrust of ‘artificial’ manipulations of reproduction, has created one of the obstacles that prohibit the widespread use of AI in wildlife species (Holt and Lloyd, 2009 and Mastromonaco et al., 2011). It should be recognized that although both these groups have differing ideologies, they share a common goal in striving for the propagation and conservation of threatened species. Increased dialogue is needed to outline the merits of AI to overcome species-specific problems (such as the highly complex social structure and hierarchy of the African wild dog) that influence the success and genetic management of captive natural breeding programs.

In Canidae successful AI using both fresh and frozen semen has been widely performed in foxes and wolves (Thomassen and Farstad, 2009). Several wolf species are threatened by inbreeding and human interference (Thomassen and Farstad, 2009). Since wolves have a complex monogamous social structure (Asa and Valdespino, 1998), AI could permit the introduction of new blood without disrupting established pair-bonds.

However, reproduction is regulated by a series of species-specific mechanisms and patterns of hormonal cyclicity. As a result of such differences, reproductive information cannot always be extrapolated between species, even if closely related (Paris et al., 2007). Even within the Canidae, there are several important differences in seasonality and reproduction (Table 3 in Wildt et al., 2010). This lack of basic reproductive knowledge in many endangered animals is often the reason why ARTs cannot be used as an effective method to help rescue a critically endangered species already on the brink of extinction (Holt and Lloyd, 2009).

The use of AI has yet to be reported in the African wild dog, and only two publications currently describe the cryopreservation of semen in this species (Hermes et al., 2001 and Johnston et al., 2007). The development and optimisation of these techniques is of immediate priority while viable populations of animals still exist. In addition, the establishment of a genome resource bank containing cryopreserved semen of genetically valuable animals, together with basic reproductive research, delivers a certain level of insurance for the future of African wild dog populations. Such banks can provide a buffer against possible threats such as fires or sudden epidemic of infectious diseases both in captivity and in the wild (Pukazhenthi et al., 2007). Indeed, an outbreak of Canine distemper virus in a captive African wild dog breeding group in 2000 resulted in the death of 49 out of 52 animals within two months (van de Bildt et al., 2002). During the late eighties in the Masai Mara and Serengeti National Parks bordering Kenya and Tanzania, disease resulted in the disappearance of 8 entire African wild dog study packs (Woodroffe and Ginsberg, 1997), although recent evidence suggests African wild dogs have persisted in the Serengeti–Mara (Marsden et al., 2012). Gene (semen) banking initiatives coupled with artificial insemination techniques, therefore, should be considered as important for conservation as disease prevention, habitat preservation or community education. Moreover, since African wild dogs have a complex social structure, with strict dominance hierarchies, AI could overcome the high levels of intra-pack aggression associated with the translocation and introduction of new genetically valuable animals (Johnston et al., 2007). Transportation of semen instead of live animals to infuse new genes into a group cannot only improve animal welfare, by reducing translocation- and introduction-associated aggression, but can also provide economic and ecological benefits. Transportation of semen is cheaper, avoids the removal of animals from the wild, and can also decrease the incidence of disease transmission. In the wild, cryopreserved semen and AI could potentially be used to facilitate meta-population management so as to avoid inbreeding in fenced reserves that are smaller than the range required for African wild dog populations to be self-sustaining, or in cases where natural dispersal is limited (M. Szykman, personal communication).

6. Conclusion

The African wild dog is an endangered canid with a dominance hierarchy and a cooperative breeding strategy. Its reproduction appears to be broadly seasonal and females are generally mono-oestrus, although lower levels of fertile mating may occur year round. Collective evidence suggests that reproductive suppression of subdominant animals primarily occurs at the behavioural level, since both male and female subdominant individuals occasionally produce a limited number of offspring but the success of raising subdominant female litters is greatly reduced. However, it is yet to be determined whether dominance and pack structure have secondary effects that reduce the fertility of subdominant individuals via other mechanisms (such as hormone-induced or pheromone-induced suppression). There are still considerable gaps in our knowledge of male and female reproductive hormone profiles and female cyclicity. Moreover, efforts need to be directed toward the management of intra-pack aggression, the development of sperm cryopreservation and artificial insemination, and the improvement of contraception as complementary strategies to genetically manage both captive and wild populations.

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