

EAZA Best Practice Guidelines

Cheetah (*Acinonyx jubatus*)



Cheetah at Cheetah Conservation Fund Namibia (CCF)

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Preamble

Right from the very beginning it has been the concern of EAZA and the EEPs to encourage and promote the highest possible standards for husbandry of zoo and aquarium animals. For this reason, quite early on, EAZA developed the “Minimum Standards for the Accommodation and Care of Animals in Zoos and Aquaria”. These standards lay down general principles of animal keeping, to which the members of EAZA feel themselves committed. Above and beyond this, some countries have defined regulatory minimum standards for the keeping of individual species regarding the size and furnishings of enclosures etc., which, according to the opinion of authors, should definitely be fulfilled before allowing such animals to be kept within the area of the jurisdiction of those countries. These minimum standards are intended to determine the borderline of acceptable animal welfare. It is not permitted to fall short of these standards. How difficult it is to determine the standards, however, can be seen in the fact that minimum standards vary from country to country. Above and beyond this, specialists of the EEPs and TAGs have undertaken the considerable task of laying down guidelines for keeping individual animal species. Whilst some aspects of husbandry reported in the guidelines will define minimum standards, in general, these guidelines are not to be understood as minimum requirements; they represent best practice. As such the EAZA Best Practice Guidelines for keeping animals intend rather to describe the desirable design of enclosures and prerequisites for animal keeping that are, according to the present state of knowledge, considered as being optimal for each species. They intend above all to indicate how enclosures should be designed and what conditions should be fulfilled for the optimal care of individual species.

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Summary

The content of this document is divided up into two sections. The Biology and Field data of the species describes the natural range, habitat, behaviour, longevity and Conservation status of cheetah “*in-situ*”. The Management in Zoos and Aquariums describes in eight chapters all captive specifications for the best practice regarding the keeping of cheetah in captivity including enclosure design, nutrition, breeding, handling and veterinary care.

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Introduction

In this report the best practice guidelines for the cheetah (*Acinonyx jubatus*) are discussed. The best practice guidelines are developed in order to help optimise the conditions for the wellbeing of the animals kept in captivity. The guideline is divided into two sections. The first section provides general information on the species biology, conservation status, ecology, diet, reproduction and behaviour. The data is obtained from different literature sources, such as books, articles and the Internet. The second section contains information on the actual management in captivity. This section presents recommendations on the enclosure, diet, social structure, breeding, behavioural enrichment, handling and veterinary considerations for cheetahs. The data to write this section is acquired from different literature sources and from the results of a survey that was conducted among EAZA zoos that participate in the EEP program for cheetahs.

Section I: Biology and field data

In this section biological information and field data are combined to give a general overview of the cheetah. Topics such as the taxonomy, geographic range, morphology, reproduction, behaviour and conservation status are discussed.

1.1 Taxonomy

Cheetahs are classified into the order Carnivora, family Felidae and genus *Acinonyx*. The genus *Acinonyx* comes from the Greek words “akaina” and “onyx”, which means thorn and claw, respectively. This refers to the dog-like claws cheetahs possess (Caro, 1994; San Diego Zoo, 2002). The species name *jubatus* is Latin, it hints to the unique mantle the cubs exhibit in their first year (San Diego Zoo, 2002).

Amongst this species there are 5 recognized sub-species:

- 1) *Acinonyx jubatus hecki* (Hilzheimer, 1913): Northwest Africa.
- 2) *Acinonyx jubatus fearsoni* (Smaith, 1834): East Africa.
- 3) *Acinonyx jubatus jubatus* (Schreber, 1775): Southern Africa.
- 4) *Acinonyx jubatus soemmerringi* (Fitzinger, 1855): Northeast Africa.
- 5) *Acinonyx jubatus venaticus* (Griffith, 1821): North Africa to Central India (Durant, 2015).

The cheetahs’ common name originally stems from the primary sacred language of Hinduism, Sanskrit. The word used was “chitraka”, which means speckled or spotted. This led to the Hindi word “chita”, which means, "spotted one". Later this term was anglicised into the name we use today (Hunter, 2003). In the following table (Table 1), the word ‘cheetah’ is displayed in different languages.

Table 1: Translation of cheetah into several languages

Languages	<i>Acinonyx jubatus</i>
Dutch	Jachtluipaard
English	Cheetah
French	Guépard
German	Gepard
Spanish	Guepardo

1.2 Geographic range

Historically the cheetah was a widespread species that could be found in 38 nations of Africa. A survey showed that in the early 1970s, around 7000 to 23000 cheetahs were living in Africa. Nowadays, the cheetah population is estimated to be around 6700 individuals distributed across 29 African countries, see figure 1 (Caro, 1994; CCF, n.d.; Durant, 2015). The Asiatic cheetah could be found in different parts of Asia, but has become extinct throughout most areas except for a small population of around 50 individuals that occurs in Iran (CCF, n.d.; Hunter, 2003). Overall, cheetah densities are low in comparison to other carnivores. They range between 0.25/100 individuals per km² to 5.0/100 individuals per km² (Caro, 1994).



Figure 1: Historic and present cheetah distribution (CCF, n.d.)

1.3 Habitat

Cheetahs are distributed throughout Africa and therefore inhabit different types of habitat. They can be located in sparse sub desert, steppe and medium to long-grass plains (Jewelers, 2017b).

1.4 Physical description

Cheetahs reach adult body mass in the wild at 49-96 months of age. Sexual dimorphism can be observed, with the average male being larger than the average female (Marker, 2003). Table 2 and figure 2 display the mean body measurements for the cheetah. When born, cheetah cubs weigh around 200-350 gram each, but in captivity can reach 460 grams (Wilson, 2009). The cheetah cubs' mantle camouflages them from possible threats. On the top they exhibit smoky white hair and on the under parts they have dark fur. Before the cubs become two months of age the under parts lighten and their characteristic spots emerge. The white mane will disappear slowly and can still be seen in animals of up to one year of age (Wilson, 2009).

Table 2: Cheetah body measurements (Marker, 2003)

Body Measurement	Male	Female
Mean weight	45,6 kg	37,2 kg
Mean head-body length	125,5 cm	120,1 cm
Mean tail length	76,7 cm	72,5 cm
Mean total length	202,2 cm	192,4 cm
Mean chest girth	71,7 cm	67,3 cm
Mean shoulder height	77,0 cm	73,6 cm

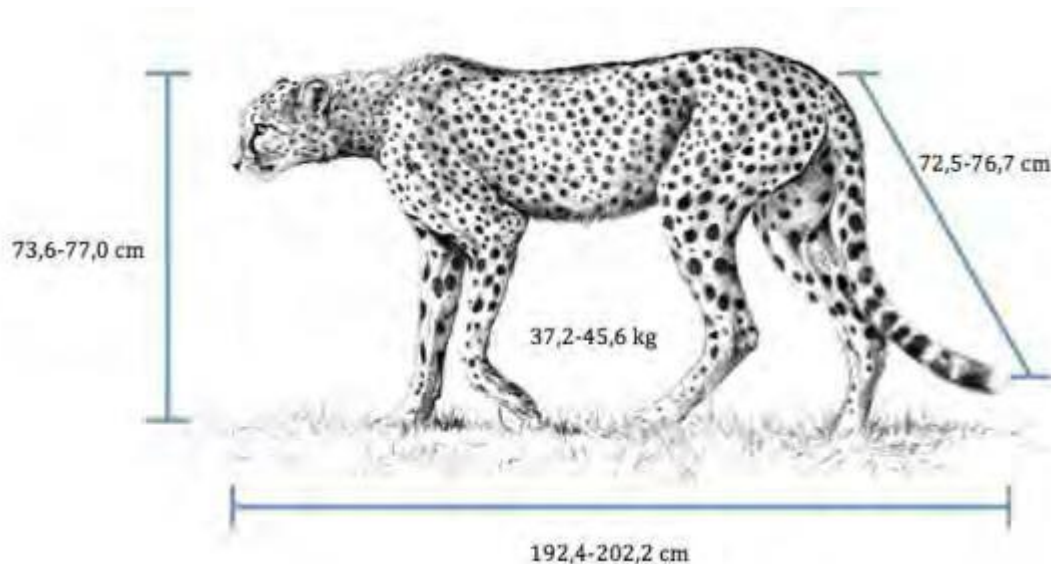


Figure 2: Body measurements cheetah (Thompson, 2013; Marker, 2003)

Adult cheetahs have orange-yellow irises with round pupils. Beneath their eyes and on their chin their fur is whiter. Their head and face have a spherical shape and their small ears are set far apart. The backs of the ears have black fur with white markings. On their face they exhibit lines that extend from the inner corner of each eye to the outer corner of the mouth called “tear lines”. Spots are also present on their head and cheeks. On the rest of the body the coat is slightly coarse with short hair. It is of yellowish colour, being darker along the mid-back with white fur on the chin, throat and belly. Additionally, it is covered all over with evenly spaced small black spots, these are only absent in the space between the ankles and the toes of the hind feet. On the tail, the spots fuse together forming two to three black tail rings, ending with a white tail tip (Wilson, 2009).

The “King cheetah” is a common cheetah that exhibits a fur pattern mutation. In order for this to happen a recessive gene has to be inherited from both parents (Jewelers, 2017a). In figure 3, a picture of a King cheetah next to a common cheetah is displayed.



Figure 3: King cheetah (African Safari Pictures, 2006)

The cheetah is the fastest land mammal on earth, reaching speeds up to 105 km/h. It is currently the only feline specialized in hunts via an extended velocity chase. In

order to reach such velocities their body has adapted, consequently straying from the basic cat form. They have the most elongated forelegs, where the radius and humours are almost identical and the lower limb bones are proportionally shorter. The elongated legs give the cheetah a longer stride. Over and above that, the bones of the lower legs and paws are thin and light. They have the longest and most flexible spine of any large cat, which helps to increase the stride length. When running, the cheetah has all his paws in the air for more

than half of every stride. The cheetah's claws can only be partially retracted and the paws miss the skin sheaths found in most other felids. At high speeds the claws can always be found fully extended; here they act like a sprinter's spikes, enhancing the grip. The dewclaw is a very important tool during the hunt. It is used by cheetahs to hook and trip the prey during the last moments of the chase. It is therefore the proportionally biggest dewclaw amongst large cats. Furthermore, the pads of the feet are very hard with ridges this increases traction. The tail is long, muscular and tubular to provide balance during fast changes in direction during the chase. Cheetahs have small canines with small roots (see table 3 for the dental formula); this creates space for the enlarged nasal passages that allow the cheetah to breathe while suffocating its prey. As well, they present enlarged lungs, bronchi, an oversized heart and muscular arteries. The normal respiratory rate of a cheetah ranges from 20-30 breaths per minute and rises up to 150-200 breaths per minute after a high-speed chase. The heart rate is approximately 120-170 beats per minute and rises to 200-250 beats per minute after a chase (Central Florida Zoo & Botanical Gardens, n.d.). All these features ensure the maximum delivery of oxygen after a chase (Hunter, 2003). The mean body temperature is $38.5 \pm 0.28^\circ\text{C}$ (Robyn, 2013).

Table 3: Cheetah's dental formula (Bloemfontein, 2017)

Cheetah's dental formula:	
$I \frac{3}{3} C \frac{1}{1} P \frac{3}{2} M \frac{1}{1} \times 2 = 30$	Incisors (I) Canines (C) Premolars (P) Molars (M)

1.5 Diet and feeding behaviour

Cheetahs are carnivores that specialize in Antelopes of medium size (between 20-60 kg) and complement their diet with hares. They kill their prey by suffocating them, but when killing hares they bite through their skull. Coalition members attempt to hunt larger prey than singletons, such as wildebeests. Other animals, which together contribute less than 5% of the cheetahs diet, include ostriches, bustards, guinea fowls, mole rats and cane rats. They exhibit limited prey versatility as a result of their physical specialisation for speed (Hunter, 2003). Young prey animals are always favoured over adults (Caro, 1994).

There are 5 different hunting methods described for the cheetah:

1. Walking slowly toward their prey in full view and brake into a sprint 60-70 m away.
2. Waiting crouched or sitting for their prey to move toward them.
3. If prey is distracted starting pursuit as far as 600 m.
4. Stalking its prey while walking semi crouched, freezing in mid-stride or dropping to the ground until it is close to launch an attack.
5. Flushing prey such as hares or neonate gazelles and pursuing them (Caro, 1994).

Cheetahs are diurnal animals that have two hunting peaks a day. The first between 7:00 and 10:00 hours and the second between 16:00 and 19:00 hours. During the rest of the day they can be found resting (Wilson, 2009). By displaying this schedule they try to avoid competing with predators such as lions and spotted hyenas. Due to their build, jaws and claws they are no match for other carnivores and will hand over their kills rather than risk injury by

defending it (Hunter, 2003). Another strategy to reduce the loss of a catch to competitors is to swallow their food quickly, they can consume up to 14 kg in one sitting. After they have suffocated their kill, they drag it to the nearest cover or shade and start by eating the hindquarters of their kill; they usually ingest around 2-4 kg/day. If water is available, cheetahs will drink every day (Caro, 1994; Wilson, 2009).

During gestation the female's diet changes to adjust to their demanding bodily requirements, such as the need to increase calcium. They start catching only smaller prey, like hares and newborn gazelles whose bones they can eat in its entirety (Caro, 1994). Moreover, as the pregnancy advances, their movements become more limited and the change in diet decreases the risk of injury. Likewise their hunting success increases, with a catch rate on newborn gazelles of almost 100% and 9 out of 10 hunts on hares end successfully (Hunter, 2003).

1.6 Reproduction

In captivity males reach sexual maturity between 1-2 years of age and females in their first 2-3 years (Ziegler-Meeks, 2009). Females then have a reproductive peak beginning on their fourth year and lasting until they are 9 years old. The youngest female to reproduce was 2 years old and the oldest female recorded was 11 years. In contrast, males are able to reproduce since their first year up until they reach 15 years of age (Bus, 2015). In the wild males have been seen to reproduce at 2,5-3 years and females at 2-3 years (Wilson, 2009). Cheetahs are aseasonal and conceive throughout the year, even though there is a conception peak during the wet season. This may happen due to differences in nutritional availability between wet and dry seasons, as there is an increase of gazelles born during the wet season (which is the main prey hunted by pregnant females) (Laurenson, 1992).

Female cheetahs are polyoestrous (they come into oestrus in multiple occasions), cycling in captivity approximately every 12 days (range 3 to 27 days) (Wilson, 2009). The onset of ovarian cyclicity is unknown. Oestrus cycle length is thought to be from 10-20 days with sexual receptivity ranging from 1-3 days and lengthy periods of anoestrous (Ziegler-Meeks, 2009). It is very difficult to detect when female cheetahs are in oestrus as the signs are extremely subtle. Females are seen spending more time on the ground resting and rolling, and they show less interest in food a day or two before the onset of oestrus. They defecate on conspicuous sites like for example termite mounds and large trees (Caro, 1994). Female cheetahs are induced ovulators (Hunter, 2003). The gestation period lasts on average 92 days, ranging from 90-98 days. The inter-birth period in the wild is 15-19 months (Wilson, 2009). Females often conceive while still caring for their last litter. However, they will separate from their cubs before giving birth to the next litter. If a female loses her litter, her cycle resumes and on average becomes pregnant within 19 days (Hunter, 2003).

After the gestation period the female gives birth to litters of average 3-4 cubs, but the number of cubs can range from 1-8 (Caro, 1994; Ziegler-Meeks, 2009). Cheetah cubs are altricial at birth. They have closed eyes, little locomotive skills and will open their eyes 4 to 11 days after birth. Young cheetahs start walking after 12 to 13 days when their eyes are open. They get milk teeth between 3-6 weeks, which are replaced by adult teeth at 8 months. They are weaned between the first 4 to 6 months of age. From 6 to 12 months onwards, the mother teaches the cubs how to hunt, even though at this time she still

provides most of the food. After the first 12 months she starts hunting together with the cubs (Eaton, 1970; Wilson 2009). The cubs become independent between 15-19 months of age (Wilson, 2009; Eaton, 1970).

Once the cubs have separated from the mother they form a sibling group consisting of females and males alike. These groups stay together until the cubs reach around 27 months of age. At this point, females will reach sexual maturity and split from the group. They will stay close to their mothers range whereas the males will leave avoiding breeding with relatives (Laurenson, 1992; Hunter, 2003). There is no evidence of mate fidelity. Over and above, females in the wild mate with different males, as a result there are high levels of multiple paternities in each litter. Polyandry may exist because it increases the genetic diversity among descendants, which itself augments offspring fitness (Jennions, 2000). It can also help to reduce conflict between males and the increase of post-copulatory sperm competition, which in turn leads to an increase of the females' reproductive success (Gottelli, 2007).

1.7 Behaviour

Cheetahs, unlike other felid species, have a very unique social organization. Females live alone, except when accompanied by cubs. Males either live as singleton or form coalitions. Coalitions can consist of 2 to 4 individuals formed by siblings from the same litter or unrelated males. A clear hierarchy can be observed in the lifelong coalitions, leading by a dominant male (Caro, 1983; Caro, 1986; Eaton, 1969).

Male cheetahs are territorial and around the age of 4 try to occupy an area that they will defend against other males. These confrontations may lead to injury or even death. The boundaries and any relevant points throughout their territory will be marked with urine and faeces. They position their territories around female hotspots, which are small defendable areas rich in resources where females are bound to pass. Territories are very important for males as they can obtain different rewards from it, such as better feeding and reproductive opportunities. However, not all males are part of a coalition, some of them roam alone and are unable to occupy, much less defend territories. Those wander over vast home ranges trying to avoid male territories. All male cheetahs begin their adult live as floaters and as they get older are forced to return to that state by younger males (Hunter, 2003).

Females do not establish a territory and are not territorial. The area they occupy is termed a home range, each home range overlaps with other females' home ranges. When encountering another female they move away, or ignore them. The home ranges vary in size and can be very vast. Females follow a characteristic pattern of travelling across it, typically spending a few days to a few weeks hunting locally in one slice of their range before travelling to another area and doing the same. As mentioned before, female ranges often overlap and this can lead to a very unusual occurring in nature: adoption. From time to time encounters between females result in the cubs from different families getting mixed up. In such situations, the unrelated cubs are usually tolerated by the female (Hunter, 2003).

Cheetahs have some unique calls used for different communication purposes. Table 4 gives an overview of the different sounds cheetahs make and for what reason (Hunter, 2003).

Table 4: Cheetah vocalizations (Hunter, 2003)

Vocalization	Sound and Usage
Yipping	This can be perceived as a high-pitched barking sound. Adults commonly use it to locate one another and to find each other if separated. Additionally, females use it when they are trying to find their cubs.
Chirping	This call sounds like a bird cheeping and is emitted solemnly by cubs, either when lost or in stress situations.
Yelp	The yelp is a variation of yipping, which is used by adults when they are fearful.
Churr	Also named stuttering, emitted during social encounters.
Yowl	This is a drawn-out moan, which is used when a threat has escalated.
Growl, hiss, spit	Cheetahs show these vocalizations when they are annoyed, frightened or in a dangerous situation.

1.8 Longevity

In the European captive population on average 30% of the cubs do not survive the first month. After surviving the first two years, mortality rate decreases and stays low until the age of 11. Not many animals pass 13 years of age and no records are kept with individuals reaching 18 years (Bus, 2015). In the wild, one of the biggest causes of cub decline is due to predation by lions and spotted hyenas (Laurenson, 1992). Female cheetahs surviving to independence have a longer lifespan than males with an average life expectancy of 6.9 years. Probably no cheetah reaches 12 years of age (Caro, 1994; Hunter, 2003).

1.9 Conservation status

There are various international conventions and different action plans put into place in order to assist with the conservation of the cheetah and its habitat. CITES listed the Cheetah on Appendix I in 1975. This means that CITES prohibits international trade in specimens of this species (CITES, n.d.). In 2014 an exception is made for an annual quota of 150 “live specimens and trophies” in Namibia, 50 in Zimbabwe and 5 in Botswana (CITES, 2014). IUCN classified the cheetah as vulnerable since 1986 on the Red List. Two of its subspecies *A. j. venaticus* and *A. j. hecki* are classified as critically endangered. Additionally, the Convention of the Conservation of Migratory Species and Wild Animals (CMS) has listed the Cheetah in Appendix I. This is the highest level of protection for a species and means that they are trying to conserve and restore its habitats (Van de Meer, 2016). All these measures are a result of the drastic population decline, as cheetahs can currently only be found on 10% of their historic range in Africa. In Asia they can only be found in the central deserts of Iran (Durant, 2015). The decline in range comes as a result of habitat loss, fragmentation and degradation, illegal trade and human-wildlife conflict (CCF, n.d.).

Section II: Management in Zoos and Aquariums

This section gives an overview on general husbandry for the cheetah in the zoo environment. It is divided into the following chapters: Enclosure, Feeding, Social structure, Breeding, Behavioural enrichment, Handling and Veterinary considerations.

2 Enclosure

When designing an enclosure, the welfare of the species, their space and social needs, must be taken account of (EAZA, 2014). Therefore, in the European climate, cheetahs need to have access to an outdoor as well as an indoor facility, depending on the geographic location on the European continent. The holding pen designs will firstly depend on the intended purpose for the enclosure; they can either be used for breeding purposes or solely to display the animals. Secondly, it will depend on the area available. If the area is of considerable size, large enclosures can be built and/or more enclosures can be made (if the desire is to hold numerous cheetahs).

Generally any enclosure, no matter the purpose, should include the following three compartments: indoor enclosure, small outside enclosure and the main outside enclosure. In case of multiple exhibits, all should be interconnected by means of sliding doors and runway systems to guarantee easier and less stressful relocations. Lastly, visual contact with other large carnivores should be avoided if possible. The following paragraphs describe specific recommendations for the indoor and outdoor enclosure.

2.1 Indoor enclosure

Every European institution holding cheetahs is expected to provide suitable accommodation for the comfort and well being of their animals throughout the year (EAZA, 2014). Considering European winters, an exhibit with solely outdoor facilities is not sufficient. Therefore, an indoor enclosure should be part of the housing facilities. Even in countries with relatively mild winters we recommend an indoor enclosure for management purposes.

Dimensions:

Every cheetah needs to be placed in an individual pen, which should be no less than 6 m². In case multiple cheetahs are being held, each pen needs to be interconnected to allow maximum flexibility in the daily management.

Boundary:

The boundary between the cheetahs and the keepers should be made out of wire mesh. The same material should be used for the separation of adjoining pens. One might consider creating a solid barrier in between one or multiple cages in case one would want to visually separate animals from each other. It is not advised to keep males and females in the same indoor facility (unless there is an agreement with the EEP) as explained further in chapter 5.

Substrate:

When building new enclosures, considerations regarding the use of biosoil in combination with a drainage system underneath are recommended. The use of biosoil provides natural warmth and less pressure on the joints of the animal. Furthermore, it has ecological and

economical benefits since the use of bedding materials reduces. Experience of keeping carnivores on biosoil is increasing, as one needs to consider the thickness of the biosoil in combination with the degrading of the typical “cat ammonia odor”. Situations where the use of biosoil is absent, the floor of the indoor facility should consist of a hard surface with a protective coating, for example, concrete with a protective coating. The coating will improve the concrete’s endurance and help facilitate the indoors’ daily maintenance work.

Furnishings and maintenance:

Biosoil makes the use of additional bedding materials unnecessary. Visible droppings need to be removed on a daily basis. However, there is no need to clean and scrub with water on a daily basis, except keeping the biosoil a little bit moist. Depending on the situation one might need to replace the top of biosoil every once in a while or renewed completely. In case biosoil is absent, bedding needs to be made possible. Some institutions place wooden platforms indoors, or have wooden frames to insert bedding inside. All furniture should be cleaned on a daily basis and if it becomes tainted with urine it should be replaced. It is advised to include bedding such as straw, hay, wooden shavings/chippings or a combination. The complete indoor facility should be cleaned and scrubbed with water on a daily basis, depending on the animal’s habits.

Environment:

Cheetahs need to have access to water ad libitum. In places where cold temperatures are reached during wintertime, heating should be provided in the form of underfloor heating¹, radiators, heat lamps, forced air, heat radiant panel heaters or hot water pipes. The temperature of the indoor enclosure should range between 10-20 degrees. This will depend on the outside temperature and the situation. During winter, animals should have unlimited access to the indoor enclosure, unless they have a heated area in their outdoor enclosure. Healthy animals may have access to the in- and outdoor enclosure even in extreme weather, whereas sick and old animals should be kept inside. If animals are kept inside more than twelve hours, artificial or natural lighting should be provided to stimulate natural cycles. Draughts should be avoided, therefore some institutions use plastic hanging strips on the doorways to allow the animals to enter and exit the indoor enclosure while minimising draughts and heat loss.

Example:

Figure 4 is an example of how an indoor enclosure for 2 cheetahs could be built. The keeper needs to be able to access the indoor enclosure from the outside, without having direct access to the animals (section 1). In this section there needs to be a hose for the cleaning routine. In addition, the appliances to clean and materials for bedding should be able to be stored there. A drainage system in section 1 is helpful with the cleaning. Each cheetah enclosure should have a door for the keeper to enter. An option is to instal a

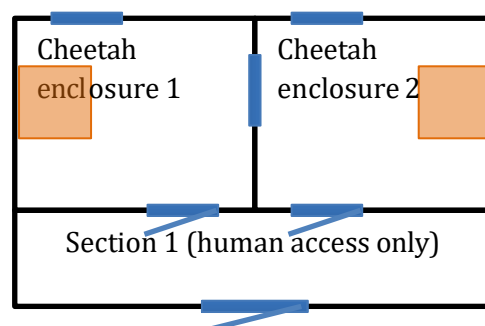


Figure 4: Indoor design

¹ When using underfloor heating it should not cover the whole enclosure. This will give the animals the opportunity to choose where to lie down. Furthermore, attention needs to be paid to the floors temperature in order to avoid unnecessary accidents, such as injury of the footpads due to high floor temperatures.

feeding access slide that is located at shoulder height (person), in order for the keeper to insert food without having direct contact with the animals. Cheetahs need to enter the inside enclosure through a sliding door. The opening and closing of this slide should be controlled from section 1. Adjacent enclosures should also contain a slide to optimize movement of the animals. Each cheetah enclosure needs to have a water dispenser.

2.2 Outside enclosures

There are two kinds of outside enclosures: small and main outside enclosure. They distinguish mainly in function and size rather than building instructions. Therefore, recommendations regarding boundary, substrate, furnishing and maintenance holds for both types of enclosures unless mentioned otherwise.

Small outside enclosure

Having a small outside enclosure provides the possibility to separate an individual from the group, either for medical reasons or agitation between groupmembers. Additionally, it can be used for introduction and training purposes. The size of the small enclosure does not need to be numerous, however not less than 20 m². To prevent cheetahs from jumping on the roof of the indoor enclosure and escape, the enclosure should have a cover that unifies with the one from the indoor enclosure. A barrier, on the roof of the indoor enclosure, described at 'Boundary' and displayed in figure 5 is also sufficient. Lastly, if animals will stay in the smaller outside enclosure without having access to the indoor or larger outside enclosure a water source and in case of extreme weather, a shelter should be provided. For a cheetah to reach the main outside enclosure a slide may be used.

Dimensions:

Solely square meters are not the deciding factor to determine appropriateness of a cheetah enclosure. It depends on the topography, furnishings, group composition, target of exhibit and many other factors. Therefore, it is impossible to recommend a minimum size for the main enclosure. Under 'Furnishing and maintenance' several topics are mentioned which should be included in the main outside enclosure.

Boundary:

Cheetahs can be housed in enclosures with different types of barrier materials. The most common are chain-link-/wire mesh, solid walls and glass windows. Materials used should be strong enough to withstand the impact of a 40 to 65 kg object smashing against it at 90 km/h. Walls and fences should be at least 2,5 m high and have an overhang towards the enclosure at a 45 degree angle (Figure 5). Electric wire can be

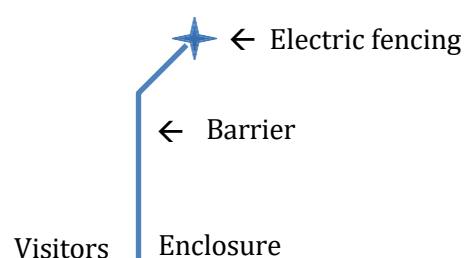


Figure 5: Barrier design

included in the corners and can also be distributed along the top of the barrier. If the mesh used as barrier is covered with plastic, another electric wire has to be present parallel to the hot wire, for grounding. Some cheetahs are known to dig holes by the barriers. Therefore fences should be placed 30-40 cm into the ground. Also cheetahs (especially young cheetah) are known to climb fences. Solid fences are easier to climb for them than chain-link/wire mesh. Bars as a barrier material are not recommended for the enclosures, as it can lead to injury to visitors or adjacent individuals due to aggression.

Substrate:

The substrate should be natural ground: grass, plants and soil.

Furnishing and maintenance:

Cheetahs, both males and females, need high points to scan their environment. In the wild, males spend the majority of their time scanning the environment, either to find prey or females (Caro, n.d.). Therefore, in captivity different raised places such as high rocks, termite mounds, tree stumps, terraces, platforms or any artificial structure that is elevated should be available. In addition, logs and timber stimulate the natural behaviour of scratching for claw wear and maintenance. Hiding areas should also be provided in the enclosure, this could be achieved with tall grasses, shrubs and shelters. Trees should be added to the enclosure as they can provide shade for animals during sunny days and give some cover when it rains. Cheetahs are known to climb trees on occasion. Therefore the trees near exhibit edges need to be made inaccessible for cheetahs to avoid escape. Placing collars on the trees can do this. Attention needs to be given to plants present in the enclosure to avoid the presence of toxic plants. Cheetahs need to have access to water in the main enclosure as well. For this, water dispensers can be provided or shallow ponds, pools or streams. Make sure they are designed for easily cleaning and sanitizing since cheetahs may defecate in the water. If cubs are to be displayed in an enclosure with ponds, pools or streams, they need to be drained, in order to avoid drowning.

The enclosure needs to be in possession of at least one shelter, depending on the number of animals. The shelter will provide shade during hot days and water and cold insulation during bad weather. The shelter should be wind proof, during colder weather they should be provided with bedding and the entrance can have refrigerator flaps in order to reduce the passage of cold temperature. If possible heated platforms and shelters should be provided so the animals have the option to stay outside but still keep warm (see figure 6). After a while, microorganism and parasites may contaminate the dirt substrate from the outside enclosure, this can lead to health problems in the enclosure residents. Therefore, contaminated substrate should be regularly removed and replaced. Faeces should also be extracted every day together with some of the soil around it.



Figure 6: Shelter with floor (and roof) heating (Colchester Zoo)

Environment:

Adult cheetahs can endure cold temperatures, but it is recommended to have shelters available for periods when temperatures reach 0 degrees. Damp conditions on the other hand are very unfavourable for them; here they need the provision of shelters at their disposal so they can maintain a dry coat (Marker, 1998).

2.3 Maternity area

If the intention is to breed cheetahs, the enclosures need to be built in a way that allows easy movement of the animals since you will need to introduce the animals, separate them and rotate them through the enclosures. In addition, the enclosures have to be prepared to house the female with her cubs once they are born. These types of enclosures should be placed off exhibit, preferably isolated from other enclosures, to prevent disturbance of the animals and to make their management easier.

Before the female gives birth, she should be able to make herself acquainted with her new enclosure and the maternity den. It is recommended to design the enclosure with the structure that is previously explained in this chapter: an indoor enclosure, small outside and main outside enclosure. Of these three, only the inside enclosure needs adjustment to facilitate breeding.

Dimensions:

The inside enclosure needs to exist of two compartment, one for the adult cheetah to isolate from her cubs and the other as maternity den. Each compartment should be no less than 6 m². Furthermore, the two compartments need to be interconnected via a sliding door.

Boundary:

The boundary of the maternity den needs to be of a solid material in order to create maximum isolation and minimum disturbance. Video monitoring equipment placed inside the den, a one-way glass or a small window on 1 of the den walls is highly suggested in order to have a better overview of what is happening, without disturbing the female and her cubs.

Substrate:

The groundsurface needs no additional adjustments. Therefore, the use of biosoil with a drainage system underneath is recommended or otherwise a concrete floor with a protective coating.

Furnishings and maintenance:

Next to the solid walls, the presence of a nest box makes an inside enclosure a maternity area. The nest box can consist out of wood or plastic (see figure 7). With plastic surpassing wood on sustainability and maintenance level. A Nest box needs to be big enough for an adult female to turn and lay down with her legs outstretched together with her cubs. It is important for the box to have an aperture in order for the cubs to be able to get out when they start walking. Furthermore, the complete maternity den needs to be well bedded at all times, with materials such as straw, hay or shavings.

Environment:

Maternity dens need to be warm and dry during cold weather and well ventilated during warm weather to prevent disease. Refrigerator flaps can be placed at the opening, in order to reduce the passage of cold temperature. Additionally, a board of around 15 cm of height should be placed in the opening to prevent the cubs from leaving the den too early (Ziegler- Meeks, 2009).



Figure 7: Plastic, foldable nest box (Safari de Peaugres)

Example:

At Safaripark Beekse Bergen the maternity area possesses one den, which is located in the indoor enclosure (see figure 8). The indoor enclosure consists of different divisions. The keeper is able to access the indoor enclosure from the outside without having direct access to the animals. In section 1, there is space to store bedding and cleaning materials together with a hose and a drainage system. Section 1 has an access to the cheetah enclosure and to the maternity den. It is

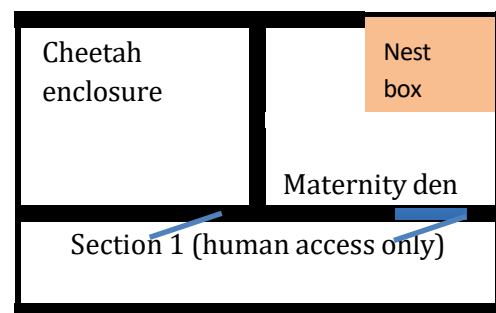


Figure 8: Maternity area

designed with a feeding access slide facing the cheetah enclosure that is located at shoulder height. The walls of the maternity den are solid and one has a small window, where keepers are able to look inside without disturbing its inhabitants (when not in use, the window is covered). There is a sliding door for the cheetah to enter the cheetah enclosure from the outside enclosure and another one letting her access to the maternity den; both slides can be controlled from section 1. On the opening from the cheetah enclosure to the outside enclosure there are refrigerator flaps. The floor of the indoor enclosure is made of concrete with a protective coating and inside of the maternity den a nest box is placed. Bedding is deposited inside of the entire maternity den.

3 Feeding

This chapter gives recommendations on the cheetahs' basic diet and looks into special dietary requirements.

3.1 Basic diet

Free roaming cheetahs consume a variety of whole vertebrate prey, while eating muscle, skin, fur/feathers, viscera and bones in the process. They get a balanced diet in the wild that is difficult to replicate in a captive setting as the prey animals fed to them in captivity have had a different diet than the ones in the wild, and are also likely to be different species (Kaiser et al., 2014). Additionally, in many institutions only muscle meat is offered, preventing the animals from feeding on other body parts (Whitehouse-Tedd et al., 2015).

In a captive setting, the husbandry and nutrition administered to cheetahs needs to be optimized, in order to maintain the welfare of the animals and increase the success of the captive breeding program (Beckman et al., 2013). Therefore, the feeding of whole carcasses and the administration of supplemented muscle meat with carcass parts presents a more balanced dietary model that augments the animal's physical and psychological wellbeing (Bond and Lindburg, 1990; Whitehouse-Tedd et al., 2015).

Digestive system morphology and physiology

The cheetah's digestive tract is composed of a short small intestine, a rudimentary caecum and a small colon. Felids consume highly digestible animal protein and are therefore metabolically adapted to a lower glucose usage, a higher protein metabolism and require a number of pre-formed amino acids, fatty acids, and vitamins in their diet. The following figure (Figure 9) displays the digestive tract of a domestic cat; the cheetah's digestive tract is similar, however the relative size of the small intestine is smaller (Depauw, 2012).

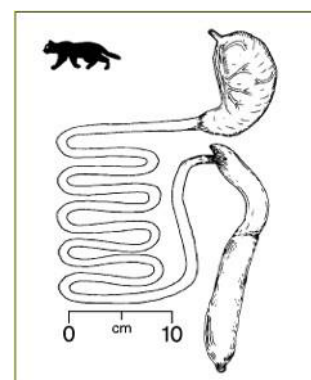


Figure 9: Gastrointestinal tract of domestic cat (Stevens, 1995)

The nutrient requirements of cheetahs are normally extrapolated from domestic cats, as cheetahs' exact nutrient requirements are not known. However the hypothesis that nutritional guidelines

developed for domestic cats are applicable to the nutrition of captive cheetah requires confirmation. Even though there are some similarities detected, such as certain analogous nutritional metabolic pathways, disparities were also found for maternal milk composition, vitamin A transport in the serum, growth and developmental features (Bell, 2010), as well as gut microbiota (Becker et al., 2014).

3.2 Practical feeding recommendations

Cheetahs are strict carnivores that specialize in the wild in antelopes of medium size (between 20-60 kg) and complement their diet with other small mammals or birds (Hunter, 2003). Typical meat sources fed to captive cheetahs include beef, chicken, turkey, rabbit, lamb, goat or horse (Whitehouse-Tedd, 2015). The nutrient composition of meat sources and whole prey has previously been reported in a range of publications and databases (Zootrition®, USDA Food Database, Dierenfeld 2002, AZA 2017 SSP ACM, Whitehouse-Tedd et al., 2017).

Both the feeding of whole prey and supplemented meat have been determined as nutritionally imbalanced if they were to be fed exclusively; therefore the feeding of a single food source is not recommended.

- **Horse and goat** were shown to increase the risk of gastrointestinal disease (Whitehouse-Tedd, 2015). However, empirical evidence is required to determine if a biological association exists between horsemeat and gastrointestinal perturbation.
- **Chicken** was found to reduce the odds of gastrointestinal disease (Whitehouse-Tedd, 2015). However, chicken has very low **copper** and high **zinc** concentrations (a mineral which acts antagonistically to copper) (Kaiser et al., 2014). Regarding copper requirements, ruminant meat is richer (2.56 mg /kg as fed) than whole poultry (up to 1.5 mg /kg as fed) (Beckman et al., 2013).
- The feeding of exclusively **whole rabbit** for 4 weeks led to increased blood levels of vitamin A (Depauw et al., 2011b). Feeding whole rabbit includes the daily consumption of liver, which contains high concentrations of **vitamin A**. In many carnivores excess vitamin A has led to liver and kidney stress, general health and reproductive issues (Bechert et al., 2002). Additionally, high levels of vitamin A intake can prevent metabolism of vitamin E and D, as well as copper (Depauw et al., 2011b; Kaiser et al., 2014), therefore daily consumption of raw liver is not recommended (Depauw et al., 2011b). Moreover, it was documented that the daily feeding of rabbit as whole prey can lead to **increased** blood **cholesterol** (Depauw et al., 2011b). However, the health implications of this on a long-term basis are unclear.

Supplemented meat versus whole carcasses

In Europe, captive cheetahs are fed supplemented raw meat (muscle meat) in 38% of institutions, carcasses in 21%, or a mixture of both in 41% (Whitehouse-Tedd, 2015). Free roaming cheetahs consume a variety of whole vertebrate prey: eating muscle, skin, fur/feathers, viscera and bones in the process. The use of solely supplemented meat in captivity leads to a marked decrease in variation of consumed animal tissue (Figure 10).

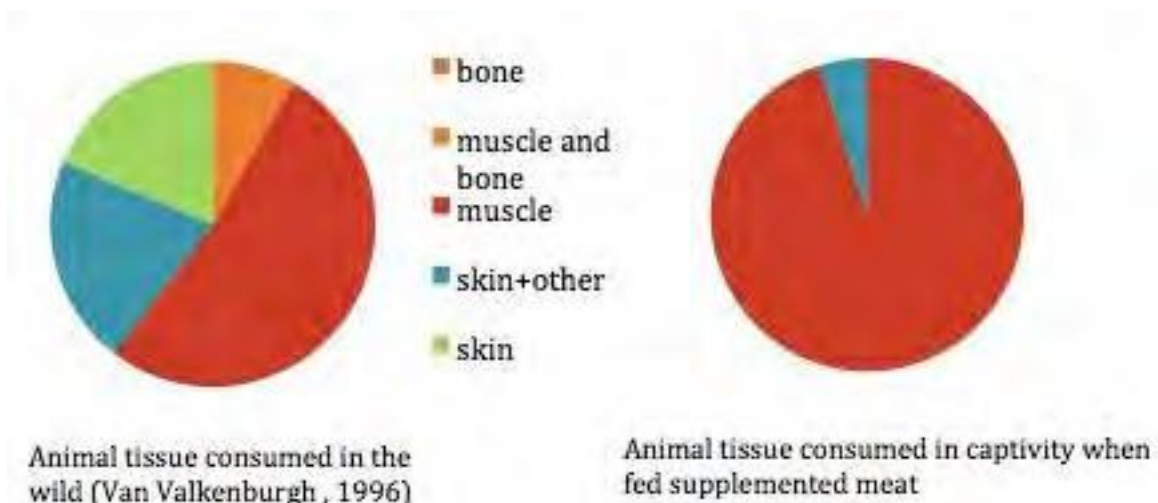


Figure 10: Variation in consumption of animal tissue from a wild diet versus a supplemented meat diet in captivity (Depauw, 2012).

- Feeding whole prey reduces the risk of gastritis and non-specific gastrointestinal disease (Whitehouse-Tedd, 2015).
- Feeding whole rabbit reduces the concentration of faecal markers for gastrointestinal inflammation, loose stool and diarrhoea compared with feeding supplemented beef. This is likely due to the higher concentrations of animal fibre in whole rabbit (indigestible animal tissue such as raw bones, tendons, cartilage skin and hairs), which might play an important role in the digestive system (Depauw et al., 2011a; Depauw et al., 2014). It is advised to regularly check the stool of the cheetahs to evaluate the suitability of the diet and detect possible digestive problems. A faecal scoring system can be found in [Appendix I](#).
- Feeding a mixture of whole prey and correctly supplemented meat reduces the risk of mineral imbalances due to the challenges of appropriately supplementing a meat-only diet (Bechert, 2002).
- The protein to fat ratio (6.6:1) of muscle meat diets is considerably higher than found in whole prey (3:1) (Bechert, 2002). In order to reduce the high ratio of protein to fat, a mixture of both muscle meat and carcasses (correctly supplemented) should be provided (Bechert, 2002).
- The high ratio of protein to fat content in muscle meat in contrast to wild prey has been suggested to contribute to chronic renal disease in cheetahs (Bechert et al., 2002). Yet, to date, high dietary protein levels are not proven to be involved in the aetiology of renal disease in cheetahs, nor in other felids. However, treatment of renal disease includes reducing dietary protein. Therefore, due to the high incidence of renal failure in captive cheetahs and its undefined cause, regular feeding of whole carcasses, having a more natural protein to fat ratio, is advised.
- Fats are important for many functions in the body and for the absorption of several vitamins from the gut. Therefore, it is advised to avoid lean meats, and to retain the fat of muscle meat (i.e. do not trim it off) in order to achieve a better protein to fat ratio (Dr. Tordiffe, A., pers. comm., 2017; Tordiffe et al., 2016).
- Research has demonstrated that healthy wild cheetahs are likely to consume more saturated fat from ruminant prey species and less unsaturated fat compared to captive cheetahs (Tordiffe et al., 2016). The reduced intake of unsaturated fat, combined with differences in fatty acid profiles between ruminant and non-ruminant prey has potential links to health implications for cheetahs (Tordiffe et al., 2016) and therefore preference towards the use of ruminant prey in feeding captive cheetahs is recommended.
- Additionally, the feeding of carcasses stimulates natural behaviour, increases oral health, and psychological wellbeing (Bond and Lindburg, 1990; Haberstroh et al., 1984; Hartstone-Rose et al., 2014).

How much to feed

In captivity, adult cheetahs are fed to maintain their body condition (AZA, 2017). The daily energy intake is reported as ranging between 335 to 875 kJ/kg BW^{0.75}/day (Dierenfeld, 1993; Depauw et al., 2011b; Kerr et al., 2013; Vester et al., 2008; 2010). Currently there is a broad range of recorded daily energy intake in captive cheetahs and more research is needed on metabolisable energy requirements in captive cheetahs. AZA (2017) suggests that the food administered needs to be increased 10-20 % during the winter months and decreased the same amount during summer days as energetic demands differ with changing temperature

and natural elements. Facilities in regions with more moderate climates and/or less seasonal variation in temperature may not need to adjust dietary provision. Feeding quantity should be assessed on an individual, case-by-case basis, according to the animals' body condition, health and reproductive status.

Obesity is rather common in cheetahs kept at zoos. Therefore, control of energy intake, in combination with stimulation of appropriate physical exercise, should be implemented to improve (or maintain) animal health status (Depauw et al., 2011b). To evaluate the adequacy of energy intake in cheetahs it is important to **monitor their body condition score (BCS) on a regular basis** (monthly). Even if it is possible to weigh the animals, it is still important to evaluate the BCS as well. Standardized body condition scores are tools to help assess how much muscle and fat content the animal carries. A standardised body condition score was developed for cheetahs in a captive environment, which can be found in [Appendix II](#).

How to evaluate energy intake with body condition score

- BCS 3 (figure 3) = ideal body weight.
→ Correct amount of food for this animal.
- BCS 2 (figure 2) = underweight.
→ On the condition that illness is excluded, this animal needs a greater amount of food.
→ Increase the daily amount with 15%.
→ Evaluate the BCS after 2 weeks.
→ If BCS = still 2; increase again the amount of food with 15% and re-evaluate BCS after a further 2 weeks. When BCS has improved after 2 weeks, maintain on revised feed quantity but monitor BCS every 2 weeks – food amount may need to be reduced very slightly once ideal BCS is reached and stabilised. Careful monitoring is required.
- BCS 4 (figure 4) = overweight.
→ This animal needs a lower amount of food and increased exercise.
→ Decrease the daily amount by approximately 15% to 20%.
→ Evaluate the BCS after 2 weeks.
→ If BCS = still 4; seek veterinary and nutritionist advice to avoid compromised welfare due to hunger or insufficient nutrient supply.
- BCS 1 and 5 are not acceptable and need veterinary care and nutritionist advice.

3.3 Mineral and vitamin supplementation

Cheetahs in the wild rely not only on muscle meat, but also organs, fat, bones and connective tissue to meet their mineral, vitamin and fatty acids requirements. This results in a balanced diet that is difficult to replicate in a captive setting. Moreover, farmed animals fed to cheetahs in captivity are not only mostly different species than hunted in the wild, but also show a different nutrient composition partly due to a difference in their diet (Kaiser et al., 2014; Dierenfeld 2002; Clum et al., 1996).

Vitamins and minerals are important, especially in young growing animals, where metabolic bone disease (calcium deficiency/Vitamin D3 deficiency), vitamin A deficiency, copper deficiency and thiamine (vit B1) deficiency are common. Most of the minerals like calcium are found in bones and the liver, while vitamins like vit A and D3 occur primarily in the liver of the prey eaten by cheetahs. Muscle meat diets are extremely low in these nutrients and this is why young animals frequently develop problems in captivity (Tordiffe, A., pers. comm., 2017). When animals reach maturity, their requirements for these nutrients is dramatically reduced and thus metabolic bone disease and these other vitamin/mineral deficiencies are rarely noted in adults.

Vitamin and mineral recommendations

- Fresh whole prey contains adequate levels of minerals and vitamins and does not require supplementation. A **regular** (minimum 3 times a week) **inclusion of whole carcasses** of small to medium size mammals (similar to a goat or smaller) is recommended.
 - Vitamins are partially lost/degraded after freezing. When feeding exclusively frozen whole prey, caution must be taken to avoid vitamin deficiencies. Fat rancidity is also a potential problem in stored meat products and care should be taken to minimise the duration of storage and ensure appropriate storage and defrosting conditions are maintained. Supplementation with fatty acids and vitamin E (an important antioxidant) may be necessary.
 - Meat-only diets are very deficient in both minerals and vitamins and require therefore supplementation of minerals and vitamins.
 - A single mineral and vitamin supplement might not be suitable across all the different feed types and misuse can lead to imbalances, excesses and/or deficiencies of specific nutrients. It is therefore recommended to **seek nutritionist advice** to determine the most appropriate supplement for the cheetah's diet.
 - Organ meat is an important source of fat-soluble vitamins and should be included in the diet on a regular basis.
-
- Correct supplementation of **Ca** may be of particular relevance to cheetahs that are not fed whole small prey since they are not capable of consuming bones from species weighing above 10 kg with the exceptions of the ribs and vertebrae of prey species ranging from 30 to 50 kg (Phillips, 1993). Even when fed meat such as horse or beef on the bone, their inability to manipulate and consume large bones often results in an exclusive dependence on Ca supplementation in captivity. However, in practice, the administered supplement is often not weighed or gets partly lost during transport or feeding. Supplemented meat can therefore still result in inadequate Ca:P ratios, which should be between 1:1 to 2:1 (Depauw et al. 2011b). In general, whole prey diets fed to cheetahs contain a more adequate concentration on minerals and vitamins than muscle meat. **This stresses the need for the regular inclusion of small whole prey, having an adequate Ca:P ratio, when feeding captive cheetahs.**
 - An additional mineral that is often neglected in cheetahs is sodium. It occurs at high concentrations in blood and blood rich organs. These carcass components are often removed after slaughter and cheetahs therefore potentially receive a diet that is deficient in sodium. If you are feeding whole or near whole carcasses (including

bones and organs), then you do not need to add any supplements (except for salt). **If carcasses have been exsanguinated, then add 2.5 grams (half a teaspoon) of iodated table salt to every kg of meat fed** (Tordiffe, A., pers. comm., 2017).

- Copper (**Cu**) is required for a number of bodily functions. Also, a lot of different vitamins (for example: A and a number of B vitamins) and minerals (for example: Ca, P, zinc (Zn)) act antagonistically against Cu (Kaiser et al., 2014). Likewise, Cu retention might be compromised in stressful situations, due to an increase in corticoid production (Kaiser et al., 2014). Chicken has unsuitably low Cu concentrations and a high level of Zn (Kaiser et al., 2014). Cheetah cub ataxia and hind limb paralysis and paresis is associated with Cu deficiency, in addition to other vitamins such as vitamin A and some B vitamins (Beckman et al., 2013). Poultry meat has been recorded to contain low Cu concentrations, therefore special attention should be given to **provide supplements containing copper (Cu) when fed poultry regularly** (Kaiser et al., 2014).
- Low fecundity and poor condition are also linked to vitamin A and E imbalances (Beckman et al., 2013). It is very important to include **vitamin A** in the diet, as felids lack the ability to convert sufficient concentrations of beta-carotene into retinol. A deficiency of this vitamin has also been associated with ataxia in cheetahs (Kaiser et al., 2014); this can happen as a result of feeding a lean unsupplemented red meat diet. However, some supplements contain high levels of vitamin A and special attention needs to be given to the amount of vitamin A ingested, as when fed more than 10,000 IU/kg it can become toxic (Kaiser et al., 2014). Also, liver contains high concentrations of vitamin A and might result in vitamin A toxicity when fed (as such, or as part of whole prey) on a daily basis (Depauw et al., 2011b). **Vitamin D:** Vitamin D can be obtained by ingesting the fat, liver and blood of prey (unlike other mammals, sunlight exposure is insufficient for bioconversion to active forms of vitamin D). This vitamin is involved in calcium and phosphorus homeostasis (Dittmer, 2010) but excessive vitamin A can interfere with vitamin D metabolism resulting in skeletal deformations (Depauw, 2011b). Fresh whole-prey diets typically provide adequate **vitamin E** levels. However, frozen and defrosted meats, or meats with high fat content are prone to oxidation and therefore vitamin E degradation may occur, thereby requiring additional supplementation. Cheetahs are able to synthesize **vitamin C**, and its concentration is controlled homeostatically, therefore deficiency or toxicity is rarely observed (Beckman et al., 2013).
- Vitamin A, E and D3 are fat-soluble vitamins that are mainly stored in internal organs and fat tissue. Fresh whole prey typically contains adequate levels of these vitamins in their bodyfat and organs (Beckman et al., 2013). Fresh carcasses, as opposed to frozen, contain a higher amount of vitamin B, E and A, but most of these vitamins are lost during freezing and thawing (Beckman et al., 2013). Vitamins are only present in low quantities in muscle tissue, thus feeding muscle meat alone requires supplementation. There are different vitamin and mineral supplements on the market that can be used. Some examples are: Carmix (Kasper Faunafood), Carnivore Supplement (Mazuri) and Carnizoo (Twilmij B.V.) (Ziegler-Meeks, 2009). However, any fat soluble vitamin, such as vitamin A, E and D3, has the potential to become toxic if provided in excess, so caution has to be taken when using powder supplements to try to balance the nutrient and vitamin composition of muscle meat. **Organ supplementation** is a good alternative to provide fat-soluble vitamins but

should be obtained from a reliable source and must be as **fresh** as possible. There is a risk of salmonella and E.coli contamination of organ meat if obtained from intensive farms (Tordiffe, A., pers. Comm., 2017).

- Dietary and circulating fatty acids (FA) provide a valuable source of energy, they provide structural components of biological membranes, help with hormone production and cellular signalling, and play a role as modulators of gene transcription (Tordiffe et al., 2016). Cheetahs are obligate carnivores and therefore require certain fatty acids such as arachidonic acid in their pre-format states in their diet to ensure health and breeding success (Davidson, 1986). Fats are made up of a number of fatty acids, which are broadly divided into saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs). The serum fatty acids generally reflect dietary intake. Wild cheetahs have high SFA:MUFA and high SFA:PUFA ratios compared to captive cheetahs, indicating that the fats consumed by wild cheetahs are primarily of the SFA variety. This makes sense in that their natural diet consists primarily of small ruminants. The rumen microbes easily convert PUFAs and MUFAs to SFAs and these are incorporated into their fat reserves. Simple stomached animals (rabbits, horses, pigs, chickens, etc.) generally have higher proportions of MUFAs and PUFAs in their bodies (Tordiffe et al., 2016).
- Unsaturated fatty acids are very unstable and deteriorate quickly during storage. Therefore, it is quite possible that a diet high in PUFAs may be problematic for cheetahs. In the wild they consume their prey rapidly after it is killed and thus there is little potential for fatty acid oxidation. In captivity, carcasses are often stored for several days or weeks before use and this may also contribute to increased intake of oxidised fatty acids. This might have health implications in regards to antioxidant status (Tordiffe et al., 2016). Symptoms of essential fatty acid deficiency (EFA) are: skin lesions, hair loss, dry skin, sores, dullness of the eyes, sperm abnormalities and loss of oestrus in females (Davidson, 1986). A higher rate of deterioration of dietary fatty acids is present, in comparison to the deterioration in meat of ruminant prey species (Tordiffe et al., 2016). Therefore, it is **recommended that cheetahs are fed ruminant carcasses (sheep, goats, antelope, beef) rather than meat from simple- stomached animals whenever possible** (Tordiffe et al., 2016).
- During periods of higher physiologic need for fatty acids (e.g. during growth, reproduction and lactation), dietary evaluation should be conducted to determine whether meat requires additional supplementation. The fatty acids can also be increased by avoiding lean meats, by providing whole carcasses and by not trimming the fat off the meat. Plant oil and fish oil mixtures at a ratio of 3:1 and 2ml per kg of feed have previously been recommended (Bechert, 2002), but should be considered in consultation with a zoo nutritionist.
- **Taurine** is an essential amino acid (organic compounds that form proteins and cannot be made by the body) in cats; a deficiency in this amino acid can lead to retinal atrophy, cardiomyopathy and lower reproductive success (Markwell and Earle, 1995). However, there are normally sufficient amounts found in meat and carcass diets. Chicken meat contains a variable taurine content and is therefore not suitable as a dietary staple for cheetahs (Dierenfeld, 1993).

3.4 Feeding schedule and food presentation

In practice, many zoos administer the food once a day, or they distribute the food intake in 2 daily rations. The feed can be administered in the outside or indoor enclosure and depending on the individuals being fed, they may need to be separated during feeding in order to reduce competition for food.

Hunting in wild, cheetahs normally occurs every 3-7 days (Mills, 2004). This is based on observations of actual hunts and kill sites, whereby small prey consumed in their entirety are less easily detected by researchers. As such, the meal frequency may be higher than observed. Moreover, cheetahs will gorge themselves on larger prey items when possible, which is likely to sustain them for longer periods between meals, but this feeding pattern is very seldom replicated in captivity. Changes in food availability can lead to enrichment. Altman et al. (2005) investigated nutritional and behavioural effects of gorge and fast feeding in captive lions. They provided bigger quantities of food that were offered less frequently (3 times a week) and with less predictability (fasting days were selected randomly and there were no restrictions on the pattern). This led to an improvement in the digestibility of fat, protein and dry matter; lions had increased appetitive behaviours and a decrease in body weight to a healthy level (Altman et al., 2005). Free roaming cheetahs do not feed every day, or at fixed intervals. Therefore, such a feeding random gorge-fast routine may also be beneficial for them. One fast day per week has been previously recommended, however there is currently no evidence that the introduction of a fasting day per week has either beneficial or detrimental (or indeed any) influence on cheetah health. It is also important to note that one fast day per week is not the same as a gorge-fast feeding regime as occurs in the wild.

Captive facilities housing cheetahs should carefully consider the use of fasting days – the implementation of a fast day without the provision of a larger than normal meal on the day preceding the fast day may actually be contrary to the best interests of the animals' welfare. As such, if a fasting day is implemented, facilities are advised to conduct behavioural observations of the animals on fasted and fed days, and a comparison made; if the practice is to be continued it must be confirmed that there is no increase in abnormal or stereotypic behaviours on fasting days. If an increase is detected, either removal of the fasting day, or increased dietary provision on the day prior to the fast may be necessary.

When supplemented meat diets are provided it is recommended that meat on the bone is fed so that the animal has to manipulate the meat first – slowing the feeding process down and necessitating the chewing action. This is important for oral health, as well as optimal digestion (Whitehouse-Tedd, K.M., pers. comm., 2017). As explained in chapter 6 "Behavioural enrichment", feeding enrichment increases natural behaviours, incorporates novelty into everyday routine and reduces pacing (Quirke, 2011a; 2011b; 2012). In captive cheetahs, unpredictability of feeding times (temporal feeding variation) and varying the location of food distribution (spatial feeding variation) has been proven to decrease stereotypic behaviour (Quirke, 2011a).

Current recommendations for cheetah feeding regimes include:

- Only implement a fasting day when it has been assured, by continued observation and statistical comparison of behavioural data that there is no increase in abnormal and stereotypic behaviour during fasting days compared with no fasting days.
- Implement unpredictable feeding times and vary the location of where the food is presented at random.
- Implement a feeding enrichment plan.
- Feed meat on the bone, to stimulate chewing, which improves oral health and digestion

3.5 Special dietary requirements

During **pregnancy**, the female's diet in the wild changes to adjust to their physiological demands, such as the need to increase calcium. Therefore, they start only catching smaller prey, like for example hares and newborn gazelles, whose bones they can eat in its entirety (Caro, n.d.). Moreover, as the pregnancy advances their movements becomes more limited and the change in diet decreases the risk of injury and increases their hunting success (the catch rate for newborn gazelles is almost 100% and 90% for hares (Hunter, 2003)).

Lactation is the most energetically costly component of reproduction. During this time energy expenditure can increase as a consequence of milk production, changes in metabolic rate and changes in activity level (foraging behaviour and care for young). Energetic requirements increase as the young grow, but steadily decrease towards the weaning age. In order to compensate for their energy demands, free roaming females whose cubs are in the lair or have recently emerged, eat more frequently with larger daily intakes (Laurenson, 1995a). Cub growth rates drop if daily maternal food intake becomes lower than 1.5 kg/day (Laurenson, 1995b). Therefore, females increase their food intake during this period by switching to larger prey, which is more abundant, in order to reduce travel time. However, they do not discriminate if they encounter small prey (Laurenson, 1995a).

In a captive environment, food intake for females should be increased during late pregnancy and maintained at approximately 2-3 times the normal intake during the lactation phase. Body condition should continue to be monitored to ensure females maintain an acceptable BCS. Vitamin and mineral supplementation should not be necessary if an adequately formulated diet is provided as the baseline, but consultation with a nutritionist is recommended. It is important that the diet contains carcasses on a regular basis.

Hand rearing:

Please refer to [Appendix IV](#) for information on the dietary requirements of hand-reared individuals.

3.6 Water

Water should be available at all times.

4 Social structure

When keeping animals ex-situ they should be able to live in social structures that mirror the life of their conspecifics in the wild (EAZA, 2014). The social organisation of the Cheetah in the wild is described in chapter [“1.7 Behaviour”](#). In this chapter, the basic social structure and introduction methods in captivity are explained, followed by the description of a successful mixed-species exhibit between cheetahs Black and White Rhinos.

4.1 Social structure in captivity

Male cheetahs can be kept alone or in coalitions. By keeping them in coalitions, males are able to express species-specific social behaviours (such as grooming or resting next to each other) that they otherwise would not. Additionally, coalition members seem more “confident”, possibly because they are able to rely on the support of their members in social situations. This confidence may improve reproductive performance (Caro, 1994; Ziegler-Meeks, 2009). Furthermore, it has been documented that several groups of male cheetah can be housed in the same enclosure when given enough space (Chadwick, 2013). Previous studies have declared that new coalitions can be made from unrelated individuals before they reach the age of 2 years. Once adult, encounters between males being introduced for the first time are unpredictable (Caro, 1993). Once a coalition is formed, animals should be kept together for their remaining life as they do in the wild. Separating them will lead to unnecessary stress since it has been proven that there is a psychological attachment between coalition members (Caro, 1994; Ruiz-Miranda, 1998).

Females can be housed alone, with offspring, or in compatible female groups (Ziegler-Meeks, 2009). When the recommendation is to breed with a female cheetah it is advised to house that individual alone. Studies in captive settings have shown that forced social living with other females may lead to ovarian suppression and compromised behaviour (Wielebnowski, 2002). Fortunately, this phenomena is reversible, as soon as female groups are separated ovarian suppression was absent (Wielebnowski, 2002). Whereas male cheetahs form lifelong coalition, this is not the case with female cheetahs.

Sporadically, cheetahs are housed in a mixed-sex group. Although some mixed-sex groups/pairs have reproduced, it is seen as a rarity (Ziegler-Meeks, 2009). Since this is an unnatural social composition, which is not found in the wild, it leads to question whether or not it is healthy for the animals. Especially for female cheetahs, living mainly solitary.

4.2 Changing group structure

Births, deaths and transfers between zoos are the most common reasons a group structure changes. Whereas births and deaths not necessary mean that animals need to be introduced to one another, transferring animals between zoos do. In this paragraph introduction with cheetahs of the same sex are explained. Introduction with cheetahs of the opposite sex is highlighted in [“Chapter 5: Breeding”](#).

Introduction of cheetah to conspecifics:

Introductions regarding either males or females will follow the same guidelines, which will be explained with the following three examples.

1) Introductions at the Binder Park Zoo:

At first the cheetahs, which need to be introduced to each other, are placed in enclosures where they are able to see and smell each other, without having close contact for several weeks. Subsequently, they are given access to adjoining pens for several hours a day. At this point some fighting may happen through the fence. This routine should be repeated until the animals seem to have accepted each other. Only then the cheetahs can be physically introduced, placing both in the same enclosure. The first physical introduction should take no longer than one hour, prolonging this with each attempt. If the introduction is between one male and a coalition, the new male is introduced to each coalition member separately.

2) Introduction at Chester Zoo:

Case study where two coalitions, consisting of two sibling males, were introduced to each other. All individuals were 20 months of age. Firstly, the two groups were placed in adjoining pens for a period of six months. This enabled visual and olfactory access. Secondly, all cheetahs were given access to both pens. Aggressive behaviour was rarely observed, they more or less avoided each other until one of the males was relocated to another institution. Subsequently, the remaining sibling joined up with the other pair and formed a triadic coalition.

3) Introductions at Safaripark Beekse Bergen:

Before the physical introduction, the cheetahs are placed in adjoining enclosures to familiarise to one another through visual and olfactory contact. This can last for a few weeks until the keepers are positive that the animals have accepted each other. Thereafter, the animals are released in the same enclosure. Depending on the interactions between the animals, they are left together or separated and introduced again the next day.

Numerous introductions have been documented. In general, an introduction follows a three-step approach:

- 1) Animals need to have visual and olfactory contact for a few weeks or months before physical access.
- 2) Once the animals seem to have accepted each other and the situation, they are introduced to one another physically.
- 3) Depending on the exhibited behaviour the keepers can decide what to do. Animals behaving extremely aggressive (as some mild aggression is expected) need to be separated. Animals accepting each other can be left together or separated and introduced daily for longer periods of time.

Introduction of cheetah to new environment:

Compared to other species, the cheetah adapts easily to a new situation, taking only a few days to acclimate to their new environment. In case the animal is transported to another institution this can take around 1 to 2 weeks. Before releasing a cheetah into its new enclosure, it is preferable to keep the animal in a smaller holding area for the animal to acclimate to its new environment and daily routine. When the animal recognizes the area as a secure place and feeding area, access to the entire exhibit can be offered. The diet for cheetahs, new to an institution, should be gradually converted from the originating zoo to the new (Kleinman, 1997).

4.3 Mixed exhibit with other species

In order to house different species, together in an exhibit, special attention has to be placed in the design and space of the enclosure. Well-planned and managed exhibits have the potential to supply the animals with environmental enrichment and the public with more interesting and educational enclosures. While planning, the following factors need to be assessed:

- 1) Risks for the animals, for example as a consequence of animals in heat. During this period animals change their behaviour and conflicts can arise between the species. Additionally, newborns may experience harassment by other species and fall prey to them.
- 2) Transmissible viral, bacterial and parasitic diseases between species.
- 3) Nutritional problems, as a result of competition between species for the provided food. In order to avoid this, it is advised to choose species with different dietary requirements.
- 4) Hierarchy problems between species.
- 5) Characteristic differences between both species.

A mixed species exhibit needs to be able to meet both physiological and psychological requirements, ensuring the welfare of all species involved (EAZA, 2014; Dorman, 2010).

Cheetahs have been successfully introduced with Black and White rhinoceros in the past. At Leipzig zoo, they currently house a cheetah female group together with a male and female Black rhino. Prior to the introduction of both species, each was placed alone in the enclosure for a few days. In the exhibit there are places where the cheetahs can retreat if they desire. Except for some mild attacks from the rhinos towards the cheetahs, there are no major conflict or injuries perceived. At Borås Djurpark located in Sweden, cheetahs are kept together with White rhinos. Here similar proceedings were followed. Another example is the combination with pygmy hippo in Bangkok Safari World, but surely multiple other examples have not been listed yet.

5 Breeding

Breeding with cheetahs is not a straightforward task. This chapter discloses several strategies to facilitate breeding and how to handle in case mating is succeeded.

5.1 Mating

Before introducing cheetahs to each other, with the purpose for breeding, there are a couple of general matters improving breeding. Firstly, special care should be taken for adjacent exhibits. For instance, housing hoofstock or other natural prey species have shown to increase reproduction, whereas housing natural competing predators has decreased reproduction success. Secondly, regarding the social structure in which the animals are placed. Zoos that house cheetahs in groupstructures similar to their wild counterparts have higher breeding success, institutions that want to breed cheetahs should try to achieve a similar constellation (Wielebnowski, 2002; Chadwick, 2014). Furthermore, it is advised to separate males and females not only physically but also visually, as they can form a sibling bond making them less likely to breed (Ziegler-Meeks, 2009). Lastly, rotating the animals to new enclosures, the introduction of new animals to each other or any novel situation can stimulate breeding activity, therefore it is recommended to do so.

Introductions between cheetahs of the opposite sex differ from an introduction with the same sex in duration and purpose. When introducing animals, with the purpose to breed, there is an internal motivation of the cheetahs, working in favour of the caretakers. Moreover, the timespan the animals need to be with each other can be as short as one hour, in order to be successful. Therefore, an introduction with the purpose of mating follows a different pathway. As said earlier, breeding with cheetahs is not a straightforward task. There are several introduction methodes and each animal can react differently. A crucial role for successful introductions is the experience of the staff in recognizing the behaviour of the animals (Bus, 2015). This is vital for preventing fights and injuries between two animals and form successful breeding pairs. First priority is recognizing the onset of oestrus. Females have very subtle key behaviours during oestrus that indicate they are ready to mate. They can be seen spending more time on the ground resting and rolling. They also defecate on prominent sites, like termite mounds and large trees. One or two days before the onset of oestrus they are less interested in food. Males that have access to females' faeces and urine during their oestrus cycle (this can be done by placing the male overnight in the outside enclosure of the female and restrict her acces

to the inside enclosure) have been observed to chirp, yelp, stutter, run and display erections. When both parties are put together in adjacent enclosures, the male's reaction normally intensifies and the female normally shows a combination of tail flagging, rolling or rubbing her head against objects. When zookeepers monitor positive behaviour of the animals it is time to physically introduce the animals to each other, there still might be some aggression (as seen in the wild). Usually, copulation occurs



Figure 11: Mating (Safari de Peaugres)

within the first hour and lasts around 30-45 seconds (Figure 11). After copulation the male exhibits a flehmen response and hisses, whereas the female is seen rolling from side to side (Frank, 2005).

There are different opinions and experiences concerning the mating of cheetahs in captivity. One issue is, whether or not a female should be introduced to a single male or a coalition. In the wild females are more likely to encounter a coalition, as they defend territories around female hotspots (Caro, 1993). However, it remains unclear if the female only mates with the dominant male or with several coalitionmembers (Gottelli, 2007). What has been proven is that females are promiscuous and have cubs with multiple paternities (Gottelli, 2007). Meaning that in one oestrus period they copulate with more than one male. Conceivable multiple singletons, several males of one coalition or they mate with multiple males from different coalitions.

Introducing a male and female together can take place via different pathways:

- 1) Introducing the female to one male.
- 2) Introducing the female to a coalition.
- 3) Introducing the female to a different male each day, during oestrus.
- 4) Introducing the female to a male of choice, by placing different males in adjacent enclosures.

There is a strong mate preference between both sexes (Ziegler-Meeks, 2009). Therefore the options that give the animals the opportunity to choose a particular (or more than one) individual may influence acceptance, and as a consequence thereof, reproductive success (Asa, 2011).

5.2 Pregnancy

The gestation period usually lasts 90 to 98 days (Laurenson, 1992). During pregnancy, meals should be increased and fasting days terminated (Ziegler-Meeks, 2009). Small, whole carcasses should be fed in order for them to receive all necessary nutrients. If the female lives in a group she should be separated as soon as pregnancy is suspected or confirmed (Fitch-Snyder, 1988).

There are three alternatives to assess the pregnancy status of a cheetah:

- 1) **Faecal progesterin tests:** This method can only be carried out after 70 days post-breeding. Progesterin levels increase post-breeding above the baseline until around 60 days. However, then a decline sets in, rising again in case the female is pregnant. If she has suffered a false pregnancy progesterin levels will not rise again. Therefore, the test needs to be taken after the 70 days timeperiod.
- 2) **Weekly weighing:** During (false) pregnancy there is a steady weight gain during the first 60 days. After that point, females having a false pregnancy start to lose weight, whereas pregnant females will continue to put on weight.
- 3) **Ultrasound:** This method should only be carried out with females that are trained for it and will not get severely stressed during the procedure (Saffoe, 2005).

5.3 Birth

A few days before giving birth, females usually stop eating (Laurenson, 1993). At first by reducing their food intake 1 to 5 days prior to parturition and stop eating altogether 1 to 2 days before giving birth. Before parturition females may seem restless, they are observed pacing around the enclosure or entering and exiting the den regularly, also smelling and scraping the bedding. Getting closer to labor, females will stay in the maternity den, grooming and an increase of respiration can be seen. Parturition can range from 30 minutes to 14 hours and can arise at any time, day and night. During labor animals are kept inside the maternity den. Disturbance should be kept to an absolute minimum. Therefore, it is recommended to install a camera inside the maternity den, have a one-way glass or a small window on one of the walls in order to check on the animals without being seen or smelled (Ziegler-Meeks, 2009). Complications may occur and if that happens the veterinarian should be notified (Ziegler-Meeks, 2009).

As soon as the females have given birth it is advised to restrain access to the main outside enclosure. The average litter size is 3 to 4 cubs, but litter sizes may range from 1 to 8 cubs. During the following days, depending on the female's behaviour (important to keep in mind that every cheetah is different and there is no golden rule) they can be given access to the small outside enclosure. The slide needs to be closed behind her, to reduce the chance of taking the cubs with her until they are old enough. Females need to be able to rest from their litter each day (Laurenson, 1993). During the night depending on the temperature and the female, she is either locked in the indoor enclosure or left to go in the indoor and the small outside enclosure. Water dispensers need to be present in each compartment of the enclosure.

5.4 Development and Care of Young

Bear in mind that each individual is different; therefore the proceedings during this period will depend on the mother's behaviour. During the lactation period, cheetahs should not have any contact with other conspecifics and all forms of disturbance should be kept to an absolute minimum after birth (Fitch-Snyder, 1988). Extreme caution is required while handling first time mothers, as keepers have no prior experience on the behaviour reaction of the mother. If staffmembers enter the den, mothers may stop nursing properly, try to move the cubs to another place or even abandon them. Therefore, zookeepers should enter the den through a different opening while the female is separated and cannot enter the den. Keepers should not touch the cubs, however, if necessarily for chipping, sexing or veterinary reasons the cubs should be handled with gloves. Females will usually stay in the den during the first 72 hours after giving birth, excluding small periods of time where they eat, drink, defecate or urinate. After this period, the mother will start going out of the den for short amounts of time (Ziegler-Meeks, 2009). From this point onwards, the bedding will need to be checked daily in order to remove food or clean if the female has urinated inside, this should be done with prudence.

The female's interaction with the cubs needs to be closely observed to avoid unawareness of neglectful behaviour. During the first 48 hours it is not necessary to offer the female food, though different institutions offer food the same day or the day thereafter. 48 hours after parturition females should eat, if not, confirm that birth has taken place and that the cubs are in good health. In the event no deviation is perceived, favourite foods may be offered at

different locations (Ziegler-Meeks, 2009). During lactation, provide whole carcasses to compensate the energetic demands during this period.

In [Appendix III](#) a poster can be found with pictures on the development of cubs. Mother and cubs should remain together for at least 1,5 years. There are two approaches of separating mother and her cubs. One is to remove all the cubs at once and place them as a sibling group elsewhere. Female siblings are allowed to stay with their brothers until their first oestrus before separating the siblings. Male siblings should remain together for life, even when transferred to other zoos (Caro, n.d.). Another approach is to remove only the males. The female cubs can stay with their mother for a longer period.

In places with warmer temperatures, some cheetahs have been observed to rear the cubs outside, without the protection of a den. Preferably, this situation should be avoided due to limited management options. Checking the health of mother and her cubs may be difficult or not possible at all. Furthermore, at 3 weeks old, the cubs will start to move with more freedom than in a den. The mother might try to continually pick them up in an effort to keep the cubs together, confining the animals to a smaller space might be necessary (Ziegler-Meeks, 2009).

5.5 Hand rearing

Factors that might trigger a discussion regarding hand rearing are maternal neglect, weight loss, sickness or single birth. Note that, parent rearing is preferred over hand rearing and therefore the latter is discouraged and should only occur when it is strictly necessary for the cub's survival. An alternative to hand rearing may be euthanasia, especially when behavioural problems are expected in the future. Before any decision making, the EEP coordinator should be consulted (EAZA, 2014).



Figure 12: Cross-fostering, the smaller cub in the middle was accepted by the female as her own (Safari de Peaugres)

Another alternative to hand rearing may be to cross-foster cheetah cubs (Figure 12). Especially, when a singleton is born it is recommended to try and find another cheetah with a litter of similar age and size. Successful introductions have been witnessed when impregnating the new cub in the urine and faeces of the females litter (Ziegler-Meeks, 2009).

If, after consulting the EEP coordinator, the decision is to hand rear the animals, see [Appendix IV](#) for the Cheetah hand-rearing protocol. The protocol contains information on maternal milk composition, the different milk formulas and the calculations to be made in order to decide the amount of formula to administer. Furthermore, it contains information about common digestive problems, weaning and housing. It is important to keep the animals as “wild” as possible; therefore cubs should not be cuddled. This behaviour may lead to the lack of specific stimulation needed for the development of species-specific behaviour, leading to social, maternal and sexual disruptions (Kleinman, 1997). As soon as possible the

cubs should be weaned and placed in an enclosure where they can see their conspecifics. At Safaripark Beekse Bergen 2 singletons were reintroduced with their mothers after being weaned and the mothers accepted the cubs immediately.

5.6 Contraception

Offspring in zoos should not be produced if the institution cannot house them under suitable conditions (EAZA, 2014). As cheetahs need to stay with their mother for at least 1½ (male cubs) to 2 years (female cubs), institutions should have the facilities to house the cubs for at least 2 years. The easiest solution to limit undesirable reproduction is holding female cheetahs separate from the males (common practice). Due to possible side effects, other more invasive methods are not recommended. Such methods include: sterilization, castration or implants with hormones for either females or males. For further information on contraception in cheetahs, “egzac” can be contacted (egzac: EAZA Group of Zoo Animal Contraception).

5.7 Population management

Captive populations are small and fragmented. Therefore, they need intensive management in order to retain gene diversity and minimize the effect of random demographic variation (EAZA, 2012). In the European region 2 separate EEP's are managed: the Southern and Northern cheetah EEP. The Southern cheetah EEP is managed by Lars Versteeg, Hilvarenbeek and the Northern cheetah EEP by Sean McKeown, Fota. Both populations are managed completely separate, as the Northern and Southern cheetah are 2 genetically distinct populations. Because of this distinction, careful planning is needed concerning target populations and carrying capacity within EAZA (Versteeg, L., pers. comm., 2017). Next to the European studbook, regional studbooks are managed in AZA, ZAA, JAZA and PAAZAB. In addition to the regional studbooks is the International cheetah studbook, which is a registry of all cheetahs kept in captivity around the globe (Versteeg, L., pers. comm., 2017).

The greatest challenge in population management still lies in achieving demographic sustainability. For this, global cooperation may be needed to aid smaller captive populations, like for example the populations in the ZAA and JAZA regions. Additionally, it is important to create and maintain a genetic sustainable population through the breeding of underrepresented individuals (Versteeg, L., pers. comm., 2017).

6 Behavioural enrichment

Enrichment techniques increase levels of activity and natural behaviour, simultaneously, reduce stereotypical behaviour and therefore enhance the welfare of animals in captivity (Quirke, 2011b). Stereotypical behaviour is defined as a repetitive, unvarying behaviour that serves no obvious function (Mason, 1991).

Quirke (2012) describes four factors that influence stereotypical behaviour in cheetahs:

- 1) Enclosure size
- 2) Ability to view adjacent cheetahs
- 3) Feeding regime predictability
- 4) Group membership

Increasing size of enclosure decreased stereotypical behaviour, whilst being solitary, being fed on a predictable feeding regime and having the ability to view other cheetahs in adjacent enclosures increased levels of stereotypical behaviour. Causes may lie in the fact that solitary cheetahs are unable to perform affiliative and other social behaviours. In extension, when cheetahs are kept in adjacent enclosures without visual and olfactory barriers between them, stereotypical behaviours such as repeatedly pacing around the edges of the enclosures are more present. The animals are able to see each other, but not perform affiliate or aggressive behaviours directed towards the other individual, leading to frustration (Quirke, 2012).

Stereotypical behaviour can be minimized with the help of different enrichment techniques and husbandry practices, examples listed below. The introduction of these enrichment techniques increases natural behaviour, incorporates novelty into everyday routine and reduces pacing (Quirke, 2011a).

- **Temporal feeding variation:** The regular feeding time is modified becoming unpredictable (Quirke, 2011a).
- **Spatial feeding variation:** By varying the feeding location, cheetahs are unable to predict where the food is coming from (Quirke, 2011a).
- **Olfactory enrichment:** Freshly collected animal faeces (from prey species) are placed at random in different locations within the enclosure (Quirke, 2011a). Other novel scents can be introduced in the enclosure such as spices (Skibieli, 2007) or adding liquefied odours onto cloths that are distributed inside of the enclosure (Wells, 2004).
- **Novel objects:** Introduction of novel objects, such as bones and frozen food, promote natural behaviours and help reduce non-wanted behavioural patterns like pacing (Skibieli, 2007).
- **Cheetah run:** This method can give the animals' mental stimulation, a chance to move and carry out hunting behaviour in a captive setting (Figure 13). Different run courses described can be found in the article: *A Comparative Study of the Speeds Attained by Captive Cheetahs During the Enrichment Practice of the "Cheetah Run"* (2013) by Quirke, T., O'Riordan, R. and J., Davensport.



Figure 13: Cheetah run, mechanical pull system for a lure as behavioural enrichment (Ree Park Ebeltoft, Henrik Nordvig)

Note that continuous use of enrichments may lead to habituation. Therefore, it is advised to introduce these methods on a random schedule to prevent habituation. In the situation habituation still occurs, removing the enrichment for sometime can restore the initial response (Murphy, 2003; Quirke, 2011b).

7 Handling

As a large carnivore, managing this species risk assessments and evaluations are necessary when working in close (direct) contact. Some guidance is given in this chapter on possibilities how to handle cheetahs.

7.1 General handling

Cheetahs need to be kept in protected contact; handling should take place through a barrier. Therefore, separation facilities need to be provided in order to ensure that keepers are able to work without having direct contact with the animals. Needless to say, direct contact of visitors with cheetah in any way is not accepted. Each institution is obligated to be in the possession of a risk assessment manual, in order for the keepers to be prepared and be able to handle exceptional situations. While on duty, keepers need to carry a radio and/or mobile phone in case of an emergency. If it is necessary to enter the enclosure, with a non-sedated animal, do so with two experienced keepers carrying protective equipment like sticks, rakes or brooms.

7.2 Individual identification and sexing

There are different identification methods used for the cheetah. The primary identification method is the electromagnetic microchip transponder. Each transponder code is unique and should be reported in ZIMS Species 360 and notified to the EEP coordinator when placed in an animal. The chips need to be implanted between the shoulder blades. The transponders are available in different sizes; all of them are suitable for cheetahs. A second identification method is a documentation of characteristic marks per individual, for daily basis use. Cheetahs can be identified by the unique spots on their faces, tail patterns, size, fur colour and acquired marks such as: scars, wounds, limps and chipped teeth.

Temporary marking can be used in order to identify cubs until they are easily recognizable between each other. Spray paint or site-specific hair clipping is suitable as temporary markings. Cubs can be microchipped and sexed at three weeks old. The recommended method of sexing is by visual means (Kleinman, 1997). Always handle cubs with gloves.

7.3 Catching/Restraining

Anaesthesia should be administered with caution, as narcotics have many side effects that in the long run affect the cheetahs' health. Therefore, it is advised to train the animals in order for them to be crated, shifted and endure medical procedures, without the necessity to anesthetize them (Kleinman, 1997).

Crush cages are very useful for routine blood collections and the administration of medication (see figure 14). They are used to restrain the animals, reducing their range of motion. It is important to habituate the animals to these devices, before using the cages for procedures (Ziegler-Meeks K. 2009). Additionally, in each enclosure, a small holding area should be available for the use of confinement. The benefits of such areas are: the ability to have closer observations, to be able to transfer the animal to a transport container, to

monitor individual food intake, administer specific medication, administer anaesthesia and collect individual urine and faecal samples (Kleinman, 1997).



Figure 14: Crush cage (Prague)

7.4 Training

Training for husbandry purposes is encouraged, as it reduces the animals stress levels that rise during physical or chemical restraining methods. However, the training methods applied should follow the following principles:

- 1) All animals need to have the freedom to participate (or not) in the training.
- 2) Only positive reinforcement techniques should be used during training sessions.
- 3) None of the training goals can be detrimental to the individual's welfare (EAZA, 2014).

Cheetahs can be trained to execute numerous behaviours. For example, holding down position with its tail outside of the holding area, for the veterinarian to take blood samples or measure its rectal temperature. Other behaviours are: opening its mouth and holding it open close to the mesh, stepping on a scale and holding position, putting its paws against the mesh, going into the desired enclosure, entering the crate and allowing the doors to close etc. However, the initial phase of learning and the maintenance of such behaviours is very time consuming. Therefore, each institution should assess the benefits and costs of such a program before decisionmaking.

Crate training:

One of the most valuable trained behaviours and highly recommended to perform, is crate training. During this process, animals are gradually habituated to a shipping crate. The crate can be placed in the enclosure with the doors removed, this way the animal can explore it when desired. In order to increase the time the animal spends in the surroundings of the crate, food can be placed inside. Another approach would be to place the crate in front of a shift door, so the animal has to walk through it on a daily basis. The next training step is

closing the doors of the crate, holding the animal inside. Over time the animal can be kept in there for prolonged periods. The next phase would be to move the crate with the animal inside. When the animal arrives at the new enclosure it should be left inside the crate with one of the doors open, this way the animal can decide when to get out (Kleinman, 1997).

7.5 Transportation

All transfers must be organized in full consultation and agreement of the EEP coordinator (EAZA, 2014). The majority of transports will be done via road, but take care that in the case of air transports there are additional regulations regarding crate use and cargo safety.

Preparations before transportation:

Start crate training several weeks before the transport, this reduces stress levels with the animals and avoids the use of sedation. It is important to communicate with the receiving institution regarding food diets offered in order to coordinate efficiently how the food-transitioning period will take place.

Provision of water and food during transport:

Under normal conditions animals do not need to be fed during the first 24 hours after the start of the transportation but watering should be possible always.

General care and loading:

Administering tranquilizers to the animals is not advised, as they influence the individual physical and psychological well being of the animal, which might lead to unexpected complications. During transport ventilation should be optimal and care should be taken that overheating does not take place.

Container requirements:

The animal needs to be able to individually lie, stand and turn comfortable inside the container. The frame should be made out of solid wood or metal with a minimum width of 12 mm, in case of short distance transfers 8 mm is sufficient. The pieces are either bolted or screwed together. The roof needs to be solid. In the interior plywood or similar materials must line the frame to give it a strong and smooth finish. It is important that the floor is designed in such a way, that neither urine nor faeces spill out. The floor can either be leak-proof and be covered by plenty absorbent material, or have a narrow slatted floor with a liquid proof tray on the bottom. A sliding door on either side of the container needs to be present. The front exit door should be made out of steel welded mesh, or strong iron bars (the space between bars cannot be big enough for the animal to pass its legs through them). The other exit should be made out of wood. Both openings need to be fastened with screws or bolts when the animal is inside, in order to avoid the doors from being accidentally opened. Ventilation openings have to be always present and have to be placed at all heights so that there is enough ventilation inside every inch of the container. Ventilation openings need to have a minimum diameter of 2,5 cm and be covered by mesh. Handles need to be present. Food and water containers must be fixed on either side of the interior without touching the floor, there has to be a safe outside access in order to insert the water or food in a secure way (Live Animals Regulations, 2004).

If the total weight of the container together with the animal, weights more than 60 kg extrusions for the forklift need to be present (for land transfers this is not required) (Live Animals Regulations, 2004).

7.6 Public demonstrations

Specific recommendations have been written regarding the use of cheetah in any kind of demonstration. In the EAZA Felid TAG Demonstration Guidelines, the details regarding the use of any felid in demonstrations are explained.

8 Recommended research

There are still many topics in this species which require more research.

- Nutrition
 - Optimising diet composition
 - Which food items NOT to feed
- Veterinary
 - Vaccination specifications
 - Immobilisation; drugs and dosages
- Breeding
 - Biological details and practical implications
 - Practicalities when wanting to breed cheetah

9 Veterinary: Considerations for health and welfare

Captive cheetahs suffer from different health problems that are not common in free ranging cheetah. Some of them due to old age, others because of differences in food and housing. Cheetahs are susceptible to gastrointestinal diseases, renal diseases, diseases affecting the central nervous system and nutrition related diseases. Additionally, they are also vulnerable to many infectious diseases caused by prions, viruses, bacteria, fungi and parasites.

In [Appendix V](#) the Cheetah Veterinary Guidelines are encompassed. These guidelines aim to give a clear overview on the different (non-)infectious diseases that affect cheetah, with information on the clinical signs, diagnosis methods and treatment measures. This is followed by an overview on medical examinations that need to be executed. Another chapter holds information on the different drugs to administer for the sedation and anaesthesia process. The closing chapter of the veterinary guidelines is dedicated to preventive medicine; here vaccination schedules and endo- and ectoparasite treatments are illustrated.

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Appendix I: Cheetah faecal scoring system

BASIC FECAL SCALE - FELIDS



1.

Hard, dry, multiple pellets that are easy to crumble or break apart into pieces. No fecal residue remains on the ground after collection.



2.

Very firm, with some moisture. Segmentation is apparent and likely occurs as more than one fecal unit. Minimal moisture residue may remain on the ground collected and form is maintained.



3. *considered ideal for most felids*

Moist, surface that is pliable and formed. Moisture on surface appears as shine. Fecal unit or units maintain shape, and only moisture residue remains on the ground after collection.



4.

Very moist, has some texture, and occurs in piles or spots. Loses form when collected and leaves fecal residue on the ground after collection.



5.

Watery liquid, that can be poured and occurs in puddles and flattens and may occur with splatter marks. Has minimal texture and leaves significant residue on the ground after collection.

Considerations:

While a basic fecal scale of 1-5 provides an initial opportunity for documentation of fecal consistency, animal managers also should describe and document the occurrence of blood, mucous, foreign bodies, off color, or off odor in relation to felid fecal excrement. Presence of those characteristics or inconsistencies in individual fecal scores over consecutive days may be indicative of serious gastrointestinal tract disturbance and should be reported to staff veterinarians and nutritionists as soon as possible.

Whole prey, bones and other dietary enrichment items provide valuable stimulation and variety to managed felids; however, animal managers should monitor these dietary enrichment activities carefully. Changes in fecal consistency are likely to occur for up to 48 hours following ingestion of enrichment items including bones and whole prey.

Fecal Consistency Following Bone Ingestion

Fecal excrement following ingestion of bones will likely occur as hard dry pellets (usually scores of 1 or 2) and appear white and may crumble into powder or may include small undigested bone fragments.



Fecal Consistency Following Whole Prey Ingestion

Following whole prey consumption, fecal consistency may vary from normal to dry, or include mucous and undigested fur, hair or bones. Enrichment days should be documented on fecal score sheets in order to account for observed variation among animals.



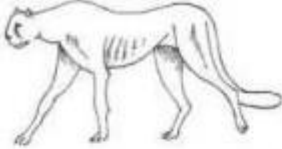
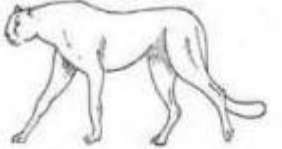
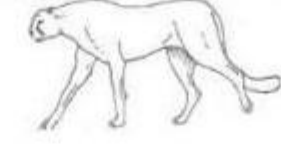
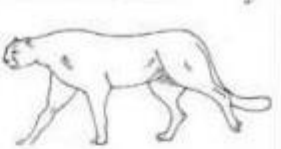
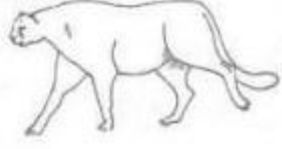
2014 Produced by Felid TAG with funding from Sustainable Swine Resources

**ASSOCIATION
OF ZOOS &
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FELID TAG
CONSERVATION SCIENCE PLUMBING

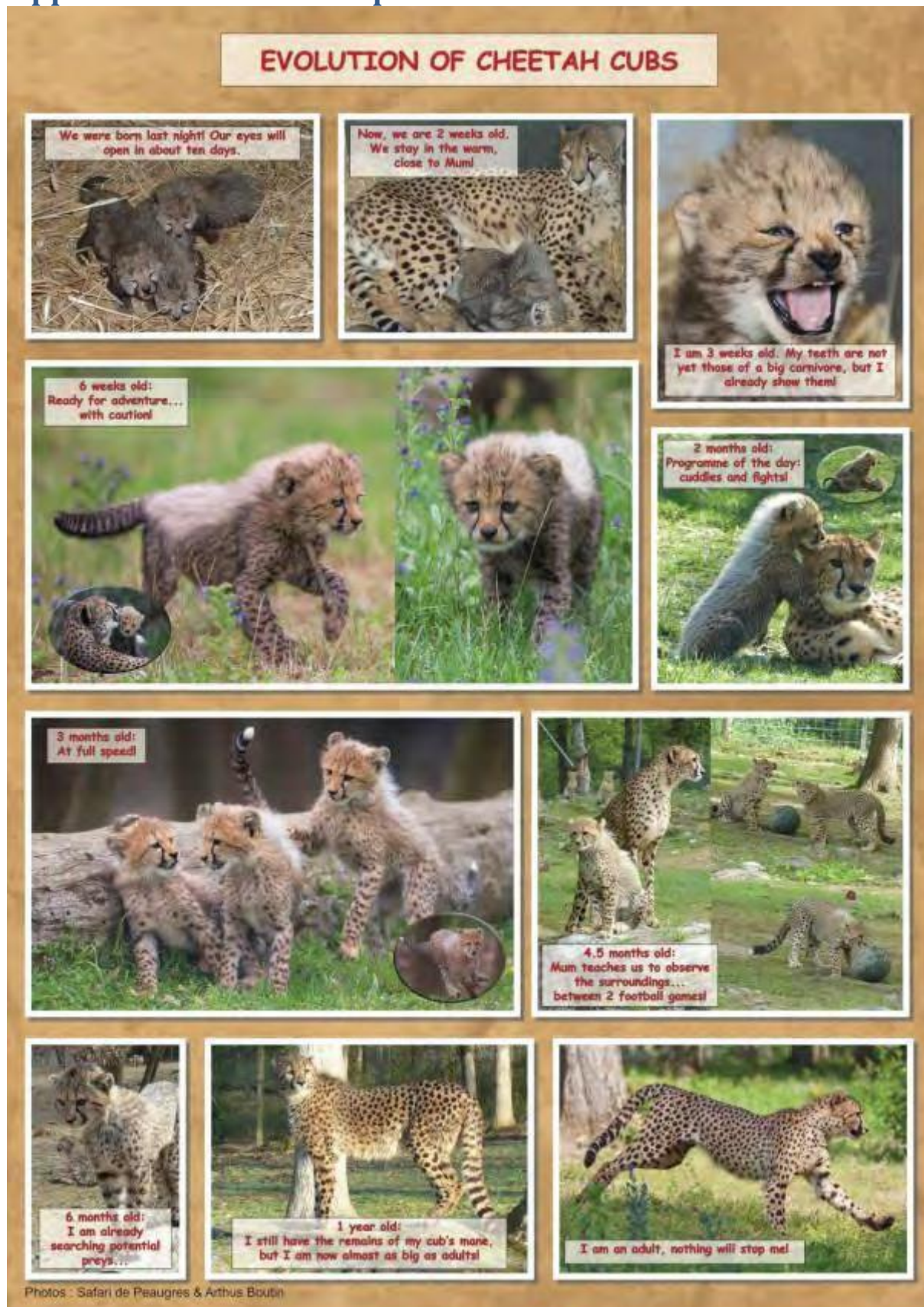
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Appendix II: Cheetah body condition scores

SCORE	1. Very Thin	2. Underweight	3. Ideal	4. Overweight	5. Obese
Outline depictions					
Overall	Loss of muscle mass. Cheek bones prominent; facial features gaunt.	Lean, exaggerated limb delineations, poor muscling. Cheek & face gaunt.	Lean and muscular appearance; obvious delineations between shoulder, stomach and pelvic regions	Stored fat present on inner thigh, pelvic and stomach regions.	Obvious fatty deposits; no definition between shoulder, stomach & pelvic regions
Neck and Shoulders	Bone structure easily visible from a distance	Thin neck	Visible shoulder bones	Shoulders rounded. Neck is thick	Neck is thick and blends into shoulders
Abdominal tuck	Severe	Prominent	Visible without fat pad. Note: pregnant females may have rounded stomach.	None. Note: pregnant females may have rounded stomach.	Large fat pad, no tuck.
Tailhead and pelvis	Very prominent bony structures	Lumbar vertebrae & pelvic bones visible	Bony structure visible but not prominent; thigh muscle obvious while walking. Rear has square appearance.	Fat deposits evident; rear and back have rounded appearance.	Obvious fat deposits over back, pelvis and tail base. Rear & back flat and/or rounded.
Ribs	Obvious	Visible. (Note: Ribs likely not visible if pregnant)	Not visible. (Note: also true if pregnant)	Not visible, fat evident. (Note: also true if pregnant)	Obvious fat deposits

Body condition score in cheetahs; physical features to evaluate are: general anatomy, neck and shoulders, abdomen, ribs, tailhead and pelvis (AZA Cheetah SSP).

Appendix III: Cub development



Cub development (Safari de Peaugres & Boutin, A.)

Appendix IV: Cheetah hand-rearing protocol

WILDLIFE SAFARI CHEETAH HAND-REARING PROTOCOL

Kerrin Grant
2004



CHEETAH HAND-REARING PROTOCOL
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The purpose of this paper is to give pertinent information to caretakers so that informed decisions can be made regarding hand-rearing programs at individual facilities. This paper will address the issues concerning a proper milk formula as well as weaning diets. Additionally, a general hand-rearing protocol is included which may be used as a reference guide for facilities needing this information. Hand-rearing wild neonates is part science and part art. It is necessary to have nutritionally sound diets from the nursing stage through weaning. But it is also important to understand that individual animals have different metabolic rates, food preferences, temperaments and health status, all of which affect the ability to successfully hand-rear an infant. The information in this manual is meant to be a guide and a starting point. It is not considered the "Right" or "Only" method, nor is it meant as a criticism to other protocols. It is just another viewpoint which will hopefully be a valuable asset in cheetah hand-rearing programs.

The maternal milk composition of cheetahs is more concentrated in solids and fat and lower in protein and carbohydrates than the domestic cat (table 1). Kitten milk replacer (KMRTM) has been used quite regularly at cheetah breeding facilities which periodically hand-rear individual cubs or entire litters. KMRTM, in the liquid form, is most commonly used and many times is diluted with water or 5% dextrose for several feedings. Anecdotal reports indicate cubs have digestive problems (diarrhea or constipation) when the straight formula is used. Facilities have indicated the powder form of KMRTM, which can be mixed with water at different dilutions, doesn't mix well and is more prone to cause digestive upset, presumably because the powder stays in a "lump" in the cub's stomach and can't be digested properly. Other facilities have chosen to use EsbilacTM, which is a puppy milk replacer. EsbilacTM is higher in fat and lower in carbohydrates than KMRTM. Taurine, an essential amino acid for felids, is not in the EsbilacTM formula, so must be added prior to feeding (250mg/cub/day) (McManamon and Hedberg, 1993). **A recent survey on hand-rearing protocols of captive felids indicated there was equal preference for EsbilacTM + taurine and KMRTM + Multi-MilkTM formulas (Hedberg, 2002).**

Table 1: Comparison of the maternal milk composition of cheetah and domestic cat. Ben Shaul (1962)¹, Abrams (1950)².

	Cheetah ¹ AF	Domestic cat ² AF	Cheetah ¹ DM	Domestic cat ² DM
Solids %	23.7	17.7		
Protein %	9.41	7.17	39.7	40.5
Fat %	9.48	4.96	40.1	28.0
Carbohydrate %	3.51	4.92	14.8	27.8
Ash %	1.3	0.65	5.4	3.7
Kcal/ml*	1.37	0.93	5.79	5.25

* Kcals were calculated. AF = as fed basis. DM = dry matter basis

Cheetah milk is higher in total solids (less water), fat and ash (mineral component) and lower in carbohydrates than domestic cat milk. In many species that are hand-fed, carbohydrate is the limiting nutrient. Many species are lactose-sensitive and lack the enzyme lactase to break down milk sugars. Because of that, milk formulas manufactured for domestic species must usually be diluted significantly to maintain a formula that doesn't exceed the carbohydrate level wild species are able to digest. Such is the case with KMR^{T*}. The liquid form (canned) provides 18.2% solids, 42.2% protein, 25.0% fat and 26.1% carbohydrates (DM basis). That formula is comparable to the domestic cat's milk, but is very different from the cheetah's. If the carbohydrate component is the limiting factor, the milk must be diluted enough to make the carbohydrate portion approximately 14-15% of the total solids (on DM basis) or 3.5% (on "as fed" basis). Diluting KMR^{T*} liquid to a 2:1 ratio of formula to water gives a carbohydrate content of 3.2%. A 3:1 ratio gives 3.6% carbohydrates, both of which would be acceptable for cheetahs. However, diluting the formula to reduce the carbohydrates also decreases the amount of protein and fat in the diet. See table 2 for the proximate analysis of KMR^{T*} canned formula dilutions.

Table 2: Comparison of nutrient composition of KMR^{T*} canned formula dilutions. Values are on an "as fed" basis. Ben Shaul (1962), Pet Ag^{T*}

	Cheetah req. ¹	KMR ^{T*} ^canned	KMR ^{T*} 3:1 dilution	KMR ^{T*} 2:1 dilution
Solids %	23.7	18.2	13.7	12.1
Protein %	9.4	7.7	5.8	5.1
Fat %	9.5	4.6	3.5	3.0
Carb. %	3.5	4.8	3.6	3.2
Kcal/ml	1.37	0.91	0.69	0.60

From the above data, it is apparent that while reducing the level of carbohydrates to the cheetah requirement, it also decreases the amount of protein to 54-62% of cheetah milk and fat provides only one-third the requirement. Felids obtain energy from protein and fat (Bechert, et al., 2002). The main effect that results from a diluted formula is delayed growth rates and/or skin and haircoat problems. Hair loss was noted in snow leopards that consumed on Esbilac^{T*} formula deficient in protein. The problem resolved after adding chicken baby food, which increased the protein level (Hedberg, 2002). It should also be noted that not only will protein and fat levels be below the requirements for cheetah cubs, but essential amino acids, vitamins and minerals, including taurine, vitamins A and D, calcium and phosphorus, will be diluted as well. The concern here is that chronic nutrient deficiencies in growing cubs may develop into serious health problems such as retinopathy, cardiomyopathy and metabolic bone disease (Howard, et al, 1987; Robbins, 1993). For that reason, it is not advisable to maintain growing cheetah cubs on a diluted milk formula for longer than is absolutely necessary during the formula initiation phase, unless suitable vitamin/mineral and taurine supplements are provided.

Another issue with diluting the milk formula concerns the amount of calories the cub receives in a 24 hour period. Growing cubs require a minimum amount of calories for basic body functions, development and growth. Many hand-rearing protocols suggest feeding a certain percentage of the body weight (e.g. 15-20%) on a daily basis. However, there can be vast differences in the caloric content of formulas, especially when diluted.

For example: say we have three 600g cheetah cubs. One is maternally raised, the other two, hand-raised. Of the hand-raised cubs, one is fed formula #1, as described below. The other cub is fed formula #2. Based on the recommendation that formula be offered in the volume equivalent to 15-20% of the body weight, each cub would receive between 90-120 ml of formula/day. Cheetah milk provides 1.37 kcal/ml. At 15-20% body wt., the cub would receive between 123-164 kcal/day. In this example, we'll use that caloric range as the target for the two hand-rearing formulas.

Cheetah milk**Provides 1.37 kcal/ml of formula****Fed at 15-20% body wt: receives 90-120 ml formula/day****90 ml x 1.37 kcal/ml = 123.3 kcal/day****120 ml x 1.37 kcal/ml = 164.4 kcal/day****Formula II (canned KMR^{T^A}, diluted w/ water at ratio of 3:1)****Provides 0.69 kcal/ml of formula****Fed at 20% body wt: receives 120ml formula/day****120 ml x 0.69 kcal/ml = 82.8 kcal/day****Formula 62 (KMR^{T*} di Multi-Milk^{T*} powders mixed w/ water at ratio 1: 1: 2) -> in table 3****Provides 1.26 kcal/ml of formula****Fed at 17-20% body wt: receives 102-120ml formula/day****102ml x 1.26 kcal/ml = 128.5 kcal/day****120 ml x 1.26 kcal/ml = 151.2 kcal/day**

The caloric content of formula 61 provides 50-67% of the calories in cheetah milk, when offered at 20% body weight. The caloric content of formula 62 falls within the range of cheetah milk, when fed at 17-20% body wt., and provides 1.8 times more calories than formula II, when the same amount (20% of the cub's body wt.) is offered. Formula 62 is more nutrient dense than formula II. In order to provide equivalent calories, formula II would have to be fed at 30-40% body wt. to match formula 62 and cheetah milk. Diarrhea has been reported in exotic felids that consume > 25% body wt/day, so no more than 20% should be offered (Hedberg, 2002). As a result, without some type of supplement, formula 62 will likely result in delayed growth rates compared to cubs raised on a more nutrient dense formula, or maternally raised cubs.

The point of the above example is to demonstrate that formulas are not equal when it comes to determining feeding schedules. Offering 15-20% body wt/day is appropriate for formulas that provide adequate nutrient and energy concentrations, but may not be sufficient in less nutrient dense formulas.

Many facilities have indicated that chicken or turkey baby food should be added to the formula early on in the hand-rearing process. The addition of baby food will provide supplemental protein and fat. Chicken and turkey are reportedly good sources of taurine (Hedberg, 2002; NRC, 1986). One jar (2.5oz.) of Gerber's^{T^A} chicken 2nd foods contains 12.9% solids, 11.8% protein, 4.1% fat, 1.47% carbohydrates, 0.6% calcium, 0.09% phosphorus and 15 IU vitamin A and provides 66 kcal (USDA 2004). Taurine was not listed in the analysis. Gerber's^{T*} 2nd foods, turkey flavored, is also a good source of protein and fat, and very low in carbohydrates. However, the calcium: phosphorus ratio is skewed towards phosphorus (1:6.5) so would not be a good choice unless another source of calcium is provided to give a Ca: P ratio of 2:1. Additionally, there is no vitamin A in the turkey baby food.

Panthers spp. have benefited from the addition of poultry-based human baby food (e.g. Gerber's^{T*} 2nd foods), as early as 1-2 weeks of age (Hedberg, 2002). The baby food provides additional protein and calories, but should be limited to less than 17% (2.5oz baby food to 12.5oz prepared formula) of the diet (Hedberg, 2002). Baby food must be added gradually over one week to prevent digestive upset. This is not considered part of the weaning diet, but as an addition to the formula which increases protein, fat and calories to otherwise dilute formulas. Knox^{T*} gelatin has also been added to formulas to increase the protein content (D. Strasser, pers. com.).

It is not advisable to add meat-based baby foods to nutrient-dense formulas such as those presented in table 3. Laurenson (1995) stated that wild cheetah cubs had physiological limits on growth even when an unlimited food supply was available. However, the addition of protein and calories may promote a faster than optimal growth rate and contribute to potential bone growth abnormalities. Cubs that are consistently growing at >10% body wt/day may need to have their formula diluted to slow their growth. Fast growth promotes bone deformities and fractures because they are not able to support the additional body weight. This phenomenon is common in giant breeds of dogs fed puppy food (Irlbeck, 1996).

Wild cheetah cubs have average growth rates of 37 - 62.4g/day (Laurenson, 1995; Beekman, et al, 1999; Wotck et al, 1991). The recommended average daily weight gain (ADG) goal for hand-reared cheetah cubs is approximately 5% body weight while on milk formula, and 8-10% increase per day after solid foods are introduced (Hedberg, 2002). Formulas and weaning diets that do not meet these goals may need to be modified in one or more ways to ensure proper growth rates of cubs.

Calculations associated with feeding schedules

The following calculations are provided to assist the caretaker in determining how much and how often the formula should be fed to provide adequate nutrition, energy and optimal growth rates.

The Basal Metabolic Rate (BMR) or Basal Energy Requirement (BER) is the amount of energy (kcal) an animal needs for basic metabolic function at rest in a thermoneutral zone. In other words, the amount of calories it needs to stay alive, without having to use energy to maintain normal body temperatures. The formula to determine the PER/BMR is:

$70 \times \text{body wt (nkg)}^{.75}$ (Kleiber, 1947) For a 600g (0.6kg) cub, the BER would be:
 $70 \times 0.6^{.75} = 47.72 \text{ kcal/day}$.

Once we have the BER, we can determine the Maintenance Energy Requirement (MER). This determines the amount of calories the animal needs to function in a normal capacity at its life stage. For adults in a maintenance life stage, the BER is multiplied by 2. For infants that have a higher metabolism and are developing and growing, the BER is multiplied by 3 or 4, depending on the species and other factors. The MER factor of 3 is appropriate for large felids (including cheetahs) that grow at a slower rate than small mammals.

The stomach capacity for most placental mammals is 5-7% of the total body weight (Meehan, 1994). Convert the body weight into grams to find the stomach volume in mls (cc's). To calculate the stomach capacity in ounces, convert body weight into the same units (30g = 1 oz). *The key is to make sure units are the same for body weight and stomach volume). The stomach capacity is the amount of formula a cub can comfortably consume at one feeding. Offering much more than this value may lead to overfilling, which may lead to stomach distension and bloat. It also prevents complete emptying of the stomach before the next feeding and promotes the overgrowth of potentially pathogenic bacteria, diarrhea and enteritis. (Evans, 1987).

The following calculations will determine the total volume and kcal to feed/day, as well as the amount of formula/feeding and the total number of feedings/day.

1. Calculate Maintenance Energy Requirement (MER): $70 \times \text{body wt (kg)}^{.75} \times 3$. See Appendix 1 for calculated MERs for various body weights.
2. Determine stomach capacity (amount that can be fed at each meal): Body weight (in grams or ounces) $\times 0.05$.
3. Divide MER (number of calories required per day) by number of kcal/ml to get the volume of formula to be consumed per day. This value can be converted into ounces, by dividing by 30.
4. Divide ml (or oz) of formula per day by volume to be consumed at each meal (stomach capacity). This gives the number of meals to be offered per day.

Example: 600 gram (0.6 kg) cub

1. $MER = 70 \times 0.6 \text{ kg} \times 3 = 143 \text{ kcal/24 hr. period}$
2. $\text{Stomach capacity} = 600\text{g.} \times 0.05 = 30 \text{ ml/feeding}$
OR: $20\text{oz} \times 0.05 = 1 \text{ oz/feeding}$

****The following calculations are based on a milk formula that provides 1.26 kcal/ml. Formulas that provide more or less energy will result in different volumes of formula per feeding and number of feedings/day. A formula that provided 0.69 kcal/ml would require 207 ml of formula per day given over 7 feedings.**

$$3. \frac{143 \text{ kcal}}{1.26 \text{ kcal/ml}} = 113 \text{ ml of formula to be offered in 24 hr. period (approx. 20\% bw)}$$

$$4. \frac{113 \text{ ml}}{30 \text{ ml/feeding}} = 3.76 \text{ feedings (round up to 4)}$$

The cub in the above example would receive 30 ml (1 oz.) of formula at each feeding and would be fed 4 times over the course of the day. The total amount offered in 24 hrs. is approximately 20% of the cub's body weight. The number of feedings would be split by whatever time period caretakers are able to feed, with a minimum of 3 hours and maximum of 8 hours between feedings.

It is not unusual for infants to feed well at one meal and consume very little at another. Whatever is not consumed at individual meals can be made up by an additional meal later in the day. However, it is important to note that if a cub is expected to consume 30 ml at one meal, but only takes in 15 mls, the deficit can not be made up by offering 45 ml at another feeding. Even if the cub wants to take more than the calculated stomach capacity volume, it must be limited to that amount. Overfeeding may cause bloating and allow for pathogenic bacteria to proliferate in the digestive tract, which will increase the risk of diarrhea, gastric distension and enteritis (Even, 1987). When cubs are hungry, many times they finish their bottle before the feeling of satiety occurs, but are sound asleep 10-20 minutes later. If the cub is still hungry after it has received its designated volume, shorten the time period to the next feeding by an hour, if necessary.

With a very young or weak cub, it would be advisable to feed smaller amounts more frequently, although it is generally not necessary to feed more often than every 3 hours. Frequent feedings that cause the cubs to be repeatedly awakened is actually more stressful than letting cubs sleep for longer periods (Meehan, 1994). Generally, healthy cubs will start to get restless when they get hungry, which can be used to gauge how frequently they need to be fed. In the wild, reports have indicated mother cheetahs may regularly stay away for nine hours between feedings without ill effect to the cubs (Laurenson, 1993).

FORMULAS

Pet AgTM manufactures KMRTM, EsbilacTM and Multi-MilkTM. Multi-milk is a formulated powder with a very low carbohydrate content. Adding it to either KMRTM or EsbilacTM will maintain high levels of protein and fat while keeping the total carbohydrate content to a minimum. Table 3 provides two formulas using Multi-MilkTM. One combines it with KMRTM, the other with EsbilacTM. The nutrient compositions are very close to cheetah maternal milk.

Table 3a: KMRTM-based recipe for a cheetah hand-rearing milk formula

Formula	Component	AF basis	DM basis
KMR (42/25): 1 part	Total solids:	22.4%	
Multi-milk (30/55): 1 part	Protein:	8.9%	39.7%
Water: 2 }— parts	Fat:	9.5%	42.4%
	Carb:	2.5%	11.2%
	Ash:	1.5%	6.7%
	Calcium:		1.4%
	Phosphorus:		1.0%
	Magnesium:		0.08%
	Kcal/ml:	1.26	5.63

AF = as fed, DM = dry matter basis

Table 3b: EsbilacTM-based recipe for a cheetah hand-rearing milk formula

Formula	Component	AF basis	DM basis
Esbilac (33/40): 1 } parts	Total solids:	23.0%	
Multi-Milk(30/55) 1 part	Protein:	7.9%	34.2%
Water 3 parts	Fat	11.2%	48.8%
Taurine: 250mg/cub/day	Carb:	2.6%	11.2%
	Ash:	0.8%	3.5%
	Kcal/ml	1.4	6.0

AF = as fed, DM = dry matter

The above formulas should be diluted for the initial feedings and gradually increased in concentration until given as a straight stock formula. Because the carbohydrate content of the full-strength formula is lower than that of cheetah milk, digestive problems should not be an issue. However, since there is no way to control the ingredients of the milk powders, there is always the potential for problems to occur. One factor that has been reported is lactobezoars (milk clots in the abdomen) of cheetah cubs. The cause of this condition is unknown. One facility indicated they thought the milk formula was too concentrated. However, at the time of the lactobezoar incident, they were feeding KMRTM liquid as their stock formula, which was high in carbohydrates. Bloating and lactobezoars in two hand-reared polar bears was associated with a milk formula high in carbohydrates (Kenny, et al, 1999). The abdominal distension in the cheetahs may have been caused by fermentation of undigested carbohydrates.

The inability to digest certain types of fatty acids might also contribute to lactobezoars. Prior to 1993, Pet AgTM used coconut oil as their fat source in the KMRTM, EsbilacTM and Multi-MilkTM recipes. In 1993, the ingredients were changed and they replaced coconut oil with butterfat. The change was made due to research indicating butterfat was more digestible in domestic dogs and cats. However, wildlife rehabilitators and zoo facilities which hand- raised infants noticed that various species were developing digestive problems, even though the caretakers were using the same recipes as before. Lactobezoars were reported in tigers and leopards (Hedberg, 2002). Coconut oil has a high concentration of medium-chain fatty acids, which are generally more digestible than the long-chain fatty acids present in butterfat (Robbins, 1993). Wild felids may not be able to digest butterfat as easily as coconut oil.

Caretakers also reported that the new formula was difficult to mix and had a greasy residue. Pet AgTM responded to the situation by marketing the Zoologic Milk MatrixTM line of milk formulas. It is essentially the pre-1993 version of their milk formulas, and contains coconut oil instead of butterfat as the fat source. The Milk MatrixTM line uses formula numbers, which refer to the concentration of protein and fat, as the product names.

KMR = Milk Matrix 42/25

Multi-milk = Milk Matrix 30/55

Esbilac = Milk Matrix 33/40

Therefore, the Milk-MatrixTM version of KMRTM, EsbilacTM and Multi-MilkTM may be preferable products to use in cheetah hand-rearing formulas, especially if lactobezoars are a concern.

From personal experience, the Milk MatrixTM line is easy to mix when the powder is added to cold water in equal parts and stirred in a "whisking" fashion. Then the additional water is added to the slurry and mixed completely. There are usually a lot of air bubbles right after mixing, but they dissipate within a few hours. The consistency is much thicker when the formula is cold, and thins out significantly when heated to 100°F. The formula must be refrigerated between feedings.

Many mammalian species lack the enzyme lactase which breaks down milk sugar (lactose) into glucose for absorption into the cells. Gas build-up in the gastrointestinal tract and diarrhea can result as the undigested sugar ferments in the small intestine. Species that have low carbohydrate levels in the maternal milk are generally considered lactose-sensitive or lactose-intolerant. Because commercial milk formulas made for domestic dogs and cats are generally higher in carbohydrates than the maternal milk of the species we're feeding, modifications to the diet are required to prevent digestive distress. Methods used to deal with this issue include:

- 4. Diluting the formula to reduce the amount of carbohydrates from being consumed**
- 5. Including Multi-MilkTM in the recipe to reduce the carbohydrate content**
- 6. Adding lactase enzyme or lactose-eating bacteria (e.g. *Lactobacillus*) to the formula**

LactaidTM is a product that contains the lactase enzyme. The dose is two drops to 100ml of formula. Lactase will begin breaking down the sugar in the formula and will be effective for 24 hours. LactaidTM must be added to the formula 24 hours prior to offering it to the cub.

Simethicone is a de-foaming agent that reduces gas build-up in the intestinal tract, a symptom associated with the inability to break down lactose. But this product does not contain lactose, so it doesn't break down the milk sugar. Trade names for simethicone include Gas-XTM, MyliconTM (pediatric formula) and PhazymeTM.

Lactobacillus spp. is a group of bacteria that produce lactase and digest lactose. It is marketed as "Acidophilus" for humans and ProbioticsTM or Bene-bacTM for animals. These bacteria live naturally in the gastrointestinal tract of mammals, and help maintain a healthy gut. They also help prevent the proliferation of pathogenic bacteria, such as *E.coli*. (Supplement Watch).

The maternal milk of cheetahs is comparatively low in carbohydrates. As an obligate carnivore, cheetahs obtain their energy source from proteins and fat, not carbohydrates (Bechert, et al., 2002). Diarrhea has been reported in *Putorius* spp. raised on milk formulas high in carbohydrates (Hedberg, 2002). It is a fair assumption that cheetahs also do not digest milk sugars efficiently. Therefore, it would be wise to add one or more products to assist in the breakdown of lactose to prevent gastric upsets, particularly in milk formulas that exceed 3.5% carbohydrates (14.8% DM). LactaidTM is commonly used prophylactically and is readily available. The main drawback of LactaidTM is that it must be added to the milk formula 24 hours in advance of feeding and refrigerated in order to be effective. But when used, it appears to be helpful in preventing the signs associated with lactose-intolerance.

Simethicone may be used to treat a cub with a distended "bloated" abdomen, or added to the formula to prevent the occurrence of a "gassy-stomach". The dose for rabbits with gastrointestinal stasis (ileus) is 67-133mg (1-2ml of pediatric formula) once an hour x 2-3 doses (Krempels, et al., 2000).

The addition of *Lactobacillus* spp. in conjunction with simethicone may be an effective alternative to LactaidTM, if it is not available. It is unnecessary to add *Lactobacillus* spp. to the milk formula several hours in advance of feeding. The milk is mainly a vehicle for the bacteria to enter the digestive tract, where it will breakdown lactose into glucose and lactic acid, and the glucose is absorbed into the cells from the small intestine.

ProbiosTM, a product manufactured for livestock, recommends a dose of 50 million bacteria (5 grams) for newborns of all sizes (lambs and calves). This product does contain vegetable oil (as the binding agent) and sucrose (in very small amounts). If given in too high of dose, it has the potential to loosen the stool, but may be helpful with cubs prone to constipation. The product comes in gel form and packaged in a calibrated syringe. The dose is squirted directly into the mouth of the animal.

ProbiosTM is not required every day. The purpose of this product is to ensure there is an adequate population of lactose-consuming bacteria in the gut. They will be self-reproducing, so it is not required that they be completely replenished on a regular basis. A daily dose until the cub is on the full-strength stock milk formula may be advisable, and then every 2-3 days after that until the cub starts consuming solid food. ProbiosTM can be discontinued during the weaning process, but given as needed if loose stool/diarrhea occurs.

Bene-bacTM is a product similar to ProbiosTM. It is manufactured by Pet AgTM for birds and small animals. It comes in powder and gel forms. The powder form does not contain vegetable oil and may be added directly to the milk formula during preparation. Like ProbiosTM, it also contains 10 million bacteria/g. Pet Ag'sTM recommendation is to give it every two days from birth until seven days (or first week of hand-rearing) and then once a week until the introduction of solid foods. The dosing of *Lactobacillus* spp. bacteria is more about infusing an adequate number of bacteria into the gut rather than being weight-related, so the product recommendations should be considered reasonable guidelines more is not necessarily better.

Acidophilus comes in tablet form and may be crushed and added to the milk formula. The dose for humans (adults and children) is one tablet (1 billion bacteria). From personal experience, I have given small mammals (rodents and rabbits) *acidophilus* at the rate of 1 - 1 tablet in a batch of formula which lasts 2-3 days and have had no ill effects from that dose. As a general guideline, one-half tablet/cub/day may be adequate. *Acidophilus* works the same way as ProbiosTM and Bene-bacTM. It is just another form, and does not contain vegetable oil or sucrose.

Growth curve in hand-reared cubs

Hand-reared animals typically have a delayed growth rate compared to maternally-raised cubs. There are many factors which contribute to that.

1. Cubs receive maternal antibodies in utero (before birth), in the colostrum and in the milk. Mother-raised cubs receive considerably more passive immunity to a variety of pathogens than the hand-reared cubs.
2. Many times, hand-reared cubs are pulled because they are poor-doers and are nutritionally and/or immunologically compromised from the start, and simply don't have the ability to make up for lost time.
3. The hand-rearing formula, no matter how nutritionally sound it appears, is restricted to the nutrients in the powder mixes. As we learn more about nutritional idiosyncrasies of each species, we find that many times the form of protein, fat or carbohydrate in the artificial formula is not compatible with those in the maternal milk, and may not be as digestible. All we can do is our best with what we know at any given time. Over the years, milk formulas have improved vastly, and will no doubt continue to improve in the future.

4. **Formulas given are not nutritionally balanced or are deficient in one or more major nutrients such as protein and fat. An average weight gain of approximately 5% body weight while on milk formula and 8-10% weight gains during the weaning process are the targets (Hedberg, 2002). There will always be some fluctuation where there may be a 2% gain one day and 8% the next. So the key is to see what the average is over a period of 3-5 days. If the cub is consistently maintaining weight for several days or only has slight gains, the formula composition and feeding schedule should be evaluated. barring any health problems to explain a delayed growth rate in an individual, low weight gains are generally related to a diet that is not meeting the caloric and/or protein requirements.**
5. **During the initiation phase of the milk formula, diluted formulas are given over the course of a few days. Weight gains generally do not occur during this period. Cubs will either maintain or lose a small amount of weight until they are consuming the formula at 80-100% full strength. This is not of concern in otherwise healthy cubs. As soon as they transition to the stock formula, they will begin gaining weight. Additional formula is not required to provide the target amount of kcal, unless cubs are regularly restless in between feedings. If this occurs, an additional feeding can be supplied, which is eliminated as the cubs consume the more concentrated formula.**
6. **Cubs that are weak may not have the energy to consume the target volume of formula at each feeding. In these cases, small, frequent feedings and the addition of LRS+ 2.5% dextrose given subcutaneously (SQ) may be more appropriate. Weak cubs may also take longer to transition onto the stock formula because of weakened organ function. Close monitoring of these cubs is warranted to ensure they begin gaining weight as soon as feasible, without stressing their immune system any more than necessary. Even in these cases, the cubs should ideally be on a formula at 80-90% full strength concentration within five days and possibly another two days to get to the full-strength stock formula. If diarrhea occurs when these cubs go onto the full-strength formula, they may do better on a 2:1 or 3:1 dilution (full-strength formula: water) as their stock formula.**

GENERAL HAND-REARING PROTOCOL

Key stages of cub development/hand-rearing process (Meehan, 1994)

7 days = eye open	28 days = introduce solid foods
7-14 days = teeth begin erupting	35 days = cease nursing from bottle, wean to bowl
14 days = walking unsteadily	70-80 days = completely weaned

NEWBORNS

Colostrum

Domestic kittens receive maternal antibodies in *utero*, and from the colostrum and milk (Casal, et al, 1996; Robbins, 1993). Colostrum contains 4000-8000 mg/dL IgG, and can be absorbed in the feline gut until 16 hrs. post-partum (Levy and Crawford, 2000; Casal, et al, 1996). IgG is the first line of antibody defense after birth to prevent and combat systemic infections in newborns. IgA provides more localized defense in the gastrointestinal, reproductive and respiratory systems and helps prevent enteritis in the newborn (Zuba, 1991). IgA can usually be absorbed for a longer period than IgG, although there is no data on the time period in felids (Miller-Edge, 1994).

Ideally the cubs should stay with the mother until at least 24 hrs. of age so they can receive colostrum and some maternal milk. Without access to maternal antibodies, the cubs will be immuno-suppressed and more susceptible to contracting various infections until they are vaccinated and can develop their own active immunity to pathogens. Additionally, weak cubs are prone to bronchopneumonia during the first few weeks of life (Hedberg, 2002). If cubs must be removed from the mother within the first day, adult cheetah (or other appropriate felid) serum may be administered to cubs at the rate of 150 ml/kg body wt. during the first 24 hours (Levy and Crawford, 2000). The authors suggest giving 75 ml/kg intraperitoneally (IP) or subcutaneously (SQ) in two separate doses within 16-24 hrs. postpartum.

Umbilical stump

Newborn cubs that are removed from the mother shortly after birth will need the umbilical stump cleaned to prevent infection. This area is the prime site of infection entry for newborns. Typically, povidone-iodine is used as the disinfectant. Chlorhexidine 0.5% solution (NolvosanTM 2% diluted with 3 parts sterile water) is a preferable choice (Hedberg, 2002). Iodine (7%) is caustic and can trap bacteria in the stump. Chlorhex is not caustic and has greater residual antimicrobial activity than iodine (Madigan, 1997). Hedberg (2002) recommends that caretakers clean the umbilical stump every 6 hours x four treatments and monitor for heat and/or swelling at the site.

FORMULA FEEDING

Initiation Phase

1. At intake, weigh cub (in grams or ounces) and calculate stomach capacity (5-7% of body weight).
2. Take rectal temperature. Normal body temp. for felids are: (Hedberg, 2002)
 0-7 days = 95-96.8°F.
 7-28 days = 97-99°F.
 >28 days = 101-102.4°F.
3. Cubs with a normal body temperature may be offered an oral electrolyte solution, at 5-7% body wt. (stomach capacity). Electrolyte choices include: unflavored PedialyteTM, Lactated Ringer's Solution (LRS), LRS + 2.5% dextrose and Normosol-R. Electrolytes should be warmed to 95-100°F. to prevent lowering the body temperature from ingesting cold fluids.
4. Cubs with body temperatures below 94°F. should be warmed in a temperature-controlled incubator or placed in a box or other comparable container with a heating pad set on LOW underneath one-half of the container. Do not place cubs directly on a heating pad or have the heating pad inside the box. Allow the cub to warm up to the normal body temperature for its age before offering anything by mouth. SQ fluids (LRS + 2.5% dextrose) may be given to slightly dehydrated cubs. Maintenance fluid level is 50-75ml/kg/day (Hedberg, 2002). The level of dehydration is added to this value. Divide the fluid volume for a 24 hr. period into 2-3 doses, with half given at the first dose and the remainder split equally between subsequent doses. Intravenous (IV) fluids (LRS with either 2.5% or 5% dextrose) may be warranted in weak or hypothermic cubs (body temp < 90°F.). Hypothermic cubs, especially those that are non-responsive, may also be hypoglycemic. It is advisable to take a small blood sample to determine blood glucose level at this time, and treat accordingly. The normal blood glucose level for most placental mammals is 90-130 mg/dL. (Fraser, 1991). Clinical signs of hypoglycemia, which include hypothermia, incoordination, cold and clammy skin, constant crying, and a poor or absent suckling response; may not be apparent until the blood glucose level drops below 50 mg/dL (Fraser, 1991).
5. The first bottle feeding of oral electrolyte solution will dehydrate the cub, strengthen the suckling response and clear the stomach of the mother's milk (Hedberg, 2002). Offer an additional bottle of oral electrolytes (5% body wt) two hours later.
6. The third feeding should be the stock milk formula diluted with WATER, not electrolyte solution, at the dilution rate of 1:4 (formula: water). Electrolytes have the potential to interfere with milk absorption with the formula (Fettman, et al, 1986; Heath, et al, 1989). If the cub needs additional oral electrolytes, they can be given in between formula feedings. Regular tap water that does not contain excess minerals, or is otherwise contaminated, may be used to mix with formulas. Distilled water may be used as an alternative.

7. At the 4th feeding, the cubs should be put onto their regular feeding schedule. At this time, the caretaker should have calculated the number of kcal the cub requires over a 24 hr. period, the volume of fluids to offer at each feeding and number of feedings per day (see page 5 for calculations). The calculations should be based on the information from the stock formula, not the dilutions. Over the course of the first 72 hrs. the cub will receive fewer calories than required, but that is part of the transition process. Offering too much too soon increases the likelihood of digestive upset. The cub will be consuming the required nutrient composition and caloric content soon enough. The problem occurs when the time frame required to transition to the full-strength stock formula is extended beyond 3-5 days. If that occurs, for whatever reason, additional feedings or vitamin/mineral supplements may be warranted.
8. In an ideal situation, the cub will receive a 1:4 dilution formula for 2-3 feedings, then 1:3 dilution for 24 hours, then the 1:2 dilution for 24 hrs, 1:1 dilution for 24 hrs, then the full-strength stock formula on the 5th day and from then on. However, in the real world, things don't always work out as planned. Cubs may periodically need to stay on a dilution a little longer, particularly when going from the 1:1 dilution to the full-strength formula. Intermediate steps may need to be added, such as going from 1:1 to 2:1, then full-strength to give the cub more time to adjust. Occasionally cubs need to take a step back if diarrhea occurs. For example, if the cub does well on 1:2 then develops loose stool on the 1:1 dilution, which gets worse at each feeding, delete the next feeding, give electrolytes (at 5-7% body wt) for 1-2 feedings, then go back to the 1:2 dilution step. Offer that formula for 2-3 feedings and progress to 1:1.5 if the stool improves. Healthy infants tend to resolve digestive upset/loose stool pretty quickly when dealt with appropriately. Compromised infants may have other issues that are compounding the problem. They may be stressed and immuno-suppressed. They may have bacterial or viral infections, particularly if they didn't receive colostrum before being removed from the mother. They may have parasites. Or there may be other factors that are adding to the cub's stress level which hampers its ability to adapt and adjust to the hand-rearing process. This is where the "art" of hand-rearing comes in, and the caretaker must make various adjustments to help an individual cub do its best.

Urogenital Stimulation

Animals that are not fully developed and mobile at birth (altricial) require manual stimulation to urinate and defecate until their eyes are fully opened. A soft towel/wash rag should be moistened with warm water and massaged along the urogenital region either immediately before or after each meal. Some caretakers prefer to do it before feeding because it awakens the cub and encourages them to nurse. Others recommend stimulating after feeding because cubs tend to get fussy during the process and may not be able to settle down enough to take the bottle, if done beforehand. It's all a matter of choice on the part of the caretaker and the temperament of the cub. The important point is consistency. Cubs will likely urinate during each stimulation, but not always defecate. This is normal, particularly if they receive several feedings throughout the day. Normal defecation should occur at least 2-3 times a day.

Bottle Feeding Guidelines

Nipple selection is important to allow milk flow at the proper rate. Nipples that have a very slow flow rate will cause the cub to suck harder and harder, and eventually suck air into the stomach, which will lead to a distended, "bloated" stomach. Nipples with holes that are too large will cause too much milk to enter the mouth, and potentially be aspirated into the lungs, leading to aspiration pneumonia. Venting the bottle by loosening or tightening the cap rings will adjust the flow rate (Hedberg, 2002).

Felids feed eternally (lying on abdomen) with the neck extended and head tilted up slightly. It is important that the bottle be tilted up enough so there is always milk filling the cap to ensure the cub doesn't suck in air while nursing.

Many times, individual cubs will require their own nipple(s) as each cub suckles with different intensities and styles. *Panthera spp.* have responded well to the Mead-Johnson standard nipple as well as the "NUK" shaped orthodontic preemie-sized nipple, which are shorter and softer than the standard human infant nipple (Hedberg, 2002). The EvenfloTM preemie nipple has been successful with very young cubs because the flow rate can be easily adjusted. However, it does collapse with strong, aggressive suckling, so would not be advisable with older cubs or particularly aggressive nursers. Smaller felids (under 500g.) may be able to use the Pet NurserTM bottle by Pet-AgTM, which come in 2oz. and 4 oz. sizes, or the elongated conical-shaped nipples from Four PawsTM (Hedberg, 2002).

Tips for feeding from the bottle (Meehan, 1994; Hedberg, 2002; Felid TAG):

1. Providing slight tension on the bottle while feeding may encourage suckling, particularly in the more difficult nursers.
2. Squirting a little bit of milk directly into the cub's mouth to start the suckling process.
3. Allow cub to push and "knead" a towel with its front paws as it suckles. This is how they stimulate milk flow when nursing from the mother. This behavior might also stimulate motor function in limbs.
4. If suckling action is weak, place the thumb and index finger on either side of the mouth to create better suction.
5. Make sure the milk formula stays warm (100°F). The digestibility of milk lowers as it cools, and some cubs will stop drinking if the formula temperature drops below a certain point. This is more of an issue with slow nursers. Having a second bottle in a warmer or cup of hot water for these cubs may be helpful. Always check the temperature of bottles sitting in hot water before feeding by shaking it a couple of times, then squirting a few drops on the back of your wrist ---- it should feel slightly warm, not hot. "" Milk formula must never be fed cold ---- it will lead to digestive problems.
6. Some cubs will nurse better if the nipple is placed in the mouth before the cub is fully awake.
7. Mimicking the maternal behavior, such as scruffing the cub and swinging it gently, as it is lifted out of the incubator may stimulate the instinct of the cub to start nursing after being carried by the mother.
8. Weak and/or very young neonates will often suckle until tired, not full. Give these infants time to rest in between more frequent feedings, if necessary.

Digestive Problems

In most cases, loose stool and diarrhea are nutritionally related, either by providing a diet that is nutritionally imbalanced or by transitioning from a diluted to a concentrated formula too quickly. Stress is a factor that may come into play at certain stages, such as having new caretakers feed or when cubs begin eating solids.

Nutritionally-related digestive upset most commonly occurs at three different stages:

1. When the first 48 hours of the initiation phase is not followed. The initiation phase consists of **offering 2-3 feedings of an oral electrolyte solution then giving diluted stock milk formulas that approximate 1:4, then 1:3, 1:2, and 1:1 dilutions (of stock formula: water) prior to giving the full-strength milk formula. Infants that are started immediately on a milk formula are more apt to have digestive problems than those that receive the electrolytes first.**
2. When the cub transitions from the 1:1 to the straight formula. Sometimes the full-strength formula, even though it is nutritionally sound, may be too rich for the cub. This is more of an issue with individual cubs that may have sensitivities to nutrients in the formula or have a temperament that is stressed more easily, or other unknown factors. There may be times when it is more appropriate to feed 2:1 or 3:1 (mixed formula: water) dilutions rather than the full-strength formula in order to maintain normal stool. Cubs in this situation may also benefit from the addition of chicken baby food to a diluted milk formula (1:1 or 1:1.5). It should be noted that cubs which are maintained on a slightly diluted milk formula long term may have a delayed growth rate.

- 3. From time to time infants may have slightly loose stool as they transition from one dilution to another.** Many times this situation clears up on its own within 1-2 feedings. Depending on the individual cubs (health status, temperament, etc.) the caretaker may want to continue offering the formula at that dilution rate for 2-3 feedings and monitor the stool color and consistency. If the color is normal and the consistency is slightly loose, but maintains that consistency or improves, the formula transitions may continue as scheduled. However, if the consistency worsens and begins looking like diarrhea, or if the color becomes pale, then it is appropriate to delete the next 2-3 feedings and offer oral electrolytes instead. The first formula feeding after that should be the dilution level just prior to the onset of diarrhea. Generally, 2-3 more feedings at the previous dilution is enough to improve the condition of the stool and it may then transition to the next step. In the past, KaopectateTM has been used to successfully treat diarrhea in young animals, including kittens. However, the ingredients of this product were recently changed and it now contains salicylates (like Pepto-BismolTM). Salicylates may cause adverse reactions in felids (Plumb, 1991), so this product should not be administered to cheetah cubs.

Infants that are receiving dilutions of a stock formula low in total solids (compared to maternal cheetah milk) may experience bouts of diarrhea, not because the formula is too rich, but because it is too low in total solids (analogous to a human living on a liquid diet). When there is an excessive amount of fluids going in, excess fluid will be excreted with the waste products and appear to be very watery. Also, diets that contain higher levels of carbohydrates than the maternal milk may also cause diarrhea.

Common causes of diarrhea: (Marcum 1997; Hedberg, 2002)

1. Introducing concentrated formula too quickly without transitional stages
2. Offering formula with a carbohydrate (milk sugar) content that exceeds that found in the maternal milk composition
3. Providing a diluted formula too long (lack of adequate total solids in diet)
4. Overfeeding (> 7% body wt/feeding and/or »20% body wt/day)
5. Sudden change in diet or addition of new food (during weaning process).
6. Nutritionally imbalanced diet
7. Unclean feeding utensils
8. Stressful conditions

Stool consistency:

Loose stool: is not well-formed and is excreted in a pile. Milk stool is naturally softer and less well-formed than stool excreted when on solid food diets because of the high water content of the formula. Loose milk stool is of pudding consistency. Diarrhea is watery stool and can lead to dehydration.

Stool color: (Marcum, 1997)

1. **Normal milk stool: tannish-yellow or light brown. First stool of the newborn is dark, greenish-brown (meconium) and indicates the cub has not nursed yet. During the hand-rearing process, after 1-2 doses of electrolytes, many times the first stool to appear will be dark brown and more solid than typical milk stool. This is because the cub hasn't eaten for an extended period and the stomach is empty. The color and consistency will become softer and lighter as the cub starts consuming the formula.**
2. **Black, tarry stool: may indicate intestinal bleeding. Cubs that are fed fresh, raw meat during the weaning stage may have black stool on occasion, which is not necessarily a concern. It may be from consuming meat with blood on it that has passed through the digestive tract.**
3. **Green stool: may be caused by enteritis (inflammation of the intestine) or by a particular food.**
4. **Red stool: may be caused by hemorrhaging or presence of blood in colon. Small amounts of blood may be seen during the weaning stage if cubs are eating bones, which can scrape the intestinal lining.**
5. **White stool: may indicate bile disorder or enteritis**

Constipation

Cubs that fail to defecate within a 24-hr period may be constipated. Massage the abdomen with a warm, dry cloth. Sometimes inserting a lubricated thermometer into the rectum will loosen stool and stimulate it to be excreted. If the cub is constipated, or if lactobezoars (milk clots in abdomen) occur, delete the next 1-2 formula feedings and replace them with oral electrolytes (5-7% body wt.) If condition doesn't resolve after the second electrolyte feeding followed by manual stimulation, consult a veterinarian for further treatment.

HOUSING

Heat Sources

Incubators provide an environment where the temperature and humidity may be controlled for severely compromised neonates. An incubator may not be a reasonable option for a litter of healthy, active cubs, but is preferable to other heat sources for very young and/or weak cubs. It is important to provide 50% humidity for cubs in incubators because the process of heating air reduces its relative humidity and can lead to dehydration (Meehan, 1994). It is also important that the temperature in the incubator not become too warm. Monson (1987) gave the following recommendations for ambient temperatures with hand-reared kittens, which are reasonable guidelines for cheetah cubs.

- 0-7 days of age = 88-92°F.
- 8-14 days of age = 80-85°F.
- 15-28 days of age = 80°F.
- 28-35 days of age = 75°F.
- > 35 days of age = 70°F.

Signs of insufficient heat are: burrowing deep into blankets, rolled up tightly in a ball. Cubs may also experience gassiness and constipation from an inability to completely digest food when hypothermic. Cubs that are too weak may lie fully stretched out, pointing with nose and mouth near incubator air vents; are fussy and may have diarrhea (McManamon and Hedberg, 1993).

Heating pads and heat lamps may be appropriate alternatives for providing ambient heat to healthy, active cubs. They are not good alternatives for very young and/or weak cubs that are unable to move away from the heat, as needed. Severely compromised cubs will tend to sleep in one position for extended period which can result in thermal burns from either a heating source directly above or below the animal. Active cubs are able to move toward or away from heat sources more easily.

Heat lamps should be positioned far enough above the animal so it can't jump up and contact the heat lamp directly or knock it down into the enclosure. Only half of the cub's enclosure (or area large enough for all cubs to have access) should be heated. An equivalently-sized unheated area must also be provided so the cub(s) can move away from heat, as needed. Ideally there would be a 5-10°F heat range from the warmest to the coolest area of the enclosure (Hedberg, 2002). Monitor the temperature of the heat lamp by placing a room thermometer in the enclosure, below the heat lamp and at approximately the same height the cub's body would be in relation to the lamp when turned on. Adjust the lamp as needed to maintain an appropriate temperature.

Heating pads provide a heat source that comes from below the animal. This may more closely simulate the heat coming from lying along side of or on top of the mother's abdomen. Heating pads should never be placed directly in an enclosure with an infant. The two problems that occur are thermal burns from direct contact with the pad, and chewing on electrical cords. The disadvantage of heating pads is the inability to regulate the temperature as well as heat lamps and incubators. Heating pads have settings of low, medium and high, and have a wider variance of temperatures within those settings. The general recommendation for using heating pads with animals is to wrap the pad in a regular bath towel (not overly thick) and place UNDER the enclosure, such as a box or pet carrier.

The heating pad is ALWAYS set on LOW. The heating pad should only provide heat to approximately half of the enclosure so the infant can move away from the heat source, as needed, to prevent overheating. When setting up, a room thermometer should be placed on the enclosure floor after the heating pad has reached its designated temperature to determine how much heat is available to the cub. Adjustments can be made with the thickness of the towel wrapped around the pad to increase or decrease the temperature. The towel should always be under the heating pad to prevent excess heat loss from the pad into the floor or table that it sits on. Providing a cloth barrier between the pad and the housing container is not always necessary. Having the container directly on the heating pad will provide additional heat, if needed. Core must be taken if the cub is housed in a cardboard container, which can become quite hot if set directly on a heating pad.

Bedding

Polyester fleece and fake fur are good sources of bedding that are easily washed and sanitized. Sewing fleeces together into pillow-shapes and stuffed with small towels can be placed in the incubator or other enclosures. They simulate the soft feel of the maternal abdomen. Cubs reportedly sleep on top of fleeces, which may reduce stress and give the cubs a sense of security (Hedberg, 2002). Care should be taken to ensure cubs do not chew on and consume fabrics, which have the potential to cause an intestinal obstruction.

Color coding feeding utensils, including nipples, is a good way to distinguish supplies for each cub when litters are hand-reared together. Applying different combinations of colored nail polish to one or more toenails of each cub will aid in identification until definitive physical characteristics are determined (Hedberg, 2002).

Physical/mental stimulation

Cheetah cubs become quite active within two weeks of age. Physical movement may assist with motor function stimulation and proper development/growth of bones, muscles and nerves. Cubs confined to an incubator or small enclosure, which limits movement, may benefit from manual stimulation of limbs several times a day. Taking a few minutes before or after each feeding to manipulate limbs or allowing cubs to run, jump, pounce and stretch forelimbs may assist with proper development. The Felid TAG recommends moving cubs to larger enclosures that allow for movement by 21 days of age.

Play behavior between cubs may serve several useful purposes, including the development of predatory skills, developing strength/endurance, social bonding and communication skills (Caro, 1995). Cheetah cubs have been observed to engage in several types of aggressive play behavior, including crouching, stalking, pouncing and chasing with littermates, which may be important motor activities related to future hunting skills. Caro (1995) observed wild, mother-raised cheetah cubs and noted that cubs engaged in contact social play behavior with littermates on a daily basis until eight months of age. Play behavior was separated into five categories: Locomotor play (running, rushing about) was the most common play behavior at two months of age, and was thought to serve the purpose of escaping predator (Caro, 1995). Social (contact and non-contact), and object play categories increased between two and three months of age. Exploratory play behavior continued to rise until one year of age. The author made connections between play behaviors and predatory skills used later in life. Stalking and crouching observed in non-contact social play was associated with approaching prey. High levels of object and contact social play were associated with higher rates of patting, biting and grasping live prey.

WEANING

There are many different viewpoints regarding when and how to initiate the weaning process in exotic felids. The Felid TAG recommends adding chicken or turkey baby food to the milk formula at 4 weeks of age with small felids. Various zoo facilities recommend weaning when the cubs show interest in solid food. Gittleman and Otte (1987) indicated cheetahs first consumed solid food at 33 days of age and weighed 1.94 kg (4.27 lbs.)

As the cubs begin consuming solid foods in a measurable amount, the volume of formula can be reduced proportionately. Many times cubs will take the bottle at some feedings but refuse it at others. Eliminating specific feedings rather than reducing the volume at each feeding allow the cubs to get hungry enough to explore other food options available, such as a bowl of meat.

It is important to weigh cubs every 1-2 days to monitor weight fluctuations during the early stage of the weaning process, especially when multiple cubs are fed together. It is common for different levels of food consumption to occur amongst individuals in a litter. You may be providing an appropriate amount of food to feed four cubs, but two cubs are eating 60-80% of the food. Monitoring the weight will help caretakers determine if all cubs are consuming appropriate amounts of the weaning diet. Another clue that a cub may not be progressing in the weaning process is that it always seems very hungry at each bottle feeding compared to the littermates. In this case, adding pureed meat to the formula for that cub, or separating it to feed from its own dish will ensure the cub gets its share.

At this point, there are no nutrient requirements established by the NRC specifically for cheetahs. Therefore, until more specific data becomes available, the domestic cat is used as the reference model on which the cheetah requirements are based. Slab meat diets typically provide much higher levels of protein than the domestic cat requirement. The cat requirements are generally considered minimum requirements, and are used as a guideline until further research on cheetah nutritional requirements become available.

Table 4: Minimum nutrient requirements for domestic kittens. (DM basis) NRC (1986).

Crude protein - 24%	Zinc = « 50mg/kg DM
Crude fat 9%	Niacin 40.9/kgDM
Calcium = 0.8% DM	Taurine = 400mg/kg DM
Phosphorus = 0.6% DM	Vitamin A = 3333 IU/kg DM
Magnesium = 0.04% DM	Vitamin D = 500 IU/kg DM
Iron = 80 mg/kg DM	Vitamin E = 30 mg/kg DM
Copper = 5 mg/kg DM	

Appropriate foods for weaning diet

Poultry-based human baby foods (chicken and turkey) are commonly used at the initial stage of weaning. The baby food is gradually added to the formula or placed in a shallow bowl with warmed formula poured over it to entice the cubs. Small amounts of baby food are provided at each feeding, and increased daily as long as the stool appears normal (Felid TAG). This is a common weaning strategy for small exotic cats. It may not be practical with the larger species, including cheetah, except in specific cases involving “poor-doers” or runts of the litter that need special attention and have chronic digestive problems. Chicken baby food has been beneficial in keeping the stool firm, whereas beef baby food is preferable in cubs prone to constipation (Felid TAG).

Poultry has the added benefit in that it is a good source of taurine, an essential amino acid in felids. The requirement for domestic kittens is 400mg/kg DM (NRC, 1986). Uncooked chicken muscle contains, on average, 99a mg/kg DM taurine. Cooking reduces the taurine concentration somewhat (NRC, 1986). However, the Felid TAG recommends cooking chicken prior to feeding to kill *Salmonella*, which can cause diarrhea.

Commercial feline diets for domestic kittens, ZuPreemTM and Nebraska BrandTM feline diets may be offered in a bowl with warmed milk formula poured on top, to entice exploration. As cubs start consuming the diet in measurable amounts, the addition of milk formula to the bowl may be discontinued.

Zoo facilities that feed slab meat to adult cheetahs may prefer to wean cubs onto a meat diet comparable to that of the adults. Muscle meat is low in calcium and high in phosphorus. See table 5 for a comparison of muscle meats and whole prey items. Chicken muscle meat has a 1:21 ratio of calcium: phosphorus, whereas whole chicken has a Ca:P of 1:1 (USDA, 2004).

Table 5: Comparison of muscle meat, organ meat and whole body prey. Values are on DM basis. Dierenfeld, et al (2002) ; USDA, (2004)a: Ullrey and Bernard, (1989)³

Meat	CP %	Fat %	Ca %	P %	Mg %	Fe mg/kg	Cu mg/kg	Zn mg/kg	Vit A IU/kg	Vit E IU/kg	Kcal/kg
Chicken, whole!	42.3	37.8	1.68	1.3	0.09	40	3.0	45	35600	51.3	5900
Chicken, muscle* meat only	77.5	22.9	0.04	0.76	0.07	39.6	3.2	50.9	3818	8.7	5382
Chicken heart*	58.9	35.1	0.05	0.68	0.06	225	13.1	249	1132	0	5774
Chicken liver*	72.3	20.4	0.03	1.26	0.08	383	20.9	114	471404	29.8	5064
Horse, meat only³	76	18	0.05	0.34	0.05	232	3.0	128	2593	0	n/a
Cow, meat only³	63	29	0.03	0.55	0.06	78	2.0	106	1428	3	n/a
Deer, meat only³	65	29	0.03	0.59	0.06	165	5.0	68	0	0	n/a
Rabbit, whole!	63.5	15.3	2.35	1.68	0.16	302	16	86	6200	16.2 - 60	5410
Rabbit, meat only*	74	20.7	0.05	0.8	0.07	58.2	5.37	58.2	0	0	1360
Rooster, whole (>50g.)!	61.8	32.6	3.45	1.91	0.15	195	7.5	92.1	35600	139	6370
Quail, whole body¹	71.5	31.9	3.43	n/a	0.06	74.9	2.6	53.0	70294	66.8	6790

n/a = data not available

Muscle meat and whole animals (birds, rats, rabbits) stripped of fur, tail, head, feet, beak, etc. can be ground up and provided in small amounts in a bowl with warmed formula poured on top at the initiation of the weaning process. Rabbits that were fed nutritionally balanced diets during their lives are considered a “complete food” in that it meets or exceeds the basic dietary nutrients of a kitten without the addition of vitamin or mineral supplements. This is with the assumption that the cub consumes the meat, bone and viscera (except stomach and intestine). Rabbit is generally accepted by cheetahs since it is similar to one of the wild cheetah's natural prey (springhare).

Vitamin A

Pre-formed vitamin A is an essential nutrient for felids, so must be provided in the diet (Irlbeck, 1996). However, as a fat-soluble vitamin, it is stored in the body, so it is not required on a daily basis. It is important not to provide excessively more than the requirement since it can accumulate in the body to toxic levels. In growing animals, vitamin A toxicity is associated with skeletal malformations and fractures, internal hemorrhage, enteritis, conjunctivitis, and reduced function of liver and kidneys (McDowell, 2000; Robbins, 1993).

In rabbits, vitamin A is contained in the organs, particularly the liver. While cubs are still consuming milk formula, additional vitamin A is generally not required (depending on the nutrient composition of the formula), so organ meat should not be provided at this stage. However, after completely weaned onto solid food, cubs must consume the liver of rabbits to meet the vitamin A requirement, if vitamin supplements or other food items high in vitamin A are not provided.

Vitamin A is relatively high in whole chicken, rat and quail, and in chicken liver. The whole animals exceed the kitten requirement by 11-21 times, and chicken liver exceeds it by 141 times, by weight (NRC, 1986). If these food items are frequently included in the weaning diet, a vitamin supplement containing vitamin A should not be provided. In addition, combining a diet of one or more of these items with another food item low in vitamin A, such as chicken meat, chicken heart and rabbit muscle meat will help offset the excess. On an "as fed" basis, one ounce (30 g.) of chicken liver provides the daily vitamin A requirement for domestic kittens.

Vitamin D

Natural sources of vitamin D are available in two forms - D₃ which is synthesized in the skin of animals with exposure to sunlight, and D₂, which occurs mainly in plant matter. Most carnivores are able to utilize both D₂ and D₃, although lions and tigers preferentially utilize D₃ (Robbins, 1993). It is unknown as to whether or not this is also the case with cheetahs. Vitamin D is not present in the milk of most mammals, with the noted exception of polar bears (Kenny, et al, 1999). Maternally-raised captive cheetah cubs have reportedly left the lair, for short periods, at 28-38 days of age (Stoecker-Horwath and Schwammer, 2003; personal observation). This may be the point at which cubs require a source of vitamin D₃.

Milk formulas based on KMRTM or EsbilacTM contain vitamin D at levels which meet or exceed the domestic kitten requirement. As long as cubs are consuming the milk formula, no supplementation is not required. In order to maintain proper bone growth, cubs that are weaned off formula at an early age may require access to sunlight (or indoor UV-B light) or a D₃ supplement if their diet contains less than 500 IU/kg DM of D₃.

Calcium and phosphorus

Whole rabbit, rat and chicken provide a balanced Ca:P ratio of 1.4:1 - 1.8:1. All other food items (muscle meat, liver and heart) have a skewed Ca:P ratio in favor of phosphorus. Not only do felids require an absolute amount of calcium and phosphorus (0.8 and 0.6% of the diet, respectively), but they also require a balanced ratio between the two minerals to promote proper calcium absorption. Ca: P ratios of 1:1 to 2:1 are the recommendations for growing infants. (Trendler, 1997). Grinding the skeleton of rats, rabbits and/or chickens and including them in the meat diet will provide a good source of calcium. Grinding must be thorough and large pieces of bone and sharp bone shards are removed before feeding. Cartilage, tendons and ligaments may be offered as a source of fiber.

Meat diets that do not contain bone require the addition of a calcium supplement. Table 6 compares various forms of calcium. It should be noted that supplements can not all be used interchangeably since they have different concentrations of minerals. In addition, calcium may have different absorption rates, depending on the form it's in. Limestone is the least available source of calcium, whereas calcium phosphate and bone meal are more readily absorbed into the body (E. Dierenfeld, pers. com). When adding a calcium supplement, it is important to provide only enough to balance the diet. Too much calcium can be as detrimental as not enough in growing animals. Excess calcium in the diet has been linked to osteochondrosis, enlarged joints, splayed feet, angular limb deformities and stunted growth (Hazewinkel, et al, 1985; Hedhammer, et al, 1974).

Table 6: comparison of calcium supplements. Values on DM basis. Kellems and Church (2002)

Supplement	DM%	CP%	Ca%	P%	Mg%	Cu mg/kg	Fe mg/kg	Zn mg/kg
Bone meal, steamed	97	13.2	30.7	12.9	0.3	-----	26700	100
Calcium carbonate	100	-----	39.4	0.04	0.05	-----	300	-----
Dicalcium phosphate (Dical TM)	97	-----	22.0	19.3	0.6	10	14400	100
Limestone, ground	100	-----	34.0	0.02	2.1	-----	3500	-----

DM= dry matter, CP= crude protein, Ca= calcium, P= phosphorus, Mg = magnesium, Cu = copper, Fe= iron, Zn = zinc

As cubs continue to consume more and more of the meat diet, eliminate formula feedings from the daily schedule, one at a time. Late night feedings can be dropped first, if a bowl of food is provided to allow for self-feeding. The a.m. formula feeding should be the last bottle feeding dropped from the schedule, and may be eliminated by 5 weeks of age, in most cases. Continue offering warmed formula in a bowl with the solid feline diet. Complete weaning from formula should occur within 10 weeks of age, but cubs will probably lose interest in the formula before then.

Cubs that are weaned onto a slab meat diet may not require a vitamin supplement while they continue to consume formula, if the meat portion of the diet is a combination of chicken meat, whole chicken and rabbit. They will, however, require a calcium/iron supplement if meat only (no bone) is offered. A taurine supplement may be warranted if red meat is offered instead of chicken. Table 7 is an analysis of a diet composed of equal parts of milk formula and chicken meat for a 3.0 kg (6.6 lb) cub.

Table 7: Nutrient analysis of a weaning diet, combining chicken meat and milk formula.

Food Item	CP %	Fat %	Ca %	P %	Mg %	Fe mg	Vit A IU	Vit D IU	Vit E mg	Kcal
Chicken meat, 175g 48g DM	38.8	11.5	0.02	0.4	0.04	1.8	172	0	0.4	265.5
Milk Formula, 180ml 40 g. DM, (table 3)	19.9	21.2	0.7	0.5	0.04	3.1	4473	361	9.1	227.0
Calcium carbonate 1g.	-----	-----	0.4	-----	-----	0.3		----		-----
Limestone, 1g.	-----	-----	0.3	-----	0.02	3.5	-----	-----	-----	-
Total	58.7	32.7	1.42	0.9	0.10	8.7	4645	361	9.5	492.5
Requirement	24-40	* 40	0.8	0.6	0.08	7.0	290	43.5	2.6	479
Difference	HIGH	OK	OK	OK	OK	OK	HIGH	HIGH	HIGH	OK

The analysis indicates that the milk formula contains an excess of fat-soluble vitamins, so should not be supplemented in the chicken meat. This diet is, however, deficient in calcium and iron unless a supplement is provided. A combination of calcium carbonate and limestone is used to provide an adequate level of iron. Calcium carbonate, while a good source of calcium, is quite low in iron. Limestone, which is a lower quality calcium source, contains ten times the amount of iron as calcium carbonate. The combination of the two supplements provides the necessary concentrations of both minerals. The diet, with supplements, provides a Ca: P ratio of 1.6:1, which is optimal.

The protein concentration of the diet is higher than the NRC requirement. But healthy captive cheetahs typically consume a diet very high in protein, and will continue to consume protein in excess of 50% of the diet in adulthood. The one concern with growing cubs is that they will grow at a faster rate than recommended (8-10% body wt/day). Fast growth rates are associated with abnormal bone growth and deformities (Irlbeck, 1996). During the weaning process, the protein content of the diet may be reduced, if necessary, by mixing the slab meat diet with a commercial feline diet, such as ZuPreemTM or Nebraska BrandTM, which contain 43 and 47% CP, respectively (on DM basis).

In general, animals feed to meet their energy needs. Felids metabolize protein and fat for their energy needs (Bechert et al., 2002). Cubs consuming high protein diets may consume fewer calories than expected, based on the MER. Cubs that maintain an ADG of 8-10% are meeting their nutritional needs on more nutrient-dense diets. If feasible, continue weighing cubs, at one week intervals, as long as possible until weaned to ensure there is consistent growth.

The crude fat requirement for kittens has not been established by the NRC. For this example, since the cub is still consuming formula, the 40% fat content of formula was used as the maximum. Bechert, et al (2002) indicated a low protein: fat ratio (2:1 to 3:1) was consistent with that found in whole prey and was preferable to higher ratios. The protein: fat ratio of this diet is 1.8:1, which may be appropriate for growing animals.

The high levels of fat-soluble vitamins can not be lowered significantly while cubs are on the milk formula. The high level of vitamin E is actually beneficial in lowering the absorption rate of vitamin A. But it is important to note that the fat-soluble vitamins should not be supplemented until cubs are weaned off the formula, since more than enough is provided. Additionally, liver should not be included in the diet until cubs are weaned off milk formula completely. It should be noted that the milk formula used in this example is the KMR formula in table 3a. Different formulas, especially those that are less nutrient dense, will have a different nutritional analysis, and may be deficient in some nutrients. The purpose of this example is to show that it is important to know the nutrient composition of the diet prior to adding supplements, and to add only those that are required.

SUMMARY OF POINTS

- 1. The initial 2-3 feedings should be electrolytes only.**
- 2. Gradually transition to the stock milk formula over a period of 3-5 days, starting at 1:4 dilution of formula: water and increasing the formula concentration by increments. 1:4 x 2-3 feedings, 1:3 x 24 hrs., 1:2 x 24 hrs, 1:1 x 24 hrs, full strength.**
- 3. Determine the number of kcal, amount of food that can be offered at each feeding and number of feedings/day on the first day and adjust every few days as the cub grows, to maintain optimal growth rates. See Appendix 1 for calculated values.**
- 4. It is preferable to provide a nutrient dense milk formula with a carbohydrate content comparable to cheetah milk (14-15% DM).**
- 5. The Zoologic Milk MatrixTM line of milk formulas may be preferable to KMRTM and EsbilacTM, especially if lactobezoars or constipation becomes an issue.**
- 6. Digestive problems (diarrhea and constipation) can initially be dealt with by offering 1-2 doses of oral electrolytes, and going back to the formula dilution recipe prior to when the problem first occurred. Give 2-3 feedings at that rate and then go to the next more concentrated level. Seek veterinary attention if problem doesn't resolve within 24 hours.**
- 7. Solid foods (in pureed form) can be introduced at approximately 1 month of age. Do not include the calories from this food in the cub's daily caloric allotment until the amount consumed is measurable.**
- 8. Start decreasing formula feedings, by a proportional amount, when solid food consumption becomes measurable.**
- 9. During the weaning process, analyze the nutritional composition of the diet to determine if vitamin/mineral supplements are warranted.**
- 10. Calcium, iron and taurine are nutrients that may need to be supplemented as soon as cubs start consuming solid food.**
- 11. With nutrient dense milk formulas, vitamin A should not be supplemented in the diet until cubs are completely weaned off milk formula.**
- 12. Weaning can be completed, in most cases, by 8-10 weeks of age.**

Appendix 1: Calculated values for Kcal/day and ml/feeding

Weight†	ME (Kcal/day) $[70 \times \text{bw (kg)}^{0.75} \times 31]$	ml/feeding (Stomach capacity)
450g. (15 oz.)	115 kcal/day	22.5 ml/feeding
500g.	125	25.0
550g.	134	27.5
600g. (20 oz.)	143	30.0 (1 oz.)
625g.	148	31.25
650g.	152	32.5
675g.	156	33.75
700g.	161	35.0
725g.	165	36.25
750g. (25 oz.)	169	37.5
775g.	173	38.75
800g.	178	40.0
825g.	182	41.25
850g.	186	42.5
900g. (30 oz.)	194	45.0 (1 oz.)
950g.	202	47.5
1.0 kg. (2.2 lb)	210	50.0
1.1 kg.	225	55.0
1.2 kg.	241	60.0 (2 oz.)
1.3 kg.	256	65.0
1.4 kg.	270	70.0
1.5 kg. (3.3 lb)	285	75.0 (2 oz.)
1.6 kg.	299	80.0
1.7 kg.	313	85.0
1.8 kg.	326	90.0 (3 oz.)
1.9 kg.	340	95.0
2.0 kg. (4.4 lb)	353	100
2.1 kg.	366	105 (3 oz.)
2.2 kg.	379	110
2.3 kg.	392	115
2.4 kg.	405	120 (4 oz.)
2.5 kg. (5.5 lb)	418	125
2.6 kg.	430	130
2.7 kg.	442	135 (4 oz.)
2.8 kg.	455	140
2.9 kg.	467	145
3.0 kg. (6.6 lb)	479	150 (5 OZ.)

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www.valleyvet.com

ZuPreemTM

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1-800-345-4767
www.zupreem.com

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Appendix V: EAZA Felid TAG demonstration guidelines

Demonstration Guidelines for felid species used in public demonstrations



The 'Guidelines on the use of animals in public demonstrations' (September 2014) provides guidance on the use of exotic animals in public demonstrations at EAZA member institutions. The EAZA Felid TAG supports the use and implementation of the 'Guidelines on the use of animals in demonstrations' document and has, thus far, used this as the model in the absence of taxa specific guidelines. To best support our members on best practice guidance, the EAZA Felid TAG has in addition to EAZA Best Practice Guidelines produced this 'Demonstration guidelines for felids species used in demonstrations' document to provide species-specific guidance for institutions performing public demonstrations with felid species.

Public demonstrations are defined as any case where an animal is demonstrating behaviours, trained or natural, while under the supervision or control of a trainer in the view of guests, with the intention of educating, inspiring, and entertaining the visitors (EAZA, 2014). A public demonstration should be designed with animal welfare as the primary factor and the visitor experience as a secondary factor and should utilize the best practice standards. Following the definition of Good Practice in the 'EAZA position statement on circus membership of the association' (April 2017) Felid demonstration programmes should focus on behaviours that are demonstrations of an animals' natural intellectual or problem solving abilities and their physical attributes, showcasing as much behavioural diversity as possible. All public demonstration programmes with felids should reflect section 1.11.2 of the 'EAZA standards for the accommodation and care of animals in zoos and aquaria' document (2014) and serve to:

- Promote an understanding and raise awareness of wild felid species.
- Allow the visitors to see the cats active and showcase felid species natural behaviours.
- Educate visitors about their special adaptations and their role as predators in their ecosystem.
- Educate the visitors about the threats that most of these species face in the wild and the role of zoos in the conservation efforts of felid species and the role of zoos generally in the global effort to preserve animals and their habitats in the wild.

- Inspire guests to connect with felid species with the hope of conservation minded behaviour change.

The Felid TAG does not support the performance of any behaviours that when implemented poses a demonstrable or probable risk towards animal and / or human health or welfare, be it physical or psychological. These include:

- Any situation where an animal, a staff member or guests' safety is unnecessarily and knowingly placed at risk.
- Any situation that is demeaning or degrading to the animal
- Any practice that requires the physical disciplining of an animal to provide protection for a staff member who is in contact with that animal for any purpose other than the preservation or improvement of its health and wellbeing.

The Felid TAG recognises any kind of behaviours which could be utilized for medical training (e.g., body parts presentation, touching targets, holding for examination and blood draw training) as part of husbandry training. The behaviours can be performed as part of a public demonstration to serve to facilitate the inspection of the animal and (in special cases) also to the drawing of samples or the treatment of a trained individual. Actual medical procedures should not be on display but the Felid TAG recognises that desensitization (e.g. fake needle touches, the use of ultrasound wands) could be part of an educational talk about veterinary care and protected contact. For information on species-specific behaviours and recommended husbandry/safety procedures check the relevant EAZA Best Practice Guidelines or contact the programme leader (EEP/ESB) for further advice.

For the purpose of this document the Felid TAG considers the following grouping of cats:

- Large cats: species of the genera *Panthera* (incl. *P. uncia*), *Neofelis*, *Acinonyx jubatus*, *Puma concolor*;
- Medium Cats: species of the genera *Lynx*, *Caracal*, *Catopuma*, *Leptailurus*, *Leopardus pardalis*, *Prionailurus viverrinus*;
- Small Cats: *Felis* species, all other *Leopardus* sp., all other *Prionailurus* sp., *Otocolobus manul*, *Pardofelis marmorata*, *Herpailurus yagouaroundi*;

The species under the TAG's remit have been sorted into three categories (large/medium/small cats) for the following section to allow more specific guidance on e.g., guaranteeing safety, usage of props and training of the animals.

The following points are strongly recommended by the Felid TAG to be followed and should be taken into account prior to setting up a public demonstration with felid species:

Training of animals

- 1) [*Large/ medium/ small cats*] The Felid TAG is strictly against any use of rearing techniques for demonstrations that directly affect the welfare and health of the animal, including the premature removal of an animal from the mother with the intention of hand-raising specifically for use in a demonstration. This could lead to unacceptable imprinting on humans and a welfare issue in them not being entirely socialized to conspecifics. (Health of animal)
- 2) [*Large/ medium/ small cats*] Training techniques used for demonstrations must be positive reinforcement only to promote animal welfare.
- 3) [*Large/ medium/ small cats*] There should be an emphasis on the use of motivating operations whereby the animal is seeking out food in a natural way, not reliant on the hunger state. (This is referred to in the use of “weight control” in the EAZA animals in demonstrations guidelines).
- 4) [*Large/ medium/ small cats*] The cats should always be able to choose if they want to participate in the demonstration. There should be no repercussions in food, choice or enrichment if they choose not to participate.

Safety and housing

The Felid TAG does not support placing cats in a performance environment that does not reflect the EAZA Standards for the Accommodation of Animals in Zoos and Aquaria (2014).

- 5) [*Large/ medium/ small cats*] The animals should not be moved outside their enclosure for the purpose of demonstrations because of animal welfare and safety reasons. Situations where animals need to be moved temporarily or permanently from their current enclosure to another enclosure / location (i.e. for veterinary purposes, crate training, socialising / reintegrating of one cat to another) should not be done as part of any public demonstration to ensure the upmost animal welfare. Situations with small cats that may have to go through rehabilitation with a keeper away from their enclosure / holding facility should always be done away from public view.
- 6) [*Large cats*] Full contact management must be excluded for all demonstrations purposes in the large cats due to safety considerations.
- 7) The Felid TAG recommends to not take members of the public into the same enclosure as where the cats are.
[*Large/ medium cats*] Members of the public should not be taken into the same enclosures that houses large or medium categorised cats as it can greatly compromise human safety and animal security.
[*Small cats*] The Felid TAG accepts that with small cat’s members of the public may be taken into the enclosures as part of demonstrations when the following conditions are met:
 - Guests must always be accompanied by at least one animal keeper.
 - Guests should be limited to a maximum of three at any one time.

- Guests must never attempt to touch the animals.
 - The enclosures must be large enough and complex enough to offer privacy and security for the cats.
 - The animal keeper must end all demonstrations if the animals show signs of stress, fear or excessive aggressive behaviour.
 - Guests must not be taken in with kittens (0 – 6 months).
 - The animal keeper must notify the guest of the correct behaviour inside the enclosure
- 8) [*Medium/ small cats*] For the smaller wild cat species the Felid TAG acknowledges the benefit for a keeper to perform husbandry training and/or feeding demonstrations, which would be best from outside, but may also according to in-house safety protocols be allowed from inside the animals' own enclosure.
- 9) [*Large/ medium/ small cats*] The physical touching of animals by the public and the demonstrator is not accepted by the Felid TAG. (Human/Animal interaction). Situations where the demonstrator has to touch an animal as part of a veterinary/ medical training procedure i.e. touch the tail to condition for blood, that is visible by the public would be accepted however the purpose of such an activity should be made clear to public via signage or personal communication. It should be made clear that the touching of animals by public/guests in the company of an animal keeper could encourage other members of the public that are viewing this activity to attempt to touch cats through fences when no keeper is present and thus compromises public safety.

Usage of props

- 10) [*Large/ medium/ small cats*] The walking of animals on a leash is not accepted by the Felid TAG (Human/Animal interaction). It sends the wrong message to the public about these wild animal species (predators) being tame or even domesticated. This action also eliminates choice for the animal once on the leash and has the potential to increase risk of human/ animal injury should the animal break or pull free from the demonstrator.
- 11) [*Large/ medium/ small cats*] The Felid TAG does not accept the use of props (any item/ object used by the animal or trainer as part of animal demonstration or training program) that compromises animal welfare or promotes negative reinforcement. Props that are added to an enclosure or are permanently part of an enclosure that are used during animal demonstrations (i.e. feed poles, zip lines, keepers feeding poles, targets, weighing scales, boomer balls, pull ropes etc) must promote natural behaviours and positive animal welfare. It is encouraged that collections should do their utmost to explain the positive use of such items during demonstrations to the public.

EAZA Felid TAG (Nov. 2017).

Appendix VI: Cheetah veterinary guidelines

Introduction

The cheetah veterinary guidelines are developed in order to help optimise the veterinary conditions, for the wellbeing of the animals kept in captivity. The data is obtained from different literature sources, such as books, articles and the Internet. Two sources, from which a fair amount of information was cited, are: the Cheetah SSP Health Chapter (Citino et al., 2007) and the veterinary guidelines for the Cheetah EEP (Tschurlovits, n.d.).

The first chapter of the cheetah veterinary guidelines describe the different non-infectious and infectious diseases. It contains information on the clinical signs, diagnosis methods and treatment measures. Chapter 2 Diagnosis is an overview on medical examinations that should be implemented. Chapter 3 contains information on the different drugs to administer for sedation and the anaesthesia process. The final chapter is dedicated to preventive medicine; here vaccination schedules and endo- and ectoparasite treatments are illustrated.

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1 Diseases

This chapter is divided in two: the different non-infectious diseases and infectious diseases. Per disease, information on the clinical signs, diagnosis methods and treatment measures are described.

1.1 Non-infectious diseases

The non-infectious diseases affecting cheetahs in captivity are categorised in five paragraphs; gastrointestinal diseases, renal diseases, neuromuscular diseases, miscellaneous non-infectious diseases and nutrition related diseases.

1.1.1 Gastrointestinal diseases

Gastritis

Severe gastritis is linked to different types of *Helicobacter spp.* and it is a major cause of morbidity and mortality in captive cheetahs (Terio, 2012). The pathogenesis is likely attributed to the cheetah's immune response and chronic stress (Terio, 2012; Lane et al., 2011). Diet affects the rate at which cheetahs develop gastritis (Lane et al., 2011).

Clinical signs

Chronic vomiting, undigested meat in the faeces, weight loss and dull hair coats can be seen. Additionally, cheetahs may develop secondary complications such as asphyxiation or pneumonia and systemic amyloidosis, which are often fatal (Lane et al., 2011).

Diagnosis

- Endoscopic biopsy (Lane et al., 2011). Should be used for initial diagnosis.
- Urea breath test (UBT) can be used to monitor recrudescence or reinfection (Chatfield et al., 2004).

Treatment

- Antibiotics (tetracycline or amoxicillin and metronidazole), bismuth compounds and omeprazole should be administered. Additionally, an evaluation of husbandry methods should be made to find a way to reduce stress (Wack, 1999). However, antibiotics should not be used unless clinical signs are clearly apparent (Citino, 2005), because gastritis is not always cured with treatment (Tschurlovits, n.d.). In cats' metronidazole genotoxicity was observed in feline PBMC (Peripheral Blood Mononuclear cells) and the T-cell lymphoma line (Sekis et al., 2009).
- No single drug is enough to treat *Helicobacter*, the best results are achieved through the combination of three or more drugs. The short-term elimination of *Helicobacter* has been achieved through the combination therapies of antibacterials and PPIs such as omeprazole/clarithromycin/amoxicillin or lansoprazole/clarithromycin/amoxicillin and tetracycline/metronidazole/Pepto-Bismol (Citino, 2005).

Prognosis

Most therapies do not provide a long-term cure (Tschurlovits, n.d.). Antibiotics provide only a short-term relief (Citino, 2005).

Focal palatine erosion

Focal palatine erosion is (as the name implies) the erosion of gums, followed by the perforation of the palatine bone. It was believed that this disorder only occurred in captive cheetahs originating from Namibia. However, this disease can also be found in free-living individuals of Namibia, in other free-living cheetah populations, as well as in 14 other species of felids (Zordan et al., 2011).

Clinical signs

Bloody-mucus, nasal discharge, halitosis, sneezing and coughing. Severe cases lead to localized osteomyelitis, intermittent chronic septicaemia and without treatment death (Zordan et al., 2011).

Diagnosis

- Oral examination (Tschurlovits, n.d.).
- Pathology (Tschurlovits, n.d.).

Treatment

In order to treat this disorder, the lower molars can be trimmed, systemic antibiotics can be administered and a surgical repair can be attempted (Zordan et al., 2011).

Foreign body ingestion

Enrichment items, or any other items present at the installations can be swallowed, leading to a gastrointestinal obstruction. Clinical signs observed can be choking, vomiting and anorexia (Tschurlovits, n.d.).

1.1.2 Renal diseases

In order to diagnose renal failure, a blood serology (kidney panel: urea and creatinine) and urine analysis can be conducted (Kaandorp, C., pers. comm., 2017). Another method for early detection of the disease is endogenous creatinine clearance (CCr). The mean value is: 1,47 +/- 0,20 ml/min/kg body weight (Holder et al., 2002). Lastly radiographs or ultrasound can be used to measure the size of the kidneys in order to diagnose renal failure. The average ratio of a healthy kidney to vertebra is around 1,81 +/- 0,14. However, cheetahs with the disease can have the same measurements as a healthy individual (Hackendahl, 2004; Kaandorp, C., pers. comm., 2017). One of the firsts indications of possible renal affliction is a noticeable increase of water intake by the animal (Stagegaard, J., pers. comm., 2017).

Amyloidosis

Amyloidosis is a group of diseases characterized by the deposition of an amyloid protein in different organs and tissues (Woldemeskel, 2012). In Cheetahs, the amyloid deposits normally strangulate over time the blood supply to the renal papilla, leading to acute or chronic renal failure (Carlisle, n.d.). If the animal is additionally suffering from glomerulosclerosis the condition further aggravates (Carlisle, n.d.). There is a high prevalence that amyloidosis is a result of chronic inflammation and environmental factors, instead of the consequence of inheritance (Campbell, 2007).

Clinical signs

The clinical signs may vary; they will range between subclinical to organ failure. It will depend on the organ that is affected and the amount of the deposition (Tschurlovits, n.d.). In cheetahs, protein loss in the urine, weight loss, non-regenerative anaemia, uraemia, polydipsia and polyuria can be observed. Other signs may include dull hair coat, elevated urea and creatinine levels, ataxia, weakness, anorexia, dehydration, vomiting and diarrhoea (Carlisle, n.d.).

Diagnosis

The early detection of renal function can be tested with the following tools: endogenous creatinine clearance and fractional excretion of electrolytes (i.e. Na, K, P and Ca). During the end stages of the disease, it can be tested by looking at the BUN-to-creatinine ratio (Tschurlovits, n.d.). Microscopic examination of biopsied tissue, Congo red staining and Masson's Trichome stain are methods used to diagnose amyloidosis (Woldemeskel, 2012; Carlisle, n.d.). Immunohistochemical staining is used to identify the type of amyloidosis (Woldemeskel, 2012).

Treatment

It consists in the treatment of the symptoms and of the chronic disease (aggressive diuresis), a change of diet and reduction of stress (Tschurlovits, n.d.). The administration of nephrotoxic drugs (aminoglycosides, NSAID's, etc.) should be avoided (Carlisle, n.d.).

Glomerulosclerosis

The cause of the disease is unknown, however, it is believed that diet and chronic stress may contribute to its appearance (Tschurlovits, n.d.). It is characterized by the gradual thickening of the glomerular basement membrane; overtime this leads to glomerular ischemia and sclerosis (Robert, 2009).

Clinical signs

Clinical signs observed will be very similar to other renal diseases.

Diagnosis

The early detection of renal function can be tested with the following tools: endogenous creatinine clearance and fractional excretion of electrolytes (i.e. Na, K, P and Ca), during the end stages of the disease it can be tested by looking at the BUN-to-creatinine ratio. A histopathological examination of the kidney can be performed: here a thickened glomerular membrane, interstitial fibrosis, nephritis, glomerulonephritis and calcifications can be observed (Tschurlovits, n.d.).

Treatment

It consists in the treatment of the symptoms and of the chronic disease, a change of diet and reduction of stress (Tschurlovits, n.d.).

Oxalate nephrosis

Oxalate nephrosis is a disease that damages the kidney through the formation of oxalate crystals deposits in this organ. It can become fatal if left untreated (Schmidt-Küntzel, 2015).

Oxalate nephrosis is not related to renal and liver disease, gastritis or enteritis. The possible causes are a genetic predisposition, diet and colitis (Mitchell, 2016).

Clinical signs

Symptoms of acute renal failure can be observed (Tschurlovits, n.d.).

Diagnosis

Blood analysis can be performed. With this condition elevated renal parameters (urea, creatinine, phosphorus, calcium and potassium) are to be expected. A renal biopsy can be carried out or a histopathological examination of the kidney. Here acute tubular degeneration associated with large numbers of birefringent crystals, renal medullary amyloidosis and glomerosclerosis can be observed (Tschurlovits, n.d.). Raman spectroscopy can be used to confirm the presence of oxalates (Mitchell, 2016).

Treatment

It consists in the treatment of the symptoms (aggressive diuresis) and dietary management (Tschurlovits, n.d.).

1.1.3 Neuromuscular diseases

Spinal myelopathy

Cheetahs affected by this disease display degenerative lesions on the spinal cord and cerebellum. There is a degeneration of the white matter of the spinal cord, together with axonal and myelin loss. Cheetahs of every age group can be affected, and normally all cheetahs of the same litter develop the disease. The exact causes of this disease are not known (Robert, 2009a). Spinal myelopathy disease is often fatal (Tschurlovits, n.d.).

Clinical signs

This disease can cause ataxia and paresis (Robert, 2009a).

Diagnosis

A differential diagnosis can be conducted with the help of a fast spin echo (FSE). However, Magnetic Resonance Imaging (MRI) is not useful. A histopathology can be conducted; here degenerative lesions of the spinal cord and the cerebellum will be visible, together with the loss of myelin (Tschurlovits, n.d.).

Treatment

It consists in the treatment of the symptoms (Tschurlovits, n.d.).

1.1.4 Miscellaneous non-infectious diseases

(Myelo) Lipomas

A myelolipoma is a nodular lesion that consists of mature adipose tissue and myeloid cells (Walzer, 1996). It has been found in the spleen and liver of cheetahs. Myelolipoma lesions are not clinically important, but it is important that they are not confused with metastatic cancer (Tschurlovits, n.d.).

Diagnosis

- Abdominal ultrasound.
- Necropsy (Walzer, 1996).

Generalized mastocytosis

Cheetahs may exhibit single or multiple firm raised skin masses, called generalized mastocytosis. They have been associated with insect bites and generally disappear on their own. Special attention is advised during diagnosis, as they should not be confused with malignant tumours infiltration (Robert, 2009b).

Some individuals display exudative dermatitis with mast cell infiltration. In this case, animals may have discomfort and suffer weight loss. These lesions can be treated with a short-term corticosteroid therapy (Robert, 2009b).

Peaugres-syndrome

This syndrome is a fatal genetic disease found in cheetahs. Affected individuals do not normally survive the first 134 days of life (Robert, 2009a).

Clinical signs

Poor hair coat, heart malformations, liver fibrosis, stunted growth, osteoporosis and encephalitis (Robert, 2009a).

Beta amyloid deposition and neurofibrillary tangles

Beta amyloid deposition, neurofibrillary tangles and cerebral atrophy was found in some aged cheetahs. These lesions were similar to the ones observed in humans who suffered from Alzheimer disease (AD). The cheetahs suffering from beta amyloid depositions and neurofibrillary tangles, displayed cognitive dysfunction. This condition arises due to genetic factors and chronic stress. In mice, environmental enrichment has not only contributed to the reduction of beta amyloid depositions but it has also lessened cognitive dysfunction (Serizawa, 2012).

Veno-Occlusive Disease (VOD)

It originates by fibrous occlusion of the efferent blood supply of the liver. This leads to liver failure and ascites. The causes of this disease are not known (Robert, 2009a).

1.1.5 Nutrition related diseases

Ulnar metaphyseal osteochondrosis

Is an osteodystrophy that affects the antebrachii during endochondral bone growth. It causes and unbalanced growth of the antebrachium, leading to a displacement of the radius curvus and valgus of the paw. Genetics and/or a dietary hypersupplementation of calcium can lead to this disorder (Allan, 2008).

Clinical signs

Joint pain and transient carpal deviation can be observed after the first 6 months of life (Allan, 2008).

Diagnosis

- Radiography of the antebrachii (Allan, 2008).

Treatment

Surgical intervention (Allan, 2008).

Carpal valgus deformity

This is a condition of the thoracic limbs, where there is an outward rotation of the lower limb. It appears as a result of an improper diet, either an excess intake of vitamin or mineral supplementation, and/or a reduced amount of amino acid or protein was administered (Bell et al., 2010). This usually is present in hand-raised cheetah cubs that were given inadequate kitten milk replacers (Ziegler-Meeks, 2009).

Calcium and phosphorus imbalance

An improper balance of Ca:P ratio can lead to secondary alimentary hyperparathyroidism (“All meat syndrome”), which can have consequences such as osteochondrosis dissecans, enlarged joints, splayed feet, angular limb deformities and stunted growth (Ziegler-Meeks, 2009). Also a skewed calcium and phosphorus ratio in favour of phosphorous can lead to a depletion of copper, a mineral required for a number of body functions (Kaiser et al., 2014).

Copper deficiency

Copper deficiency can lead to lateral head tremors, ataxia, partial collapse, loss of balance, paralysis of the hind limbs and staggering gait. Cubs can even enter into respiratory distress and die. Affected cubs are treated with dietary copper. Pregnant females need to receive a diet containing an adequate copper proportion (Ziegler-Meeks, 2009).

Vitamin A

Too much vitamin A in the body can lead to skeletal malformations and fractures, internal haemorrhage, enteritis, conjunctivitis, and reduced function of liver and kidneys (Ziegler-Meeks, 2009).

- More on the background of nutritional requirements and results of improper diets can be found in chapter “3 Feeding” of the EAZA Best Practice Guidelines for Cheetah (*Acinonyx jubatus*).

1.2 Infectious diseases

The infectious diseases affecting cheetahs in captivity are categorised in five paragraphs; prions, viruses, bacteria, fungi and parasites.

1.2.1 Prions

Feline Spongiform Encephalopathy (FSE)

FSE is a neurodegenerative transmissible disease that affects humans and various mammalian species; cheetahs are one of them. Feeding transmissible spongiform encephalopathy-contaminated feed transmits this disease. Animals can also become infected through medical procedures and it cannot be ruled out that females can pass this

disease to their litter before or during parturition. This disease has a long incubation period, in cheetahs it is estimated to be between 4,5 to 8 years (Eiden, 2010; Iowa State University, 2016).

Clinical signs

Clinical signs observed in a cheetah kept at the zoological garden of Nuremberg were: Ataxia, progressive bilateral hindlimb lameness and tremor, dyskinetics and dyspositions of the head (Eiden, 2010). Other clinical signs observed in felines are: excessive salivation, decreased grooming, polyphagia, polydipsia, dilated pupils, somnolence and convulsions. Behavioural signs are: uncharacteristic aggression, timidity and hiding (Iowa State University, 2016).

Diagnosis

Only post mortem tests can be performed. Pathological findings are:

- Finding scrapie-associated fibrils (SAF).
- Immunoblotting.
- Histopathology.
- Immunohistochemistry (IHC).
- Protein misfolding cyclic amplification (PMCA).
- Quaking-induced conversion (QuIC) (Eiden, 2010; Iowa State University, 2016).

Treatment

No treatment is known to this day (Iowa State University, 2016).

Prognosis

After the first signs appear, the disease is progressive and fatal (Iowa State University, 2016).

1.2.2 Viruses

Canine Distemper Virus (CDV)

This virus is highly contagious; the main route of infection is through aerosol droplet secretions from infected animals or by contact with other bodily excretions and secretions. This virus affects species from the following families and orders: canidae, mustelidae, procyonidae, viveridae, ailuridae, ursidae, elephantidae, primates and felidae. However, not all felids develop the disease. Mortality for this virus has been reported in tigers, lions, leopards, jaguars, lynx and bobcats (Creevy, n.d.; Miller, 2015). In a study made in Namibia, antibodies of CDV were found in free-ranging cheetahs between the years 1995-1998. These results suggest that cheetahs are able to survive the disease (Munson et al., 2004a).

Clinical signs

As clinical signs for this virus are unknown in cheetahs, the clinical signs observed in dogs are described. Infection might be imperceptible or it can lead to severe signs. Dogs may exhibit fever, leukopenia, anorexia, nasal discharge, mucopurulent ocular discharge, lethargy, GI, respiratory signs, secondary bacterial infections, encephalomyelitis, hyperkeratosis of the foot pads, epithelium of the nasal planum, enamel hypoplasia in incompletely erupted teeth, neurologic signs and sometimes pustular dermatitis (Creevy, n.d.).

Neurologic signs are for example: circling, head tilt, nystagmus, paresis to paralysis and focal to generalized seizures (Creevy, n.d.).

Diagnosis

- Isolation of virus (IV).
- Reverse transcription polymerase chain reaction (qRT-PCR) of tissues.
- ELISA.
- Indirect fluorescent antibody (IFA) test.
- Immunofluorescence of conjunctival scrapings.
- Antibodies in cerebrospinal fluid (CSF) (Miller, 2015).

Treatment

Currently no recommendations can be made on how to treat cheetahs for this disease. However, the treatment used on CDV-positive dogs is aimed at limiting secondary bacterial invasions, supporting fluid balance and controlling neurologic manifestations. In order to do this, broad-spectrum antibiotics, balanced electrolyte solutions and parenteral nutrition is administered, in addition to antipyretics, analgesics and anticonvulsants (Creevy, n.d.).

Management

- Direct and indirect contact between cheetahs and potential carriers, like for example: dogs and raccoons should be avoided.
- This virus is very unstable outside the host and can be inactivated with most disinfectants (Creevy, n.d.).
- In a study at a German zoo, 2 cheetahs were vaccinated with an inactivated CDV vaccine, but the vaccines failed to produce measurable antibody titers. As a consequence of this, plus due to the low apparent disease risk, vaccination is not recommended (Ziegler-Meeks, 2009; Maack, 2000).

Prognosis

It is unknown if CDV has the potential to be as fatal in cheetahs as it is in lions and tigers (Munson et al., 2004a; Ramsay et al. 2016).

Feline Corona Virus (FCoV)

FCoV has two forms:

1. Feline enteric Corona Virus (FeCV), which infects the intestines.
2. Feline Infectious Peritonitis Virus (FIPV), this form is mutated and the fatal variant of FeCV, which causes the disease Feline Infectious Peritonitis (FIP) (Miller, 2015).

There are 2 serotypes of FCoV. Type I is the original FCoV and type II is thought to be the recombination between type I together with the Canine Corona Virus (CCV). Both types can cause FIP. The most common type to affect felines is type I (Benetka et al., 2004; Kipar et al., 2006).

This virus is highly contagious between cats that are in close contact. It is transmitted by the faecal-oral route through direct contact or by fomites (fomites are objects contaminated with infectious organisms that serve in their transmission). Asymptomatic cats can also pass the disease to other cheetah. After exposure most cats develop an immune response and

recover (Miller, 2015). But if the virus is not cleared, continuous infection is present and intermittent shedding may result. Intermittent shedding may occur due to reinfection, continuous shedding is the result of the animal having been unable to clear the virus (Kipar et al., 2006).

Clinical signs

1. FeCV: may be asymptomatic or result in mild diarrhoea that can become chronic, signs can last 2-5 days.
2. FIPV: fever, vomiting, diarrhoea and transudate effusions into body cavities with high protein content (Miller, 2015).

There are two types of FIPV variants, the “wet” form and the “dry” form. The “wet” form causes leakage of protein-rich fluid from the blood vessels into the abdominal cavity and vascular lesions surrounded by proliferation of inflammatory cells (Sharif et al., 2010). Other signs for this form are dyspnoea, mild pyrexia, muffled heart sounds, uveitis, keratic precipitations, changed colour of the iris, effusions, antibiotic non-responsive fever, anorexia, depression, lethargy and weight loss (Tschurlovits, n.d.). The “dry” form causes pyogranulomatous or granulomatous lesions in multiple tissues (Sharif et al., 2010). Further symptoms are vague with a slow onset and depend on the organs affected (Tschurlovits, n.d.).

Diagnosis

To detect a cheetah infected and actively shedding detectable FCoV, RT-nPCR testing should be done on five individual consecutive faecal samples in a time period of 30 days. This can correctly identify 90% of cheetahs with detectable FCoV in faeces. In order to have a more reliable result this examination should be combined with serology for FCoV, even though the results will still not be 100% reliable (Gaffney et al., 2012). Once an individual has a positive result RT-nPCR testing should be retested 3 times each month for 6 months (Tschurlovits, n.d.). To detect FIPV the previous tests should be made and confirmed by Immunohistochemical or Immunofluorescent staining. The current best procedure available to confirm the presence of FIPV is to perform a histopathology on animals with effusions (Gaffney et al., 2012).

Diagnostic Methods

- The medical history of the animal should be revised and the present clinical signs.
- Antigen detection
 - Reverse transcriptase polymerase chain reaction (RT-nPCR). Testing should be done on five individual consecutive faecal samples in a time period of 30 days.
 - Electron microscopy.
 - Immunohistochemical staining (Gaffney et al., 2012).
- Serology
 - Antibody titres: Titers >1:1600-3200 are suggestive of FIP. This testing method is a screening tool to detect presence or absence of the virus. And it can aid with the detection of FIP (Miller, 2015). Some individuals presenting the “wet” form may have low titers or no antibodies against FCoV. This is because large amounts of virus inside the cats’ body render them unavailable or because the antibodies are lost in effusions (Sharif et al., 2010).

- Competitive ELISA: This test only predicts correctly positive results 67% of the time and there are many false positives, therefore this test method is not very reliable (Sharif et al., 2010).
- Effusions
 - Anti-FCoV antibodies.
 - Rivalta's Test.
 - Immunofluorescent staining of FCoV Antigen in Macrophages is performed in effusions (FCoV).
- Haematology and blood chemistry.
- Radiographs: can reveal pleural, pericardial, or peritoneal effusion and hepatomegaly or renomegaly (Sharif et al., 2010).
- Ultrasonography: can be used to confirm the presence of abdominal fluid in cats with minimal fluid volumes and to evaluate the pancreas, liver, lymph nodes and kidneys (Sharif et al., 2010).
- Magnetic resonance imaging (MRI): can reveal periventricular contrast enhancement, ventricular dilatation, and hydrocephalus in FIP affected cats (Sharif et al., 2010).
- Histopathology is the gold standard for FIP diagnosis (Miller, 2015). Here effusions or yellow to white foci or nodules can be found in different organs. Additionally histologic lesions are present. In the "wet" form lesions consist of an arteriole or venule with central necrosis and perivascular infiltration with macrophages, neutrophils, lymphocytes and plasma cells. The "dry" form has focal accumulations of inflammatory cells and fibrinous necrotic-proliferative lesions (Tschurlovits, n.d.).

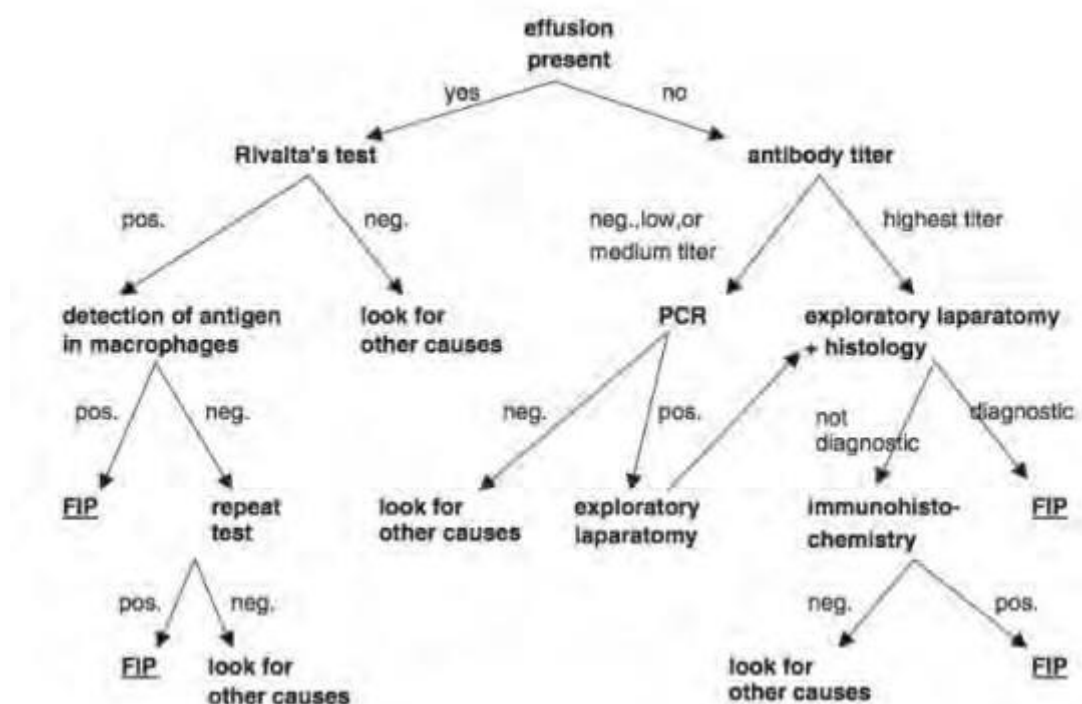


Figure 1: Algorithm for the diagnosis of FIP in domestic cats (Hartmann, 2005)

Management

Animals that recover will remain carriers. Prevention can be accomplished by limiting the exposure to infected cats and their faeces. Detergents and disinfectants can inactivate the virus, but it can survive up to 2 months in a dry environment (Miller, 2015). Currently there is no effective vaccination available, or a curative treatment for FIP (Sharif et al., 2010). The

population should be managed as endemically infected with FCoV. And individuals positive for this virus should not automatically be excluded from exchange between institutions. However, persistent shedders should be kept as exhibit animals only (Tschurlovits, n.d.).

Treatment

(These are the treatments administered to domestic cats)

1. Healthy but FCoV positive: There is no specific treatment for these cases. In order to help the animals' stressful situations should be avoided.
2. FeCV enteritis: It is self-limiting in domestic cats. Supportive care should be supplied in the form of fluid therapy and diet.
3. FIP: An effective antiviral treatment against FIP has not been found yet, but high doses of immunosuppressive and anti-inflammatory drugs may slow down the progression of the disease (Tschurlovits, n.d.).

Prognosis

Once FIPV has developed the course of this disease results in death (Hartman et al., 2003).

Feline Herpes Virus (FHV), Rhinotracheitis

FHV 1 is a highly contagious Alphaherpesvirus, shed in saliva and ocular and nasal secretions. Transmission of the virus occurs through direct animal-to-animal contact, other transmission sources are animal handlers, feed containers and flies. This virus is often self-limiting and may resolve on its own in 14-28 days. It can have co-infections with Calicivirus, *Chlamydophila psittaci* and *Mycoplasma ssp.* Vaccination does not protect against infection, but the administration of a dead vaccine has shown to reduce the severity of clinical signs, reduce the viral shedding and prevent recrudescence (Flacke, 2005; Miller, 2015; Vuuren, 1999).

Clinical signs

FHV 1 affects the ocular and upper respiratory system. Clinical signs are: Corneal ulceration, ulcerative rhinitis and conjunctivitis, ocular and nasal discharge, wheezing, sneezing, anorexia, salivation, glossitis, pneumonia, abortion, keratitis and general infection (Flacke 2015; Hargis, 1998; Vuuren, 1999).

Ulcerative dermatitis is less common, but can also develop and if it does, it is difficult to heal. Lesions occur in periocular, perionasal and perioral areas, occasionally they can be found on the extremities. The affected areas exhibit ulceration, crusting with purulent discharge, necrosis and prominent eosinophilic inflammation (Flacke, 2015).

For most cubs that contract the virus after the first month of life, the initial signs (sneezing and watery eyes) will be self-limiting. On the other hand neonatal cubs develop the worst and most persistent lesions and may die from acute infection. Lesions observed in neonatal cubs can be corneal ulcers or scars, chronic keratitis, blindness, prolapsed third eyelids, chronic epiphora and ulcerative dermatitis. Most cubs contract the infection from their mothers (Ziegler-Meeks, 2009).

All infected animals become latent carriers and may have a recrudescence later in life. Additionally they will intermittently shed infectious viral particles. Reactivation occurs if the

animal has to endure stressful situations such as for example a change of housing, parturition and lactation (Flackes, 2015; Hargis, 1998; Ziegler-Meeks, 2009).

Diagnosis

A presumptive diagnosis can be made based on the exhibited clinical signs; this diagnosis is then confirmed with the help of different methods. Current methods available to diagnose FHV are:

- Taking swabs of the conjunctiva, nasal or oropharyngeal regions for viral isolation (VI) (Miller, 2015).
- Polymerase chain reaction. However, PCR has resulted in many false positives, therefore if used the results should be confirmed with the help of another method (Persico, 2011).
- Immunohistochemistry (IHC) is the gold standard for FHV diagnosis (Persico, 2011).
- Histopathology (Persico, 2011).
- Fluorescent antibody (FA) (Miller, 2015).
- Electron microscopy (Hargis, 1999).

Management

- Cheetahs should be kept isolated a minimum of 7 days after all symptoms have cleared (including lesions). The virus is viable in the environment for less than 72 hours after the animal has been removed from the enclosure.
- Females should be vaccinated before breeding and again 2-3 weeks before parturition. During the pregnancy and the development of the young cubs, they should be kept isolated as much as possible from other animals to prevent transmission.
- Early detection in cubs can minimize severity of lesions. Therefore, a visual check-up should be made, as soon as the behaviour of the female allows it. Keeping in mind that cubs will not develop herpesvirus signs until a minimum of 3 to 4 days of age. If lesions are present, keep an eye on it and if severe lesions develop, the cubs might have to be removed from the female. It has been documented that as soon as the cubs are removed from the female the lesions begin to recede.
- If a cheetah has this disease it should be treated with L-lysine for several weeks before and after shipment. However if animals are displaying active lesion before shipment, the animals should not be transported.
- If a cheetah is affected, isolation measurements should be implemented, as the virus is difficult to contain (Infected animals should be physically separated from the rest. Additionally, keepers should use maximum hygiene, separate tools and come in contact with the infected animals after having finished all the other tasks with the healthy animals).
- Captive cheetah populations should be managed as infected and efforts should be made to minimize disease and disease transmission between conspecifics and other susceptible species (Ziegler-Meeks, 2009).

Treatment

- L-lysine can be administered as a dietary supplement. For cubs a dosage of 120 mg/day orally is recommended. Whereas for a full-grown cheetah a dosage of 2,500

mg/day is sufficient. It may reduce lesions and/or shedding of the virus (Ziegler- Meeks, 2009).

- Antibiotics such as amoxicillin or doxycycline are used to prevent secondary infections. After two weeks of treated symptoms should vanish (Tschurlovits, n.d.).
- An ophthalmic preparation of cidovovir has been proven effective in domestic cats. Here cidovovir is compounded in 0.5% of carboxymethyl cellulose and administered twice a day. Idoxuridine has also been shown to be useful. However, trifluridine and viroptic seem to be painful and are therefore not recommended (Ziegler-Meeks, 2009).
- Skin lesions can be treated with cryotherapy (Ziegler-Meeks, 2009).
- Tetracycline can be applied to lesions (Munson, 2004b).
- Famciclovir is recommended for the treatment of this illness (Ziegler-Meeks, 2009).
- Interferon omega has also showed to be helpful (Flacke, 2015).
- On the other hand guanine analogue antiviral drugs such as aciclovir have not been proven to be helpful and carry the risk of bone marrow suppression (Ziegler-Meeks, 2009).

Prognosis

It is a self-limiting disease, which can become chronic. Many individuals experience recrudescence later in life (Tschurlovits, n.d.).

Feline Immunodeficiency Virus (FIV)

FIV is a lentovirus, which causes an acquired immunodeficiency syndrome in domestic cats. It is transmitted through saliva, sexual contact and from mother to offspring. With this said, the primary form of transmission is through bites (Miller, 2015; Troyer et al., 2008). Cheetahs seem to become infected very rarely with FIV. Wild cats show little evidence for immune suppression or mortality. It appears that FIV evolved independently within its host species, but there may be also cross-species transfers in unusual situations (Grisham, 1997; Miller, 2015; Troyer et al., 2008).

Clinical signs

Animals are usually asymptomatic. If not, they can show oral cavity disease, chronic infections of the upper respiratory tract, anaemia, skin infections, weight loss, vomiting, diarrhoea or neurologic disease (Grisham, 1997; Miller, 2015).

Diagnosis

The following methods can be used to confirm the presence of the disease:

- ELISA.
- Western blot is the confirmatory test, as it is more sensitive.
- Isolation of virus (IV) (Grisham, 1997; Miller, 2015).

Treatment

Different treatments have been tested on domestic cats, therefore in the unlikely event that a cheetah is seropositive and shows some of the symptoms specified in “clinical signs”, research papers on FIV infections in cats can be searched for treatments.

Management

- Routine testing is recommended.
- This virus is labile outside of the host and is inactivated by common disinfectants (Miller, 2015).
- If an institution possesses a FIV-positive animal the EEP coordinator must be informed of the animal's status (for reproductive and transportation purposes) (Grisham, 1997).

Prognosis

Wild cats show little evidence for immune suppression or mortality (Grisham, 1997).

Feline Leukemia virus (FeLV)

Feline leukaemia virus is normally transmitted through saliva or blood, via grooming, sharing of water and food or fighting. Further the virus can be found on tears, urine, semen, vaginal fluids and faeces. This virus can be transmitted from the mother to her cubs either during pregnancy, birth or while nursing. Cheetahs are infected by the direct, or indirect contact with seropositive FeLV cheetahs and domestic or feral cats. Felines are mostly able to clear the virus, yet additional stressors like for example due to enclosure relocation or the administration of immunosuppressive drugs can help the development of the disease (Hartmann, 2011; Marker et al. 2003; Miller, 2015).

Clinical signs

Animals are usually asymptomatic and some are able to clear the virus. However, if the virus becomes persistent, signs such as immunosuppression, anaemia, anorexia, depression, chronic, inflammatory conditions, enlarged lymph nodes secondary infections, persistent fever, neutropenia, lymphoid or myeloid tumour, or reproductive problems are visible. Usually persistently viremic cats develop fatal disease. A FeLV positive pregnant female cat normally suffers from fetal resorption, abortion and neonatal death. If the kittens are born and infected, they normally suffer from “fading kitten syndrome”, which ends with an early death (Hartmann, 2011; Miller, 2015).

Diagnosis

The following methods can be used:

- Immunofluorescent antibody (IFA)
- ELISA

In both, false-positive and false-negative results can occur, therefore the disease needs to be confirmed with:

- Virus isolation (VI) or
- Polymerase chain reaction (PCR) on blood, bone marrow, and tissues (Miller, 2015).

Other used methods are: Histology and immunohistochemistry (Tschurlovits, n.d.).

Treatment

The following treatments are used on domestic cats:

- Interferon.
- Chemotherapy.
- Supportive therapy. The cats are administered hematologic agents such as vitamin B12, folic acid, anabolic steroids, erythropoietin and blood transfusions.

- Immunosuppressive therapy, this may be able to help with autoagglutinating hemolytic anaemia, but may activate virus replication (Tschurlovits, n.d.).

Management

- Routine testing is recommended (Miller, 2015).
- Vaccination is available in North America and can therefore be used as a prevention measurement. However if there are no seropositive cases inside of the institution, and direct and indirect contact between cheetahs and domestic or feral cats can be avoided, then vaccination might be superfluous (Miller, 2015).
- This virus is unstable outside the host, nevertheless it is able to survive for up to a week in dried biologic deposits. It can be inactivated with detergents and common disinfectants (Miller, 2015).

Prognosis

Some cats are able to clear the virus, but others may remain persistently viremic. The last mentioned succumb to the disease (Miller, 2015).

Feline Panleukopenia Virus (FPV), Parvovirus

FPV is a common cat disease related to Mink Enteritis Virus (MEV) and Canine ParvoVirus (CPV). It is very contagious and is transmitted by direct or indirect contact of affected individuals, their secretions and excretions. The virus can be shed in faeces up to 6 weeks after recovery. The illness usually lasts between 5-7 days, it can become fatal in less than 24 hours and mortality in cubs under 5 months of age is high. This virus is very resistant and can survive for longer than a year in a suitable environment (Lane et al., 2016; Miller, 2015).

CPV mutated into additional strains, and even though the original form does not infect felids, the new strains (CPV types 2a, 2b and 2c) do. Due to vaccination, parvovirus outbreaks are less common. As inactivated vaccines induce high persistent levels of serum antibodies to FPV in cheetahs. However, cases of CPV2 have increased, because vaccination is focused on FPV strains (Lane et al., 2016).

Clinical signs

The illness can be subclinical. Per acute cases may show the following signs: fever, depression, anorexia, dehydration, vomiting and diarrhoea (Miller, 2015).

Diagnosis

An initial diagnosis can be made based on the exhibited clinical signs; this diagnosis is then confirmed with the help of the following methods:

- Polymerase chain reaction (PCR) on faeces (Lane et al., 2016).
- Electron microscopy on faeces (Lane et al., 2016).
- Elisa. For example SNAP a Canine Parvovirus Antigen Test Kit is able to detect FPV antigen during the acute phase (Abd-Eldaim, 2009).
- Immunofluorescent antibody staining (IFA), detecting antigens in cryostat tissue sections (Krueger et al., 2004).
- Virus isolation (Ziegler-Meeks, 2009).
- Pathology (Lane et al., 2016).

Treatment

Affected animals can be treated with antibiotics to prevent secondary bacterial infection. Nutritional support can alleviate symptoms and fluid therapy should be used to replace fluid loss as a result of diarrhoea (Tschurlovits, n.d.).

Management

- Cheetahs need to be vaccinated every year with an inactivated vaccine. Additionally, pregnant females should receive a booster near the end of the term (Miller, 2015).
- The virus is resistant to inactivation and can survive more than a year in a suitable environment. 6% Household bleach is known to inactivate the virus (Miller, 2015).

Prognosis

High mortality in cubs under 5 months (Miller, 2015).

Rabies virus

All mammalian species are susceptible to this virus and it can be transmitted to humans. Transmission usually occurs through bites from infected animals, also through contact of saliva with mucous membranes or open wounds. The incubation period last from 3 weeks to several months. The virus is not stable in the environment and is inactivated by common disinfectants.

Clinical signs

Signs observed are salivation, abnormal behaviour, auto-mutilation, neurologic signs such as paresis and seizures, and sudden death. Hyper aggression and biting can sometimes also be observed (Miller, 2015; Kaandorp, 2010).

Diagnosis

Diagnosis is done post mortem with brain tissues and serum (Kaandorp, 2010). This is the only way to confirm the disease.

Treatment

There is no treatment. Infected animals should be euthanized (Kaandorp, 2010).

Management

- Vaccination with a killed vaccine is recommended in endemic areas. The first course of vaccinations is given at 6 months and at 12 months of age. Afterwards an annual booster or a booster every three years (depending on the registration of the vaccine used) should be administered (Tschurlovits, n.d.).
- Exposure to wild carnivores and bats should be avoided.
- If an animal has been infected, it has to be reported.
- Euthanasia is recommended and the head should be shipped to a qualified laboratory for testing (Miller, 2015).

Prognosis

It is a fatal disease and individuals infected will die within 2 to 7 days after the onset of clinical signs (Miller, 2015).

1.2.3 Bacteria

Anthrax

Cheetahs are susceptible to Anthrax. Anthrax is not endemic in Europe, however it remains common in countries near the Mediterranean Sea (Turkey, Greece, Balkan countries, Italy and Spain) (Schmid, 2002). It is endemic to Namibia, Botswana and Zimbabwe. Therefore, control measures such as vaccinations should be taken if necessary in such locations (Turnbull et al., 2004). Other susceptible animal groups affected are ruminants, primates and occasionally other species, including humans. It can be transmitted percutaneously, perorally (when soil is contaminated by spores) and aerogenously (Kaandorp, 2010). Additionally transmission can also happen through the ingestion of contaminated meat (Jäger et al., 1990). Incubation period is between 3-5 days (Kaandorp, 2010).

Clinical signs

In chimpanzees clinical signs are sudden weakness, vomiting and death within hours. Humans can display cutaneous ulcers, intestinal anthrax and pulmonary anthrax (Kaandorp, 2010). Cheetahs exhibit an increased respiratory rate, vomiting, apathy and death (Jäger et al., 1990).

Diagnosis

- Blood smears
- Cultivation
- Ascoli-reaction
- PCR
- Fluorescence assays using bacteria cell wall and capsule antigens (Kaandorp, 2010).
- Histopathology, cheetahs affected by anthrax have lung edema and blood tinged hydrothorax (Jäger et al., 1990).

Treatment

Antibiotics such as penicillin and tetracycline can be used. And chloroquine can be employed for antitoxic treatment (Kaandorp, 2010).

Management

- (In endemic areas) Vaccination: 0.3 ml spore vaccine in *M. mulatta* (Kaandorp, 2010). Two months after the first vaccination cheetahs should receive a boost. Afterwards they should receive a yearly booster, as more than one dose is needed to induce a substantial protective immunity (Turnbull et al., 2004).
- It can be inactivated with heat, 120 degrees for the duration of 3 minutes is needed (Kaandorp, 2010).
- If an animal is found dead with blood draining from any orifices and there is bloating in the abdomen in ruminants or swelling of the head in carnivores, the body should not be opened. First special precautions need to be taken in order to not further spread the bacteria.

Prognosis

If not treated it becomes fatal (Tschurlovits, n.d.).

Bartonella

This genus currently consists of 16 species, where 7 of them are associated with human diseases. For example: “cat-scratch disease”. Here transmission from cats to humans occurs through scratches and bites (Molia, et al. 2004). Bartonella species cause long-lasting bacteraemia in hosts. Cats can be infected for weeks to months (Chomel et al., 2006).

Clinical signs

Mainly felids are asymptomatic carriers (Tschurlovits, n.d.). However, there are cases of uveitis, endocarditis, kidney disease, urinary tract infections, stomatitis and lymphadenopathy observed in infected felids.

Diagnosis

- TaqMan PCR (Molia, et al. 2004).

Treatment

In domestic cats this disease can be treated with antibiotic therapy (Perez et al., 2010).

Clostridium perfringens

This bacterium affects animals and humans.

Clinical signs

Chronic, intermittent to continuous, bloody and/or mucoid diarrhoea can be observed. Also stool with blood and mucus drops. Yellowish stool or tenesmus can also be found. There is no evidence of systemic illness or weight loss (Citino, 1995).

Diagnosis

- Histopathology: Endoscopic colon biopsy
- Faecal smears

Treatment:

This bacterium can be treated with tylosin, metronidazole and psyllium fiber (Citino, 1995).

Ehrlichia

Ticks transmit most Ehrlichia species in endemic areas (Southern Europe). In cats the disease can develop chronically over the years (Tarello, 2008).

Clinical signs

General signs are poor appetite, lethargy, fever, wasting, vomiting, diarrhoea, polydipsia, dehydration and respiratory symptoms such as polypnea, wheezing and sneezing. Medical signs observed in a group of cheetahs were: deterioration of health, decreased food intake, weight-loss and death (Tarello, 2008).

Diagnosis

- Clinical signs.
- Cytological demonstration (Blood smear).
- Inclusion of bodies in leukocytes.
- PCR (Tarello, 2008).

Treatment

Doxycycline, enrofloxacin and imidocarb (Tarello, 2008).

Prognosis:

If left untreated it can lead to fatal consequences (Tarello, 2008).

Haemobartonella felis

Haemobartonella felis or heamotrophic mycoplasma can cause disease as a primary pathogen or through an opportunistic infection. The latter affects animals that suffer from Immunosuppressive viruses like for example: Feline Immunodeficiency Virus (FIV), Geline Leukaemia Virus (feLV), or Canine Distemper Virus (CDV). This bacterium is mainly transmitted through bite wounds, fleas and other bloodsucking arthropods. Kittens can become infected through intrauterine transmission, parturition or lactation (Haefner, 2013; Krengel et al., 2013; Torkan et al., 2013)

Clinical signs

The following are clinical signs observed in domestic cats: Tachypnea, depression, weakness, anorexia, weight loss, pale mucous membranes, dehydration, icterus, splenomegaly and death (Krengel et al., 2013, Torkan et al., 2013). A cheetah was discovered to have heamotrophic mycoplasma, however no symptoms were observed and the individual seemed not to be impaired by the infection (Krengel, 2013).

Diagnosis

- Blood smear (Giemsa staining).
- Complete blood count.
- Immunofluorescence.
- Western Immunoblot analysis.
- PCR assay (preferred method)(Haefner, 2013).

Treatment

It is not clear if non-domestic cats need treatment (Haefner, 2013). In cats however, positive results were observed through the administration of oxytetracycline together with dexamethason (Fathi, 2010). Enrofloxacin together with prednisolone and fluid therapy (based on the dehydration status of the cat) has also been proven to be a successful treatment (Saqib et al., 2016).

Salmonellosis

Transmission of this bacterium occurs perorally. It has zoonotic potential. And incubation period can be between 8 to 48 hours (Kaandorp, 2010). Malnutrition could immunosuppress carriers, leading to infection (Venter et al., 2003).

Clinical signs

Diarrhoea with or without fever, sometimes it can lead to vomiting, abortion or osteomyelitis too. Asymptomatic carrier stages can develop (Kaandorp, 2010).

Diagnosis

- Enzyme- immunoassays and immunofluorescence.

- Slide- or tube agglutination.
- Enrichment media: MacConkey agar, S-S agar, Drigalski agar, etc.
- Planting media: Tetrathionate medium and selenite F-broth, etc. (Kaandorp, 2010).
- PCR (Venter et al., 2003).

Treatment

Revision of diet, elimination of parasites, supportive treatment (volume and electrolyte substitution). If needed antibiotic treatment after antibiogram (Kaandorp, 2010).

Management

- Strict hygiene.
- Rodent control.
- Exclusion of raw chicken meat from the diet.
- No contact with infected animals (separation) (Kaandorp, 2010).
-

Prognosis

Not often it can become fatal (Kaandorp, 2010).

Campylobacteriosis

Transmission of this bacterium occurs perorally. It has zoonotic potential. The incubation period is between 2 to 10 days (Kaandorp, 2010).

Clinical signs

It is mostly asymptomatic, if symptoms occur secretory diarrhoea can be observed. In severe infections mucohaemorrhagic diarrhoea can occur (Kaandorp, 2010).

Diagnosis

- Cultivation.
- ELISA-test.
- PCR (Kaandorp, 2010).

Treatment

This bacterium is usually self-limiting; volume and electrolyte substitution can be given for support. It can be treated with antibiotics such as erythromycin and tetracycline; some strains have become resistant to quinolones, aminoglycosides and macrolides (Kaandorp, 2010).

Management

- Strict hygiene (Kaandorp, 2010).

Prognosis:

Not fatal (Kaandorp, 2010).

Tuberculosis

Nearly all of the mycobacteria from the “tuberculosis complex” can infect wild species and it has zoonotic potential. The course of disease and symptoms will vary among species (Kaandorp, 2010). Transmission can occur aerogenously or perorally (Kaandorp, 2010). At

Kruger National Park (KNP) tuberculosis caused by the mycobacterium bovis was diagnosed in a cheetah. Here tuberculous granulomatous lesions in the lungs were observed (Keet et al., 1997).

Clinical signs

Signs are rarely seen before death occurs, if cough and dyspnoea appear, the animal has already entered a late and irreversible stage of the disease. Chronic weight loss can also be observed (Kaandorp, 2010).

Diagnosis

- Gold standard: culture (takes a long time) (Kaandorp, 2010).
- Microscopic examination (Kaandorp, 2010).
- Mycobacterium DNA amplification (Kaandorp, 2010).
- Pathology (Kaandorp, C., pers. comm., 2017).

Treatment

A choice needs to be taken with veterinary officials and all related actors (TAG, EEP) to either euthanize the animal or treat it. Treating implies following strict rules of drug administration, pharmacokinetic check, observance and excretion follow-up to decrease emergence of resistant strain. A successful treatment leads to an animal with a latent stage of this disease and reactivation is always possible, therefore the affected animal needs to be strictly monitored for the rest of its life (Kaandorp, 2010).

Management

- Strict quarantine and testing programs (Kaandorp, 2010).
- Strict hygienic protocols need to be followed due to its zoonotic potential (Kaandorp, C., pers. comm., 2017).
- Because of its infectious character, euthanasia should be taken into consideration.

Prognosis

If clinical signs are observed, death quickly follows (Kaandorp, 2010).

1.2.4 Fungi

Dermatophytes

Infection occurs through direct contact with the spores or hyphae of any of the three genera *Microsporum*, *Trichophyton* and *Epidermophyton*, and it can be transmitted to people. Dermatophytosis occurs more frequent in countries with hot and humid climates (Bentubo et al., 2006).

Clinical signs

The animal may not manifest any clinical symptoms (Tschurlovits, n.d.). It can also display mild and superficial lesions with circular frames, desquamation, alopecia and erythema of the edges (Bentubo et al., 2006). Or it can display a highly inflammatory reaction with scarring, hair loss and formation of granulomas (Tschurlovits, n.d.).

Diagnosis

Collection of samples

- Carpet technique (Bentubo et al., 2006).
- Collection material from ear canal (Albano, 2013).

Diagnosis methods

- Wood's lamp.
- Light microscopy (50% of false negative results).
- Culture.
- Serology.
- PCR (Tschurlovits, n.d.).

Treatment

Terbinafine or itraconazole are successful oral therapies administered to cats (Nuttall et al., 2008). Also topical antifungal products like lime sulphur and enilconazole solutions can be used (White-Weithers, 1995).

Systematic candidiasis

Systematic candidiasis was found on a geriatric captive cheetah during post-mortem examination. The cheetah had a clinical history of intermittent chronic gastritis and chronic renal failure, which were treated with broad-spectrum antimicrobial therapy (La Perle et al., 2000). Candidiasis was also found in cats and dogs. All the individuals affected suffered from a compromised immune system, which likely made them more predisposed to this type of infection (Pressler, 2003).

Diagnosis

- PCR of blood samples (Avni, 2010).
- Fluorescent antibody testing of yeast, pseudohyphae and hyphae in affected tissue (La Perle et al., 2000).

Cryptococcus neoformans

Cryptococcus neoformans is a non-contagious and opportunistic organism that affects animals and humans. It is life threatening and usually develops in immunocompromised hosts. Initial infection happens through inhalation, afterwards it disseminates from the nasal cavity or lungs to the skin, eyes, central nervous system and other organs where granulomas form. Adult male cheetahs are more susceptible (da Silva, 2017; Millward, 2005).

1.2.5 Parasites

Toxoplasmosis

All mammalian (zoonotic potential) and avian species are susceptible to this parasite. However, felids are the definitive hosts. Low or elevated temperatures destroy the parasite, but it cannot be destroyed by disinfectants. It can be transmitted through three different ways: either transplacental infection, infection through ingestion of feline contaminated faecal matter or after ingestion of affected tissues (Kaandorp, 2010).

Clinical signs

Lymphadenopathy, headache, and muscle ache are common signs, however affliction can occur to any organ, so clinical signs may vary. Other signs may be retinitis, uveitis, central nervous system (CNS) involvement, pneumonia, respiratory insufficiency, neurologic signs and incoordination. In sheep and goats abortion is common and acute death can occur to highly susceptible species (Kaandorp, 2010).

Diagnosis

It is difficult to diagnose on signs alone. But it should be contemplated if major organ systems are affected (lung, liver, CNS) and domestic or wild cats are in the area.

Detection of antibodies in serum through:

- Methylene blue dye binding (MBD).
- Indirect immunofluorescent antibody (IFA).
- Indirect haemagglutination.
- Enzyme linked immunosorbent assay (ELISA).
- Direct and modified agglutination (DAT, MAT).
- Latex agglutination (LA) (Kaandorp, 2010).

Identification of organism through:

- Biopsy, Necropsy.

Treatment

There is no treatment to effectively eliminate the infection. In small domestic animals clindamycin (25-50 mg/kg for 14-21 days) is used (Dubey, 2016). Other treatment is the combination of sulfonamides (30-60 mg/kg PO q12 hours) with pyrimethamine (0.25-0.5 mg/kg PO q12 hours) during therapy folinic acid must be used. Other less effective drugs, but with fewer side effects are chloramphenicol, tetracycline and doxycycline (Kaandorp, 2010).

Management

- Meat fed to cheetahs should be frozen for at least 3 days at -12°C to kill *Toxoplasma* cysts.
- Hands and cutting material should be washed with soap after handling uncooked meat.
- The entrance of feral/domestic cats and rodents in the enclosures and in the feed storage areas should be prevented.
- Daily removal of faeces can reduce transmission (Kaandorp, 2010).

Prognosis

Although infection is common, clinical disease is rare in cheetahs (Spencer, 2003).

Dirofilaria immitis

Dirofilaria immitis is a parasitic nematode that leads to cardiopulmonary dirofilariasis in both domestic and wild hosts, and has a zoonotic potential. It is transmitted through the bite of an infected mosquito. Initially the pulmonary vasculature and the lung becomes affected, this is then followed by the affliction of the right chamber of the heart (Morchón et al.,

2012). Leading to feline heartworm (HW) disease. In felids, the parasites seldom develop into their adult stage (Otranto et al., 2015). It takes the parasites around six months from when they enter the feline body until they mature and are able to reproduce. Eight months after the initial infection they produce microfilaria that stay in the cat's bloodstream for a month (Cats are resistant and therefore only very few microfilaria are found). It will take around two years for the infection to be eliminated from the cat (Ward, 2008a).

Clinical signs

There are no specific clinical signs. Some observed are coughing and rapid breathing, weight loss and vomiting, an apparent healthy cat can suddenly be found dead, or develop a sudden overwhelming respiratory failure (Ward, 2008a).

Diagnosis

A combination of the following tests is needed to diagnose heartworms:

- Heartworm antibody test.
- Heartworm antigen test.
- Blood sample/blood smear tested for presence of microfilariae.
- Eosinophil count.
- Radiographs.
- Cardiac ultrasound/Echocardiography (Ward, 2008a).

Treatment

Currently there is no drug for treating heartworms in domestic cats. The drug used for dogs has serious side effects in cats, as dying worms can lead to sudden death of the cat. Other possible treatments are to surgically remove the heartworms and to treat the symptoms hoping that the cat outlives the worms (Ward, 2008a).

Management

In domestic cats, a year-round preventative is given in areas where mosquitoes are present all year round (Ward, 2008a). Therefore in places where *dirofilaria immitis* is present, cheetahs can receive as a preventative ivermectin, moxidectin, or milbemycin at standard feline doses e.g. 0,1-0,2 mg/kg monthly (Tschurlovits, n.d.).

Prognosis

It is a severe and life-threatening disease (Morchón et al., 2012).

Toxascaris spp. or Toxocara spp.

Are roundworms (intestinal parasites), which are often found in felids and can lead to death in kittens. They are large worms (8-15 cm) that are found in the intestines. They can be transmitted during nursing, through the ingestion of faeces from infected cats and ingestion of affected paratenic hosts (Ward, 2008b).

Clinical signs

Not very pathogenic in adult cats, however they can lead to life-threatening problems in kittens and older cats when present in large numbers (Ward, 2008b). Captivity imposes a stressful environment for felids, leading to a bigger probability of infestation (Ramesh et al.,

2009). Common signs of infestation are: pot-bellied appearance, abdominal discomfort, depressed appetite, vomiting, diarrhoea and poor growth (Ward, 2008b).

Diagnosis

- Microscopic detection of eggs present in faeces (Ward, 2008b).

Treatment

Anthelmintic or deworming medication (Ward, 2008b).

Management

- Breeding females should be dewormed prior to mating and again during late pregnancy (Ward, 2008b).
- Cubs should be dewormed (Ward, 2008b).
- A prophylactic protocol should be established in the institution. Faecal sampling and deworming should be done regularly (Kaandorp, C., pers. comm., 2017).
- Faeces of animals with roundworms need to be properly removed, as eggs remain viable in the environment for long periods of time (Ward, 2008b).

Prognosis

If medication is given promptly the infection can be fought off, however extremely debilitated kittens may die (Ward, 2008b).

Ollulanus tricuspis

Are worms that live in the stomach of cats, they are small (females between 0.8 to 1mm long, males between 0.7 to 0.8 mm long) and difficult to see with the naked eye. The infection is transmitted when an animal ingests the vomitus of an infected individual. The parasites stay viable for up to 12 days in the vomitus. They have no paratenic or intermediated hosts (Collett, 2000).

Clinical signs

This infection is associated with gastritis, vomiting, anorexia and diarrhoea (Collett, 2000).

Diagnosis

- Examination of vomitus, gastric washings or gastric mucosal scrapings (dissecting microscope).
- Gastric endoscopic biopsies.
- Examination gastric content and washings by repeated dilution and sedimentation.
- Examination vomitus with Baermann's funnel technique.
- Post-mortem: peptic digestion of samples of stomach wall. (Collett, 2000)

Treatment

- In domestic cats tetramisole 2.5% at 5 mg/kg has been found effective. On the other hand morantel (0.5 g/cat) and dichlorvos (10.3 mg/kg) were found ineffective. However there are some who found dichlorvos at 11mg/kg to be effective, when applied during 2 treatments with an interval of 1 month between them.
- Fenbendazole was described as 99% effective at 10 mg/kg as a 10% oral suspension.

- A severely infected tigress was administered levamisole (20 mg/kg) to control her infection.
- Oxfendazole (10 mg/kg) administered twice daily for 5 consecutive days was found to reduce, but does not eliminate the infection (Collett, 2000).

Further research needs to be conducted to find an effective treatment.

Management

Infection by *Ollulanus* should be suspected if any felid displays the following signs: vomiting, loss of condition with or without diarrhoea (Collett, 2000).

***Aelurostrongylus* spp.**

Aelurostrongylus abstrusus is a lungworm parasite whose definitive host is the cat. Cats become infected through the ingestion of intermediate or paratenic hosts, such as rodents, birds, etc. Then larvae migrate to the lungs where they reach sexual maturity approximately 4 weeks post infection. After mating, females produce eggs, and once hatched the larvae are released to the environment via faeces, where they will infect an intermediate host (Taubert et al., 2009). This parasite has no zoonotic potential (AAVP, 2014).

Clinical signs

The symptoms can range from minimal respiratory signs and cough in mild infections, to interstitial bronchopneumonia, open-mouthed abdominal breathing, intense coughing, sneezing, muco-purulent discharge, dyspnoea, and hydrothorax in heavy infections (Taubert et al., 2009).

Diagnosis

- Examination fecal samples with Baermann's funnel technique (Taubert et al., 2009).

Treatment

- Cats have been treated with fenbandazole with varying results. Fenbandazole (55 mg/kg) was administered on a daily basis for 21 days with successful results. Fenbandazole (20 mg/kg) was also administered for 5 days, followed by a second 5-day treatment after a 5-day hiatus and had favourable results. However 15 cats were treated with fenbandazole (50mg/kg) on a daily basis for three days, and even though in the beginning it showed promising results, 14 days after treatment larvae reappeared in faeces (AAVP, 2014).
- A cat was administered ivermectin (200 µg/kg), followed by a second treatment of ivermectin (400 µg/kg) and the infection was eliminated (Kirkpatrick, 1987).

Prognosis

Occasionally it can lead to a fatal respiratory failure (Di Cesare, 2016).

Ectoparasites

Ectoparasites that are known to infest cheetahs are: fleas, lice, ticks, mites (*Cheyletiella* spp., *Octodectes* spp., *Notoedres* spp., *Sarcoptes* spp. and *Demodex* spp.) chiggers, biting flies and hippoboscids. Also myiasis can occur as a result of open wounds and lesions (Citino et al., 2007). Information on treatment can be found in "[Chapter 4: Preventive medicine](#)", paragraph "[4.3 Ectoparasites](#)".

2. Diagnosis

This chapter contains examples of medical examinations, which should be followed during diagnosis. Furthermore, a table with reference values is included, as procedures regarding shipping, quarantine and necropsy.

2.1 Standard health evaluation protocol

Physical examination

(Citino et al., 2007), “A thorough evaluation of each organ system, body weight, and assessment of body condition is essential. The oral cavity should be inspected for papillomatous plaques under the tongue and other lesions such as ulcers. The use of a flea comb may help identify mild flea infestations. Foot pad lesions (superficial ulcers) can be characteristic of calicivirus infection in cheetahs.”

Dental examination and prophylaxis

The cheetahs dental formula is I 3/3, C 1/1, P 3/2, M 1/1 (Bloemfontein, 2017).

(Citino et al., 2007), “The teeth and soft tissue structures of the mouth and throat should be examined for abnormalities. Odour from the mouth may indicate dental problems. The area of the hard palate adjacent to the carnassial teeth should be examined for erosions and punctures of soft tissues and possibly the underlying bone that extends into the nasal cavity (palatine erosions). These lesions can be treated by rounding off and shortening the points on the mandibular molar without exposing the root canal. This can be done prophylactically once the permanent teeth have erupted. Foreign bodies lodged between teeth, such as bone fragments, sticks, etc., can predispose oral disease. These should be removed and infections or traumatic lesions treated as indicated. As in domestic cats, calculus accumulation should be removed in cheetahs while under anaesthesia; here care must be taken to remove material from the subgingival sulcus. Regular dental care is important in preventing bacteremia of oral origin that can contribute to, or promote systemic disease. If an ultrasonic scaler is used, the scraped surfaces should be polished to smooth the surfaces. This will deter future calculus accumulation.”

In addition the accumulation of dental calculus can be partially prevented through the regular feeding of whole prey (Stagegaard, J., pers. comm., 2017).

Vaccination

(Citino et al., 2007), “Vaccination status should be reviewed and necessary vaccinations should be given as needed. Severely stressed animals may not mount appropriate titers and should be revaccinated if conditions indicate.” More information on vaccines can be found in [“Chapter 4: Preventive medicine”](#).

Urine collection

(Citino et al., 2007), “Urine can be collected by expressing the bladder, catheterizing the bladder, or by cystocentesis. Urine samples should be submitted for routine urinalysis and sediment exam.”

Faecal collection

(Citino et al., 2007), “Routine screening should include faecal flotation and direct smear.” Monitoring of gonadal and adrenal steroids in captive animals can give us information on their physiology, health and reproductive status. This can be done through non-invasive faecal steroid assays. For this procedure faecal samples can be stored up to 14 days at -20°C, or immersed in 95% ethanol at room temperature. Followed by lyophilization and extraction. Other methods used to dry samples are solar or conventional ovens, however with these methods variations in the measured concentrations of steroid hormones (except for androgens), where observed (Terio et al., 2002).

Blood collection

Blood collection and analysis needs to include:

- Complete blood count.
- Serum chemistry panel.
- Serum banking.
- Serologic testing.
- Testing through blood smear for hemoparasites (Citino et al., 2007).

Standard values

The following reference values for captive cheetahs were acquired from Species360:

Table 1: Reference values for captive cheetahs (Teare, 2013)

Physiological Reference Intervals for <i>Acinonyx jubatus</i>								
Test	Units	Reference Interval	Mean	Median	Low Sample ^a	High Sample ^b	Sample Size ^c	Animals ^d
White Blood Cell Count	*10 ⁹ cells/L	5.43-19.02	10.19	9.51	1.22	24.30	2225	485
Red Blood Cell count	*10 ¹² cells/L	4.99-9.18	7.07	7.05	2.47	10.60	2118	488
Hemoglobin	g/L	88-167	129	128	42	209	2179	484
Hematocrit	L/L	0.263-0.521	0.393	0.390	0.148	0.670	2482	508
MCV	fL	44.8-66.3	55.6	55.6	37.0	75.1	2075	480
MCH	pg	15.1-21.0	18.2	18.3	12.9	23.8	2033	475
MCHC	g/L	285-370	328	329	237	422	2139	480
Segmented Neutrophils	*10 ⁹ cells/L	2.81-14.43	6.91	6.35	0.02	18.50	2223	485
Neutrophilic Band Cells	*10 ⁹ cells/L	0.02-0.18	0.06	0.05	0.00	0.22	2048	463
Lymphocytes	*10 ⁹ cells/L	0.67-4.06	1.97	1.84	0.04	5.67	2198	483
Monocytes	*10 ⁶ cells/L	66-982	343	288	14	1340	1897	465
Eosinophils	*10 ⁶ cells/L	94-2380	805	653	40	3195	2020	470
Basophils	*10 ⁶ cells/L	4-330	104	93	0	348	137	103
Platelet Count	*10 ¹² cells/L	0.157-0.797	0.393	0.372	0.008	0.961	910	272

(Continuation table 1) Test	Units	Reference Interval	Mean	Median	Low Sample ^a	High Sample ^b	Sample Size ^c	Animals ^d
Nucleated Red Blood Cells	/100 WBC	0-4	1	1	0	5	315	180
Reticulocytes	%	*	0.8	0.6	0.0	3.2	35	27
Glucose	mmol/L	4.99-12.94	7.84	7.41	0.28	16.12	2348	495
Blood Urea Nitrogen	mmol/L	8.3-20.8	13.3	12.9	1.4	25.7	2330	494
Creatinine	μmol/L	90-334	209	208	0	424	1544	436
BUN/Cr ratio		9.0-31.7	16.3	15.2	4.2	37.0	1508	426
Uric Acid	μmol/L	0-65	13	6	0	83	559	193
Calcium	mmol/L	2.29-3.25	2.66	2.62	1.95	3.58	2300	500
Phosphorus	mmol/L	1.17-3.42	1.98	1.79	0.26	4.85	2223	497
Ca/Phos ratio		1.1-2.8	1.9	1.9	0.4	3.9	2214	496
Sodium	mmol/L	144-165	156	156	135	177	2278	482
Potassium	mmol/L	3.7-6.0	4.5	4.4	2.8	6.6	2281	482
Na/K ratio		24.4-43.0	34.8	35.2	18.7	51.6	2275	482
Chloride	mmol/L	112-131	122	122	101	138	2182	471
Total Protein	g/L	50-79	65	65	37	93	2120	470
Albumin	g/L	25-45	36	36	18	51	1617	416
Globulin	g/L	20-47	31	31	5	58	1789	463
Fibrinogen	g/L	0.00-4.78	1.77	2.00	0.00	6.00	252	80
Alkaline Phosphatase	U/L	4-60	17	12	0	81	1703	451
Lactate Dehydrogenase	U/L	19-218	73	56	4	262	800	263
Aspartate Aminotransferase	U/L	20-122	51	44	2	152	2066	470
Alanine Aminotransferase	U/L	35-218	93	82	0	266	2177	485
Creatine Kinase	U/L	63-969	323	224	2	1364	1317	351
Gamma-glutamyltransferase	U/L	0-8	2	1	0	14	1287	326
Amylase	U/L	757-2120	1261	1192	15	2546	1392	330
Lipase	U/L	0-53	14	9	0	63	225	105
Total Bilirubin	μmol/L	1.3-10.0	3.9	3.4	0.0	15.4	2073	492
Direct Bilirubin	μmol/L	0.0-1.7	0.6	0.0	0.0	1.7	328	137
Indirect Bilirubin	μmol/L	0.0-7.7	2.9	3.4	0.0	10.3	301	138
Cholesterol	mmol/L	2.95-8.15	4.84	4.65	2.07	9.97	2120	472
Triglyceride	mmol/L	0.15-1.20	0.47	0.41	0.01	1.49	999	263
Bicarbonate	mmol/L	12.4-23.9	18.6	18.8	6.4	27.4	376	127
Magnesium	mmol/L	0.458-1.216	0.794	0.837	0.284	1.192	78	53
Iron	μmol/L	1.2-19.0	8.7	8.4	0.7	22.2	189	86
Carbon Dioxide	mmol/L	11.9-50.5	24.0	19.9	7.0	64.2	745	232
Body Temperature	C	36.8-40.5	38.6	38.7	34.9	41.7	1473	312

^a Lowest sample value used to calculate the reference interval.

^b Highest sample value used to calculate the reference interval.

^c Number of samples used to calculate the reference interval.

^d Number of different individuals contributing to the reference interval.

* Sample size is insufficient to produce a valid reference interval.

Cortisol (ng/ml) mean value 39,3 +/- 27,8 (Tschurlovits, n.d.).

2.2 Pre-shipment examinations and quarantine

Health certificate

A health certificate needs to be present before any animal transport. This certificate should include: past medical history, blood values, immunizations, faecal examinations and serology results (Citino et al., 2007).

Quarantine

Transported animals between two BALAI listed zoos do not need to follow quarantine once they have arrived in the new installations according to the directive 92/65/EEC. However, if an animal is transported from a non-BALAI listed zoo to a BALAI listed zoo, it has to undergo a quarantine period.

A quarantine period should be long enough to cover the incubation period of most infectious diseases. Therefore the quarantine period should be at least **30 days long**. During this period the animals behaviour and health status should be monitored. If required, tests can be taken during this period. Diet transition should be made gradually to avoid food refusal and/or gastrointestinal upset (Citino et al., 2007).

2.3 Necropsy

After the death of every cheetah a necropsy complete with gross and histopathologic examination needs to be carried out. The EEP Cheetah Necropsy Protocol to be followed can be found in [Appendix I](#). Also, the spinal cord removal during necropsy in ataxic EEP cheetahs can be found explained in [Appendix II](#).

3. Management

This chapter contains information on the sedation and anaesthesia process.

3.1 EAZA BioBank

There is a call from the EAZA BioBank to collect extra blood, tissue and/or serum samples whenever an animal is sedated. This helps supporting population management and conservation research. The sampling protocol can be found in [Appendix III](#).

3.2 Drugs potentially causing adverse reactions

- **Griseofulvin:** can lead to severe bone marrow suppression, drug induced anaemia and death. Treated animals should be monitored carefully (Tschurlovits, n.d; Wack, 1992).
- **Zuclopenthixol acetate:** can cause inappetence, ataxia, extrapyramidal reactions like for example head swaying, akathisia, and prolapsed third eyelid. Therefore it is not recommended to use on cheetahs. Instead perphenazine enanthate (3.0 mg/kg) should be used; the dosage needs to be adjusted to the individual, it will depend on its temperament and age. In old or very young animals, lower dosages should be administered (Huber, 2001).
- **Haloperidol:** May result in extrapyramidal effects (Citino et al., 2007).
- **Levamisole:** After four 3-month-old cubs were dewormed with Levamisole (subcutaneously at a dosage of 5 mg/kg), they all showed severe respiratory distress and seizures, finally dying despite resuscitation attempts (Oevermann, 2004).

3.3 Sedation and anaesthesia

3.3.1 Preparation

The animal should be fasted 8-24 hours. And should not receive water for 6-12 hours prior to sedation. If the temperatures are very high, or the animal suffers from a specific disease, water may be withheld for a shorter period of time. If possible, a venous access is preferred. The administration of the drug should be done in a small, quiet, safe area (Tschurlovits, n.d.). If the cheetah lives in a group, it should be separated prior to being anaesthetized (CCF, 2002).

3.3.2 Injectable anaesthetics drug combination

The following drugs can be administered intramuscularly, unless specified otherwise:

Table 2: injectable drug combinations for cheetah (Tschurlovits, n.d.),

Drug dose	Partial antagonistic dose
0,05 mg/kg Ketamine + 1,5 mg/kg Medetomidine	5 mg/kg Atipamezole per each mg of Medetomidine administered. With these doses no waiting time is required (will not lead to convulsions). (Kaadorp, J., pers. comm., 2017).

(Continuation table 2)		Partial antagonistic dose
Drug dose		
2,5 mg/kg Ketamine + 0,05-0,07 mg/kg Medetomidine		No antagonistic drug available 0,3 mg/kg Atipamezole
3,0 mg/kg Ketamine + 0,03 mg/kg Medetomidine + 0,3 mg/kg Butorphanol		No antagonistic drug available 0,15 mg/kg Atipamezole + 0,3 mg/kg Naltrexone
2,0 mg/kg Ketamine + 0,02 mg/kg Medetomidine + 0,1 mg/kg Midazolam		No antagonistic drug available Atipamezole Flumazenil or Sarmazenil (Stagegaard, J., pers. comm., 2017)
0,035 mg/kg Medetomidine + 0,15 mg/kg Midazolam + 0,2 mg/kg Butorphanol		0,175 mg/kg Atipamezole + 0,03 mg/kg Flumazenil + 0,2 mg/kg Naltrexone
5 mg/kg Ketamine + 0,02 mg/kg Dexmedetomidine		No antagonistic drug available 0,1 mg/kg Atipamezole
5-10 mg/kg Ketamine + 0,5-1,1 mg/kg Xylazine		No antagonistic drug available 0,1 mg/kg Atipamezole
3-4 mg/kg Ketamine + 0,75-1,5 mg/kg Xylazine + 0,03-0,04 mg/kg Midazolam		No antagonistic drug available 0,1 mg/kg Atipamezole + 0,03 mg/kg Flumazenil or 0,1 mg/kg Sarmazenil
3-5 mg/kg Tiletamine-zolazepam		0,03 mg/kg Flumazenil or 0,1 mg/kg Sarmazenil
1,6 mg/kg Tiletamine-zolazepam + 0,03 mg/kg Medetomidine		0,03 mg/kg Flumazenil or 0,1 mg/kg Sarmazenil + 0,15 mg/kg Atipamezole
1,3-1,5 mg/kg Tiletamine-zolazepam + 0,013(?) - 0,15 mg/kg Medetomidine + 1,3-1,5 mg/kg Ketamine		0,03 mg/kg Flumazenil or 0,1 mg/kg Sarmazenil + 0,75 mg/kg Atipamezole
1,0-1,3 mg/kg Tiletamine-zolazepam + 0,4-0,52 mg/kg Xylazine + 1,6-2,1 mg/kg Ketamine		0,03 mg/kg Flumazenil or 0,1 mg/kg Sarmazenil + 0,1 mg/kg Atipamezole
0,5-4 mg/kg Propofol IV		No antagonistic drug available

Ketamine + medetomidine

(Tschurlovits, n.d.), "If reversals are abnormally long (i.e. >20 minutes) the atipamezole dose may be increased to 0,5 mg/kg."

Tiletamine-zolazepam

- (Tschurlovits, n.d.), "Avoid in cats with known or suspected renal disease!"
- Prolonged recoveries may be possible. Flumazenil can be used to antagonize the zolazepam fraction."

Combinations with ketamine

- The use of ketamine alone is not recommended in cheetahs! It may be necessary to treat resulting seizures with benzodiazepines (diazepam, midazolam) (Tschurlovits, n.d.)
- Avoid in cats with known or suspected renal disease! (Tschurlovits, n.d.)

- (Tschurlovits, n.d.), “Ketamine cannot be antagonised! Therefore you have to wait for at least 30 minutes after ketamine administration before you may antagonise the other components of the drug combination. Otherwise the animal will experience a recovery phase solely under influence of ketamine! This may result in uncontrolled body movements possibly combined with severe hyperthermia, which can lead to injuries, or in the end to the death of the animal.” However to avoid this, ketamine could be administered in lower dosages (approx. 1 mg/kg – 1,5 mg/kg). When ketamine is administered in lower dosages, the waiting period to antagonise the other components is not required (Kaandorp, C., pers. comm., 2017).

Xylazine

(Tschurlovits, n.d.), “Possible urine contamination during electro ejaculation. Avoid xylazine in late term gestation!”

Propofol

(Tschurlovits, n.d.), “(Rapid) administration may result in apnoea! Apply oxygen before administration and intubate as quickly as possible to maintain sufficient oxygen supply.” Propofol can only be administered intravenously! (Kaandorp, C., pers. comm., 2017).

3.3.3 Intubation

- For procedures longer than 20 min the cheetah should be intubated and oxygen should be supplied to optimise O₂ saturation (Kaandorp, C., pers. comm., 2017).
- Topical anaesthesia of the larynx is not necessary (Tschurlovits, n.d.).

3.3.4 Inhalation anaesthesia

- (Tschurlovits, n.d.), “For prolonged procedures inhalation anaesthesia is recommended after induction with injectable anaesthetics.
- Isoflurane is recommend but sevoflurane and halothane also can be used safely.
- Normally spontaneous respiration with occasional assisted respiration is sufficient but ventilation may be useful because of subclinical hypoxia.”

Table 3: Tranquilizers to administer (Tschurlovits, n.d.)

Drug	Dosage
Diazepam	0,5-0,2 mg/kg p.o. SID-TID (also for long term use)
Acepromazine	0,5-1,0 mg/kg p.o.
Perphenazine enanthate	3,0 mg/kg i.m. (long acting for 5-10 days)

Table 4: NSAIDs to administer (Tschurlovits, n.d.)

NSAIDs		
Drug	Dosage	Comment
Meloxicam	0,1-0,2 mg/kg p.o./i.m. SID	p.o. for retreated treatments, 0,2 mg/kg for single application
Carprofen	1,2 mg/kg p.o. SID	
Etodolac	6 mg/kg SID	

In cats with renal disease NSAIDs should be used with caution! (Kaandorp, C., pers. comm., 2017).

Table 5: Opioids to administer (Tschurlovits, n.d.)

Opioids		
Drug	Dosage	Comment
Butorphanol	0,2-0,4 mg/kg s.c./i.m.	κ -agonist and μ -antagonist
Fentanyl	50 μ g/h s.c. or 100 μ g/h patch	μ -agonist, short term post-op
Morphine	0,1 mg/kg epidurally	Administer 45 min pre-op for hind limb orthopaedic.
Tramadol	2,0-2,5 mg/kg p.o. BID	μ -, κ -, δ -agonist, short term and long term.

3.3.5 Monitoring

During anaesthesia the heart rate, respiration and body temperature need to be monitored. The following values are normal vital rates for cheetahs under anaesthesia:

- Heart rate: 120-140 beats per minute.
- Respiration rate: 15-20 breaths per minute.
- Body temperature: 38,5 °C (CCF, 2002).

Hypothermia

The animal's body temperature can drop due to low environmental temperature or through a long surgical procedure. If the cheetah's temperature drops to 37 °C or less, it has to be warmed up. Blankets, heat pads, thermal heaters, warm water enemas and warmed IV fluids should be used to increase the animals' body temperature (CCF, 2002; Tschurlovits, n.d.).

Hyperthermia

If the animals temperature goes up to 40 °C (due to high environmental temperature, exposure to direct sunlight, pre-anaesthetic excitement, etc.), its temperature has to be brought back down. Ice packs wrapped in blankets can be placed where the legs join the body, cold water can be rubbed into the fur and fans can be used. If the temperature reaches 40,6 °C the cheetah will suffer from severe hyperthermia, this has to be treated aggressively, cold enemas and IV fluids need to be administered and the cheetah needs to be rubbed with cold water (CCF, 2002; Tschurlovits, n.d.).

3.3.6 Antagonists

- (Tschurlovits, n.d.), "Naltrexone: 1 mg Naltrexone per 1 mg of Butorphanol.
- Atipamezole: 5 times the Medetomidine dose.
- Flumazenil, Sarmazenil: There is no difference between these two antagonists, the use of both is recommended."

4. Preventive medicine

Prevention is nine-tenths the cure. Therefore, this chapter contains information on preventive medicine. Vaccinations for infectious diseases are described, as prevention for endo- and ectoparasites.

4.1 Vaccination

Vaccination programs for felids are often designed after recommendations for domestic cats. Core vaccines are those that the animal truly needs. For felines these include:

- Feline parvovirus (panleukopenia virus) (FPV).
- Feline calicivirus (FCV).
- Feline herpesvirus (FHV), Cheetahs are at a high risk of contracting this disease, therefore it is recommended to vaccinate cubs at 6 weeks of age.
- Rabies (Miller, 2011)

Parenteral killed vaccines are recommended, because modified live (ML) vaccines can lead to vaccine-associated disease.

Table 6: Virus abbreviation and full name

FPV	Feline panleukopenia virus
FCV	Feline calicivirus
FHV	Feline herpesvirus
FelV	Feline leukemia virus
FIV	Feline immunodeficiency virus
CDV	Canine distemper virus
FCoV	Feline corona virus

Preliminary considerations:

- (Tschurlovits, n.d.), "Type, serial number, expiry date, volume, source and administration site should be documented (Tschurlovits, n.d.)."
- **Some drugs may interfere with the vaccination.**
- When for example the animal is darted, one needs to make sure that the full dose was administered.
- **Animals that are showing clinical signs from disease cannot be vaccinated.**
- If there is a disease outbreak, all susceptible animals should be vaccinated and a booster should be administered 14-21 days later.
- **A lot of different vaccines have not been approved for non-domestic species; there are cases where cheetahs have become sick (vaccine-induced disease) after FeLV and FPV vaccines where administered."**
- Cheetahs should be tested for FeLV and FIV. If individuals are found positive the EEP should be contacted. A possible advice could be to house negative and positive individuals separately, instead of using vaccination (Miller, 2011).
- **At this moment vaccinations for FCoV, CDV, FeLV and FIV are not recommended** (Citino et al., 2007).

4.1.1 Cubs

Vaccinations (FPV, FCV, FHV) should be administered at 6-9 weeks of age. They should be re-administered every 2-4 weeks until the cubs are 14-16 weeks of age. Then the animal should receive a booster after 12 months. Core vaccines should only be administered every one or three years thereafter (it will depend on the vaccination used) (Miller, 2011).

Rabies vaccination should be firstly administered when cubs are around 6 months (they can be vaccinated at earliest 3 months). Revaccination should occur one year later. A booster should be administered every year or every three years, this will depend on the registration of the vaccine used (Miller, 2011).

4.1.2 Adults

As previously mentioned, core vaccines should be administered every one or three years (it will depend on the vaccination used). A Rabies booster should be administered every year or every three years (Miller, 2011). Vaccination protocols used in different institutions and countries vary a lot. In endemic areas rabies vaccination is recommended (Kaandorp, C., pers. comm., 2017).

This is an example of vaccination schedule used in several institutions in Europe:

Table 7: Example of vaccination schedule (Kaandorp, C., pers. comm., 2017).

Vaccine	First Vaccination	Booster	Booster	Adult Cheetah
Fel-O-Vax®-5 Vaccine Inactivated (Chlam) (Boehringer Ingelheim), Fevaxyn Quatrifel® (Zoetis) Vaccine Inactivated	9 weeks	12 weeks	16 weeks	Once a year
Rabies (dead vaccine) in endemic areas	-	Normally first vaccination given at 6 months (earliest 3 months)	Booster possible at 12 months	Repeat once a year (or once every 3 years, depending on vaccine registration)
Cheetahs must be vaccinated with inactivated vaccines!				

4.1.3 Pregnant females

Killed vaccines need to be used in pregnant females (Miller, 2011). If it is known when females are due, they should be re-vaccinated for FPV, FCV and FHV with a killed virus vaccine, three weeks before giving birth (Citino et al., 2007).

4.2 Endoparasites

The different endoparasites known to infest cheetahs are described under “[1.2.5. Parasites](#)”. Regular faecal examinations should be done and action should be taken according to the results (presence or absence).

Faecal examination

- Faecal examinations should be made 4-6 times a year (Tschurlovits, n.d.).
- Negative results do not mean that there are no parasites; keep in mind that latency periods and intermittent shedding occur (Tschurlovits, n.d.).
- In practice institutions have an established prophylactic protocol for feral cats. Here animals are regularly treated for parasites. However, in theory treatments should only be prescribed after positive results, to avoid creating parasite resistance (Kaandorp, C., pers. comm., 2017; Tschurlovits, n.d.).

Treatment

Although treatment against endoparasites should be prescribed after positive faecal testing, many institutions have preventative protocols to treat their animals with antiparasitic drugs due to the risk of re-infection in the often highly contaminated enclosures (Kaandorp, C., pers. comm., 2017). See table 8 for examples of drugs to treat adult cheetahs for endoparasites.

Table 8: Drugs to treat cheetahs for endoparasites (Tschurlovits, n.d.)

Drug	Parasite class	Dosage
Pyrantel	Nematodes	20 mg/kg p.o., for 3-5 days.
Fenbendazole	Nematodes, Giardia	50-100 mg/kg p.o., single application for 3-5 days, for giardia 10-14 days.
Ivermectin	Nematodes, Heartworms	0,2 mg/kg s.c./p.o. 0,1-0,2 mg/kg (or 10mg/individual adult cheetah), monthly for ascarid elimination or heartworm prophylaxis.
Praziquantel	Cestodes Trematodes	5,5-6,6 mg/kg s.c./p.o. single application higher doses as needed (e.g. <i>Spirometra</i> spp.).
Sulfadimethoxine*	Coccidia	50 SID p.o. the first day, then 25 mg/kg SID p.o. for 14-20 days.

* Information acquired from Plumb (1995).

Finding the best protocol for preventative treatment of cheetah cubs for endoparasites is difficult since the opinions, experiences and publications vary a lot, depending on country and medicine used. In the following table, on the next page, some options are displayed.

Table 9: Drugs to treat cheetah cubs for endoparasites (Kaandorp, C., pers. comm., 2017)

Deworming						
Drug	Cheetah Cubs					Adults
Flubenol®, flubendazol (22 mg/kg)	4 weeks	6 weeks	8 weeks	4 months	6 months	Every 3 months
Banminth®, Pyrantel (20 mg/kg)	3 weeks	5 weeks	7 weeks	9 weeks	12 weeks	Every 3 months
Ivomec® (Ivermectin, 0,2 mg/kg)				9 weeks	12 weeks	Every 3 months
Panacur® (fenbendazol 50mg/kg)	No schedule found					

4.3 Ectoparasites

In the following table different ectoparasites that can infest cheetahs are displayed, together with the agents used to eradicate them. These agents have been used in cheetahs with similar dosages to domestic cats without apparent side effects (Citino et al., 2007).

Table 10: Ectoparasites in cheetah based on cat medicine (Tschurlovits, n.d.)

Ectoparasites	Fipronil	Methoprene	Imidacloprid	Luferon	Nitenpyram	Ivermectin	Selamectin	Indoxacarb
Fleas	X	X	X	X (fleas infertile)	X (Short acting)	(X)	X	X
Ticks	X		(X)			X		
Mites	X					X	X	
Chiggers*	X							
Lice	X	X	X			X		
Flies		X				(X)		

* Information acquired from Hnilica (2010).

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Appendix I: EEP Cheetah necropsy protocol

EEP CHEETAH NECROPSY PROTOCOL

Date		Prosector		Location	
Conditions					

Cheetah Name		ARKS ID		Other ID	
Captive		Free-Ranging		Hunted	
EEP	(Yes) (NO)	Studbook-Number		Studbook-Name	
Sex		Date of Birth/Age		Offspring	
Date of Death		Time of Death		Cause of Death	
Date of Necropsy		Time of Necropsy		Degree of decomposition	
Total Weight (kg)		<i>dead</i>	<i>alive</i>	<i>including coat</i>	<i>without coat</i>

History	<p>Enclosure structure:</p> <p>Group composition:</p> <p>Nutrition:</p> <p>Vaccinations:</p> <p>Deworming:</p> <p>Clinical signs:</p> <p>Clinical disease:</p> <p>Other remarks:</p>
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Gross Examination Worksheet

General Condition	(nutritional condition, physical condition)
Orifices	(eyes, nares, mouth, anus, genitals)
Skin	
Fur	

Musculoskeletal System	(bones, joints, tendons, muscles)
Body cavities	(serosa, fat stores, abnormal fluids)
Hemolymphatic System	(spleen, lymph nodes, thymus)
Respiratory System	(nasal cavity, larynx, trachea, lungs, regional lymph nodes)
Cardiovascular System	(heart, pericardium, great vessels)
Digestive System	(oral cavity, oesophagus, stomach, intestines, liver, pancreas, mesenteric lymph nodes)
Urinary System	(kidneys, ureters, urinary bladder, urethra)
Reproductive System	(gonads, uterus, vagina, penis, prepuce, accessory glands, mammary glands, anal glands, placenta)
Endocrine System	(adrenals, thyroid, parathyroids, pituitary)
Nervous System	(brain, spinal cord, peripheral nerves)
Sensory Organs	(eyes, ears)
Preliminary Diagnosis	

Notes:

Clinical Pathology

Laboratory Studies	Date	Sample	Examination	Result
Serology				
Blood chemistries				
Hematology				
Bacteriological cultures				
Viral cultures				
Parasitological screen				
Fungal cultures				

X-Rays

Positioning	Date	Result

Other examinations:

Fixed Tissue Check List

Preserve the following tissues in 10 % buffered formalin at a ratio of 1 part tissue to 10 parts formalin. Tissues should be no thicker than 1 cm (excluding brain and spinal cord — fix in total). Include sections of all lesions and samples of all tissues on the required tissue list.

✓	Required tissues (Formalin)	Tissue sampling procedure
	Adrenal glands	Both entire glands with transverse incision
	Brain ¹	In Toto — see below!
	Diaphragm	Representative section
	Eyes	Leave intact
	Gastrointestinal tract	3 cm long sections of oesophagus, stomach (cardia, antrum and pylorus), duodenum, jejunum, ileum, cecum, colon, rectum and omentum. Open carefully along the long axis
	Heart	Longitudinal section including atrium, ventricle and valves from both right and left heart
	Kidney	Sections from both kidneys including cortex, medulla and pelvis
	Liver	Sections from 3 lobes with capsule and gall bladder
	Lungs	Sections from several lobes including a major bronchus
	Lymph nodes	Cervical, anterior mediastinal, bronchial, mesenteric and lumbar with transverse cut
	Pancreas	Sections from 2 areas, 1 including central ducts
	Parathyroids + Thyroid	Leave glands intact
	Peripheral nerve ²	In separate container with topographic name
	Reproductive tract ³	Entire uterus and ovaries with longitudinal cut into lumen. Entire testis with transverse cut, entire prostate with transverse cut
	Skeletal muscle*	Cross section of M. biceps brachii and M. quadriceps femoris
	Skin	Full thickness of abdominal skin and lip
	Spinal cord ¹	Sections from cervical, thoracic and lumbar cord in separate containers
	Bone	Cross section Femur w . bone marrow and distal femur including growth plate (Epiphysis)
	Spleen	Cross section including capsule
	Thymus	Representative section
	Tongue	Cross section near tip including both mucosal surfaces
	Trachea	Representative section
	Urinary bladder/ureter/urethra	Cross section of bladder and 2 cm sections of tubular structures

A very careful preparation of these tissues is required!

Remove the entire brain place small frontal section (max 25% of Brain) at — 20 or lower if possible and place rest in toto in formalin.

Method description for spinal cord removal under field conditions:

Separate the spinal column from the remaining carcass; remove the paravertebral soft tissues and muscles. Transect the spinal column at the level of the intervertebral discs into approximately 15-cm-long segments. Do not confuse the individual segments in order to preserve an accurate description of the lesion distribution. Insert the provided metal blade carefully laterally to the spinal cord and into the spinal canal and move it dorsally and ventrally within the canal transecting the segmental nerves. It should be attempted to separate the dura mater from the epidural attachments in order to remove the spinal cord with the intact dura. Following this circumferential preparation, the spinal cord is grasped at one end with forceps and pulled out of the spinal canal while carefully removing persisting attachments. If possible, the spinal cord should be grasped by the dura mater to reduce artefacts. Repeat the process for each segment. The part grasped by the forceps is unsuitable for histologic examination, but should be frozen at -20 °C or more if available. The cranial part of each spinal cord segment is marked with a small incision and stored in formalin (use separately marked containers!).

² Place peripheral nerve and muscle tissue on a clean cardboard piece so that it adheres before submerging in the formalin.

* For semen assessment in male cheetahs follow the guidelines provided by the IZW!

Frozen tissue checklist

Preserve the following tissues in separate freezable plastic bags by at least — 20 °C.
Include sections of all lesions and samples of all tissues on the required tissue list.

	Required tissues (Frozen)	Tissue sampling procedure
	Brain ¹	left frontal lobe/ olfactory bulb
	Eye chamber liquid	Aspirated from camera anterior
	Femur with marrow	Freeze 1/2 part of the femur
	Gastrointestinal tract	3 cm long sections of oesophagus, stomach (cardia, antrum and pylorus), duodenum, jejunum, ileum, cecum, colon, rectum and omentum. Samples from stomach contents, ingesta and feces in separate small containers
	Heart	Longitudinal section including atrium, ventricle of left and right heart and septum
	Kidney	Sections from both kidneys including cortex, medulla and pelvis
	Liver	Sections from 3 lobes with capsule
	Lungs	Sections from several lobes including a major bronchus
	Skeletal muscle	Cross section of M. biceps brachii and M. quadriceps femoris
	Spinal cord*	Sections from cervical, thoracic and lumbar cord in separate plastic bags
	Spleen	Cross section including capsule
	Urine sample	Aspirated from intact urinary bladder

Neonatal Necropsy Protocol

Please follow the adult protocol in addition to the following:

- Examine of malformations (Cleft palate, deformed limbs)
- Assess hydration (tissue moistness) and evidence of nursing (milk in stomach)
- Fix umbilical stump and surrounding tissues
- Determine if breathing occurred

Shipping Tissues

Please obtain necessary CITES, Export-Import and veterinary/agriculture Permits before shipping tissues!

For further information please contact

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Appendix II: Cheetah spinal cord removal

A simple field method for spinal cord removal in ataxic EEP cheetahs (*Acinonyx jubatus*)

Extracted from: Walzer, C., Kübber-Heiss, A., and Robert, N. 2002. A simple field method for spinal cord removal - demonstrated in the cheetah (*Acinonyx jubatus*). J. Vet. Diagn. Invest.14: 75-78.

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Anna Kübber-Heiss, Institute of Pathology and Forensic Veterinary Medicine, University of Veterinary Medicine, A-1210 Vienna, Austria.

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Abstract

Removal of the spinal cord is considered time consuming and difficult. A delay in the necropsy procedure, especially in the central nervous system can result in significant tissue autolysis and subsequent diagnostic difficulties. In the field where many necropsies are performed, suitable electric saws are mostly unavailable. A technically simple and rapid method for spinal cord removal, requiring only a straightforward tool has been devised. No necropsy induced structural damage has been noted on histo-pathological examination.

Following standard necropsy procedures and evisceration of the carcass, the brain is removed and transected from the spinal cord at the level of the foramen magnum. The spinal column is separated from the remaining carcass and the paravertebral soft tissues and muscles are removed. The spinal column is then transected at the level of the intervertebral discs into approximately 15 cm. Long segments (fig.1).

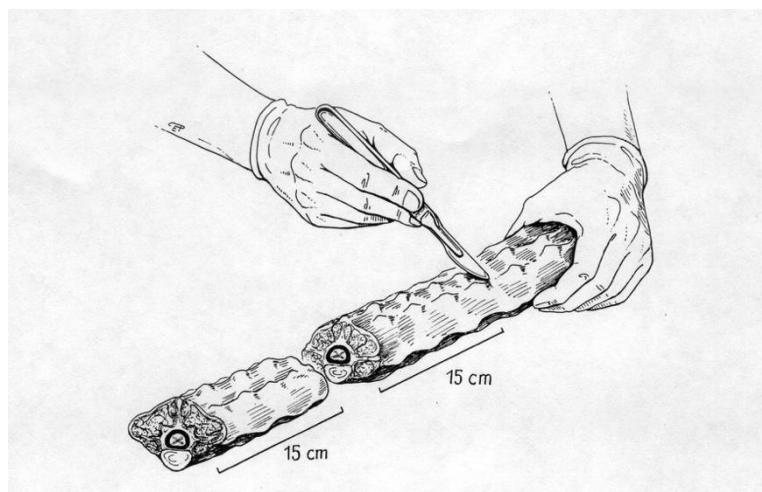


Figure 1. Spinal column, cheetah, Transected at the level of the intervertebral discs into approximately 15 cm. long segments.

It is imperative at this stage not to confuse the individual segments in order to preserve an accurate description of the lesion distribution. Individual segments now allow cranio – caudal visualization of the spinal cord within the spinal canal. In adult cheetahs a 250 mm long, 5-mm wide and 1 mm thick sterile, blunt metal blade is carefully inserted laterally to the spinal cord and into the spinal canal (fig.2).

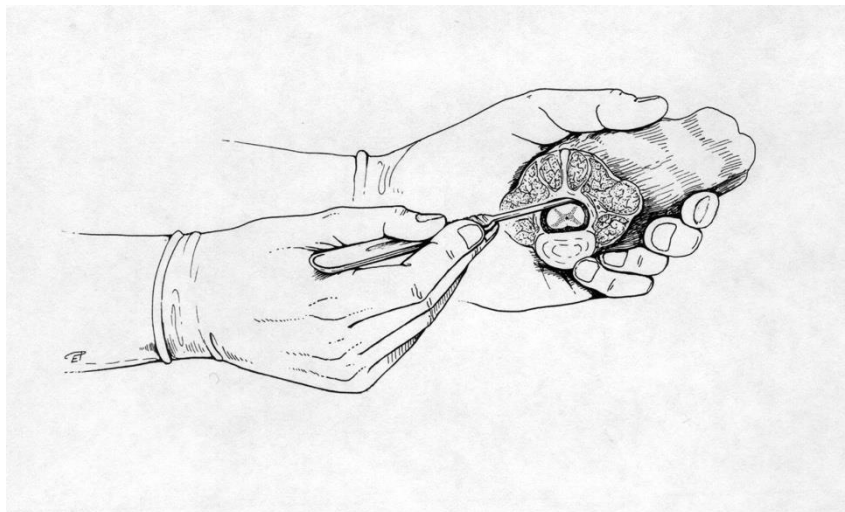


Figure 2. Spinal cord, cheetah, A 25-cm long, 5-mm wide and 2-mm thick sterile, blunt metal blade is carefully inserted laterally of the spinal cord, into the spinal canal. The blade is moved dorsally and ventrally within the canal transecting the segmental nerves.

The blade is moved dorsally and ventrally within the canal transecting the segmental nerves. Though not possible in all cases, it should be attempted to separate the dura mater from the epidural attachments in order to remove the spinal cord with the intact dura. Following this circumferential preparation, the spinal cord is grasped at one end with forceps and gently pulled out of the spinal canal while carefully removing persisting attachments (fig.3).

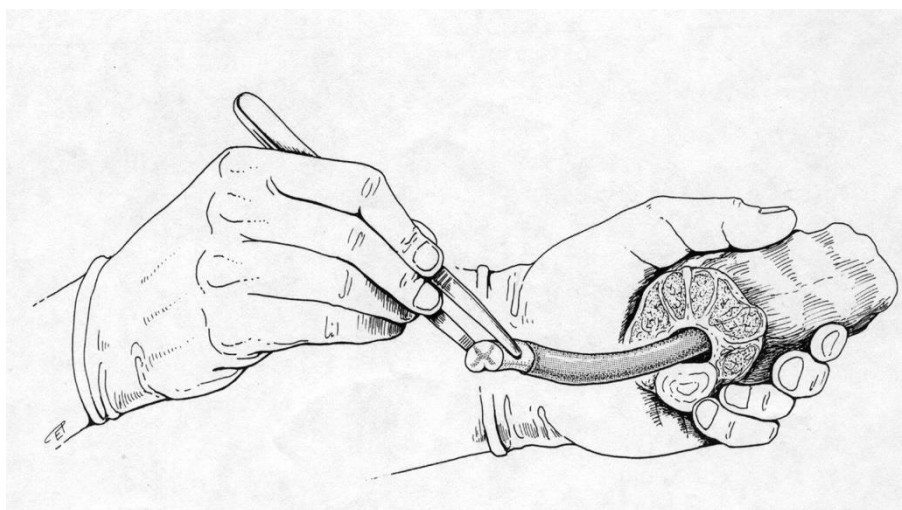


Figure 3. Spinal cord, cheetah, The cord is grasped at one end with anatomic tweezers and gently pulled out of the spinal canal whilst carefully removing persisting attachments. If possible the spinal cord should be grasped by the dura mater.

If possible the spinal cord should be grasped by the dura mater to further reduce the possibility of artifacts. The process is repeated in each segment until the entire spinal cord has been removed. Once removed the spinal cord can be processed as required for further examination.

The fragment of the spinal cord grasped by the forceps is unsuitable for histological examination. However, this fragment should be frozen for possible viral isolation or biochemical and molecular studies. The cranial aspect of each spinal cord segment is marked with a small incision and placed in 10% buffered formalin. Small tight fitting containers, with an adequate volume of formaldehyde, help in avoiding post necropsy transport trauma to the cord. Special fixatives may be required for subsequent electron microscopy, immunocytochemistry or in-situ hybridization studies. Actual cutting in of the tissue, traditionally transversely at 0.5-2.0 cm intervals, should be carried out after adequate fixation. Initial fixation can be enhanced if the formalin is changed after 24 hours. The nervous tissue in juvenile animals contains more water and fewer lipids than in adult animals and therefore does not fix as well.

The described tool is easily constructed from a flat stainless steel sheet in a simple workshop. In adult cheetahs the recommended blade is 180 – 250 mm long, 5 mm wide and 0.5 - 1 mm thick. Through variations in the size of the blunt edged blade this method can be adapted for various species and juvenile animals.

If you have additional questions please do not hesitate to contact:

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Appendix III: EAZA BioBank sampling protocol

Live animals

Samples should be taken in accordance with national legislation.

Whole blood (max 5 ml), in plastic EDTA or PAXgene blood collection tubes. Invert 15 times to mix.

Or

Tissue (max 1 cubic centimeter) from e.g. skin, muscle, or umbilical cord. Placed in a plastic tube (2ml screw cap) containing 70% ethanol or frozen in a plastic bag.

Do not use formalin or methylated alcohol.

Serum (1-10 ml) in plastic tubes. Must be spun and separated. **Should only be provided if it is accompanied by a blood or tissue sample.**

Dead animals

Samples should be taken in accordance with national legislation.

Tissue (max 1 cubic centimeter) from internal organ, skin or muscle. Placed in a plastic tube (2ml screw cap) containing 70% ethanol or frozen in a plastic bag.

Do not use formalin or methylated alcohol.

Serum (1-10 ml) in plastic tubes. Must be spun and separated. **Should only be provided if it is accompanied by a tissue sample.**

CITES

Samples from some species will require CITES permits.

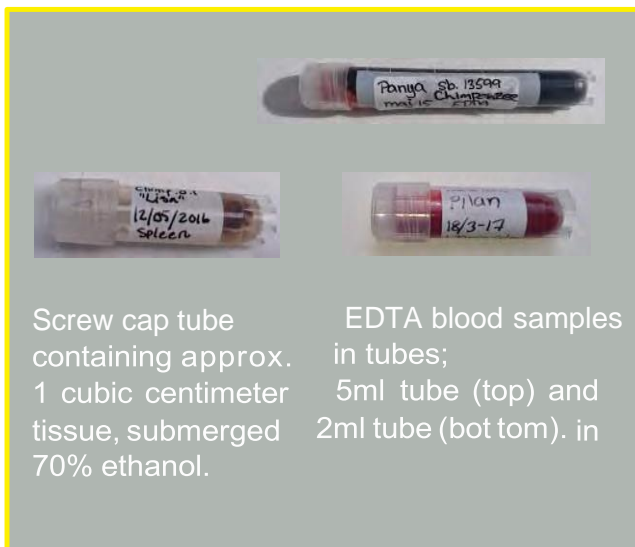


Within EU, according to CITES regulations there is no need for CITES export and import permits.* Outside EU, then CITES export permits must be applied for at the national CITES office. **Remember to apply for CITES export permits in due time before sending the samples.**

CITES exemption is possible for scientific institutions (see article VII, §6 of the CITES convention). Your institution may apply. All four EAZA Biobank hubs have the CITES exemption.

When CITES export permits are obtained, please send a scanned copy to the contact person of the receiving Biobank hub, who will proceed to obtain CITES import permits.

* Exemptions may apply.





BIOBANI

Shipping



Label the sample with animal identifiers (transponder, ring GAAP or local IDA, species name, 4iS5ue hgQB0 and dBfe When sample was taken).

Encase in a Ziplock bag with ZIMS specimen report and contact details of handler, otherwise your sample will not be processed.

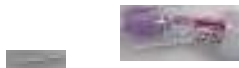
Identification, species name, date of sampling and institution.

Storage

If you cannot ship the samples within 12 hours, then store them in the freezer until shipment is possible. Ship the samples as soon as possible and please avoid arrival at the hub on a weekend.



be samples in tubes



g. Use a sturdy package

Plastic container or bag for the samples with enough material



Reinforced envelope or cardboard box. ZIMS specimen report. Enclose in a pocket in the envelope or box if the fissure is small or from.



Shipping

The package should be labeled on the outside with the diamond 'UN337' logo and the text 'Exempt animal species' and 'Exempt animal species' upon arrival.

The 'UN337' label can be provided by one of the bio hubs.



If you would like to send us your samples then please send them to the biobank hub relevant for your country.



Shipping country: UK, Ireland, Qatar, UAE, Kuwait

Edinburgh hub

ATT: Dr. Helen Senn
Address: Royal Zoological Society of Scotland
WildGenes Laboratory
134 Corstorphine Road
Edinburgh EH12 6TS, UK
E-mail: HSenn@rzss.org.uk



**Leibniz Institute for Zoo
and Wildlife Research**
IN THE FORSCHUNGSVERBUND BERLIN E.V.

Shipping country: Germany, Austria, Croatia, Czech Republic, Hungary, Poland, Russia, Slovakia, Slovenia, Switzerland, Ukraine

Berlin hub

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E-mail: fickel@izw-berlin.de



Shipping country: Belgium, Luxembourg, The Netherlands, France, Greece, Israel, Italy, Turkey

Antwerp hub

ATT: Dr. Philippe Helsen
Address: Centre for Research and Conservation
Royal Zoological Society of Antwerp
Koningin Astridplein 20-26
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Shipping country: Denmark, Estonia, Finland, Latvia, Lithuania, Norway, Sweden, Portugal, Spain

Copenhagen hub

ATT: Dr. Christina Hvilsom
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