

# Veterinary guidelines for reproduction-related management in captive female elephants

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The breeding process in elephants needs monitoring of several parameters in both males and females. The main tools for the determination of the estrous cycle, the evaluation of the genital tract in both sexes, the determination of the number of fetuses and finally the moment of parturition are hormonal assays (progesterone and in some cases LH assays) and ultrasonography.

The first part of this document briefly describes aspects of the estrous cycle and the monitoring tools that can be used in female elephants. The second part describes a protocol for veterinary intervention in elephant parturition. The third part describes the vaginal vestibulotomy procedure. This is not a static document. Colleagues are strongly encouraged to report any experience that may add useful information to these guidelines.



Calcium given as a drip to an Asian elephant during an interrupted parturition (second parturition)



Ruptured uterus of an Asian elephant after a failed parturition that lasted for more than 10 days. The calf had malformed legs.



Fetal membranes being removed by a group member after a successful parturition in a group.

## I. Main aspects for monitoring and assisting female reproduction in elephants

### Determination of the estrous cycle:

The estrous cycle can be divided in 2 phases, the luteal phase and the non-luteal or follicular phase. The luteal phase starts after ovulation and is characterized by elevated progesterone blood levels. The follicular phase starts when the progesterone blood concentration has dropped to what we call in this document the “base line” value. Most laboratory techniques fail to measure any progesterone in the blood during this phase in elephants. Some machines calculate these low concentrations rather than measure them exactly. Moreover, the progesterone concentrations that are provided by the different laboratories, vary greatly depending on the machine and technique used. *It is therefore very important to use the same technique and machine for monitoring the estrous cycle of an individual elephant.* During the follicular phase, approximately 18-20 days prior to ovulation, a 1-day LH-peak concentration can be distinguished, which may result in a temporary rise of progesterone. A second LH-peak occurs just prior to ovulation (see figure 1).

To monitor the estrous cycle in elephants gestagens should be determined in blood (progesterone) or urine (pregnanetriol). Frequency of sampling should be once a week.

Note: when using the urine test, one must be aware that the pregnanetriol concentration in the urine is always compared with creatinine. If the creatinine level is too low, a new sample should be submitted. Even when monitoring the cycle by urine-pregnanetriol, much effort should be made to train all elephants to allow blood sampling.

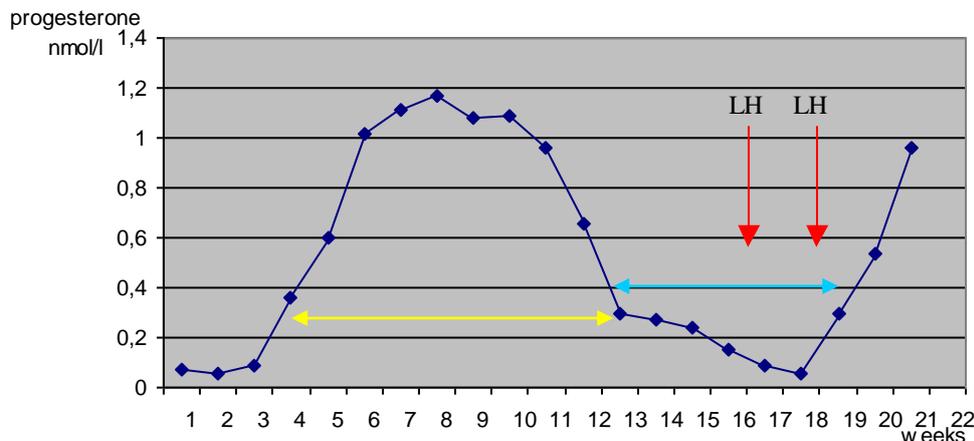


Figure 1. Progesterone blood concentration in nmol/l during an estrous cycle in an elephant. Note the 2 LH-peaks.

←→ : luteal phase  
←→ : follicular phase

### Measuring of blood progesterone in elephants:

Monitoring the progesterone concentration pattern in blood and/or pregnanetriol in urine is relevant for:

1. Determination of the right moment for mating
2. Pregnancy confirmation
3. To determine the moment that progesterone production has stopped at the end of pregnancy. In this case a quick result is needed in order to be able to act accurately.

Therefore, measuring the progesterone levels at the end of pregnancy is preferred in blood rather than in urine, because the number of laboratories that can measure (urine) pregnanetriol unfortunately is limited to one (Primate Center in Göttingen, Germany). When blood samples are submitted for the determination of blood progesterone, it is essential to be aware of the differences in the outcome. The most accurate results are obtained by using a Radio Immuno Assay (RIA). However, not many laboratories using this technique will be able to provide a daily service. Tests that are based on other immunological reactions (EIA/ELISA) are routinely used in human hospitals but they need to be validated for elephants before the results can be trusted. Figure 2 shows the differences in test results obtained by using different machines and different test units (1 nmol/l corresponds with approximately 0.32 ng/ml).

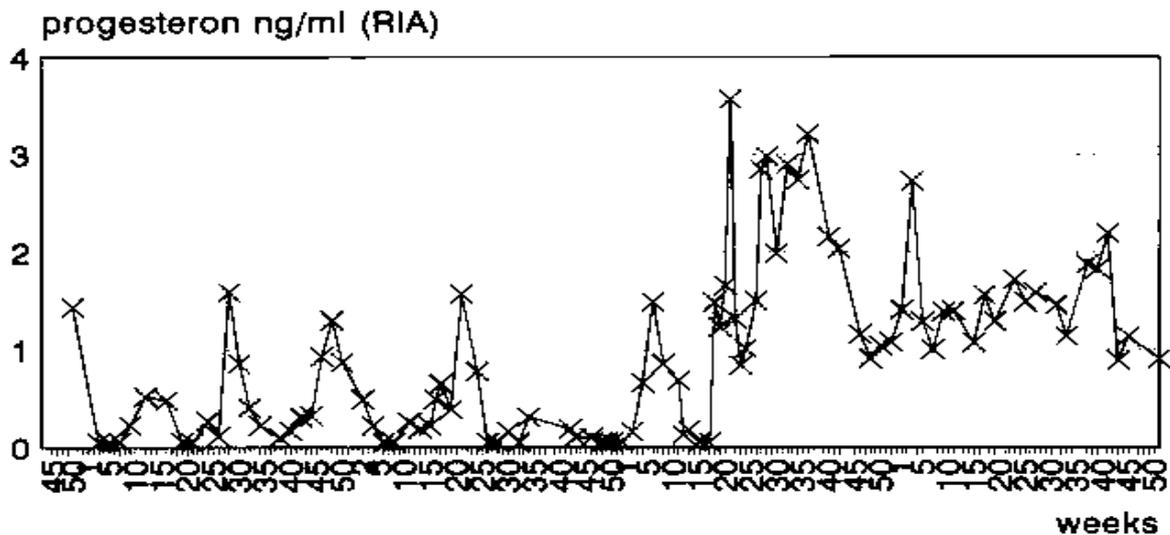


Figure 2a. Blood progesterone concentration in an Asian elephant, determined by a RIA. Seven follicular phases, six complete luteal phases and the major part of the gestation period are clearly identifiable. Note that two of these luteal phases express themselves in relatively low progesterone concentrations. This lab measured the progesterone concentrations in nng/ml..

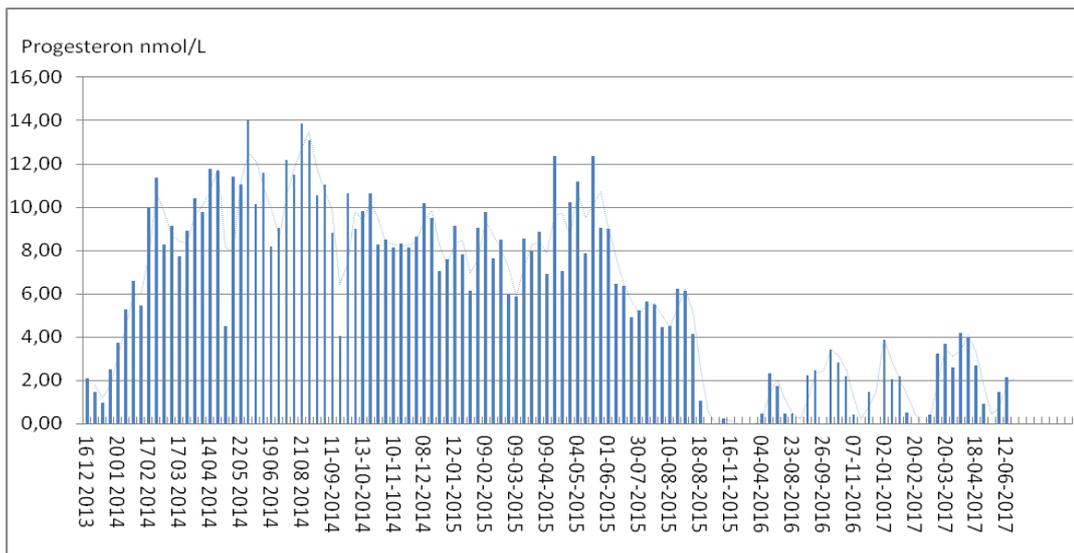


Figure 2b. Blood progesterone concentration in an Asian elephant, determined by an ELISA ( ADVIA Centaur CP – Siemens ). This graph clearly shows the terminal part of the gestation period with a sharp drop of progesterone (within 48 hours) to the baseline value specific for this elephant. The calf was born within 48 hours after the progesterone concentration had dropped to base line value.

**Progesterone base line values:**

The progesterone concentrations that are found during the non-luteal phase in elephants are often too low to be exactly measured by most techniques used currently. Some machines can calculate these values, rather than measure them. These low levels are found during the follicular phase, at the end of pregnancy and during the lactation and “menopause” anoestrous. The turning-point between luteal and non-luteal phase is what we call the “base line” value. The height of this base line value is rather arbitrary, but should be estimated for each individual animal. It should also be determined in relationship with the type of the assay used. Figure 2b and 2c make clear that the absolute values given by the laboratory may significantly differ between the machines that are used. This has already resulted in wrongly interpreted values: a progesterone concentration of 4 nmol/l was found in an a-term Asian elephant. As no blood samples were taken previously, this single value was considered a high concentration of progesterone. However, in reality it was a baseline value for this particular elephant measures on this particular machine. No action was taken as this signal of a stagnating calving process was not recognized. Consequently the calf in this case was born dead.

**Preventing pathologic conditions:**

- Elephants kept in zoos generally enter puberty at a much younger age than in the wild. This may have negative consequences for the reproductive organs. The occurrence of endometrial or ovarian cysts (cystic hyperplasia, frequently found in African and Asian elephants) or benign myometrium tumors (leiomyomas, exclusively found in older Asian females) may be the result of (normal) cyclic changes in the reproductive organs, if the cycle is not “rewarded” with pregnancy. To avoid these pathologic conditions, zoos should focus on a husbandry system that delays puberty as much as possible. The best way to achieve this condition is by raising young elephants (especially females) in intact families which may contribute to a natural way of social depression. The oestrous cycle should be monitored starting when the animal is around 3 years old. To reduce the risk of future pathological conditions in the reproductive organs, mating should take place when the young animal has gone through 2 years of normal oestrous cycles at maximum.
- For the same reasons as described above, the inter-calving period should be restricted to maximal 7 years.

**Oestrous period and pregnancy:**

- The occurrence of mating is not an indicator for impending ovulation; some animals mate even during advanced pregnancy or outside the estrous period.
- Fertilization can only result from mating during estrous preceded by the pre-ovulatory (=second) LH-peak. Monitoring LH needs daily blood sampling for at least 10 days (A test kit is available presently: LH ELISA KIT, which can be ordered from Lenora Bruce, University of California Davis, Central Storehouse Receiving, California and Larue, Davis CA 95616 USA, phone +1-530-752-0663)
- The fertile period is restricted to the time around the second LH-surge, which occurs immediately prior to the rise in progesterone. For the prediction of the next ovulation, follicular phase length has to be determined and therefore it is important to know the moment of the preceding fall of progesterone/pregnanetriol. For reliable accuracy, weekly samples throughout the cycles have good predictive value.
- LH monitoring daily during the non-luteal phase is necessary for artificial insemination programs. Determination of the post-luteal phase (=first) LH-peak can help predict the first

opportunity for (natural) breeding, as the interval between both LH-peaks is rather constant.

**Pregnancy confirmation:**

- Immediate increase of progesterone/pregnanetriol around the time of mating is suggestive for the right timing for breeding.
- Continuation of high progesterone/pregnanetriol level continuing for at least 16 weeks after mating is highly suggestive of pregnancy.
- Transrectal ultrasonographic examination at 8 - 20 weeks after mating allows visualization of single or twin calves. Between 10 and 20 weeks, the larger mature animal may need to lay down on its side for reliable ultrasonographic examination of the uterus. Uterine vascularization can be visualized to determine viability of the fetus, to exclude embryo absorption and mummification.
- Transcutaneous ultrasonographic examination (both flanks have a small "window" where visualization of fetal movements may be seen) may help to determine the status of the fetus in the last months of pregnancy.
- At 6 months post-breeding pregnancy can be confirmed by elevated serum prolactin concentration. When submitting a serum sample to a lab (some human hospitals have a prolactin assay), this should always be accompanied by a serum sample of a non-pregnant elephant for comparison.

**Behavioral observation at the end of gestation:**

- 24 hours observation including the use of a (time-lapse) video recorder starting in week 85 may add to information about relevant events prior to parturition: night pacing, kneeling down, climbing, short periods of separation from the group, beating the vulva with the tail, frequent production of small-sized feces and small quantities of urine, loss of mucous plug, playing with mucous plug, rupture of the allantois sac, labor waves.

**Predicting the time of parturition, measures and observations:**

Daily blood sampling from week 89: every other a day progesterone assay, until week 91 (637 days). From week 91 (637 days): daily assays and as soon as progesterone starts to decrease: sample twice daily.

Daily monitoring of progesterone is only possible if you have a nearby facility that runs these assays on a daily basis. Find out from your nearby hospital in the early stage of pregnancy! Many veterinary labs do not have tests that are sensitive for levels of progesterone (P4) in elephants 1-3 days prior to delivery. Also check the availability during the weekend.

NOTE: a decrease of progesterone to baseline level is not always occurring. A 23-yr-old Asian elephant at the Rotterdam zoo (with a baseline progesterone level < 0.6 nmol/l) delivered a healthy calf while her progesterone remained above 0.65 nmol/l.

Signs of an approaching parturition may include:

- Loss of mucous plug (not seen in many facilities)
- Pre-and post-parturition ventral edema may be noticed.
- Group members may react differently (vocalizations, restlessness)
- In most cases (75%) rupture of allantois sac and loss of allantois fluid (looks like urine) is seen within 2 hours prior to birth.
- The size of fecal balls may get smaller towards the end of pregnancy.
- Frequency of urination may increase around parturition, resulting in more "watery"

- consistency of the urine, resembling more like allantois fluid .
- Development of mammary gland and production of milk shortly before birth is often seen, however this may also occur in a much earlier stage of pregnancy . Milk accumulation can be visualized using transcutaneous ultrasonography several hours prior to parturition.
- Softening of the pelvic ligaments (due to estrogen surge) may result in slightly abnormal locomotion of the hind legs.

#### **Parturition/Preparations for calving:**

- Training and/or desensitizing of the pregnant elephant for veterinary intervention, like blood sampling, injections, IV-infusions, milking and rectal manipulations
- If possible, store some colostrum (freezer) or store plasma obtained from the dam in weeks prior to parturition.
- Have artificial milk available (Salvana GmbH, Germany; hand raising has been done at Emmen Zoo and Berlin Zoo)
- Check restraint chains and fixation points for the legs and one extra fixation point between the hind legs for pulling devices. Soft ropes for pulling the calf away if needed should be available. The use of a calf harness has been described. Check the stable and place bars where a calf could possibly escape. Block all possible escaping routes for the calf (not for staff!!).
- Take out all obstacles.
- Be prepared for closing the elephant house for the public (sign post, etc.)
- Make sure there is a good stock of commercial cat litter or saw dust to be used on a concrete floor as soon as the calf is born. This will absorb much of the allantois fluids and prevent the animals from slipping on the wet floor.
- 2 or 3 pairs of keeper-gloves (soccer) to get a better grip on the wet, slippery calf when needed
- Plastic hose pipe (with pump, if necessary) for rectal cleaning with lukewarm water
- 3 birth-chains with proper handles (2 for the legs, 1 for trunk or tail); find a way to avoid back sliding when manual extraction (vaginal vestibulotomy) is required.
- Drugs to be kept in store:
  - Ca-borogluconate for I.V. infusion
  - Estradiol gel (EstroGel® 0.06%)
  - Oxytocin
  - Lidocain
  - Xylazine, detomidine or medetomidine
  - Butorphanol
  - Azaperone
  - Atipamezole
  - Doxapram
  - Oxygen
  - (Betadine®-)iodine solution for navel disinfection (umbilical infection is a major cause of perinatal complications)
  - Lubricant (many liters). J-lube®, a concentrated lubrication powder, has proven to be very useful.

The normal calving process should take place within 2 hours after rupture of membranes (release of fetal fluids).

If the plasma progesterone level decreases to below base line concentration, this is a signal that calving should take place within 48 hours. Prolonged intervals have been reported (as long as

14 days) still resulting in the birth of a living calf, but it is very likely that this is due to a disturbance of the normal birth process that should be corrected before the health of the calf is jeopardized. There is one report of a parturition taking place without a complete drop to base line level.



The normal calving process in elephants is a family happening. All efforts should be directed to making this possible. Even when veterinary intervention is required



## II. Guidelines for veterinary assistance around the parturition time

Preceding any recommendation in this chapter, the following remarks need to be made:

1. It is a myth to think that a multiparous elephant does not need to be monitored and assisted during parturition according to this protocol. Too many calves have been born dead or very weak because of the fact that parturition had started unnoticed and stopped unnoticed. In our view, this protocol should be followed in ALL cases of elephant parturition as much as the elephant management allows.
2. It is a myth that oxytocin is a harmless drug to be used in elephant parturitions. In at least 3 cases the administration of oxytocin has been associated with the occurrence of an uterus rupture because of insufficient cervix dilatation. It should never be used without ultrasonographic examination of the cervix uteri. The visible presence of the allantois sac as a subcutaneous bulging mass under the tail is not a guarantee for a sufficiently dilated cervix.
3. Many elephants, including very experienced multiparous elephants may just stop the parturition process, often unnoticed. This will compromise the health of the calf. Don't relay only on what you see from the outside! For a proper judgment about the presence of labor activities or the progress of parturition, your professional judgment should be based on the results of progesterone tests and ultrasonographic examinations.
4. If you do not believe in the above mentioned statements, you may find yourself confronted with a dead elephant calf or a dead elephant mother or both. So, discuss this item with your staff and make your decision before you start breeding your elephants.
5. If the management of elephants in your zoo does not allow blood sampling or ultrasonographic examinations, you must be aware of the risks associated with a silently interrupted birth process, no matter the reproductive experience of the animal involved. Safety for the personnel should never be challenged by unacceptable risks.
6. Finally, the calving process is a natural process. Elephants should give birth in their own social environment, excluding any external disturbing factors (e.g. noisy building in the neighborhood, visits of unknown individuals, etc.). With a well trained animal, blood sampling and ultrasonographic examinations can be done while the animal is temporarily separated from the group. Immediately afterwards, the animal should return to its group. All efforts should be made to make sure that the calf is born in the group while the mother is NOT chained! This will stimulate the acceptance of the calf by the mother and group members and is an investment for future breeding successes for the entire elephant group.

To determine the right moment when calving starts, 2 parameters are essential: the progesterone blood level and the relaxation of the cervix, monitored by ultrasonographic examination.

Progesterone: the sensitivity of the equipment and the time needed to run the assay are the bottleneck for using the progesterone concentration as a reliable tool. Today many human hospitals use advanced equipment with a very low detection level that can provide results in less than 2 hours. Make sure that you have made arrangements with a lab long before you expect the parturition.

Ultrasonography: to use this technique as a reliable tool, it is indispensable for the veterinarian to gain experience long before the elephant birth is expected. This will enable the veterinarian to distinguish a normal cervix (fig. 3a - d) from the relaxed cervix (figure 4a + b) from the normal cervix. Preferably a 3.5 MHz probe should be used transrectally.

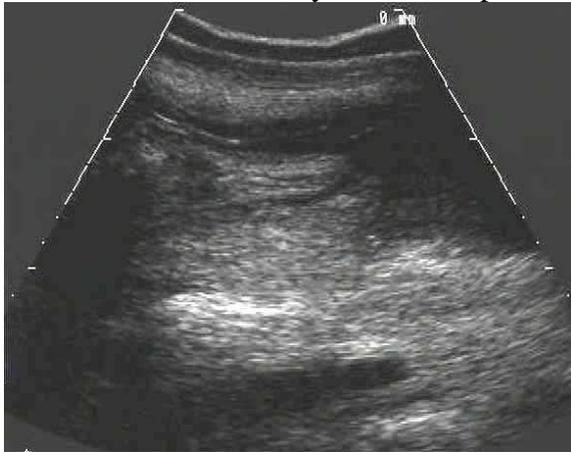


Figure 3a. Longitudinal, transrectal ultrasonographic image of the vagina and the closed cervix of a pregnant elephant.

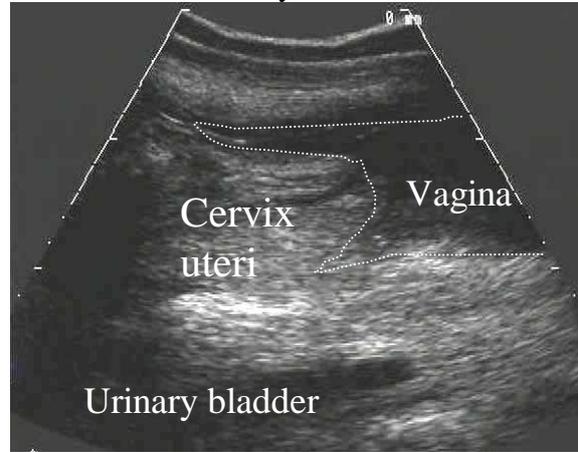


Figure 3b. The same image as figure 3a with the vagina and cervix indicated with white lines.

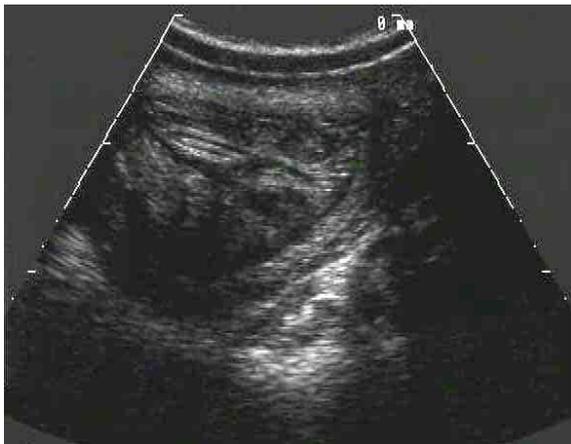


Figure 3c. Transverse, transrectal ultrasonographic image of the vagina and the closed cervix of a pregnant elephant.

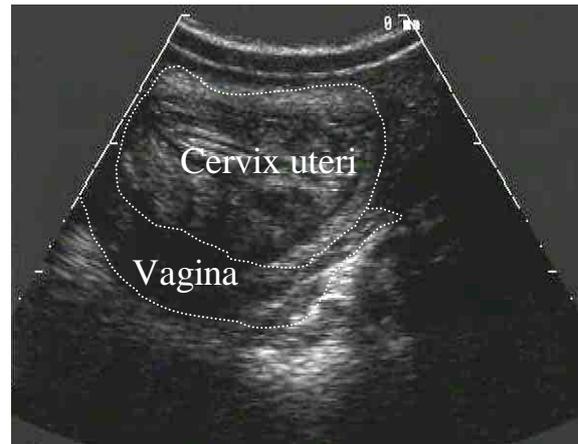


Figure 3d. The same image as figure 3c with the vagina and cervix indicated with white lines. Note the folds in the cervix uteri.



Figure 4a (transverse) and 4b (longitudinal). Allantois sac with cloudy fetal fluid in the (partly) dilated cervix of an Asian elephant 12 hours prior to delivery (white arrows: allantois sac, open arrow: pelvic bone)

During the last 2 weeks of gestation, the mucous that is present in the vagina during gestation will be discharged gradually. This is a clear indication for a pending parturition.

Recognizable onset of parturition occurs normally within 24-48 hours after progesterone has dropped to below base-line level. In these guidelines the absence of visible signs of parturition in the same time frame is considered an abnormal condition; this status requires veterinary intervention. At this point 2 situations may occur: the parturition process has started but has been interrupted without or with spontaneous rupturing of the allantois sac.

### **1. No rupture of allantois sac noticed:**

If the calf is not born by natural way 24 hours after blood progesterone has dropped to baseline level, rectal palpation and ultrasonographic examination of the cervix is highly recommended. This will demonstrate the rate of relaxation of the cervix, the presence of the allantois sac or parts of the fetus in the cervix or vagina and should be repeated at least every 8 hours.

A blood sample should be taken to measure the calcium level. If below 2.5 Mmol/l, calcium should be administered as an IV-infusion (NB: when given in an ear vein, it should be given strictly IV in order to avoid damage to the vein) or orally (suggestion: calcium syrup concentrate for human use, enveloped in the carton core of toilet paper, covered and sealed with fresh tamarind paste has worked well; most elephants will eat it, including the carton material). The effect of the calcium administration should be confirmed by rectal palpation (increase of contractibility of the uterus) and determination of the blood calcium level. Store an EDTA and heparine sample for herpes virus diagnostic purpose (both cells and plasma in freezer after separation).

Transrectal ultrasonographic examination at 48 hours:

A. No relaxation of the cervix at 48 hours: search for calf movements and nail position of the fetus (palpation and ultrasound) and blood flow in fetal vessels (ultrasound).

Transcutaneous ultrasonographic examination (both flanks have a small "window" where visualization of fetal movements may be seen) may help to determine the status of the fetus. Apply estrogens rectally. Good results have been obtained by the rectal and transdermal (perineum) application of an estradiol containing estrogen gel (Estrogel 0,06%, total dose 700-800 mg estradiol). The effect on the cervix dilatation should be monitored closely by transrectal ultrasonography! At this time, at least 1 hour after the local application of estradiol, rectal massage should be applied to test and stimulate the contractibility of the uterus.

Technique: remove feces from rectum, flush out the rectum, use abundant lubrication, keep both gloved hands (NB: the rectal mucosa is vulnerable due to estrogens) with the fists joined in a firm grip and press with the wrists or the dorsal sides of the hands against the pelvic ring to stimulate the pelvic receptors until strong labor waves appear or at least 10 minutes. When labor waves occur, continue this massage for 3 hours (if needed change operator). Check regularly by means of ultrasound the condition of the cervix. If there is still no cervix relaxation, continue monitoring the viability of the calf. If no fetal parts can be detected, consider the presence of pseudopregnancy (ovarian tumor, dysfunction of the pituitary gland, etc.). The application of estradiol gel (total dose: 400-500 mg 17- $\beta$ -estradiol) as described above may be repeated 3-4 hours after the first treatment if the cervix dilatation is still incomplete.

B. Partial or complete relaxation of the cervix (figure 4) at 48 hours or later as a result of the situation described under 1A:

Apply rectal massage to test contractibility of the uterus. If limited or no reaction, the administration of oxytocin is contra-indicated. In this case, the administration of calcium is recommended (even when blood calcium level is within normal ranges). After 2 hours the use of estradiol as described under 1.A is recommended.

Only if uterus contraction can be provoked by the rectal massage, the use of oxytocin may be considered using the following dosage: 25-50 IU oxytocin s.c. or i.m. (if needed use a blow dart).

*Oxytocin should be used with care, as it may dramatically exhaust the contractibility of the uterus muscles as well as the general condition of the female. There might also be the risk of reduced blood circulation in the umbilical chord, due to the spasms in the myometrium.*

*Prostaglandin E (dinoprost) has been used on a few occasions for cervix dilatation (after the administration of estradiol) and to stimulate uterus contractions. As there is still limited knowledge of its efficacy after transrectal administration and the risk of stormy uterus contractions, it should only be used when any obstructions or abnormalities of the calf can be ruled out.*

Depending on the progress obtained, rectal massage and the administration of oxytocin are the 2 major treatments to follow from this point. In between these treatment events, the animal should be exercised to relieve the pain and stimulate position changes of the calf and preferably it should be kept in the group. Only when the animal cannot be separated whenever required, the cow should be kept separated from the group, but with as much physical contact as possible. Oxytocin should be given in intervals of at least 2 hours for a maximum of 12-24 hours under the guidance of ultrasound to evaluate the progress.

Continue this approach of treatment until parts of the calf have entered the pelvic cavity. If the efforts remain unrewarded and no access to fetal parts is possible, not much can be done. Continue monitoring the viability of the calf. If the calf has died, while the membranes are still intact, the risk of intoxication is limited, but immunosuppression could be a complication for the cow. To date, no proper data are available.

The dosage of oxytocin may only be increased to 100 I.U. after parts of the calf have entered the pelvic area and progress is clearly observed. At this time, a bulge containing parts of the body under the tail of the dam should be visible. Progress of parturition must be monitored strictly at this stage. If this increased dose of oxytocin does not result in parturition a vaginal vestibulotomy should be performed soon to get better access to the calf. See next chapter. Expulsion of the calf should follow soon after the bulge appears under the tail of the dam. The allantois sac usually ruptures during the (induced) passage through the pelvic canal. One complication described at this stage, is reduced passage space as a result of edema in the urogenital canal resulting in a “catching effect” of the head and/or shoulder of the calf inside the soft part of the distal (vertical part) genital tract. Suffocation of the calf is a realistic complication. This condition has been observed several times in primiparous elephants of more advanced age. Elephants in this category should be prepared by the local application of J-Lube in the distal part of the vaginal vestibulum and massage of oestrogen/creme in the skin between anus and vulva. If this condition occurs, quick intervention is required by applying firm manual pressure from the outside on the calf in the sliding direction of the calf. Be aware of the risk of kicking by the mother.

## **2. Ruptured allantois sac:**

A significant event in the parturition process is the rupturing of the allantois sac, which – when intact - acts as a hydraulic dilatator for the cervix, a natural lubrication for the dam and a pressure protection for the calf.

*NB: The amniotic sac that covers the body directly, usually remains intact during the expulsion of the calf and ruptures during the final passage through the birth canal and is actively removed by the dam.*

*NB: a chained dam, may not be in the position to remove these membranes, possibly resulting in suffocation of the calf.*

Differentiation between urine and fetal fluids is extremely difficult; smell, creatinin test strips and possibly protein concentration could be helpful.

If no progress in parturition is observed, major complications should be considered, like a dead calf, malposition of the calf (which is often dead), oversized calf, malformation and twin pregnancy. Because of the urgency of this situation, the calf should be born within 2 hours after rupture of the allantois sac and loss of allantois fluid. If not so, veterinary intervention has to take place. Two situations may occur:

No fetal parts positioned in the pelvic area: treatment should aim on the urgent relaxation of the cervix. Calcium status should be determined and treated accordingly (see above). The further approach is according to 1A, however the situation is more critical for both the dam and the calf.

Fetal parts have entered the pelvic area: Calcium status should be determined and treated accordingly (see above). Ultrasound is essential to determine which fetal parts have entered the birth canal, determine the position of the calf (visualization of the nails, posterior or anterior position, number of nails, trunk) and viability.

Malposition (e.g. only one leg in birth canal, no head while in anterior position) is an indication for vaginal vestibulotomy or fetotomy.

If no abnormalities are found during ultrasound, 50-100 I.U. of oxytocin should be given i.v. or i.m. and rectal massage should be practiced. Birth should be completed within 1 hour.

### **Other drugs used**

Uterine laxants have been used in elephants on rare occasions. There are some anecdotal reports about the use of denaverinehydrochloride (Sensiblex®, Veyx) at a dose of 0,04 – 0,05 mg/kg BW (i.m.). Isoxsuprinelactate (Duphaspasmin. Fort Dodge Animal Health, 6290AA Vaals, The Netherlands) was used in a fetotomy case at 0,15 mg/kg BM (i.m.). Carbetocine (Depotocin®, Veyx), a long acting oxytocine has been used in 3 occasions dosage (0,09 µg/kg BW) i.m.

### **Post-partum care**

Disinfection of the navel with Betadine® iodine is strongly recommended (if the mother allows its application).

The afterbirth usually comes off within 12 hours. There are a few reports on retained (parts of) placenta for several weeks, without major complications for the dam. Hygienic measures should be applied to reduce the infectious burden for the calf.

Be aware of the fact that a second calf can still be present in the dam. There are reports that

second calves were born between several hours up to 3 months (the prevalence of twins in elephants is 1:3000).

The calf should drink (colostrum) as soon as possible, at least within 24 hours. If not, or when the calf makes a weak impression, the banked serum (or freshly taken serum) should be given to it orally. Try to find out the reason why the calf is not drinking successfully: e.g. too small, weak, painful mammary glands, malbehaviour of the dam.

If for any reason the mother is rejecting the calf, lactating herd member can take care of the calf. There is evidence that the mother takes over from this surrogate mother during the first 72 hours. If no lactating elephant is present in the herd, training for bottle feeding should start after 12 hours and continued for 72 hours. If the mother still rejects her calf, the best option for the calf is to move it to another herd with a lactating elephant. If introduction to this herd fails too, bottle feeding is the last option. To date very few bottle-raised elephants have reached the age of puberty.

### III. Vaginal vestibulotomy in elephants

Vaginal vestibulotomy is a surgical procedure, in which the vertical part of the uro-genital tract (vestibulum vaginae) is exposed by a percutaneous approach.

Indications for a vaginal vestibulotomy:

- No progress in calving despite of treatment according to the guidelines for veterinary assistance around parturition.
- Malposition

Contraindication for a vaginal vestibulotomy:

- No fetal parts in the birth canal (confirmed by ultrasonographic examination and rectal palpation)

Preparations:

- A. 1 extra ring in the floor between the hind legs should be present in any calving box for elephants. A pulley can be attached to it in order to provide optimal conditions for pulling the calf out (in a ventral direction) if needed. If this is no option, the calving chains (after passing through the entire vestibulum vaginae) can be attached to a long steel pipe or wooden log. Traction on the chains can be realized if 2 persons on each side of the elephant push this device downwards.
- B. If not strictly needed, don't use any sedation in the cow, as you will need the cow's straining support during the extraction. If the animal is completely hands off and restraint in a chute or otherwise is not an option, use a reversible sedation (xylazine, detomidine or medetomidine/atipamezole, preferably in combination with butorphanol) to chain the elephant on 4 legs. The sedation should be antagonized (if safety permits!) as soon as straining support from the elephant is needed. Remember that the behavior may have changed during this phase of the calving most likely in favor of the veterinary intervention: the animal is probably more interested in getting the calf out than in attacking the keepers around her. But there is not sufficient information about this point. The situation has to be evaluated for each animal separately.
- C. Use local anesthesia only. The incision side should be injected subcutaneously and intracutaneously with lidocain in sufficient deposits to anesthetize the entire incision site (4-5 injections of 20 ml lidocain over a distance of 20 cm) under the tail. The use of epidural anesthesia is recommended in order to reduce the movements of the tail and to support the local anesthesia. Lidocain can be injected easily with a long hypodermic needle in the intercoccygeal space. The depth of the epidural space lays approximately 6.5 cm below the skin surface. To localize the best insertion place, the tail should be moved up and down. The intervertebral space that lies in between the most cranial coccygeal vertebrae that can be moved should be palpated. The needle is inserted here making an angle of approximately 60° in cranial direction. In a 3000 kg elephant, 30 ml of lidocain was sufficient to induce relaxation of the tail. The movements of the tail were minimal while the animal remained standing. A higher dose might have been tried in this case, but was not considered necessary.
- D. After cutting through the skin, place a flexible plastic tube (5 cm diameter or a rumen tube) retrograde into the vestibulum vaginae to locate the incision side. To facilitate orientation, a 10x2 cm "window" should be made in the tip of the tube, 3 cm from the end.

Once the skin has been incised, this window can be palpated through the subcutaneous tissue structures including the vaginal vestibulum which can be easily incised over this “window”. The length of the incision should not exceed 15-20cm. This should allow the passage of your arms and the chains for pulling the calf.

- E. Use normal calving chains. Don't use more than 3 people on each leg. Pulling to advance the calf in caudal direction should be done through the incision wound (a “calving pulley” used in cattle has been used successfully). Once the legs of the calf have reached the wound, the chains should be advanced through the vaginal vestibulum and pulling should be realized in downwards direction allowing the passage of the calf to take place through the natural birth canal.
- F. When the calf is in posterior position and can't pass through the pelvic girdle, try to rotate the calf (90° longitudinal axis) during extraction. In cattle, active rotation is the normal way of a live calf to pass its pelvis through the pelvic canal of the mother. Figure 5 (Rotterdam Zoo, 1998) shows the birth of an elephant calf after stagnation of the delivery caused by hypocalcemia, demonstrating clearly that the calf has rotated 90°. This had happened already at the time both hind legs could be palpated in the vagina. When the calf is dead, the absence of this phenomenon might be responsible for stagnation of the calving process. In the experience of one of the authors (W.S. Rotterdam Zoo, 1993) it was only possible to remove a dead calf in posterior position by vaginal vestibulotomy after 90° rotation of the calf (figure 6). Please note that at that time the calf was delivered through the surgical incision as it was not attempted to pass it through the normal birth canal.



Figure 5: Birth of an Asian elephant calf, showing the sideward rotation of the calf. (hypocalcemia, Rotterdam Zoo 1998)



Figure 6: Vaginal vestibulotomy in an Asian elephant. Extraction was only successful after 90° rotation of the dead calf. Note the umbilical chord twisted twice around the hind leg. (Rotterdam Zoo, 1993).

- G. Don't hesitate to push the calf back in the direction of the uterus; it might return in a better position. During this action, both legs and the trunk or tail should be well connected to the chains. Refill the genital tract with at least 5 l. of lukewarm water (“artificial embryonic fluid”) by using a pump (aquarium-type). Lots of lubricants (J-lube) should be used.
- H. Don't pull at both legs in one direction at the same time. You must cross the chains and pull alternatively on each leg. It is more likely that each leg will pass the pelvic canal bit by bit, rather than that the two legs will pass in simultaneously.

- I. When the calf is out give 50 IU of oxytocin i.v., flush the uterus with at least 100 l of 0.09% saline water (add 90 g kitchen salt to 100 l of tap water and use an aquarium pump with a clean hose pipe for flushing), preferably until the placenta has come off.
- J. At this time (only if needed) the cow can be given a xylazine sedation (if sedated and reversed previously, use azaperone)
- K. On 2 occasions a long balloon catheter was inserted into the urinary bladder and stitched to the internal wall of the vestibulum vagina. In both cases, the catheter came out within 3 days.
- L. Suturing the wound is very questionable. In all known cases the wall of the vaginal vestibulum was closed in 2 layers with PDS, Vicryl or another resorbable material. In all known cases the sutures ruptured through the wound edges within 2 weeks. Repeated suturing has been attempted in most cases and has finally resulted in closure of the wound, which may take more than 1 year. In one case (after a fetotomy) a huge urinary fistula developed after 10 days located between the skin and the vestibulum vagina that drained into the outer lip of the vulva. The manipulations of the arms and chains through the wound irritate the wound edges which also does not favor natural adhesion of the sutured wound edges. Although it seems contrary to any veterinary principles, after having performed 2 vaginal vestibulotomies, it is my (WS) strong advice to initially encourage secondary healing without any suturing of the vestibulum vagina or the skin. Post-operative wound care should consist of daily flushing of the wound and the vestibulum vaginae with 50-100 l. of 0.09% saline water (900 g kitchen salt in 100 l. tap water) using a clean silicon or plastic tube fixed on a small aquarium pump. This gives minimal discomfort to the animal and will result in diminishing of the wound size by retraction of the connective tissue. In the advanced stage of natural wound healing that will certainly take place, the epidermis may adhere completely to the mucosa of the vaginal vestibulum (figure 7). In that case splitting these 2 layers from each other under local anesthesia is required in order to enable further wound healing per secundam.



Figure 7: mucosa and epidermis are fused together, impeding further wound healing

M. Reverse the sedation, wash your hands, clean up the mess and have a beer. But **DON'T CLOSE THE SKIN**. Forget everything you learned about surgery. Whatever fancy suture technique you may use, the skin wound will open again. Don't think that your special suture will work, because every possible suture technique has already failed. Even when closing the skin, feces will still contaminate the wound, because the skin property does not allow a watertight wound closure; suturing the skin has proven to result in discomfort for the elephant, the need to repeat the procedure several times and finally a permanent fistula just as large as in case the skin was not sutured. The skin will heal per secundam; periodic refreshing of the wound edges may be required during the healing process.

N. Give antibiotics during at least 7 days.

O. If you were not able to remove the calf: **DON'T TRY TO PERFORM A CESAREAN UNLESS YOU ARE ABSOLUTELY SURE THAT THE COW WILL NOT SURVIVE CONSERVATIVE TREATMENT**. All cesareans performed till now have resulted in the death of the mother. If you see yourself facing the critical situation that the calf can not be removed despite of the vestibulotomy and fetotomy is either no option or it has failed, the first advice is to leave the wound of the vestibulotomy open completely and see what happens during the days to come. Flush the uterus frequently with large amounts of 0.09% saline water with a disinfectant like Betadine® iodine to prevent the occurrence of Bandl's rings that may cause necrosis of the uterus. An antibiotic treatment is recommended.

#### Fetotomy

Only very few cases have required fetotomy. Till today, only one elephant survived this procedure. If you decide that a fetotomy is needed, you are strongly advised to contact Thomas Hildebrandt and/or Willem Schaftenaar.

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