THE JAGUAR HEALTH PROGRAM MANUAL

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Field Veterinary Program

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(Revised)

Field Health Manual Guidelines

The main objectives of the Jaguar Health Program are: 1) to provide standardized methods to assess the overall health status of jaguars in the wild; 2) to determine disease threats to jaguars including both direct threats (e.g., infectious diseases - intraspecific and conspecific via domestic animals, livestock, other free-ranging felids, prey items) and indirect threats (e.g., habitat fragmentation and degradation that may increase disease risks); and 3) to provide recommendations, based on findings from the health assessment, for the long-term management and conservation of the jaguar.

The following manual is intended only for field biologists and veterinarians with previous wildlife experience that are working in association with the Wildlife Conservation Society (WCS) Jaguar Conservation Program (JCP) (www.savethejaguar.com). This manual is not intended for inexperienced workers. It has been developed by veterinarians with the WCS Field Veterinary Program (www.fieldvet.org). The purpose of this manual is to provide a standardized and safe and ethical approach to capture, handling, and sampling protocols to ensure that the Jaguar Health Program is carried out in a consistent fashion throughout the jaguars' range. The WCS Field Veterinary Program staff do **NOT** support the immobilization and handling of any wild animals by inexperienced personnel.

These guidelines have been developed to be "living documents" and will be updated annually as needed. In an effort to make the document more useful, users are encouraged to submit their comments to improve the layout, content, and/or presentation of the material as they work with these guidelines. Any errors or sections that are unclear should also be highlighted. Please direct these comments to the JCP Program Coordinator, Kathleen Conforti, at the Wildlife Conservation Society, 2300 Southern Boulevard, Bronx, NY 10460 – 1099 USA. Telephone: 718-220-2189; Fax: 718-364-4275; email: kconforti@wcs.org .

The highest priority for the Jaguar Health Program is the safety of the biologist and the safety andwelfare of the animal. It is strongly recommended that the field biologist consult with a veterinarian prior to field work. The veterinarians with the WCS Field Veterinary Program are available for consultation or training sessions for field personnel and consultation on immobilization, handling, and health surveys of jaguars through the JCP. Please contact the Program Coordinator, Kathleen Conforti, at the Wildlife Conservation Society, 2300 Southern Boulevard, Bronx, NY 10460 – 1099 USA. Telephone: 718-220-2189; Fax: 718-364-4275; email: kconforti@wcs.org.

All diagnostic and genetic testing will be performed at selected laboratories in the U.S. that are known for their expertise in non-domestic felids. The ability of the Jaguar Health Program staff and personnel to complete these tests successfully is contingent on the exportation of samples from various countries within the jaguars' range and their importation into the U.S. Jaguars are listed on CITES Appendix I, therefore a CITES import and export permit are required for transporting tissue samples into the U.S. The JCP is developing a blanket permit for these samples, however, in the meantime, it is imperative that appropriate research permits are in place **before** transporting any samples. Researchers that collect samples for health analyses should ensure that they are able to store samples in country if there are any delays in permit processing. Permits must be issued by both the importing and exporting countries. In most instances, an export permit will not be issued until a valid import permit is presented to the office issuing the export. The U.S. Fish and Wildlife Service's Office of Management Authority is the Governmental agency which currently issues CITES import permits for the United States. In general, written proof of permission to work in the host country is required when applying for an import permit. For more information on permit requirements, visit http://international.fws.gov/permits/permits.html or refer to permit guidelines on the Field Veterinary Program Website (www.fieldvet.org) under "Resources / Technical Pages".

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I. INTRODUCTION

One important component of the Jaguar (*Panthera onca*) Conservation Program is a standardized approach to health and disease surveillance of free-ranging jaguars. Threats to the health of wildlife from anthropogenic influences are often associated with increased contact that wildlife have with livestock, domestic animals and humans, as well as habitat fragmentation and contamination of their habitats. Although the specific health threats to free-ranging jaguars are largely unknown at this time, the situations listed above are believed to be of importance in the long-term conservation of free-ranging jaguars throughout their range.

Standard guidelines will ensure that this health program is carried out in a consistent fashion throughout the jaguars' range. Many field researchers are currently immobilizing free

ranging jaguars with little or no training in veterinary skills, such as anesthesia and biological tissue collection. This fact alone suggests a strong need for standard guidelines to ensure the safety and welfare of jaguars being handled in the field. The following guidelines are intended for field biologists and veterinarians with wildlife experience that are working associated with the WCS Jaguar Conservation Program. These guidelines are not intended for inexperienced personnel. The veterinarians with the Field Veterinary Program, WCS are available, through the Global Carnivore Program Manager, for training sessions for field personnel and consultation on immobilization, handling, and health surveys of jaguars, as a component of the Jaguar Conservation Program (see appendix 1).

The main objectives of the jaguar health program (JHP) are the following: 1) provide standardized methods to assess the overall health status of jaguars in the wild; 2) determine disease threats to jaguars including both direct threats (i.e., infectious diseases – intraspecific and conspecific via domestic animals, livestock, other free-ranging felids, prey items and indirect threats (i.e., habitat fragmentation and degradation that may increase disease risks); and 3) provide recommendations, based on findings from the health assessment, for consideration in long-term conservation policies for jaguars.

II. CAPTURE AND IMMOBILIZATION

The information provided in this manual is intended for use by field researchers with previous experience in performing felid immobilizations. Field Veterinary Program, WCS staff do NOT support the immobilization and handling of any wild animals by inexperienced personnel. It is imperative that persons handling jaguars consider the safety of humans and jaguars above all else before, during and after any jaguar immobilizations.

1. Standard format and recording of capture and immobilization techniques

All field personnel should use one standard form for recording capture and immobilization events. This will ensure that all researchers are collecting the same data for every immobilization so that these data can be compiled and compared. Ultimately, this will allow us to determine the safest and best immobilization protocols for free-ranging jaguars.

We recommend the use of the Medarks software program (ISIS, 12101 Johnny Cake Rd., Apple Valley, MN 55124 USA) for compiling the immobilization data. (This software could also be used for data collection related to all aspects of the health program.) A hard copy representing a medarks standard anesthesia form is provided in appendix 2.

2. Capture Methods

There are a variety of capture and immobilization methods for free-ranging felids (De Wet, 1993). Wilson et al. (1996) provide an excellent overview of capture methods for medium to large sized mammals. Methods that have been used to capture free-ranging jaguars include treeing the animal using dogs, padded foot-hold traps, snares (i.e., Aldrich snares) and cage or box traps. The later two of these methods may or may not include bait to lure the animal to the trap. Once the jaguar is treed or trapped, it can then be darted. It is best to not dart a jaguar above 5 meter high in a tree to avoid traumatic falls. A comprehensive overview of capture methods for jaguars is currently being developed.

3. Anesthetic administration

Anesthetic agents should only be administered to free-ranging wild jaguars using remote drug delivery systems (RDDS). There are a variety of RDDS available for the practitioner, nicely reviewed by Bush (1992) and Nielsen (1999). A blowpipe, or possibly a pole syringe, may be used for immobilizing jaguars in a cage, foot-hold trap or snare. In all other field situations, it is best to use a rifle or pistol (i.e., Telinject™, Cap-Chur™, Dan-Inject™) (Appendix 3). It is beyond the scope of these guidelines to cover the principles of all these products. The

practitioner must be familiar with the instrument he/she chooses for use in the field. Darting animals is always associated with some degree of risks. Serious damage to the animal (and human participants) can and does occur if inappropriate instruments are used and / or if instruments are used inappropriately.

Dart and needle selection is also important in preparing for a jaguar immobilization. Darts that are too heavy and needles that are too long / thick can cause serious damage on impact. (Damage is also possible if the charge of the dart or the charge of the rifle / pistol is too high.) Needles available include collared, plain and barbed. A collared dart is often employed during immobilization procedures because it remains in the animal and ensures total drug injection. Unfortunately, if the jaguar is not adequately immobilized and cannot be restrained, the dart will remain in the animal and may cause problems.

We recommend the use of 1.5 X 30 mm (18 gauge X 1 ¼ inch) collared needles for immobilizing free-ranging adult jaguars. However, if the jaguar is treed or trapped prior to darting, a non-barbed (plain) needle can be used. Non-barbed (plain) needles cause less trauma to the tissues but often do not remain in the animal as long as collared needles and thus may not inject all the drug prior to falling from the animal.

When darting a jaguar, it is safest to aim for the proximal (closest to body) region of a rear limb (Figure 1). Some researchers recommend darting the triceps region of the front arm. If the anesthesiologist elects to use the front limb, it must be remembered that the thoracic region and head are in very close proximity to the intended site. Serious harm can be inflicted on the cat if the dart hits one of these regions. It is for this reason that we recommend the hind leg unless the anesthesiologist is darting the jaguar at close range (i.e., in a box trap). When aiming for the rear limb, darts should be placed in the caudal most aspect of the muscle mass to avoid the femoral bone and the sciatic nerve. Needles and darts must be disinfected prior to use on the next animal to prevent the spread of diseases. Although disinfection is often the only available means of equipment care in the field, it is best to sterilize needles between animals.

4. Anesthetic Regimens

In this section, we provide a recommended "standard protocol" for use by those people in the field with minimal immobilization experience. This protocol should provide an adequate plane of anesthesia with minimal respiratory and cardiovascular compromise. Additionally, we list anesthetic regimens that have been successfully used in captive and free-ranging jaguars based on the literature and personal experience. Please note that drug doses in this handbook are given in milligrams (mg) and micrograms (mcg). **Doses are NOT in milliliters (except where noted in table on Page 7).** Drug administration route is intramuscular (IM) unless stated otherwise.

Calculating drug dosages – Persons using anesthetic agents to immobilize jaguars must be familiar with how to calculate the volume (ml) of drug to deliver based on the recommended dose (mg/kg) for the species and concentration (mg/ml) of the drug.

1. mg needed = recommended dose (mg/kg) X animal's body weight (kg)
2. ml needed = mg needed (see 1 above)) drug concentration (mg/ml)
   = mg needed X (ml/mg)

Example:

1. 90 kg jaguar
2. Drug concentration is 100 mg/ml
3. Recommended dose is 5 mg/kg

mg needed = 5 X 90 = 450 mg

ml needed = 450) 100 = 450 mg X ml / 100 mg = **4.5 ml**

Note: 1 kg = 2.2 lb (Recommended doses may be given in mg/kg OR mg/lb. Be sure to make the proper conversion. For instance, if the weight of the animal is estimated in lb and the recommended drug dose is given in mg/kg, then you must convert the dose.)

1 kg / 2.2 lb X mg / kg = mg / lb

A. Recommended "standard protocol" for the immobilization of free-ranging jaguars

Currently, we recommend (except for the immobilization of treed jaguars; see below) the following anesthetic regimen for use by field personnel with little experience in immobilizing free-ranging jaguars. This regimen should provide an adequate plane of anesthesia for shortterm work on the jaguar (i.e., radio-collar application, morphometric measurements, biomaterial collections) while requiring a minimal level of technical skill in anesthesiology.

Telazol 6 - 10 mg/kg intramuscular by dart as the dose for immoblization in free-ranging jaguars. The darter has the option to include 150 mg ketamine in the initial dart based on work by Cavalcanti and Hoogesteijn, unpublished data. Supplemental ketamine at a dose of 1-1.5 mg/kg, intravenous or 1-2 mg/kg intramuscular, as needed to maintain an adequate level of anesthesia. (No supplemental ketamine should be delivered for at least 10 minutes after the telazol dart.) Atropine at a single dose of 0.04 mg/kg either subcutaneous or intramuscular may also be administered if the cat has excessive salivation.

NOTE: Supplemental drug is defined as the administration of any additional immobilization drug delivered following the initial drug(s) (see II D below). Supplement drugs are necessary when the animal is only partially anesthetized from the original drug(s) and/or is getting light (i.e., anesthetic agents are wearing off) during the procedure.

TREED JAGUARS: The use of telazol in treed jaguars may result in the cat falling from the tree during induction. This has been documented with both treed mountain lions and jaguars using this drug (K. Quigley, personal communication). For this reason, it is recommended that jaguars captured by treeing with dogs are darted with ketamine (7 mg/kg) for initial immobilization. Once the jaguar is safely on the ground, xylazine (0.7 mg/kg) i.m. should be administered to improve muscle relaxation and analgesia. Supplemental anesthesia is provided by administering ketamine (0.5 – 1.5 mg/kg) i.v. or (1 – 2 mg/kg) i.m. The use of ketamine does not eliminate the potential of the cat falling. However, due to the retention of a higher level of muscle rigidity under ketamine, more time is available to get to the animal.

TELAZOL [100 MG/ML] – is supplied as 500 mg dry powder that is usually reconstituted with 5 ml of sterile water making a solution with the concentration of 100 mg/ml. Telazol is a combination drug that contains equal parts of a dissociative anesthetic, tiletamine (50 mg/ml), and a benzodiazepine tranquilizer, zolazepam (50 mg/ml). In some countries the same combination is called "Zoletil" rather than Telazol but the total amount of drug in the bottle may be different. The 250 mg bottle of Zoletil makes a solution concentration of 50 mg/ml when reconstituted with 5 ml sterile water. Telazol and Zoletil can be mixed with a smaller amount of sterile water to increase their concentration and allow a smaller volume of liquid to be used in a dart. For example, using 2.5 ml of sterile water in a 500 mg bottle of Telazol gives a concentration of 200 mg/ml. We do not recommend using this drug at a concentration higher than 200 mg/ml because it may not dissolve completely and cause unpredictable absorption, clogging of needles, and/or crystallization.

Amount (mls) of Telazol (100mg/ml) at a dose of 4-8 mg/kg based on jaguar body weight (kg).

BODY WEIGHT (KG)	AMOUNT OF DRUG (ML)
50	2-4
60	2.4-4.8
70	2.8-5.6
80	3.2-6.4
90	3.6-7.2

100	4-8
110	4.4-8.8
120	4.8-9.6

Visual observation and physiologic monitoring of the jaguar during immobilization is imperative(see III 2 below). Jaguars immobilized with telazol (and ketamine) usually will have increased salivation, open eyelids, whole body muscle rigidity (including jaw tone), and intact reflexes (i.e., corneal, pedal). Jaguars should maintain swallowing and coughing reflexes. There should not be muscle tremors and seizure-like activity.

B. Literature Review of Captive Jaguars - Captive is defined here as those jaguars held in zoos and other confined situations (i.e., captivity).

1. Telazol

a. 5 mg/kg telazol (with 2 mg/kg ketamine as supplement) (Kreeger, 1996)
b. 3.5 - 4.4 mg/kg telazol (Boever et al, 1977)
c. 2.0 - 4.0 mg/kg telazol (Shobert, 1987)
d. 3.5-4.4 mg/kg telazol (Shobert, 1987)
e. 2-4 mg/kg telazol (Gray et al, 1974) - reference notes an insufficient chemical restraint at lower dosage

Note: Telazol should only be administered in the initial dart/dose. Telazol should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures. Ketamine is the supplemental drug of choice in these situations. (See Anesthetic Supplementation below).
Note - Flumazenil is the antagonist for zolazepam (the benzodiazepine component of telazol) and can be administered, once all procedures are completed, at an intramuscular dose of 1.0 mg of flumazenil for each 20 mg of zolazepam used. Flumazenil should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that the

tiletamine component of telazol is nearly completely metabolized. There should also be at least 30 minutes between the administration of any supplemental ketamine administration and flumazenil.

2. Ketamine + Xylazine

a. 4 mg/kg ketamine and 2 mg/kg xylazine
   antagonist = 0.125 mg/kg yohimbine (Kreeger, 1996)

b. 15-20 mg/kg ketamine and 1-1.5 mg/kg xylazine or 1-2 mg promazine
   supplement with 3-5 mg/kg ketamine as needed and 2-5 mg diazepam for muscle relaxation (Seal and Kreeger, 1987)

c. 2-2.5 mg/kg ketamine and 2-2.5 mg/kg xylazine (Yates, unpublished data, 1999)

Note: Xylazine should only be administered in the initial dart / dose. Xylazine should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures. The sedative effects of xylazine are easily overridden with external stimuli (i.e., noise, movement). It is especially important that people remain calm and quiet while animals are sedated with xylazine.

Note: Yohimbine is the antagonist for xylazine and should be administered at 0.125 mg/kg IM 10 and should only be delivered once the procedure is completed and at least 30 minutes after the last dose of the cyclohexane (ketamine) was given.

3. Ketamine + Medetomidine

a. 2.5 mg/kg ketamine and 70 **mcg**/kg medetomidine (Kreeger, 1996) (**note mcg** = micrograms which is a 1000th of a mg)

b. 2.5 mg/kg ketamine and 60-80 **mcg**/kg medetomidine (Jalanka and Roeken, 1990)

c. 2 mg/kg ketamine and 40 **mcg**/kg medetomidine (Spelman, 1997 unpublished data)

Note: Medetomidine should only be administered in the initial dart / dose. Medetomidine

should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures.

Note - Atipamezole is the antagonist for medetomidine and can be administered once all procedures are completed, at a dose of 4-5 X the medetomidine dose. For example, if 40 mcg/kg of medetomidine was used for immobilization, reversal with atipamezole should be at a dose of 160 - 200 mcg/kg. This should be delivered intramuscularly. Atipamezole should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that ketamine is nearly completely metabolized.

4. Ketamine and Midazolam

a. 10 mg/kg ketamine and 0.25 mg/kg midazolam intramuscular followed by thiopental 4.4 mg/kg intravenous 15 minutes later for intubation and isoflurane anesthesia (McLaughlin and Kuzma, 1991)

Note: Midazolam should only be administered in the initial dart / dose. Midazolam should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures.

Note: Flumazenil is the antagonist for midazolam and can be administered, once all procedures are completed, at an intramuscular dose of 1.0 mg of flumazenil for each 20 mg of midazolam used. Flumazenil should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that ketamine is nearly completely metabolized.

C. Literature Review of Free-Ranging Jaguars

Note: many of the doses published in the literature are based on very small sample sizes (n) of jaguar immobilizations. Sample sizes are presented when known.

1. **Ketamine**

a. 10-12 mg/kg ketamine (Hoogesteijn and Mondolfi, 1992)

b. 7 – 40 mg/kg ketamine (Crawshaw, 1992) (n=9)

c. 22 mg/kg ketamine (Rabinowitz and Nottingham, 1986) (n=7)

2. **Ketamine and Diazepam**

a. 11.8 mg/kg ketamine and 0.25 mg/kg diazepam (Hoogesteijn and Mondolfi, 1992) (n = 2)

3. **Ketamine and Xylazine**

a. 3 mg/kg ketamine and 0.6 mg/kg xylazine with 0.05 mg/kg atropine (Hoogesteijn and Mondolfi, 1992) (n = 1)

b. 7 mg/kg ketamine and 0.5 mg/kg xylazine with 10 mg diazepam (Hoogesteijn and Mondolfi, 1992) (n = 1)

c. 10.6 – 11.5 mg/kg ketamine and 1.3 – 1.4 mg/kg xylazine (Lopez de Buen and Sanchez, 1986) (n = 2)

d. 11 mg/kg ketamine and 1 mg/kg xylazine (Quigley, 1987) (n = 8)

e. 6.6 mg/kg ketamine and 0.66 mg/kg xylazine (Kathy Quigley, unpublished data)

Note: Xylazine should only be administered in the initial dart / dose. Xylazine should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures.

Note - Yohimbine is the antagonist for xylazine and can be administered, once all procedures are completed, at a dose of 0.125 mg/kg intramuscular . Yohimbine should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that ketamine is nearly completely metabolized.

4. Ketamine and medetomidine

a. 1.46-3.48 mg/kg ketamine and 36-87 **mcg**/kg medetomidine reversal with 122-163 **mcg**/kg atipamezole (Hoogesteijn et al, 1996) (n = 2)

Note: Medetomidine should only be administered in the initial dart / dose. Medetomidine should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures.

Note - Atipamezole is the antagonist for medetomidine and can be administered, once all procedures are completed, at a dose of 4-5 X the medetomidine dose. For example, if 40 **mcg**/kg of medetomidine was used for immobilization, reversal with atipamezole should be at a dose of 160 - 200 **mcg**/kg, intramuscular. Atipamezole should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that ketamine is nearly completely metabolized.

5. Telazol

a. 4-8 mg/kg telazol (**recommended dose in this manual**)

b. 6.6-16.4 mg/kg telazol (Morato, et al, in press) (n = 11)

c. 10 mg/kg telazol (Morato, 1997)

d. 3.9 mg/kg telazol (Hoogesteijn and Mondolfi, 1992) (n = 11)

e. 3.5 – 9.1 mg/kg telazol (Crawshaw, 1992) (n = 6)

f. Note: Telazol should only be administered in the initial dart / dose. Telazol should NOT be used as a supplemental dose if the jaguar is only partially anesthetized after the first dart and/or if the jaguar becomes light prior to completion of the procedures. Ketamine is the supplemental drug of choice in these situations. (See Anesthetic Supplementation below).

Note - Flumazenil is the antagonist for zolazepam (the benzodiazepine component of telazol) and can be administered, once all procedures are completed, at an intramuscular dose of 1.0 mg of Flumazenil for each 20 mg of zolazepam used. Flumazenil should not be administered for a minimal of 30 minutes after initial anesthetic agents have been delivered to ensure that the tiletamine component of the telazol is nearly completely metabolized.

6. Xylazine

a. 8 mg/kg xylazine (Bauditz, 1972)

Note - Yohimbine is the antagonist for xylazine and can be administered, once all procedures are completed, at a dose of 0.1 - 0.2 mg/kg intramuscular.

WE DO NOT RECOMMEND THIS PROTOCOL. Xylazine is a sedative, analgesic and muscle relaxant. In non-domestic species, such as jaguar, xylazine alone does not produce sufficient restraint. Common side effects of xylazine are vomiting and RESPIRATORY DEPRESSION. Additionally, animals are extremely sensitive to external stimuli (i.e., noise, movement) and can be easily aroused from a sedated state.

D. Supplemental drugs for administration during immobilization procedures

1. Anesthetic supplementation

There will be occasions when the initial anesthetic agent(s) does not provide adequate immobilization or when the effect of the anesthetic agent(s) begin to wane (i.e., increased animal movements, increased respiration and heart rate) prior to all procedures (i.e., radiocollar application, sample collection) being completed. In these cases it may be necessary to administer additional drugs for adequate anesthesia to allow safe handling. The following should be kept in mind if one is faced with either of these situations.

a. Telazol should NEVER be administered as the supplemental drug. If telazol is the initial immobilizing agent and it has not provided adequate anesthesia or if its anesthetic effects have worn off, it is best to supplement with ketamine either intravenous or intramuscular. The dose of ketamine to deliver will depend on the plane of anesthesia prior to supplementation.

Supplemental ketamine at a dose of 1-1.5 mg/kg intravenous or 1-2 mg/kg intramuscular, as needed to maintain an adequate level of anesthesia should be a safe dose in MOST adult jaguars.

b. Xylazine, medetomidine and midazolam should NEVER be administered as the supplemental drug. They should only be administered in combination with another drug (i.e., ketamine) for induction of anesthesia. It is best to supplement with ketamine either intravenous or intramuscular. Supplemental ketamine at a dose of 1-1.5 kg/mg intravenous or 1-2 mg/kg intramuscular, as needed to maintain an adequate level of anesthesia should be a safe dose in MOST adult jaguars.

c. Diazepam (valium) at the dose of 5 – 10 mg / jaguar should be administered slowly I.V. to any jaguar with extreme muscle rigidity, muscle tremors, and/or seizures. Diazepam can be administered I.V. after 3 minutes if no response to the initial injection. If there the jaguar still does not respond following the second injection, another cause of the seizure activity should be considered. If a vein cannot be located (i.e., moving animal), diazepam can be injected I.M. Caution should be exercised in administering a second dose of diazepam following an I.M. injection due to a potentially slower rate of metabolism with intramuscular injections.

d. If you are not sure of how much of the original drug(s) was successfully administered (i.e., poor dart placement, dart bounced in and out quickly), you should wait at least 10 minutes prior to administering any additional agents.

2. Anticholinergics

Some authors recommend the addition of atropine to the anesthetic protocol for the anticholinergic property of decreasing salivary secretions. However, atropine can be associated with negative side effects, most commonly on the heart and gastrointestinal tract. In field situations it may be more appropriate to administer atropine only to those cats that are displaying excessive salivation during the immobilization procedure. A single dose should be administered:

Atropine - (0.04 mg/kg) subcutaneously (SQ) or intramuscularly

3. Supportive medications

a. Ivermectin - 200 mcg / kg SQ for screwworm - in areas known to have *Cochliomyia hominivorax*.

b. Lactated Ringers solution 1-2 liters SQ for rehydration especially if the jaguar was trapped for an extended period and/or was highly stressed / hyperthermic.

c. Penicillin G benzathine 40,000 IU/kg I.M. (long-acting antibiotic) especial for use in jaguars that have significant trauma from the dart, a fractured tooth, vomited during the procedure, and / or any other active lesions.

d. Topical triple antibiotic for placement at the dart site or any other active skin lesions.

e. Topical fly strike ointment for placement at the dart site or any other active skin lesions.

f. Topical triple antibiotic eye ointment for placement on the eyes.

III. HANDLING IMMOBILIZED JAGUARS AND TROUBLE SHOOTING FOR ANESTHESIA EMERGENCIES

Any person who immobilizes a wild jaguar must remember that he/she is solely responsible for the health of that animal from the time the drug is administered (or from the time the animal is captured or treed) until the animal has fully recovered from the anesthetic agent(s). It is imperative that anyone engaged in the immobilization of free-ranging jaguars know how to handle the anesthetized cat, monitor physiologic parameters, and respond to medical emergencies should they arise. Although many anesthetic agents are relatively safe in felid species, anesthetic emergencies can and DO occur even under the best circumstances.

These guidelines are not intended to be comprehensive for all aspects of anesthesia related veterinary emergencies. They are however intended to provide the bare minimal of knowledge necessary for all people immobilizing jaguars in hopes that emergencies are

minimized, and for those cases when emergencies do occur that the researchers are equipped to better handle these. It is strongly recommended that researchers performing jaguar immobilizations do further reading on this subject (Wildlife Restraint Series, 1991; Fowler, 1995; Evans, 1996; Kreeger, 1996; Nielsen, 1999). Those inexperienced in jaguar immobilization should seek the assistance of more experienced colleagues.

1. Handling

Immediately after the animal is darted (see above information on immobilization drugs) and an initial assessment of the respiratory rate and heart rate are deemed within normal limits,

Respiratory Rate (RR) 8 - 24 breathes/minute

Heart Rate (HR) 70 - 140 beats/minute

The dart should be collected (avoid handling the needle) and put in a safe place. (It is best to have one person immediately take the physiologic parameters while a second person is in charge of the dart.) The dart site on the animal should not be touched to avoid contact with drug residues and blood. People who will have contact with the immobilized animal should wear latex gloves during the immobilization procedure to avoid the transmission of infectious diseases between the animal and his/herself, as well as minimize contact with drug residues at the injection site.

The animal should be placed in a position that allows it to breathe easily (Figure 2). Preferably, the jaguar should be placed in lateral recumbency (lying on its side). The head and neck should be placed in a position that allows air to flow through the mouth and trachea (windpipe). The mouth should be kept lower than the back of the throat and neck so saliva flows out of the mouth and not into the trachea.

Once the animal is anesthesized and placed in the proper position, the eyes must be protected. Triple antibiotic eye ointment should be applied in both eyes to prevent them from

drying due to the lack of the normal blink response, as seen with some anesthetics. A towel (nonabrasive material preferably) should then be placed over the eyes to protect them from the sun and dirt, as well as to minimize stressful stimulus to the animal. It is important to minimize wounds due to the high risk of screwworm infestation. Topical betadine (iodine solution) and a fly strike ointment can be applied to the dart site (see the section on trouble shooting – wound management).

All handling equipment (i.e., towels, non-disposable gloves) should be disinfected prior to use on another animal to prevent the spread of diseases.

2. Monitoring

During all jaguar immobilizations, the physiological parameters (i.e., respiratory rate, heart rate, and temperature) MUST be monitored. If these values fall outside the normal range, the immobilization team should be alerted to a potential emergency and be ready to respond in the appropriate manner. The normal physiologic parameters for an immobilized free-ranging jaguar are the following:

Temperature (T) 37 – 39.5^0 C (98.6 – 103.1^0 F)
Respiratory Rate (RR) 8 - 24 breathes/minute
Heart Rate (HR) 70 - 140 beats/minute

Both respiratory rate and heart rate should be monitored every 5 minutes and the temperature should be taken every 10 minutes.

Monitoring these parameters can best be done by use of a thermometer, visual observation of chest wall expansion, and either palpation of the femoral pulse or use of a stethoscope. A rectal thermometer coated with KY jelly should be placed in the anus (digital thermometers are the best and easiest to use in the field) and the temperature monitored at 10

minute intervals during anesthesia. Respiration can be monitored by watching the thorax move when the animal breathes. The easiest way to determine the respiratory rate per minute, is to count the thoracic movements during 15 seconds and then multiply this number by four. If one does not have a stethoscope in the field, then light digital pressure over the femoral artery (this artery is located on the inside region of the thigh) will provide a measure of the heart rate. You can practice finding the location of this artery on a domestic dog. Alternatively, a stethoscope can be used to auscultate the heart directly over the lateral aspect of the cranial thorax (under the elbow).

Note: We highly recommend that all jaguar immobilizations are conducted only when a stethoscope is available. The stethoscope can also be used for monitoring the respiratory rate.

The recognition of what to consider normal jaguar responses to anesthetic agents is also imperative. Jaguars immobilized with telazol (and ketamine) usually will have increased salivation, open eyelids, whole body muscle rigidity (including jaw tone), and intact reflexes (i.e., corneal, pedal). Jaguars should maintain swallowing and coughing reflexes with these agents, but should not have muscle tremors and seizure-like activity.

3. Trouble shooting common anesthetic emergencies

Table 1 contains the drugs most commonly used for preventive measures and to treat emergencies during field immobilizations.

A. Respiratory depression and arrest results in tissue hypoxia caused by inadequate oxygenation of blood hemoglobin and is probably the number one anesthetic emergency encountered in the field.

Diagnosis of respiratory depression / arrest is based on

1. The jaguar taking few or no breathes (i.e., less than 4) (no chest expansion) per

minute;

2. Mucous membrane (gums) color is blue/gray;

3. Oxygen saturation is < 80% on pulse oximetry (if available);

During field immobilization there are a number of causes for respiratory depression/arrest including 1) drug-induced depression of the respiratory center; 2) airway obstruction due to malpositioning, excessive salivation or regurgitation, laryngeal edema; 3) pressure on the diaphragm from gastrointestinal contents; and 4) excessive build up of carbon dioxide which alters normal respiration.

Treatment of respiratory depression / arrest should include the following:

1. **DO NOT PANIC** (this is true for all anesthetic emergencies!).
2. Do not administer any additional immobilization drugs.
3. Be sure the head and neck are in good positions (extended with no objects compressing them) so air can move through the mouth and trachea. Be sure there is no vomit or foreign objects blocking the trachea (see below).
4. Intubate immediately if an endotracheal tube (ETT) is available. Administer oxygen through the ETT using an ambu bag, your own breath, or an oxygen tank.
5. If no ETT or supplemental source of oxygen is available, use intermittent pressure on the chest to attempt to move air through the lungs. The jaguar should already be in lateral recumbency. Push down firmly on the chest at regular intervals (i.e., press for 1 second, wait for 1 second, press for 1 second and so on). Alternatively, you may attempt mouth-to-mouth or mouth-to nose resuscitation. Exhale into the jaguar's mouth or nose for a count of 2 sec and then inhale away from the cat's mouth/nose for a count of 2 sec.
6. Administer 1 - 2 mg/kg doxapram I.V. (or I.M. in the tongue muscle if one cannot quickly find a vein). This is approximately 80-160 mg (4-8 mls) per 80 kg adult jaguar.

Note: Doxapram can cause arousal, especially cat is immobilized with telazol, and caution for human safety must be considered if one elects to use this drug as a respiratory

stimulant. Some veterinary anesthesiologist no longer recommend the use of this drug. If respiratory arrest is not corrected with steps 1 –5 above, we recommend the use of doxapram as a last attempt for resuscitation. If a person must inject the drug into the tongue, she/he should be very careful not to traumatize the oral cavity.

7. Administer appropriate anesthestic antagonist if available (i.e., yohimbine, atipamezole). However, do this cautiously as the antagonist will only reverse the drug it antagonizes and the jaguar may be semi-anesthetized and difficult to handle after the antagonist is administered.

B. **Cardiac arrest** is usually preceded by respiratory arrest and is defined as the loss of effective cardiac function resulting in cessation of circulation. This is the **most serious** anesthetic emergency encountered during field immobilization.

Diagnosis of cardiac arrest is based on

1. Weak or absent pulse or heart sounds
2. Blue/gray mucous membranes (gums)
3. Poor capillary refill time measured by applying digital pressure to the mucous membrane until the mm turns pale and then releasing the pressure and monitoring the seconds it takes until the mm color returns to normal (this value should be < 2 sec)
4. Dilated pupils
5. Cold extremities
6. Loss of consciousness

The most common causes of cardiac arrest during field immobilization are 1) drug-induced; 2) respiratory failure leading to hypoxia; and 3) acid-base or electrolyte imbalance.

Treatment of cardiac arrest should include the following:

1. Do not administer any additional immobilization drugs.

2. Be sure the animal can breathe prior to starting cardiac massage (see above).

3. Begin external cardiac massage. The jaguar should already be in lateral recumbency. Apply firm pressure downward over the heart. Compression of the heart should be for a count of 1 and release for a count of 1 with 60-100 cycles/minute. If an assistant is available he/she should palpate the femoral pulse to ensure adequate pressure, to circulate blood, is being applied during cardiac compressions.

4. Administer 0.02 mg/kg of 1:1,000 (1.0mg/ml) epinephrine I.V. or intracardially and continue with external cardiac massage. This dose is approximately 1.6 mg (1.6 ml) per 80 kg adult jaguar. Only veterinarians should attempt intracardial injections.

5. Administer 20 ml/kg cool Lactacted Ringer's Solution as an IV bolus (i.e., a single rapid infusion).

6. If no response, repeat 4 above at 5 minute intervals indefinately.

C. **Hyperthermia** is defined as an increase in body temperature to a point where oxygen demand exceeds supply due to increased metabolism.

Diagnosis of hyperthermia is easily determined by rectal thermometer.

1. **Temperatures > 41$_0$ C (105.8 $_0$ F) are true emergencies.**

Causes of hyperthermia in field immobilization include 1) internal heat production due to excessive physical exertion; 2) external heat absorption; 3) drug-induced compromise of thermoregulation; and 4) inability to use behavioral thermoregulation.

Treatment of hyperthermia includes the following:

1. Make sure the jaguar is in the shade

2. Use portable "cold" packs that can be placed in the groin, axillae (armpit) and belly of the jaguar.

3. Cool the jaguar by applying water over the body and/or alcohol to the extremities (legs and feet).

4. Administer cold water enema if tubing is available.

5. Administer 20 ml/kg cool Lactacted Ringer's Solution as an IV bolus (i.e., rapid fluid infusion).

6. Take the temperature every 5-10 minutes to determine if the temperature is decreasing. Continue to wet the animal if the temperature remains high.

7. Administer antagonist I.V. (I.M. if a vein is not readily identified). However, do this cautiously as the antagonist will only reverse the drug it antagonizes and the jaguar may be semi-anesthetized and difficult to handle after the antagonist is administered.

8. If it is believed that the hyperthermia is due to muscle rigidity and a light plane of anesthesia, diazepam at a dose of 5 – 10 mg / jaguar TOTAL can be administered slowly I.V. to decrease muscular activity.

Note - Hypothermia (<35^0 C = <95 ^{0} F) - decreased body temperature to point of cellular death - is much less likely under most field conditions in which jaguar will be immobilized. However, this may occur (i.e., high altitude regions) and should be treated by warming the animal.

D. **Vomiting and aspiration** are defined as the ejection of stomach contents through the esophagus and mouth and inspiratory sucking into the airways of foreign material, respectively.

Diagnosis of aspiration of vomit is not always straight forward. Clinical signs suggestive of aspiration are

1. Blue/gray mucous membranes (gums)
2. Choking and gasping
3. Gurgling sounds during respiration
4. Presence of material in the larynx and trachea
5. Respiratory arrest

Causes of aspiration during field immobilizations include 1) drug-induced vomiting (i.e., xylazine) with subsequent aspiration; 2) stress; 3) excitement; and 4) positioning of the head lower than the stomach.

Treatment of vomiting and aspiration include the following:

1. Do not administer any additional anesthetic agents.
2. Clear the airway.
3. If the jaguar is not breathing on its own, begin artificial ventilation (see respiratory depression/arrest above).
4. If the laryngeal region is KNOWN to be irreversibly blocked with vomitus, a tracheotomy may be attempted in the distal trachea to allow the passage of oxygen. (This procedure should only be performed by veterinarians that are familiar with the surgical procedure.)
5. Administer long-acting antibiotics (i.e., Penicillin G benzathine 40,000 IU/kg I.M.)

Aspiration of vomit can be an acute life-threatening situation due to the initial blockage of the respiratory tract and asphyxiation. However, it should be noted that a more chronic result of aspiration is pneumonia, which may also be life-threatening. Any jaguar known to have aspirated vomit is susceptible to developing pneumonia. The use of a long-acting antibiotic will help to decrease the likelihood of pneumonia developing but is often of little help in those cases
in which a large amount of vomit has been aspirated.

E. **Shock** is defined as ineffective blood perfusion of tissues resulting in cellular hypoxia. Shock is typically classified into three categories: hypovolemic, cardiogenic, and distributive. Shock associated with the capture and immobilization of free-ranging jaguars can be any of these three categories but typically is distributive or cardiogenic.

Diagnosis is made based on clinical signs that include

1. Rapid, weak/thready heart rate

2. Slow capillary refill time

3. Hyperventilation

4. Depressed mentation in those animals not anesthetized.

Causes include 1) prolonged physical exertion; 2) prolonged physiologic stress; 3) prolonged psychological stress; and 4) severe blood loss.

Treatment should include the following:

1. Do not administer any additional anesthetic agents.

2 Administer 4 mg/kg dexamethasone I.V. (unless vein is not readily available; then use I.M.)

3. Administer 30 ml/kg Lactated Ringer_s solution I.V.

4. If the jaguar is not breathing on its own, begin artificial ventilation (see respiratory depression/arrest above).

F. **Seizures** are defined as disturbances of cerebral function characterized by a violent, involuntary contraction or series of contractions of the voluntary muscles.

Diagnosis is made based on clinical signs that include the following

1. Uncontrolled muscle and/or whole body spasms

2. Rigid extension of the limbs

Causes include 1) drug-induced (i.e., ketamine and tiletamine); 2) trauma; and 3) hypoglycemia.

Treatment includes the following:

1. Administer 10 mg diazepam I.V. slowly over 10-15 seconds.

2. Repeat one above if no improvements within 3 minutes.

3. Monitor body temperature to determine if secondary hyperthermia results from the seizure activity.

G. **Wounds** are often associated with the dart site as well as by trap or chase injuries. (Be especially cognizant of any oral lesions and/or broken teeth.)

Diagnosis is based on clinical signs. The severity of the wound will dictate the treatment modality chosen.

1. Physical examination to evaluate for traumatic lacerations and lesions
2. Oral examination to evaluate for oral lesions and broken teeth

Treatment should always include:

1. Clean the wound with a povidone-iodine or 2% chlorhexidine solution. If neither of these are available, use soapy water.
2. If necrotic tissue is present and the field personnel are familiar with veterinary surgical techniques, debride the dead tissue and repeat step 1.
3. Only suture those wounds that you KNOW are fresh (i.e., caused by the dart) and that require sutures to minimize further tissue damage. Again, only field personnel who are familiar with veterinary surgical techniques should attempt to suture any wounds.
4. Apply topical antibiotic and fly strike ointment to wound site.
5. Administer long-acting antibiotic I.M. (i.e., Penicillin G benzathine 40,000 IU/kg I.M.)
6. Administer Ivermectin 200 mcg/kg SQ (to prevent screw worm infestation at site of broken skin)

Treatment of broken teeth: It is imperative that a fractured tooth (most commonly a canine is broken during jaguar captures and immobilizations) be repaired to minimize pain and infection associated with the tooth. A calcium hydroxide product (i.e., Dycal ®) can be used to cap the tooth pulp. Instructions for application come with tooth repair kits.

H. **Capture myopathy** is defined as a complex alteration of metabolic processes that may cause peracute lethal acid-base and electrolyte imbalances or acutely produce necrosis of cardiac and striated muscles. Although rarely ever encountered in felids (it is most commonly a problem in ungulates), field researchers should be familiar with this condition.

Diagnosis is based on clinical signs that include

1. Ataxia (abnormal walk) and weakness
2. Paresis and paralysis
3. Brownish urine
4. Death

Causes include 1) prolonged physical exertion; and 2) prolonged physiological and/or psychological stress.

Treatment is often unrewarding and the backbone of treatment is PREVENTION.

1. Minimize the stress of any immobilization procedure.
2. Administer 5 meq/kg sodium bicarbonate IV
3. Administer 30 ml/kg Lactated Ringer's solution IV

I. **Dehydration** defined as the reduction of the normal body fluid is often associated with immobilization of free-ranging wildlife.

Diagnosis is based on clinical signs that include

1. Weak pulse
2. Dry mucous membranes
3. Skin lacks pliability
4. Depressed mentation in those jaguar not anesthetized

Causes include 1) decreased water intake; 2) hyperthermia; and 3) chronic water loss (ie., diarrhea, vomiting, polyuria).

Treatment should include the following:

1. Do not administer any additional immobilizing drugs.
2. Administer Lactated Ringers solution at a rate of 20 ml/kg, preferably I.V. but S.Q. can be used as the second choice. Alternatively, it would be best to calculate fluid deficit based on a scale of 5% (mucous membranes dry; skin tacky) to 8% (mucous membranes dry and reddened; skin tacky and remains tented when pinched) and administer fluids at a rate of - (% fluid deficit) X (body weight (kg)) / 100 = volume in liters

Example for 80 kg jaguar with 5% dehydration 5% X 80 kg / 100 = 4 liters

4. Medical kit for field immobilization

In addition to a strong knowledge base in immobilization and handling of jaguars, it is imperative that researchers have the proper equipment with them in the field. Devices for monitoring physiologic parameters will ensure that the researcher is alerted to possible anesthetic problems / emergencies. Additionally, a few instruments and drugs are necessary for handling emergencies should they occur. A standard medical kit for all researchers to take to the field is presented in table 2.

IV. POST ANESTHETIC RECOVERY

The recovery period is just as important for proper handling and monitoring as the induction and maintenance periods; in fact, most anesthetic complications occur during induction and recovery. It is not uncommon for anesthetic related morbidity and mortality to occur during recovery. Although there are reversal drugs for the zolazepam component of telazol (flumazenil), xylazine (yohimbine), and medetomidine (atipamezole), jaguar recoveries can not be completely reversed with one specific antidote as is available for narcotic (i.e., carfentanil, etorphine) immobilizations commonly used in hoofstock. For this reason, it is important to ensure the jaguar

does not cause injury to itself or to people involved in the immobilization during the recovery period.

During recovery, the jaguar should be positioned so that it can breathe easily and will not harm itself on nearby objects. The animal should be placed in lateral recumbency with the head and neck extended. Abrasive material should not be under the head due to possible head movements that could lead to corneal abrasions. Additionally, the jaguar should be placed in a shaded area to protect if from excessive heat and from the sun damaging the cat's eyes. People in the area must remain quiet and should NOT stimulate the jaguar. It should recover at its own pace as it metabolizes the anesthetic agent(s).

If the jaguar was originally captured in a box trap, it may be beneficial to let the animal recover in the cage where it is dark and quiet. However, it must be remembered that when the cat is awake enough for release, the danger to field personnel may be significant when opening the cage. While in the cage and recovering, the animal may also be aggressive and cause harm to itself. Thus, if one is to use a box trap for recovery, it requires judgement to be sure the jaguar is awake enough prior to release, but does not cause harm to itself while still in the cage.

Alternatively, when no trap is available (i.e., treed by dogs or darted from a blind), the animal can be placed in a quiet, padded (i.e., with leaf litter), and protected (i.e., not near ledges, hard structures) area to recover on its own. Risks are involved with both recovery methods. People should move far away from the recovering cat with one or two observers remaining only as close as needed to visually observe the animal.

V. ANIMAL SAMPLING

The number one priority for the JHP is related to the safe handling of all jaguars during immobilization procedures. The second most important aspect of the JHP is that researchers utilize proper sampling techniques. It is imperative that all samples are collected, stored and

transported appropriately so that viable tissues are available for diagnostic testing. Samples can be collected from immobilized live jaguars (i.e., blood, feces, ectoparasites, hair), field samples (i.e., feces, urine, hair) and tissues salvaged from dead jaguars.

In this section we present the types of samples that should be collected, equipment necessary to collect them, how to collect the samples, and what diagnostic tests can be performed on these samples. Field researchers will have different equipment available, as well as different levels of training and experience in the technical skills related to biological tissue sample collection. For this reason, the type of samples collected will vary from one immobilization event to another.

1. Sample collection, storage, and transport

A. Physical examinations - All jaguars that are immobilized, or observed free-ranging, should be clinically evaluated. A physical examination of the jaguar_s health will provide valuable information about its health status. Visual observations are useful but more specific physical examination techniques (i.e., thermometer, stethoscope, palpation, etc.) should be performed when feasible. Morphometric measurements are also an important component of the physical examination. An example of a standard physical examination form is provided in appendix 4.

B. Blood samples - Blood should be collected from jaguars during immobilizations when field staff are familiar and comfortable with the procedure.

1. Collection - Vessels for collecting blood from jaguars include the medial and lateral saphenous veins, femoral vein, cephalic vein, lateral tail vein, and the jugular. We recommend that biologist with no training in veterinary techniques should not attempt to collect blood. The needle and syringe size will depend on which vein is used for collecting blood. Generally, the

needle size should range from 18 - 22 gauge and 1 - 1 _ inches and syringes should be 6 - 25 ml. Ideally, a total of 25 ml whole blood per jaguar should be collected.

Additionally, a sample should be collected from a peripheral ear vein to prepare blood slides for hemoparasite identification. This sample should be collected using a sterile 18 or 20 gauge needle to puncture the vessel. Microcapillary tubes should be used to draw blood from the punctured site and slides prepared as described in Appendix 9.

2. Storage - Blood must be immediately transferred to red top tubes containing no anticoagulant and purple top tubes containing EDTA anticoagulant. Once blood is placed in purple top tubes, these tubes must be gently inverted a few times to mix the blood with the anticoagulant. This will help to prevent the blood from clotting. Red top tubes should be kept at room temperature and purple top tubes are bested placed under refrigeration (i.e., cooler with ice). Blood in both tubes should be centrifuged within 4 hours of collection to separate blood fractions. A small amount of whole blood (blood in anticoagulant) should be put into a few microcapillary tubes. This blood will be used for packed cell volume (PCV), total solids (TS), white blood cell (WBC) counts, filter paper dot blots and blood smear slides (appendices 5 - 9). The remainder of the blood should be spun at 3,500 rpm for 10 minutes and the separated serum decanted into cryotubes for long-term storage. Slides should be air dried and fixed with slide fixative. (Additionally, the quality of slides is improved if they are stained in the field.) Slides should be placed in a protective holder (slide tray) and stored at room temperature. Cryotubes should be stored frozen, preferably in liquid nitrogen but alternatively can be kept on ice.

3. Transport – Serum and plasma should be transported in liquid nitrogen or on dry ice to ensure that they remain frozen. Slides can be shipped at room temperature.

C. Fecal samples should be collected from all jaguars during immobilizations, from fresh fecal samples located in the field, and fresh carcasses.

1. Collection - In live immobilized jaguars, feces should be collected directly from the animal's rectum, using a gloved hand, and then placed in appropriate media in an airtight container (see storage below). Five to 10 grams of feces is adequate to place in each type of storage media.

2. Storage - Feces can be stored in a variety of media, depending on what diagnostic tests are to be performed. Feces should be placed in 5% formalin for parasite ova and larvae identification.

Additional feces can be placed into 70 - 95% alcohol. This can be used for DNA analysis, hormone analysis (i.e., cortisol, estrogens, progesterones, testosterone), and possibly polymerase chain reaction (PCR) testing for bacteria and viruses.

Empty cryogenic container. This sample should be kept frozen either using dry ice or liquid nitrogen. This may be used for testing reproduction and stress hormone levels, and virus isolation.

3. Transport - Fecal samples should be transported in above mentioned containers as follows:

1) Feces in formalin and alcohol should be transported at room temperature.
2) Feces in cryogenic containers should be transported frozen either in liquid nitrogen or on dry ice.

D. Ectoparasites samples should be collected from immobilized live jaguars, as well as fresh carcasses.

1. Collection - Ticks and other ectoparasites (i.e., hipposcid flies) should be removed from jaguars using either forceps or gloved hands. One should make every effort to collect the head of

the tick to minimize post-removal lesions. Ticks commonly attach around the ears, groin region, and axillae.

2. Storage - Place all ectoparasites in airtight containers with 70 - 95 % ethyl alcohol, and maintain at room temperature.

3. Transport - All ectoparasites can be transported in above containers at room temperature.

E. Hair samples should be collected from immobilized live jaguars, as well as from carcasses. (Please refer to the genetic handout.)

1. Collection - Hair samples can be plucked using forceps or fingers or cut using a knife or scissors. Plucked hair is preferable since it is more useful in genetic studies.
2. Storage - Hair samples should be placed in empty, dry paper envelopes and stored at room temperature.
3. Transport - Hair samples should be transported at room temperature in above mentioned containers.

2. Permits and laws for transporting samples internationally

All diagnostic laboratory testing will be performed at a few select laboratories in the USA that are known for their specialty in non-domestic felids. Our ability to do this is contingent on exporting samples from various countries and importing them into the US. Jaguars are listed on CITES appendix I. Therefore, a CITES import and export permit are required for transporting tissue samples into the US. The JCP is working to obtain blanket CITES permits for the work.

The importing country (USA) must issue an import permit and the exporting country must issue an export permit. In general, an export permit will not be issued until a valid import

permit is presented to the office issuing the export. The U.S. Fish and Wildlife Service's Office of Management Authority is the agency which currently issues CITES import permits. In general, they require written proof of permission to conduct the work in the host country to accompany the application for an import permit. . For more information on permit requirements, visit the technical pages on the FVP website at www.fieldvet.org.

3. Diagnostic tests that MUST be performed in the field

Blood tests that must be performed in the field include the use of whole blood, prior to centrifugation, for PCV, TS, and WBC count. After the whole blood has been separated, using the centrifuge, these test can no longer be performed on the resultant plasma. These tests require a centrifuge, capillary tubes, PCV chart, and refractometer for the PCV and TS and a microscope and WBC Unopette kit for a WBC count. Methods for performing these field based tests are presented in appendices 5, 6 and 7.

4. Diagnostic tests to be performed in a laboratory

Most diagnostic testing will be performed in laboratories. The following is a list of the tests we recommend for a complete health assessment of jaguars. These tests include an assessment of parasitic and infectious disease agents to which they have been exposed.

A. Blood

1. White blood cell differential counts (in conjunction with blood test performed in the field)
2. Chemistry profile
3. Serology for infectious and parasitic agents which will include -

Viral agents

FeLV - feline leukemia virus

FIP - feline infectious peritonitis (coronavirus)

FCV - calicivirus

FHV - feline rhinotracheitis (herpesvirus)

FPV - feline panleukopenia (parvovirus)

FIV - feline immunodeficiency virus

Puma lentivirus

CDV - canine distemper virus

Pseudorabies

Rabies

Bacterial agents

Leptospirosis (17 serovars)

Bartonella henselae

Hemobartonella felis

Parasitic infections

Toxoplasmosis gondii

Babesiosis

Dirofilaria immitis

Cytauxzoon felis

Hormone levels (i.e., cortisol, testosterone, estrogen, progesterone)

Toxins (i.e., mercury, organophosphates, carbamates, PCBs, hydrochlorinated carbons)

Vitamins / Minerals

B. Feces

1. Parasites
2. PCR test for bacteria and viruses
3. Feline coronavirus RNA antigen

Hormone levels (i.e., cortisol, testosterone, estrogen, progesterone)

C. Ectoparasites

All ticks should be identified to the species level by an acarologist. Other ectoparasites should be identified by an entomologist.

D. Necropsy protocol

Appendix 10 contains a hard copy of the necropsy manual that can also be located on the internet at http://www.vetmed.ucdavis.edu/whc/necropsy/toc.html.

The necropsy manual was written by Dr. Linda Munson of the University of California, Davis and translated to Spanish by Dra. Marcela M. Uhart and to Portugese by Dra. Paulo Rogeria Mangini. This manual provides practical information for performing necropsies in the field. It is imperative that all researchers that collect samples from a dead jaguar record the following: age, sex, GPS reading, date and time located, and condition of body. It is also extremely important that the stomach contents be collected for studies on prey preference for jaguars across their range. These contents may be dried and stored at room temperature or placed in 70 – 95% alcohol.

E. Centralized laboratories - The Field Veterinary Program, WCS is establishing agreements with a large number of experts for laboratory testing in order to standardize the data from jaguar samples.

VI. DATA COLLECTION, ANALYSIS, AND DISTRIBUTION

One of the largest benefits of a collaborative health program throughout the jaguar's range is the ability to compile and correlate data from various habitats and populations of jaguars. A standard format for data collection is invaluable for compiling the data in any meaningful way. Appendix 2 is an example of the medarks form for collecting anesthesia data. Additionally, a standard form to collect physical examination data would be beneficial. One example is presented in appendix 4. This data will require statistical and non-statistical analysis to allow us to achieve our primary objectives of assessing the overall health status of jaguars across their range, determining the disease threats to jaguars, and using both these findings for providing recommendations for consideration in long-term conservation policies for jaguars.

These data will be presented for the scientific community, governmental officials, and lay persons in English, Spanish, and Portuguese.

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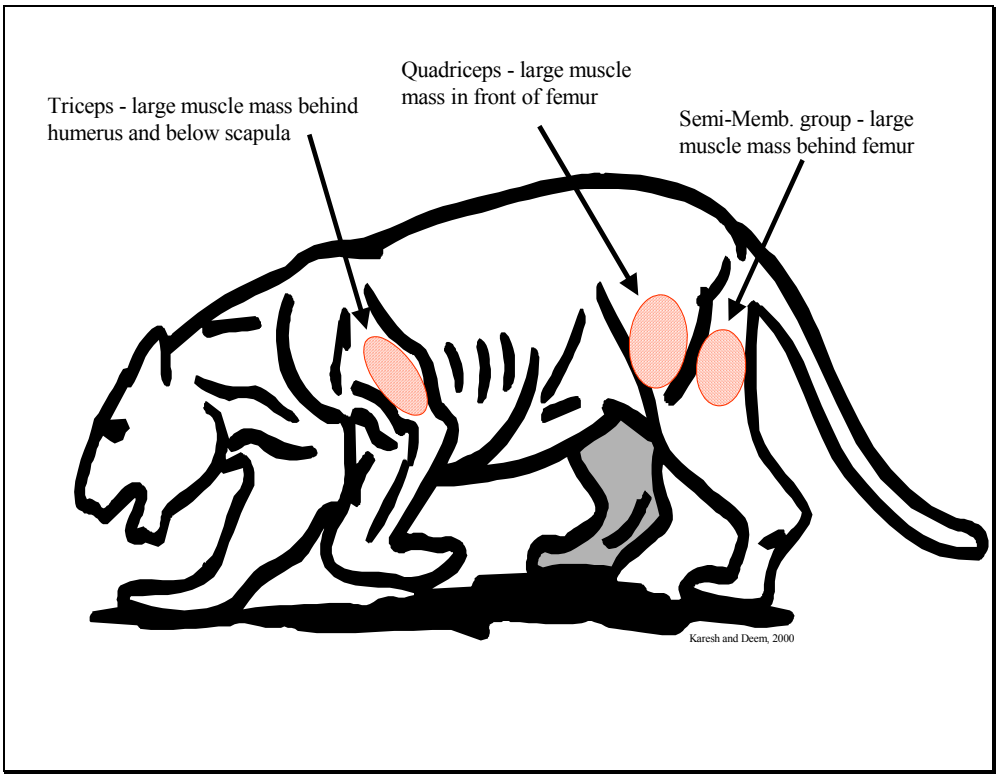
VIII. FIGURES

1. Dart placement sites for darting free-ranging jaguar.



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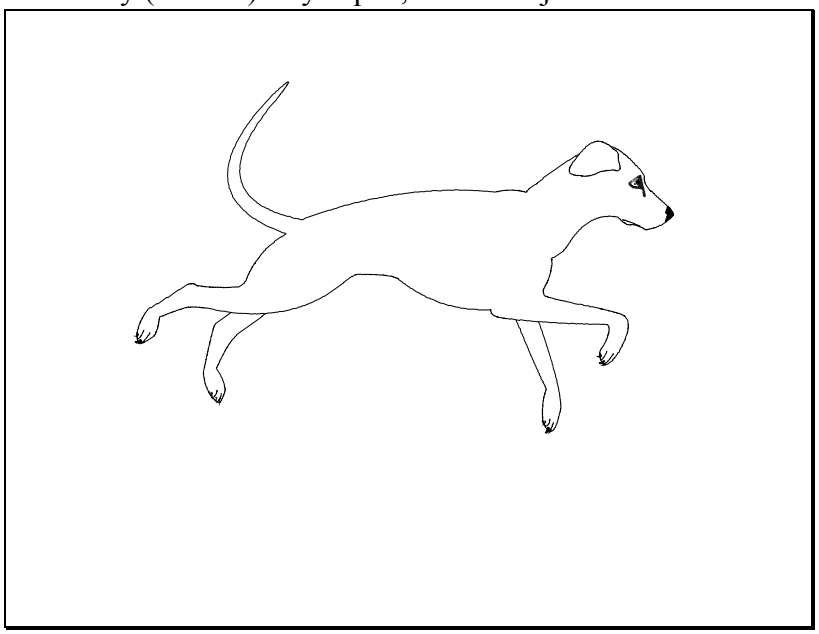
2..Example of a carnivore in lateral recumbency during anesthesia. Note extended neck ensuring the airway (trachea) stays open, and no objects or material are obstructing the mouth or nose.



IX. TABLES

Table 1 Quick reference table for administration of emergency drugs covered in this manual – based on an 80 kg jaguar

DRUG	CONCENTRATION	DOSE (MG/KG)	AMOUNT (ML)
Atropine	2.25 mg/ml	0.04 mg/kg	1.4 ml
Diazepam	5 mg/ml	0.1 mg/kg	1.6 ml
Dexamethasone	4 mg/ml	2 mg/kg	40 ml
Doxapram	20 mg/ml	1.5 mg/kg	1.5 ml
Epinephrine	1:1,000	0.02 mg/kg	1.6 ml
Lactated Ringers Soln	Non-applicable	20 ml/kg	1,600 ml
Sodium Bicarbonate	1 mEq/ml	1 mEq/kg	80 ml

Table 2 Basic medical kit for monitoring and trouble shooting during anesthetic emergencies

Monitoring equipment
Stethoscope
Thermometer
Pulse Oximeter

Emergency equipment
Laryngoscope
Endotracheal Tubes
Ambu Bag or Oxygen
Anesthetic Reversal Agents (i.e., yohimbine for xylazine; atipamezole for medetomidine)
Supplemental Drug Agents
- Atropine [2.25 mg/ml] (for excessive salivation and bradycardia)
- Doxapram [20 mg/ml] (for respiratory depression)
- Epinephrine [1:1,000] (for cardiac arrest)
- Sodium Bicarbonate [1 mEq/ml] (for capture myopathy)
- Diazepam [5 mg/ml] (for seizure activity)
- Dexamethasone [4 mg /kg])
- Lactated Ringer's Solution

Portable Ice Packs
Dental Repair Kit (i.e., Dycal ^{R})
Surgical Pack
Bandage Material

X. APPENDICES

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Appendix 2: Anesthesia form for data collection during free-ranging jaguar immobilization

Identification #:

Date: d d / m m / y y

Health Status:
1. [ ] Normal
2. [ ] Abnormal

Fasting Time:
1. [ ] < 8 hours
2. [ ] 8 – 24
3. [ ] 24 – 48
4. [ ] > 48 hours

Activity:
1. [ ] calm
2. [ ] active
3. [ ] excited

Demeanor:
1. [ ] depressed
2. [ ] alert
3. [ ] aggressive
4. [ ]

Physical Status:
1. [ ] Class I
2. [ ] Class II
3. [ ] Class III
4. [ ] Class IV
5. [ ] Class V

Immobilizing Conditions:
1. [ ] Free ranging
2. [ ] Large enclosure
3. [ ] Small enclosure
4. [ ] Squeeze cage
5. [ ] Manual restraint

1. [ ] isolated
2. [ ] in group

Body Condition:
1. [ ] obese / fat
2. [ ] good
3. [ ] fair/thin
4. [ ] poor/emaciated

Environ.
1. [ ] C
2. [ ] F

Genus/Species: _____

or

Common Name: _____

Sex: _____

Birthdate or age: _____

Housename: _____

Tattoo: _____

Tag/Band: _____

_____

_____

_____

Dose	Drug Given	Amount mg or %	Route	Time Given	Success of Delivery	Effect (stage)	Time of Effect	Bottle #
____	_____	_____	_____	____:____	_____	_____	___:___	_____
____	_____	_____	_____	____:____	_____	_____	___:___	_____
____	_____	_____	_____	____:____	_____	_____	___:___	_____
____	_____	_____	_____	____:____	_____	_____	___:___	_____
____	_____	_____	_____	____:____	_____	_____	___:___	_____
____	_____	_____	_____	____:____	_____	_____	___:___	_____

% Humidity:

Initial Effect Time: :

Recumbency Time: :

Weight: 1. [ ] kg 1. [ ]
2. [ ] lb. 2. [ ]
3. [ ] gm

Endotracheal

Complications:
1. [ ] None
2. [ ] Minor
3. [ ] Major
4. [ ] Fatal

Recovery:
1. [ ] Normal
2. [ ] Abnormal
3. [ ] Prolonged
4. [ ] Stormy
5. [ ]

Anesthesia Ratings:

	Excellent	Good	Fair
Induction	1. [ ]	2. [ ]	3. [ ]
Muscle relaxation	1. [ ]	2. [ ]	3. [ ]
Overall	1. [ ]	2. [ ]	3. [ ]

Recovery Data:

Time: : Effect:

Time: : Effect:

Recorded by:

Veterinarian:
[ ]

[ ]

Dose:	Preanesthetic	
	Immobilizing	
	Supplemental	
	Maintenance	M = intramuscular
	Antagonist	V = intravenous
	Other	P =
		S = subcutaneous
Route:	Polesyringe	
	Blowdart	
	Metal Dart	
	Hand syringe	
	Non-metal Dart	
	Oral	
	Facemásk	
	Chamber	
	Endotrachael Tube	
	Venous catheter	Physical Status
		1 = normal health
Success	Complete	2 = mild disease
	Partial	3 = severe disease
	None	4 = chronic severe disease
Effect:	0 = no effect	5 = may not survive anesthesia
	1 = mild sedation	
	2 = heavy sedation	
	3 = light anesthesia	
	4 = surgical anesthesia	
	5 = excessively deep	
	6 = death	

Blood Sample Data:
Time Collected: ____: ____

[ ] Cardiac
[ ] Cephalic
[ ] Ear
[ ] Femoral artery
[ ] Femoral vein
[ ] Jugular vein
[ ] Metatarsal vein
[ ] Saphenous vein
[ ] Other: _____

Hematology:
2. [ ] EDTA (liquid)
3. [ ] EDTA (dry)
5. [ ] Heparin (liquid)
6. [ ] Heparin (dry)
_____

Chemist
1. [ ]
2. [ ]
3. [ ]

Collecte
[ ]
[ ]
[ ]

[ ] Tail vein
[ ] Ulnar (wing) vein

[ ]
[ ]
[ ]
[ ]

Physiological Data:

Time	Body Temp.	Heart Rate	Resp. Rate	O_2 Sat	Blood Pressure Sys / Dia / Mean	
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	Rx:
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________
___:___	_______ F/C	______	______	_____%	____ / ____ / ____	______________________________

Comments:

Appendix 3. Companies that supply darting equipment

Advanced Injection Systems
705 Fourth St.
P.O. Box 1001
St. Joseph, IL 61873 USA
Tel: 217-469-2690
(Dart guns, pole syringes)

Palmer Chemical & Equipment Co., Inc.
P.O. Box 867
Palmer Village
Douglasville, GA 30133 USA
Tel: 404-942-4395
(Dart guns, darts)

Pneu Dart, Inc.
P.O. Box 1415
Williamsport, PA 17703 USA
Tel 570-323-2710
(Dart guns, darts)

Telinject USA, Inc.
9316 Soledad Canyon Road
Saugus, CA 91350 USA
Tel: 805-268-0915
(Dart guns, darts, blow pipes)

Wildlife Pharmaceuticals, Inc.
1401 Duff Drive
Suite 600
Fort Collins, CO 80524 USA
Tel: 970-484-6267
(Dart guns, darts, blow pipes, pole syringes)

Appendix 4: Jaguar physical examination form

Animal ID #	Date:
Location:	
Sex:	Body Weight:
Body Temperature:	Pulse Rate:
Pulse Character:	
Membrane Color:	Respiratory Rate:
Nutritional Status:	Superficial Lymph Nodes:
Skin and Ears:	
Eyes:	
Musculoskeletal System:	
Respiratory System:	
Cardiovascular System:	
Endocrine System:	
Alimentary System (incl. ORAL):	

Appendix 4. Continued.

Urinary System:
Genital System:
Nervous System:
General Comments:

Appendix 5 – Packed cell volume – field method

The packed cell volume (PCV) also called hematocrit is the percentage of whole blood made up of red blood cells (RBCs), also called erythrocytes. Whole blood is collected in an anticoagulant, such as EDTA or heparin, and placed in a capillary tube. Microhematocrit tubes should be filled three-fourths, one end sealed with clay sealant, placed in a centrifuge with the plugged end facing down. Centrifuge at 3,000 – 3,500 rpm for 5 minutes. RBCs have the highest specific gravity of the blood cells and gravitate to the bottom of the tube, appearing as a dark red layer. The PCV is then measured by using a hematocrit tube reader. The bottom of the RBC layer should be at the 0 line and the top of the plasma on the top line. The percentage can then be read as the line level with the top of the RBC layer.

A white to gray layer directly above the RBC layer is called the buffy coat and consists of WBCs (leukocytes) and platelets. The plasma is the clear to yellow fluid at the top. Plasma obtained by this method can be used to determine the plasma protein concentration by refractometry (see appendix 6). Plasma color and clarity may be helpful in determining a diagnosis and should also be noted.

Appendix 6 – Total solids – field method

A refractometer, or total solids meter, is used to measure fluid protein concentration or specific gravity. The most common uses of the refractometer are determination of the protein concentration of plasma and the specific gravity of urine.

Likewise, the plasma protein concentration can be determined from the small amount of plasma in a spun microhematocrit tube. The tube is broken above the buffy coat (thin white layer just above the packed RBCs) after reading the hematocrit, and the plasma is placed on the refractometer prism. Likewise, the specific gravity of a urine sample can be determined with just a drop of urine.

Procedure:

Examine the prism cover glass and cover plate of the refractometer. Clean with sterile water and dry as necessary.

Place a drop of sample fluid on the prism cover glass.

Direct the refractometer toward bright artificial light or sunlight.

Focus the light-dark boundary line by turning the eyepiece.

Read and record the result using the appropriate scale.

Appendix 7 – White blood cell count – field method

The manual Unopette hemacytometer method (Becton-Dickinson, Rutherford, NJ) of counting WBCs can be used for determining the WBC count in the field. Unopette WBC test kits contain diluting reservoirs and capillary pipettes for making a 1:100 dilution of blood in ammonium oxalate or acetic acid, both of which lyse red blood cells (RBCs). Cells are counted using a hemacytometer and cover glass. A complete, well-written instruction booklet is included in the kit.

Briefly, an appropriately measured amount of blood is added to the diluent (lysing solution) and gently mixed. After 10 minutes, the reservoir is gently inverted several times to evenly mix the cells and is converted to a dropper assembly. The iris diaphragm of the microscope must be closed to a point where the cells are most visible. The hemacytometer with coverslip applied, is filled with the solution, allowed to settle for a few minutes, and placed on the microscope, and the counting grid is found, using the 4X objective. Cells are counted in the 9 primary squares, using the 10X objective.

The number of cells counted is multiplied by dilution and volume factors (total cells counted plus 10% of those counted, multiplied by a dilution factor of 100). Counts are expressed as cells per microliter (ul) of blood. For example, if 80 cells are counted, 80 + 8 = 88 x 100 = 8800/ul.

Appendix 8 - Making a filter paper dot blot – field method

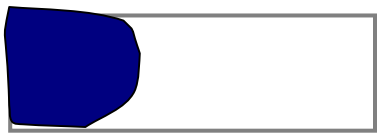
Small amounts of blood (0.1ml / dot) should be placed around the perimeter of a filter paper (medical grade 3 mm product is best). The filter paper should be labeled using a pencil with the jaguar id number and date of collection. The blood on the filter paper should then be allowed to air dry. Once dry it should be stored in a zip lock baggie with an internal silicon dry bag. The sample can then be stored and transported at room temperature.

Appendix 9 - Making a blood smear slide – field method

1) Blood collection:

- EDTA should be used for collection of hematology samples for jaguars.
- All samples should be mixed by gentle rocking for 5 minutes before making a smear.
- A drop of blood can be laced on the slide using a microcapillary tube.

2) A quality blood smear should



- Consist of evenly distributed cells.
- Be approximately ¾ of the length of the slide in length.
- Have an evenly distributed feather edge.
- Use the entire drop of blood.
- Be the entire width of the slide at the base, tapering to a nice curve at the opposite end of the smear.

3) Once the smear is made it should be:
    - Dried as quickly as possible. A hair dryer is most effective. If a hair dryer is not available, the slide should be picked up and rapidly waved in the air until it is dry.
    - The smear should be immediately placed into a slide box. In humid conditions, a desiccant can be placed into the slide box to help absorb extra moisture. Placing the smear immediately into the slide box also prevents ingestion of the blood from flies and cockroaches. Keep the box closed at all times.
    - If the smears are to be fixed, which we recommend, make sure the smear is COMPLETELY dry before placing it into the fixative. Fixing too early can cause serious streaking and uneven staining of the smear. Care should be taken to close the fixative jar while the slides are sitting in it as the fixative can also take on moisture from the air. Once the slide is removed from the fixative it should be dried standing it on end, feather edge down. Move the slide to the slide box as quickly as possible.
    - Slides can also be stained in the field by following instructions that come with stain solutions (i.e., Diff-Quick®). It is best to make 4 slides/jaguar and only stain 2 in the field.

4) Transporting the slides:

The slide box should be transported away from any other chemicals or **fumes** from those chemicals. Formalin in particular can damage blood smears. Packing blood smears **even in the same crate** with formalin can be detrimental to the quality of the smears.

Appendix 10: Necropsy protocol written by Dr. Linda Munson and located at the website http://www.vetmed.ucdavis.edu/whc/necropsy/toc.html.

Also available in Spanish and Portuguese.