

Capture, Handling and Release of Bison Standard Operating Procedure

Wildlife Care Committee Government of the Northwest Territories

Owen M. Slater

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1. Purpose

To provide guidelines for the safe and humane capture, handling, sampling and release of bison for research or management purposes, while minimizing risks to both staff and animals. In many cases, there is discussion of multiple options in terms of drug combinations and recommendations. Items listed as **must** are considered to be mandatory as part of best practices, while those listed as **should** are considered to be recommendations for best practice. It is understood that there can be circumstances and instances where practice may fall outside of the Standard Operating Procedures (SOP) and best practices. It is understood that the lead investigator would be able to justify any deviances from the best practice recommendations.

2. Application

This SOP applies to employees of the Government of the Northwest Territories (GNWT) and any other personnel involved with the capture and handling of bison in the Northwest Territories (NWT). This SOP was reviewed by the 2019/2020 NWT Wildlife Care Committee (NWTWCC), as well as external experts with experience with bison immobilization and handling.

3. Background

- Wood bison (*Bison bison athabascae*) are the only species of bison occurring in the NWT.
- They occur in three populations of wood bison that are managed by the GNWT: Mackenzie, Nahanni and the Slave River Lowlands.
- Wood bison were listed as Threatened under the federal *Species at Risk Act* (SARA) in 2004.
- Wood bison were re-assessed by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) as Special Concern in 2013.
- In 2016, they were assessed as Threatened by the NWT Species at Risk Committee.
- Under the territorial *SARA* (NWT), wood bison were listed as Threatened in 2017.
- As of 2018, the federal Minister of Environment and Climate Change has not made a recommendation regarding down-listing wood bison to Special Concern under *SARA*.
- A national recovery strategy for wood bison was published in 2018 and can be found at the *SARA* Public Registry (www.registrelep-sararegistry.gc.ca).
- It is up to the investigator to ensure all appropriate additional permits and permissions are in place prior to any bison capture and handling.
- Investigators must ensure their capture and handling protocol meets all requirements
 of the GNWT Department of Environment and Natural Resources (ENR), Wood Buffalo
 National Park (if appropriate) and the NWTWCC.
- For consistency, this SOP is modeled after existing ENR SOPs (Cattet 2017) and follows a similar format.
- ENR should view this SOP as a summary of current best practices and available information for the capture and handling of wood bison. Therefore, this SOP is subject to revision as new techniques are developed.

4. Methods

4.1 Live Capture

- Methods for live capture of free-ranging bison in the NWT include remote drug delivery, net gun or a combination of net gun coupled with hand injection of drugs after the animal is physically restrained.
- Other methods for handling captive bison, such as moving bison through a handling facility, are not covered in this SOP. Guidance for these situations will be considered on a case-by-case basis, and through other existing guidelines and recommendations.

4.1.1 Considerations

- Capture, management and research methods must be selected that minimize trauma and stress to bison, as well as limit the number captured to meet management or research objectives (CCAC 2003).
- While the primary focus during bison captures **must** be on the safety of personnel, animal safety and welfare also **must** be a high priority (CCAC 2003).
- Investigators **must** be familiar with the advantages and disadvantages of the different capture methods and select the method most suitable for their situation.
- Capture operations **must** be directly supervised by experienced individuals that have participated in at least one bison immobilization.
 - With guidance from ENR's Wildlife Veterinarian, the NWTWCC will determine if individuals have the necessary training and experience to perform procedures required for the capture and handling of bison.
 - Less experienced personnel who have relevant training may apply capture techniques only if they are under the direct supervision and mentorship of an experienced individual(s).
- Individuals administering wildlife immobilization drugs **must** have recognized and recent training in the chemical immobilization of wildlife and **must** use methods of drug delivery and immobilizing drugs that are appropriate for bison (CCAC 2003).
 - Recognized and recent training involves successful completion of the Canadian Association of Zoo and Wildlife Veterinarians (CAZWV) chemical immobilization of wildlife course or another recognized course by the GNWT, within the last five years.
- Live capture of adult female (cow) bison **should not** take place in the third trimester to six months post-calving unless justified due to management reasons such as human-bison conflict, disease or animal welfare concerns.
- Captures **must** be conducted within temperature ranges that are safe for both field personnel and bison.
 - Extreme cold poses greater safety risk to field personnel and must be factored into the decision-making process.
 - A suggested lower temperature limit for capture by aerial remote drug delivery or net gun is -30°C, with wind chill (Cattet 2017), although temperatures >-20°C are considered to have a wider margin of safety for the animals and humans.
 - A suggested upper temperature limit for capture by aerial remote drug delivery or net gun is -5°C (Cattet 2017).

- For ground-based darting, temperatures **should** be within normal seasonal ranges and if during the summer, captures **should** only occur during the coolest periods of the day to reduce the likelihood of hyperthermia and associated complications (Caulkett 2014).
- Procedures **must** be taken to prevent or detect and treat potentially harmful changes in bison body temperature during capture, handling and recovery.
 - Adult bison with winter hair are prone to hyperthermia even after relatively short periods of exertion.
 - Preventative measures **should** include keeping chase times as short as possible, using natural barriers to reduce wind and sun exposure, providing supplemental oxygen and using reversible or partially reversible drug combinations.
 - Young bison may be susceptible to hypothermia or frost-bite in extreme cold and preventative measures **should** include close monitoring of body temperature and the use of heating pads and/or sleeping bags to maintain normal body temperature ranges.
- An experienced team leader **should** be identified and tasks assigned to each team member prior to commencing captures (CAZWV, 2009).
 - This will help to avoid confusion in the field and assist in completing captures efficiently while ensuring all data and samples are taken and recorded.
- Capture of more than one animal per capture event **must** be avoided unless additional, experienced personnel are available to assist with the second animal (Cattet 2017).
- A capture team (three persons/adult animal) **must** be present to ensure all captured bison are restrained and handled immediately, while having the ability to move the animal if needed.

4.1.2 Techniques for Live Capture

4.1.2.1 Remote Drug Delivery

- The administration of wildlife immobilization drugs using projectile equipment that delivers the drugs without requiring direct contact with the animal (e.g. short or long range projectiles and darts).
- This is a suitable capture technique for longer-duration handling (>20 minutes).

- Darting **should** only be attempted by experienced personnel with well-maintained equipment that is sighted in (if applicable) and only after sufficient practice is obtained under environment conditions and at distances similar to those that will occur in the field (Kreeger and Arnemo 2018).
 - Compressed, gas-powered (air or CO_2) pistols or blowpipes (if applicable) can be used to deliver drugs to target animals over short distances, (≤ 10 m).
 - These types of remote projectiles propel darts at lower velocities than long-range projectiles and are less likely to cause trauma (Kreeger and Arnemo 2018, Valkenburg et al. 1999).
 - Blowpipes are classified as prohibited weapons under the Criminal Code of Canada, with the only exemptions being for public officers such as peace or conservation officers (CAZWV 2009).

- Blowpipes **must not** be used to deliver ultra-potent narcotics (e.g. carfentanil or etorphine) or other highly concentrated sedatives (e.g. 30-40mg/ml medetomidine), unless they are powered by compressed gas. This is to avoid the unreasonable risk of mucous membrane drug exposure.
- For remote drug delivery over longer distances (10-30 m), standard longrange projectiles (CO₂ or powder-charged) with power adjustment dial **should** be used to reduce the potential for excessive tissue trauma by the dart body (Kreeger and Arnemo, 2018)
 - Skin penetration by the dart body is less likely to occur with lightweight (e.g. 0.50-calibre; 12.5 mm diameter) darts and small drug volumes that impact the animal at a velocity <50 m/s (Cattet et al. 2006). However, the above darts still have the potential for skin penetration especially as dart mass and velocity increase (MacPherson 1994, Kreeger and Arnemo 2018).
- Longer distance ground darting (>30-70 m) **should** only be attempted with customized long-range projectiles on adult bison and only under ideal environmental conditions (no wind or extreme cold).
- Accurate distance determination should occur with a laser range finder to reduce the risk of misses or dart trauma from overestimation of the power needed to deliver the dart (Kreeger and Arnemo 2018).
- Slow injection darts coupled with needles with side-ports should be used in preference to rapid injection darts with end-ports only.
 - Examples of suitable darts and needles for bison include any Dan-inject darts and needles or slow-inject PneuDarts with tri-port needles.
- The dart needle must be long enough to ensure intramuscular (IM) drug injection, but not too long to cause injury to deeper tissues (e.g. bone) or too short to result in injection into the subcutaneous fat only.
 - Needle length in the range of 20-30 mm, **should** be used in bison (Haigh and Gates, 1995). Longer needle length (40 mm) may be appropriate for adult bison in good body condition and with thick hair coats.
- Needles **should** be barbed to reduce the likelihood of it coming out of the animal before immobilization. This reduces the risk of a lost dart (Kreeger and Arnemo, 2018).
- The position of the barb **should** be marked on the dart before use to facilitate quicker and easier removal from the animal.
- Darts must be directed into large, superficial, muscle masses with minimal fat covering, at a 90-degree angle to ensure proper placement, good drug absorption and avoiding injury to other tissues (CAZWV 2009).
 - The gluteal, semimembranosus or semitendinosus muscles (i.e. rump), shoulder muscles or the hump are all good dart sites in bison depending on the angle of approach. The hindquarters are often preferred due to less heavy hair coat in the region.
- Remote drug delivery **should** typically be employed from a vehicle (e.g. helicopter or truck) to facilitate control of animal movements before or afterwards and for human safety.

- Darts must be pre-loaded with drug to an appropriate volume prior to the vehicle moving and stored in a container to avoid accidental discharge or exposure (Kreeger and Arnemo 2018).
- Darts must not be loaded in a moving vehicle.
- The darter **must** maintain muzzle control of the remote delivery device (e.g. rifle, blow pipe or pole syringe) and treat it as a firearm.
- Darts must be loaded into the remote delivery device only just prior to the darting attempt.
 - This is particularly important during cold weather, as the drug may freeze inside the dart if left in the barrel for too long. Consider having hot packs or other warming device on hand to prevent the drug from freezing.
- When loaded, the projectile **should** be pointed down and outside the vehicle.
- It is important to be able to observe and control animal movement away from hazardous terrain or other non-target bison during pursuit and induction.
 - Induction is the time between drug administration and immobilization of the animal.
- Ground based darting via foot pursuit of bison should only be used if the operator can quickly reach a safe location if bison become aggressive or stampede and if the operator is familiar with signs of agitated bison.
 - An experienced guard with a rifle or shotgun **should** accompany the darter to ensure safety.
- In areas with reduced visibility or near urban areas, to avoid losing the darted animal or dart, remote drug delivery **should** only be attempted with telemetry darts (Kreeger and Arnemo 2018).
- The chase period **should** be < 1 minute and **must** be kept to <2 minutes
- The chase must be terminated when target or non-target animals show signs of exhaustion.
 - Best practice is to isolate the darted animal from the rest of the herd when aerial darting and if possible when ground darting
 - Bison are prone to capture myopathy (exertional rhabdomyolysis), hyperthermia and hypoxia (low tissue oxygen levels; Caulkett 2014), all of which increase in risk as chase time increases.
 - Trauma to the darted animal during induction is not uncommon and efforts **must** be made to control the movements of the herd to protect the darted animal from non-target bison during this period (Caulkett 2014).
- When more than one dart is required to safely immobilize a bison, adequate time **should** be given between injections to allow drug effects to occur.
 - In most situations, allow 10-15 minutes to elapse from the time of the first dart injection until the second dart is administered (Kreeger and Arnemo 2018).
 - If using Butorphanol-Azaperone-Medetomidine (BAM), allow 20 minutes (Kreeger and Arnemo 2018).
- Avoid unnecessary stimulation of the target animal during induction.

- For example, if the bison was darted from a helicopter, the helicopter should move far away from the animal while still watching for drug effects.
- If the animal shows some drug effect but remains standing or stands on approach, 50% of the original dose **should** be given (Kreeger and Arnemo 2018).
- If the animal shows no drug effect after 10-20 minutes of the first dart, 100% of the original dose **should** be administered (Kreeger and Arnemo 2018).
- If there is little or no drug effect after two complete dart injections the capture attempt **should** be aborted as this implies a problem with either the darts (e.g. failed injection) or drugs (e.g. inappropriate storage or concentration) (Kreeger and Arnemo 2018).
 - The animal **should** still be monitored to determine the degree of drug effects and administration of reversal drug (remotely or via hand injection) **should** occur if the risks to the animal and personnel can be minimized.
 - The animal **should** be monitored until recovered and at least once more over the next 24 hours (CAZWV 2009).
- If a darted animal is lost, effort **must** be made to track the animal and assure its welfare without compromising human safety (Cattet 2017).
- Effort **should** be made to find darts that miss their target, especially in areas frequented by people (Kreeger and Arnemo 2018).
 - Telemetry darts **may** be considered in these circumstances.

Drug Considerations:

- Drugs used to immobilize bison are very potent and can cause severe complications or death to people if not handled or stored appropriately.
- All members of the capture crew must be briefed on the drugs being used, potential side-effects if exposed, location and proper use of reversal drugs (if appropriate) and be trained in first aid and CPR (CCAC 2003).
 - Given the remote locations of bison capture work, advanced wilderness first aid training (40 hours) and certification from a course recognized by medical professionals **should** be a requirement for at least two members of the capture team, with re-certification every five years.
- Only unexpired drugs should be used in free-ranging wildlife.
- Drug combinations that can be reversed (partially or completely) with an antagonist drug (reversal agent) **should** be used instead of drugs that cannot be reversed (CCAC 2003).
 - Reversible drug combinations allow quick antagonism of immobilizing drug effects in animals showing adverse physiological responses as well as enabling quicker recovery (Kreeger and Arnemo 2018).
- Drug protocols as described in the chemical immobilization literature (e.g. Kock and Berger 1987; Haigh and Gates 1995; Caulkett et. al. 2000; Caulkett 2014; Wolfe et al. 2017; Harms et al. 2018), **should** be consulted. However, current (2018) drug combinations available and effective in bison are limited. Consideration of current drug availability, the darters experience

- and comfort level, as well as the collective experience of other **should** impact the decided protocol. Pros and cons of a select number of drug combinations are discussed below.
- Historically, carfentanil-xylazine has been a commonly used combination in both wood and plains bison at a dosage of 0.004-0.008 mg/kg carfentanil and 0.05-0.1 mg/kg xylazine (Haigh and Gates 1995; Kock and Berger 1987).
 - However, carfentanil is no longer available in Canada, which removes this combination from consideration at this time.
 - Etorphine is another ultra-potent opioid that can be used instead of carfentanil but it is also no longer available from most compounding pharmacies in Canada, making this option also impractical.
 - Given the limited availability of ultra-potent opioids and safety concerns, alternative drug combinations are recommended at this time.
- Xylazine-Telazol (XZT) has been used, but this combination is not considered optimal in free-ranging bison due to the larger drug volumes leading to decreased dart accuracy, and increased tissue trauma, in addition to longer recoveries due to the higher dose of Telazol required (Caulkett 2014). Therefore, this drug combination is not recommended.
- Medetomidine-Telazol (MZT) is preferable to XZT due to an approximate 50% reduction in volume of drug needed and lower dose of Telazol required (Caulkett et al. 2000). The lower dose of Telazol facilitates quicker and smoother recoveries (Caulkett et al. 2000). Dosages range from 0.6-0.8 mg/kg medetomidine and 1.2-1.4 mg/kg Telazol (Caulkett et al. 2000; Caulkett 2014; Caulkett personal comunication 2018). Reversal of the medetomidine portion of the combination with atipamezole at five times the medetomidine dose via IM injection is recommended. This drug combination remains a viable option for free-ranging bison (see Appendix A).
- BAM comes in pre-mixed vials (Wildlife Pharmaceuticals, Fort Collins, CO) containing 27.3 mg/mL butorphanol, 9.1 mg/mL azaperone and 10.9 mg/mL medetomidine and has been trialed in plains bison with mixed results (Harms et al. 2018). A combination of the drugs with appropriate doses could also be used from constituent drugs. For ground-based darting, 3.5 mL of the pre-mixed BAM for an adult female bison was generally effective but inconsistent inductions, particularly for helicopter-based darting, limit the usefulness of this combination at the doses reported (Harms et al. 2018). Reversal of medetomidine with atipamezole at five times the medetomidine dose is recommended. Butorphanol can be reversed at 1 mg naltrexone per mg of butorphanol, but it may not require reversal in each case.
 - The small sample size and limited effectiveness of this combination at the stated doses currently makes this combination not ideal for helicopterbased captures.
 - Further refinement of this combination for helicopter darting of bison **should** occur before it can be recommended.
- Given the limited drug options currently available for darting free-ranging bison, research into new combinations or dosages that do not require ultrapotent opioids and that are safe and effective for free-ranging, helicopter-

- based bison captures are required and **should** be a research priority for this species.
- Regardless of drug used, all drug vials must be clearly labeled with drug name(s), concentration, and date of preparation/reconstitution (CAZWV 2009).
 - If a drug combination is mixed in a vial obtained from a drug manufacturer, a highly visible label **should** be placed over the original to avoid confusion. Example: original Telazol label is replaced with a MZT label, plus the date of reconstitution and concentration
- Drugs must be protected from temperature, humidity, and light extremes (CAZWV 2009).
 - The drug storage instructions provided on the drug box or vial **must** be followed to ensure the effectiveness of the drug.
 - Azaperone will start to crystalize at temperatures just below room temperature and **must** be kept warm at all times.
- When not in use, drugs at field sites **should** be stored in a labeled, locked, crush-proof, leak-proof container that is lined with absorbent material (CAZWV 2009).
- Personnel in possession of drugs must be approved by the GNWT and have taken a recognized immobilization course within the past five years.
- All used drugs must be recorded, including amounts lost in missed darts or discarded (see Appendix C; CAZWV 2009).
- Unused, preloaded darts should be emptied at the end of each day using proper safely protocols to avoid potential drug exposure.
- If cleaning used darts that require disassembly, a venting pin **must** be used to vent the air chamber in case the drug chamber is still under pressure.
- Adequate steps must be taken to ensure that drugs used in bison are not abused by people or enter the human food chain via contaminated water or meat (CAZWV 2009).
- Animals must be clearly marked with an ear tag or collar that provides contact information to call before consumption (Kreeger and Arnemo 2018). Redundancy of marking methods appropriate for the study purpose is advised as the tags can fall off.
- Researchers must consult with communities within the study area and inform them that immobilization drugs, collars and/or tagging will be used during capture and what to do if they harvest a previously captured bison (Cattet 2017).

4.1.2.2 Net Gun

- A suitable capture technique for short-duration handling (<20 minutes).
- Enables rapid capture and release of the target animal.
- In most situations it will require the use of a helicopter.

- Ensure that the capture crew is experienced
- See Appendix B for detailed information regarding net guns and routine maintenance required.
- Physical trauma is the most common source of injury during net-gunning (Ferguson 2015) and steps **must** be taken to minimize the risks.
- The capture crew **should** evaluate the conditions at the anticipated capture area prior to initiating a capture.
- Stampeding while hazing **must** be avoided (Cattet 2017).
 - Hazing **must** be done in a controlled manner so that animals move at a slow pace as the target animal is gradually isolated.
- Animal pursuit (chase period) and capture must occur on smooth, open terrain with good footing and whenever possible, deep (30-60 cm) snow should be used (Kreeger and Arnemo 2018).
 - This will help reduce injuries (broken limbs, etc.) to the target animal and conspecifics and slow down fleeing bison.
 - Hard ground, packed snow, open water or ice **should** be avoided due to the higher risk of injury.
 - If bison are standing in heavily treed areas and are reluctant to move, devices such as cracker shells may be used to encourage movement (Cattet 2017).
- The chase period **should** be kept to <1 minute and **must** be kept to <2 minute of strenuous running.
- The chase **must** be terminated when either target or non-target animals show signs of exhaustion (e.g. panting, stumbling, etc.).
 - The risk of injury or death is significantly increased when animals are exhausted (Spraker 1993, Valkenburg et al. 1983) especially bison, which are prone to capture myopathy (Caulkett 2014)
 - Any additional attempts at capture of a fatigued animal **should** be postponed until the following day at minimum (Cattet 2017).
- Chases **should** be limited to ≤3 per larger group. Where multiple individuals are captured from the same group, the capture crew **must** avoid causing fatigue in non-target animals from prolonged hazing and **must** terminate chases if animals show any signs of exhaustion (Cattet 2017).
- The entire capture crew **must** ensure that chase limits are adhered to.
- Chase times (hh:mm:ss) **must** be recorded for each capture attempt.
 - Chase Start = Active, close range pursuit begins
 - Chase Stop = Time of capture/restraint, or when chase is broken off, e.g. due to exceeding time limit
- At least three capture guns with loaded nets, or a gun with detachable barrel and at minimum three nets, should be within arm's reach of the net gunner for each capture.
 - Back-up nets help to minimize the chase time if the first net misses or does not provide enough restraint (Cattet 2017).

- If a bison is not netted properly (i.e. head only) and does not go down shortly after netting, an additional net(s) **should** be deployed until the bison is properly restrained.
- The net gunner **must** only capture one bison in a net at a time.
 - Capturing two or more bison within a single net greatly increases the risk of injury or death and **must** be avoid.
- A capture team (four persons/adult animal) must be present to ensure all
 captured bison are restrained and handled immediately, while having the
 ability to move the animal if needed.
- Animals must be hobbled, blindfolded, handled, and released as quickly as possible following capture (Kreeger and Arnemo 2018).
- Attempts to capture and hobble two bison with separate nets prior to handling and sampling must not be done unless there are additional handlers present to attend to the first bison while the second bison is being captured.
- In the event of physical injury, the crew must re-evaluate if the techniques used and environmental conditions are suitable for additional captures (CCAC 2003).
- Administration of sedative or anesthetic-type drugs should be considered in situations where invasive and/or painful procedures will occur, or the handling time exceeds 20 minutes (CCAC 2003).
 - Drugs can be used to reduce capture stress, especially if bison excessively struggle or are severely entangled in the net, and can also be used before painful procedures are performed or for longer duration handling (Cattet et al. 2004).
 - This technique is described in more detail below.

4.1.2.3 Combination of Net Gun and Drugs

- The target animal **should** be netted as described above.
- Prior to administration of the immobilizing drugs, the crew should verify that the correct animal was captured.
- If the animal had a longer haze or chase period, a rectal temperature **should** be taken immediately and prior to administration of the drugs.
 - If the rectal temperature is \geq 41.0°C, measures should be taken to decrease the animal's body temperature, which may include immediate release.
 - If the rectal temperature is between 38-40.5°C, immobilizing drugs can be administered.
- Administration of short acting and/or reversible tranquillizer /sedative-type drugs (e.g. azaperone and/or xylazine/medetomidine; Joly 2001) should be considered to alleviate stress of capture and handling, especially if painful procedures are being performed or handling time will exceed 20 minutes (CCAC 2003).
- Intramuscular hand injection should occur into a large, superficial muscle mass and typically takes 10-15 minutes for sedation to occur (CAZWV 2009).

- Intranasal administration of drug and has been used effectively in elk and caribou (Cattet et al. 2004; Oakley et al. 2004) and **should** be considered as an alternative to IM administration.
 - Absorption of drug across the nasal mucous membranes results in quicker sedation than IM injection (5-10 minutes) and does not require the use of a needle. Accidental needle sticks are the most common method of human drug exposure (Petrini et al. 1993).
 - The use of a long atomizer (e.g. Teleflex MADgic® MAD720) with adjustable inner stylet is recommended to coat the mucous membranes with a fine mist.
- This reduces the probability of the animal swallowing a large percentage of the drug (and not becoming adequately sedated) if given via a red-rubber tube or plastic catheter.
 - The preloaded drug syringe with luer-locked atomizer **should** contain 1-3 ml of air behind the drug to ensure complete expulsion of drug through the atomizer.
 - Before insertion, the tip of the atomizer **should** be bent so it points dorsolateral to maximize the amount of drug coating the nasal mucous membranes.
 - The atomizer **should** be inserted at least half way along the nasal passage but not past the level of the medial canthus of the eye.
 - It is important that head movement is controlled (i.e. in lateral recumbency) if this method is utilized.
 - To reduce the likelihood of human drug exposure the nostril **should** be temporarily occluded to prevent expulsion of the drug if the animal sneezes.
- Short acting, reversible (partially or complete) drug combinations should be used and could include medetomidine, medetomidine-azaperone or BAM.
 Dosages will require further investigation before included in this SOP.

4.1.3 Recommendations for Approach

- Bison captured by net gun, must be approached and subdued as quickly as possible to avoid injury.
- When using remote drug delivery by helicopter, the pilot **must** land or hover at a suitable distance while maintaining a clear view of the animal to reduce stimulation during induction (Cattet 2017).
- In some situations where the animal may quickly become compromised (e.g. animal's head buried in deep snow), landing close by to quickly address this issue **should** occur if safe to do so.
- Bison immobilized by remote drug delivery must be approached cautiously and slowly to assess the animal's response to noise and touch and plane of anesthesia (Kreeger and Arnemo 2018).
 - Look for signs such as ear twitch or tail movement. If there are no voluntary movements, cautiously approach from the rear while providing the animal with easy escape options away from you.

- Initial approach **should** be from behind and/or away from the eyes with an experienced person on point with a loaded rifle (CAZWV 2009).
- Minimal stimulation of the immobilized animal is critical because peak drug effects generally occur sometime after immobilization (Caulkett et al. 1994).
- Excessive stimulation during induction or the initial approach may override the action of the drugs and cause an unwanted response by the animal (Nielsen 1999). Quick retreat to a safe location may be required.

4.1.4 Recommendations for Initial Handling

- Time of first handling (hands on time) **should** be recorded on the capture form.
- Sensory stimulation (visual, touch and auditory) must be kept to a minimum during handling (CCAC 2003).
 - Team members restraining and handling bison should stay clear of the striking area of the limbs and horns to prevent injury.
- All personnel involved with handling bison should wear nitrile or latex gloves to protect themselves from exposure to drugs and reduce risk of disease transmission (zoonoses).
 - In extreme cold, having nitrile gloves under a pair of mitts will prevent the gloves from tearing. When more delicate movements are required, remove the mitts while still wearing the nitrile gloves. This will protect the hands from drugs that can be absorbed across the skin and present serious health risks (Kreeger and Arnemo 2018).
 - As the animal is being untangled from the net, blindfold, hobbles and ear plugs should be applied.
- The eyes of anesthetized bison **must** be lubricated with eye ointment and covered with a blindfold to protect the eyes and reduce visual stimulation.
 - Drugs reduce tear production and therefore, eye ointment serves to prevent drying of the corneas (and potential ulceration) and provides a protective layer against debris, blindfold fabric and hair.
 - Apply a non-medicated eye ointment (e.g. methylcellulose) to the corneas without directly touching them.
- Hobbles should be applied after net gunning or in situations where they are required to reduce the potential for injury to the capture crew.
 - Hobbles should be placed just above the fetlocks on the canon bones to reduce the potential for trauma to underlying nerves and vessels that pass over the fetlock and pastern joints.
 - Ear plugs should also be used to reduce auditory stimulation (Arnemo and Evans 2017)
 - Large sponges cut to size work well for this purpose.
 - Sedative-type drugs with good pain-killing effects or local anesthetics **should** be provided to alleviate stress or pain in situations where painful procedures are employed or injuries have occurred during capture (Chinnadurai et al. 2016).
 - In addition, analgesics (e.g. Meloxicam) that provide several hours of pain relief should be used when pain or inflammation caused by capture or sampling occurs (Chinnadurai et al. 2016).

- Dart(s) **must** be removed from anesthetized bison at the onset of handling (CAZWV 2009).
 - Extra caution is required if ultra-potent narcotics are used.
 - Gloves **must** be worn and it is strongly recommended that long sleeves and protective eyewear be worn to further limit any potential drug exposure.
 - If using darts that require some disassembly (e.g. Dan-Inject darts), first remove the tailpiece and vent the air chamber before removing the dart. This eliminates any possibility of drug spraying from the dart during removal if the needle port was blocked during injection (Kreeger and Arnemo 2018).
 - Flushing of the dart wound **should** occur with sterile water, which cleans the wound and removes any drug contamination on the skin and fur (Kreeger and Arnemo 2018).
 - Avoid the use of topical antibiotics as a one-time application of a short acting topical antibiotic is not effective at preventing infection and the ointment will prevent adequate draining of the wound.
 - Only a licensed veterinarian **should** treat more serious dart wounds, (e.g. wounds that require sutures or needles that have penetrated the thorax and administration of injectable antibiotics).
- Anesthetized or restrained bison must be positioned so breathing is not impinged, i.e. keep neck straight, ensure nostrils and mouth are not blocked while keeping the head elevated and the mouth and nostrils pointed down to avoid aspiration.
 - Sternal recumbency is ideal to facilitate adequate ventilation while reducing the risk of ruminal tympany (bloat) (Caulkett 2014).
 - If on a hill, the head **must** be pointing uphill to prevent compression of the diaphragm from the rumen that can lead to reduced cardiopulmonary function as well as preventing regurgitation and potential aspiration.
- Reposition the animal or remove any rocks, branches or other hard objects to reduce the chance of injury.
- Repositioning the animal **should** only be done by rolling the animal over the chest with legs kept flexed, while keeping the spine aligned. Avoid rolling the animal over on its back/dorsum or grasping the skin and fur to move the animal.
- If lateral recumbency is required, the amount of time the animal is in lateral must be minimized.
 - Placing the animal in right lateral recumbency (with the right side down) is preferable to prevent compression of the rumen and potential regurgitation and to facilitate ease of monitoring for bloat over the left flank.
 - Monitoring physiologic responses (vital signs) to chemical immobilization should occur concurrently as other team members apply the blindfold, eye ointment, hobbles, +/- ear plugs.

4.1.5 Recommendations for Monitoring Physiologic Functions

- If using physical restraint only (i.e. net gun), the rectal temperature is usually the most important physiologic function to monitor.
 - This is especially important if the chase time was prolonged.
 - Hyperthermia can rapidly lead to severe complications.

- If an increasing rectal temperature trend is observed, particularly at 40-41^oC, consider shortening the procedures or reversing in order to prevent complications, such as hypoxia, capture myopathy and possibly sudden death.
- When sedation or anesthesia is performed, several physiologic functions (vitals) **must** be monitored closely and recorded throughout the handling event until the animal has recovered.
 - Assessments should be made and recorded in the animal capture form every 5 minutes (CAZWV 2009).
 - More frequent monitoring is required if vitals are outside recommended ranges or risky trends are observed.
- Attention to vital signs can provide advanced warning of developing complications (e.g. hypoxemia, hyperthermia, etc.) and provide opportunity for preventative measures or treatments.
 - Preventing complications is easier than treatment.
- A pulse oximeter **should** be used as an aid to monitor cardiopulmonary function but **must not** be used as a substitute to direct animal assessment.
 - Pulse oximeters allow continuous monitoring of cardiopulmonary function by measuring the percent hemoglobin oxygen saturation of blood as well as pulse rate (CAZWV 2009, Kreeger and Arnemo 2018).
 - Oxygen saturation values are often inaccurate because the devices are not designed specifically for wildlife, but monitoring trends is valuable.
 - If readings steadily decline, it is likely the animal's cardiopulmonary function is becoming compromised (Kreeger and Arnemo 2018).
 - When using a pulse oximeter, the probe should be applied at a consistent location (e.g. the tongue) and left in place until a stable and strong signal is obtained before recording the values.
 - Oxygen saturation **should** ideally be between 90-95%
 - Concurrent evaluation of mucous membrane colour will enable detection of hypoxemia, (oxygen saturation <85%, pulse rate increasing and blue mucous membranes).
 - Hypoxemia (<85%) arises more frequently in captured wildlife as a result of elevated body temperature and strenuous exercise, when the oxygen demand of tissues exceeds the supply, rather than from respiratory depression (Caulkett 2014).
- Supplementary oxygen should be available to treat hypoxemia (low blood oxygen levels) and hyperthermia and reduce the likelihood of the animal developing hypoxia, hypercarbia (elevated carbon dioxide) and capture myopathy.
 - Oxygen therapy is the most effective treatment for hypoxemia (Caulkett et al. 2000).
 - Compressed oxygen in pressurized cylinders (various sizes available) coupled with a regulator and nasal tubing can be used to deliver oxygen to animals in the field (CAZWV 2009).
 - The helicopter pilot **must** be consulted prior to field work to ensure they are comfortable carrying compressed oxygen on board.
 - The cylinders **should** be placed in a container fastened to framing in the helicopter to prevent them from moving during flight.

- A flow rate of 8-10 liters per minute is typically required for most adult bison (Harms et al. 2018) and the efficacy of treatment **should** be monitored with a pulse oximeter coupled with regular monitoring of mucous membrane colour.
 - If oxygen saturation exceeds 95%, the flow rate can be decreased to conserve oxygen tanks in the field.
- Supplemental oxygen must be provided to hyperthermic animals due to increased tissue demand for oxygen and to help reduce the likelihood of the animal developing lactic acidemia, hypercarbia and associated complications.
- Most ambulance supply companies provide this equipment and it should be standard equipment in field operations (Cattet 2017).
 - It can also be used for supportive care of field personnel following significant drug exposure.
- A summary of vitals to monitor and actions to take are listed below:
 - <u>Reflexes</u>: The presence/absence and strength of reflexes **must** be used to evaluate the level (depth) of immobilization. Reflexes provide early indicators of the animal's status and if additional drugs or reversal is required (CAZWV 2009).
 - Reflexes to monitor **should** include: ear twitch, palpebral, corneal, tongue and limb tone and withdrawal, jaw tone and anal sphincter reflexes.
 - At a medium depth of anesthesia (ideally for most situations), ear twitch, jaw tone and withdrawal reflexes **should** be absent with a weak or absent palpebral (drug dependent).
 - Respiratory system: Monitoring respiration is the most important factor when determining the status of an immobilized animal (CAZWV 2009) and must be monitored closely.
 - Evaluated by respiratory rate, depth or quality of breaths, and respiratory sounds.
 - Respiratory rate is affected by many factors (e.g. chase period, drugs, etc.) but, **should** remain ≥8 breathes/minute in an anesthetized bison (Caulkett et al. 2000). Each breath **should** be quiet, with full expansion and relaxation of the rib cage.
 - If the respiratory rate is <8 breaths/minute and other vitals point toward respiratory depression (i.e. blue or gray mucous membranes, oxygen saturation <85%), ensure the airway is not blocked due to head position, the blindfold or other obstructions and reposition the animal into sternal.
 - Supplementary oxygen should be provided, or flow rate increased. If these measures are not effective, reversal of anesthesia should be performed.
 - In emergencies, artificial ventilation via chest compressions performed by using knee thrusts down onto the ribcage can be used to mechanically ventilate large animals (Kreeger and Arnemo, 2018).
 - A needle or other sharp object can also be used to target the acupuncture point (Governing Vessel-26; GV-26) used in veterinary medicine to stimulate the cardiopulmonary system and treat apnea (Goe et al. 2016; Still 2001). The needle **should** be inserted into the upper lip, along the

- midline and just below the nostrils (see Kreeger and Arnemo 2018 for a picture).
- Ventilation via endotracheal tube is not typically performed in the field. If this equipment is available and personnel have been trained on its use, a mature bison will require a size 24–30 endotracheal tube (Caulkett 2014).
- <u>Cardiovascular system</u>: Evaluated by peripheral pulse, heart rate, blood pressure, mucous membrane color and oxygen saturation.
 - Pulse and heart rate are also affected by many factors as noted above but **should** remain between 50 and 80 beats per minute in an anesthetized bison (Caulkett et al. 2000, Caulkett 2014). In addition, mucous membranes (i.e. gums, anus, vulva) **should** be pink. Crude measures of blood pressure include capillary refill time (<2 seconds is normal) and strong pulse strength in the extremities (e.g. tail artery) that matches the heart rate.
 - If cardiovascular parameters increase or decrease outside of these ranges, respiratory function **should** be re-assessed immediately followed by assessing reflexes and then corrective actions taken if necessary.
 - Reversal drugs should also be available to administer but should not be given unless other signs indicate cardiovascular distress or collapse, i.e. blue or gray mucous membranes, prolonged capillary refill time (>2 seconds), dilated pupils, decreased reflexes, weak or absent peripheral pulses.
- <u>Body temperature</u>: Is heavily influenced by the chase period and respiratory rate and should be evaluated rectally using a digital thermometer.
 - A safe rectal temperature range is considered to be between 38°C and 40.5°C. A rectal temperature **should** be taken as soon as it is safe to do so and if outside the normal range, taken at more frequent intervals to monitor trends.
 - Administration of a reversal drug is the most effective field treatment if hyperthermia develops, because it enables the bison to use its normal cooling mechanisms, via panting (CAZWV 2009).
 - IV fluid administration is also effective at rapid cooling and **should** be considered if practical and the capture crew is trained in this technique.
 - Other methods, such as dousing with cold water in inguinal or axillary regions and cold-water enemas, may or may not be effective depending on access to water, animal size, body condition, fur thickness and rate of temperature increase.
 - Supplemental oxygen **should** always be provided to hyperthermic animals due to increased oxygen demand of tissues and potential to develop complications associated with lactic acidosis (e.g. capture myopathy).
 - Hypothermia (<37°C) may develop in some bison, especially smaller animals that are captured under extremely cold conditions.

- If possible, treatment **should** be directed toward active warming (heating pads, drying wet fur) of the animal until the body temperature has returned to >37°C before administering reversal drug.
- A spare thermometer **should** be carried as backup.
- All vitals **must** be recorded on data forms at the time of capture and handling.
 - These data are crucial in helping identify health complications occurring during or following handling or in the unlikely event of the death of a bison (CAZWV 2009). A template is provided in Appendix C.

4.2 Sample Collection and Measurements

- Sample collection and measurements **must** be completed efficiently with the goal of releasing the animal as soon as possible.
- Samples and measurements must be consistent with those approved by the animal care committee.
- It is recommended that a standardized set of biological samples be collected from each bison, processed, tested and/or banked for future use.
 - Properly processed and banked samples provide an invaluable resource for future analysis and research and may reduce the need for additional captures.
- Teeth **must not** be extracted from an immobilized animal.
- Bison can be aged via horn characteristics (see Appendix C) and therefore, this method **must** be used on live bison rather than removal of incisor teeth.
- The incisor teeth **should** be extracted from dead or euthanized bison only and used in combination with horn characteristics for accurate age classification.
 - Collection of the entire lower jaw is also acceptable.
- Local anesthesia **should** be used for sampling procedures that are likely to cause pain, e.g. ear punch, skin/fat biopsy (CCAC 2003).
 - In situations where analgesia is required, infiltration of the biopsy/punch site with 2-3 ml of lidocaine (with 2% epinephrine, if not at an extremity) at minimum 5 minutes before tissue collection will provide sufficient pain control (Cattet 2017).
 - Additional analgesia should be provided if indicated (e.g. Meloxicam)
- Non- or minimally invasive procedures **should** be used for genetic sampling, (e.g. ear punch, feces, hair, or oral swab).
- If an ear tag is applied, the hole **should** be made with a sterile biopsy punch (4-6 mm diameter) and the tissue plug **must** be preserved for any analyses that require tissue samples (see Appendix E).
 - To perform this procedure, a small portion of the fur on the ear **should** be clipped and wiped with dilute chlorohexidine until clean (no dirt on gauze wipe).
 - Effort **must** be made to avoid any major blood vessels in the ear. If bleeding occurs, apply pressure with gauze for a few minutes.
 - The ear tag stud is manually directed through the biopsy hole prior to securing the tag with the applicator (Cattet 2017).
 - Application of a topical antibiotic ointment must be avoided.
 - There is no scientific evidence that a one-time application of a topical antibiotic ointment is effective at preventing infection.

- Additionally, it is not considered prudent use of antimicrobials (Silley and Stephan 2017).
- Sampling of blood and tissue **should** be performed only after appropriate training and experience. Proper collection and handling and preservation protocols **must** be followed to obtain useful samples (see Appendix E).
- Given the challenges and potential risks to personnel, justification for the need to weigh bison in field conditions **should** occur prior to starting the project.
 - If the handling protocol requires weighing captured animals, weighing **must** be done in the least stressful manner possible.
 - If the animal is sedated/anesthetized, weighing **should** occur when the animal is at a medium plane of anesthesia. Manipulating an animal when it is lightly sedated/anesthetized (generally at the start or end of the immobilization) will stimulate it and could cause a spontaneous recovery, compromising the capture crew safety.
 - Animals **must not** be suspended upside down by the hobbles. This increases the risk of regurgitation and fatal aspiration (Kreeger and Arnemo 2018). Instead, it is recommended that bison are weighed by first positioning them on their sternum in a handling blanket and then suspend the animal while keeping it sternal.
- The animal must receive a standardized physical examination, checking for wounds, injuries, disease and general condition. This information must be recorded in Appendix C.
- Antibiotics must not be administered routinely to captured bison. Antibiotics must
 only be prescribed by a veterinarian and only if a diagnosis of a bacterial infection is
 made.
 - The effectiveness of these drugs in free-ranging wildlife is often unproven and largely unknown (Pietsch et al. 1999). In addition, antibiotic residues (metabolites) often remain in tissues for long periods of time posing a public health risk to persons consuming bison meat (Cattet 2002).

4.3 Identification, Marking and Telemetry Collars

- Identification or marking techniques **should** be non- or minimally invasive and **must not** compromise animal welfare (CCAC 2003).
- If applicable, they **should** remain visible (ear tag/collar) and/or functional (microchip) for the duration of the study.
 - Methods and unique ID numbers must be recorded on the animal capture form.
- Investigators **must** minimize any adverse effects of identification or marking procedures on the behaviour and physiology of the animal (CCAC 2003).
- Telemetry collars **should** be as light as possible (ideally less than 2% of body weight).
 - They **should** be selected based on the needs of the research or management project while maximizing animal welfare.
 - Collars **should not** be left on the animal indefinitely.
 - Collar size requirements will be different for bulls vs. cows.
- All collars **should** incorporate a drop off mechanism to release the collar without requiring recapture.

- All collars **should** have a rot-off section to ensure they will eventually be shed.
- Electronic, programmable break-away mechanisms can be used, but a rot-off feature should be incorporated into these collars in case the electronic break-away mechanism fails.
- A "drop-off schedule" **should** be developed in conjunction with collar deployments. This schedule **should** be used to monitor and confirm that collars have dropped from the animals (Cattet 2017).
- Collar materials and design **must** be appropriate to avoid causing debilitating injuries to study animals (CCAC 2003; Krausman et al. 2004).
- Experienced personnel **must** apply the collars to minimize risks (e.g. too tight or loose).
- Collar size **should** be based on several factors including animal age, sex, condition and time of year.
 - If not already collected, data on animal neck size ranges (by age, sex and season) **should** be collected and shared among researchers to provide a basis for selecting collar size (Cattet 2017).
 - Neck sizes from immobilized Yukon bison are as follows (100+/-10 cm for females and 150 +/-20 cm for males) (Jung 2019 personal communication).
- Ear tag transmitters **should not** be used on bison unless sufficiently justified in the animal care protocol.

4.4 Reversal Drugs and Releasing Bison

- Although moving a bison is not likely to be feasible under field conditions, the ideal position for the animal is to be facing the general direction of the herd and/or away from potential hazards, with a clear escape route visible to the animal.
- Non-essential crew members must move to a safe location and all equipment removed from the capture site prior to the reversal drug being given (CAZWV 2009).
- Hobbles and ear plugs must be removed and the blindfold strap released (but blindfold still covering the eyes) prior to giving the reversal drugs.
- Reversal **should** typically be given via IM injection, immediately followed by removal of the blindfold and allowing a few minutes to retreat to safety. Only in emergencies, where the animal is severely compromised, **should** partial or full reversal via IV injection be considered. If IV reversal is given, the blindfold **must** be removed beforehand and all personnel **must** be able to reach a safe location in <1 minute (Caulkett et al. 2000).</p>
- Once the reversal drug is administered, the person(s) administering it **must** retreat to a safe location to monitor the recovery.
 - During this time, monitoring respiratory rate and movements should still occur and be recorded.
 - Every effort **must** be made to observe the animal until it is ambulatory, with purposeful, coordinated movements and able to defend itself or flee from predators or conspecifics.
- Non-anesthetized bison **must** be provided with a clear path for retreat before the hobbles (and ear plugs) are removed.

• The person(s) releasing the hobbles **must** position themselves to avoid being kicked or gored. Remove the blindfold as soon as the hobbles are released and quickly retreat to the safety of the hovering helicopter.

4.5 Post-Capture Monitoring

Recommendations:

- Animals with radio collars **should** be observed within 24 hours of capture to detect any issues or mortalities (Kreeger and Arnemo 2018).
 - This is especially important if complications occurred during the capture operation (e.g. hyperthermia, physical injury).
 - Death may be delayed for hours, days or weeks following capture and any mortalities that occur within two weeks of capture **should** be investigated as capture related (Kreeger and Arnemo 2018).
- Bison **should** be observed from a distance that minimizes stress associated with the vehicle or personnel.
- When observing bison is not possible (e.g. dense tree cover), the animal **should** be confirmed alive and moving via changes in VHF locations.
 - Activation of the mortality sensor may not always occur if periodic movements of the collar are caused by animals feeding on the carcass.

4.6 Humane Killing (Euthanasia) of Bison

- The investigator **must** be prepared to humanely kill (euthanize) any animal that is suffering due to intolerable pain, irreversible injury or distress that is the result of capture, handling and sampling procedures or interventions (CCAC 2003).
- The technique used **must** be humane and reduce pain and distress as much as possible (CCAC 2003).
- Investigators **must** be trained in the method(s) to ensure effective, humane kills and always carry the appropriate equipment while working with bison.
- Consideration **must** be given to techniques that do not interfere with collection of important diagnostic samples during the post-mortem.
- While a variety of humane killing methods can be used, only acceptable methods for killing bison in field conditions are include here:
 - a) Gun Shot
 - An effective method and often the only option in field situations.
 - The capture crew **must** carry a firearm and ammunition that meets legal requirements for hunting bison in the NWT (i.e. a minimum 30 calibre rifle or shotgun with a bullet that results in at least 2,800kJ of energy at the muzzle).
 - A properly placed head shot should pass through the mid-brain and brainstem to produce instantaneous loss of consciousness and humane death (AVMA 2013). This shot placement **should** generally be limited to when the animal is physically or chemically immobilized (AVMA 2013).

- If the brain is required for testing, a neck shot through the spinal cord can also be used if the animal is adequately restrained (AVMA 2013; Kreeger and Arnemo 2018).
- In situations where the bison is not restrained and intact brain is required, a gunshot to the heart and lungs is acceptable (AVMA 2013).
- Due to the skull thickness of mature bulls, a centerfire rifle is required for a head shot; shotguns **should not** be used for these animals (acceptable for yearlings and cows only). For a list of acceptable rifles, cartridges and target areas for humanely killing bison, refer to the National Farm Animal Care Council: Code of Practice for the Care and Handling of Bison (available at www.nfacc.ca/bison-code-of-practice).

b) Exsanguination (Bleeding)

- This method **must** only be performed on an anesthetized animal (AVMA 2013).
- Performing this technique on a conscious animal is not considered humane.
- Both carotid arteries **should** be cut to ensure rapid exsanguination (Kreeger and Arnemo 2018).

4.7 Food Security and Carcass Disposal

Recommendations:

- Any bison carcass suspected or known to be contaminated with drug residues (immobilization drugs or antibiotics) **should** be deeply buried or removed from the field to prevent consumption of meat by either people or wildlife. Consult regional waste management regulations and procedures for appropriate disposal.
- Whenever feasible, non-lead-based ammunition **should** be used (e.g. copper) to euthanize animals to prevent contamination of meat (Arnemo et al. 2016).
 - Lead shot in wildlife is considered the largest contributor to lead contamination in the environment and poses risks to both human and animal health (Arnemo et al. 2016). Therefore, lead **should not** be present in any meat left on the landscape or offered to communities for consumption as this is a potential liability issue.
 - If lead-based ammunition is used, the lead contaminated tissues must not enter the food chain.

4.8 Morbidity and Mortality

Recommendations:

All significant injuries (e.g. broken leg or neck) observed during or following capture or handling (two weeks minimum) **must** be documented and reported to the ENR Wildlife Veterinarian as soon as possible.

- Whenever possible, the veterinarian **must** be contacted via satellite phone at the time of the incident.
- An investigation must be conducted and if the ENR Wildlife Veterinarian is unavailable, another wildlife veterinarian delegated by the ENR Wildlife Veterinarian should be contacted.

- Investigation should include collection of standardized samples for banking and/or submission to a veterinary diagnostic laboratory for further evaluation. Pictures should also be taken to document the incident for future reference and for educational and training purposes.
- Minor injuries (e.g. superficial wounds) **should** be recorded but do not require immediate reporting at the time of the incident.
- All mortalities observed during or following capture or handling (two weeks minimum)
 must be reported to the ENR Wildlife Veterinarian as soon as possible. Whenever
 possible, the ENR Wildlife Veterinarian should be contacted via satellite phone at the
 time of the incident.
- All dead bison (even if the cause of death is obvious) **should** receive a necropsy with collection of standardized samples and data collection.
 - Important exceptions to this are any bison where anthrax is suspected. In these cases, the NWT Anthrax Response Plan **must** be followed (Elkin et al. 2013).
 - Diseases or other health concerns are often noted in animals that have died from other causes and therefore, opportunities to conduct health surveillance and sampling on any bison mortalities is important and will improve our knowledge of this species at risk.
 - If the necropsy is performed in the field, standardized tissue samples **should** be collected and kept cool and/or frozen (less ideal) for submission to a veterinary diagnostic facility. Appropriate tissue samples as well as processing and storage are listed in Appendices D and E. Investigators **should** receive training in performing a wildlife necropsy from either the ENR Wildlife Veterinarian or through further training or reading (Terio et al. 2018).
- A template for a field necropsy data form is provided in Appendix D. Documentation **must** include a detailed history and images of the field necropsy to assist the veterinarian diagnosing the cause of death.
- If there are two or more significant injuries or mortalities during a bison capture operation, the ENR Wildlife Veterinarian (or their delegate), should be contacted, captures suspended and the incidents must be discussed prior to resuming captures. The ENR Wildlife Veterinarian will contact the Regional Superintendent and Director of Wildlife and Fish.
- All meat not contaminated with drugs or lead must be collected from the capture site
 for potential distribution to communities. The crew must be equipped to properly
 salvage the meat. The project lead needs to identify the most appropriate place to take
 the meat prior to the capture operation. The crew must be aware of gross lesions for
 common conditions (e.g. bovine brucellosis and tuberculosis) and be able to take
 appropriate samples to rule out those conditions.
- Photo documentation can help with case work up.

4.9 Human Safety

Recommendations:

• Refer to Appendix B for more detailed information on human safety during capture events.

- If a crew member feels their safety or the safety of others is at unreasonable risk, steps **must** be taken to address these concerns before the project starts or resumes.
- Appropriate handling and restraint techniques outlined above **must** be used for bison.
 - Personnel must have appropriate training and experience in their use to avoid injury.
 - Bison are capable of inflicting serious injury and transmitting zoonotic pathogens to persons handling them.
 - An experienced guard with a suitable firearm and ammunition **should** be on hand when working with bison.
- The risks involved in using drugs for the capture and immobilization of bison must be identified and communicated to all personnel involved in the project, including the helicopter pilot (CAZWV).
 - If ultra-potent opioids are used, the human reversal drug (naloxone or naltrexone)
 must be drawn up and the syringe labelled and placed in a location that is known to capture crew members to facilitate quick administration if required.
 - Given the short half-life of naloxone and need for repeated injections and multiple vials to reverse an ultra-potent opioid exposure, written consent from capture crew members to be administered naltrexone instead of naloxone should be considered prior to starting any fieldwork.
- The investigator **must** ensure that an emergency action plan is in place.
 - This plan provides step-by-step directions on what to do in an accident or emergency (e.g. human drug exposure, downed aircraft) and if well designed and implemented, can reduce the severity of emergencies and save lives.
- An evacuation plan to medical facilities **must** be discussed prior to fieldwork.
- At least two people on the capture team **must** have current training and certification in first aid and cardiopulmonary resuscitation (CPR).
- It is recommended that local medical authorities be informed of the potential hazards (e.g. drugs and potential zoonoses) prior to starting the project (CAZWV 2009)
- Personnel handling drugs **must** have current and recognized training (within five years) and inform other members of the team of the risks of human exposure and procedures for addressing this.
- Helicopter pilots and net-gunners **must** conduct a safety briefing at the start of the capture operation to outline the safety features of the aircraft including the location of the fire-extinguisher, emergency beacon and first aid kit.
- Helicopter pilots and net-gunners **must** have demonstrated skills in minimizing stress on target and non-target animals via gradual hazing and effective and efficient chase of target animals while also maintaining human safety.
- The lead investigator **must** ensure that potentially hazardous conditions involved in fieldwork are identified to all personnel involved and steps are taken to minimize these risks.
- Specific experience or training working around aircraft or with firearms **must** be considered prior to starting the project (CCAC 2003).
- A safety harness anchored at two points to the helicopter **must** be used by the crew member performing the darting or net gunning.

- The use of appropriate safety clothing for the weather conditions (e.g. Nomax coveralls and flight helmet) and equipment (e.g. headphones and gloves) **should** be used.
- Clothing, field gear, and equipment should be routinely cleaned during and after capture to prevent pathogen transfer to individual animals and to people.
- Communication (either verbal or via radio) **must** occur between the pilot and other crew members to ensure safety of all members.
 - For example, the pilot **must** be aware of when a bison will be given the reversal drug to ensure that a quick departure can occur if needed.
- Personnel involved in capture and restraint **must** have current training and proficiency in the use of pertinent equipment, e.g. firearms, dart rifles, etc. (CCAC 2003).
- Following completion of a training course, inexperienced personnel **should** develop and refine their skills by working with a mentor.
- While bison safety and welfare must be optimized, at all times human safety must take precedence.

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Appendix A - Drug Volume Table for Bison

Drug P	rotocol	MZT ^A		
Dru	ıgs	Medetomidine (M) + Zolazepam + Tiletamine (ZT ^B)	Atipamezole	
Formu	Ilation	1 ml M (@ 30 mg/ml) + 1 ml sterile water per vial of ZT	10 ml per vial	
Concentrat	ion (mg/ml)	13.5M + 230ZT	20	
Dosage	(mg/kg)	0.06 – 0.08M + 1.2 – 1.4ZT 0.4		
Body	Mass	Total Vol	ume (ml)	
kg	lbs	Total Volume (ml)		
100	220	0.4 - 0.6	1.4 - 2.0	
200	440	0.9 – 1.2	3.0 - 4.0	
300	660	1.3 – 1.8	4.4 - 6.0	
400	880	1.8 – 2.4	6.1 – 8.0	
500	1100	2.2 – 3.0	7.4 – 10.0	
600	1320	2.7 – 3.6	9.1 – 12.0	
700	1540	3.1 – 4.2	10.5 – 14.0	
800	1760	3.6 – 4.8	12.2 – 16.0	
900	900 1980		13.5 – 18.0	
1000	2200	4.4 – 6.0	14.9 – 20.0	

 $^{^{\}rm A}$ Adapted from Caulkett et al. 2000; Caulkett 2014 and Government of Alberta 2016. $^{\rm B}$ Sold commercially as Telazol® or Zoletil®.

Appendix B - Net Gun Maintenance, Use, and Inspection

Contributed by Ian Ellsworth, Trinity Tactical Consulting Ltd. Reviewed December 2018.

1.0 Net Gun Standards:

Various makes and models of net guns are currently used for wildlife capture operations. However, regardless of differences among types of net guns, existing or new, all net guns being used in the Northwest Territories **must** meet the following minimum standards:

- 1. Have removable barrels that contain net canisters for quick reloading;
- 2. Be equipped with a minimum of three barrel/canister units and appropriate-sized nets;
- 3. Have a minimum of four spare weights complete with 0-rings and tethers;
- 4. All weight tethers must utilize rubber tubing (capable of withstanding cold temperatures) at the junction of the weight eyelet; and
- 5. Have a safety mechanism that prevents the gun from being fired.

2.0 Maintenance:

2.1 Pre-Capture:

Net guns and accessory equipment **must** be inspected by operators prior to capture operations to ensure everything is fully functional and reliable.

- Gun:
 - Inspect for cracks or loose parts.
 - Ensure action is clean and free of oil or grease.
 - Ensure breech is clean and free of obstructions.
 - Ensure action operates smoothly and firing pin engages.
 - Check safety mechanism for proper operation.
 - Check trigger for proper operation.

Canister Units:

- Ensure barrels are clean and free of cracks and obstructions.
- Check retaining bolts for tightness.
- Inspect canister for cracks or defects.
- Inspect flaps and Velcro fasteners.

Nets and Weights:

- Check for holes or tears in nets, and repair or replace as needed.
- Ensure weights are not damaged and file burrs, if required.
- Check O-rings for wear.
- Check weight tethers for fraying or other damage.
- Check rubber sleeves and fasteners for wear.

2.2 Capture:

The net-gunner **must** regularly check nuts and bolts that may loosen with continued use. Similarly, the canisters and weights **must** be checked after each capture. Fiberglass canisters can break, rivets may tear away from closure flaps, and Velcro may lose its

effectiveness. It is also necessary to inspect weights as O-rings will wear and weight tethers will fray and break with repeated use, and may need to be replaced. Nets **must** be dried out at the end of each capture day.

2.3 Post-Capture:

Use the pre-capture list as your guide. In addition, you **should** thoroughly clean the gun and weights. Lubricate metal parts (except the bolt/firing pin) to inhibit rust. If you wish to lubricate the action, use graphite or an equivalent product that will not freeze in the cold. Dry out, clean, and disinfect all nets prior to storage.

2.4 Equipment Inspection:

All net-gunning equipment to be used during a capture season **must** be inspected by an experienced gunner prior to use. If any issues arise, the equipment **must** be repaired by a qualified technician. All inspections **must** be recorded, problems noted, and corrective actions listed on the Inspection Checklist.

3.0 Safety Equipment:

The safety of all personnel during capture operations is paramount. So, all team members **must** receive a safety briefing prior to departure. Further, all personnel **must** wear an approved helicopter helmet and clothing appropriate to the weather during capture operations. Net gunners **must** wear a harness or safety belt with a minimum of two lanyards with safety clips attached to the harness or belt and secured to fixed anchoring points in the aircraft. Lanyard tension **must** be assessed and, if necessary, altered prior to each capture. Harnesses or safety belts **must** also be adjusted to the appropriate tension to limit forward and side-to-side motion. Harnesses, safety belts and lanyards **must** also be inspected daily and replaced at the first sight of any fraying or wear. Safety equipment that has passed an expiration date **should not** be used.

4.0 Ammunition:

All net-gun ammunition **must** be hand loaded by competent personnel or purchased from a commercial distributor if available. Used brass **should not** to be used for reloading of net-gun ammunition.

5.0 Net Gunning Sequence and Considerations:

5.1 Pre-Capture:

The gunner will ensure:

- All equipment is ready.
- Net weights are plunged into barrels and properly seated (0-rings help to secure the weights in the barrels).
- Spare canister units are accessible if a second or third shot is required.
- Harness/safety belt and lanyards are properly adjusted.
- Door is removed from the helicopter, unless it is a sliding door.

5.2 Capture:

The gunner will:

- Communicate with pilot to see if airspeed is sufficiently slowed to open the sliding door.
- With the door open (or removed), place a cartridge into the chamber but **do not** close the action.
- Get into shooting position and select a target animal, while controlling the muzzle direction at all times to ensure it is pointing outward from the cabin, but never directed toward the rotor blades or skid.
- Once chase begins, close the action, disengage the safety, and keep finger outside of the trigger guard until ready to fire.
- When animal is safely within range, deploy the net.
- If a second shot is required, re-engage safety, open action, change canister, reload and follow preceding steps.
- If unable to shoot during the first pass and extended time is required to get into range, re-engage safety and open action to prevent accidental discharge. Keep muzzle pointed in safe direction and finger outside the trigger guard.
- If a misfire occurs, wait a minimum of 30 seconds before ejecting the cartridge. Always ensure the gun is pointed in a safe direction in case of delayed detonation (also called hang fire).

5.3 Post Capture:

The gunner **must** inspect the net, weights and O-rings prior to reuse. Any worn parts **must** be replaced. It is quite common for O-rings to compress and fail to hold the weight in the barrel. This can result in a net unexpectedly falling out of the canister during a chase, resulting in a dangerous situation. If this should occur, the gunner **must** immediately release the canister unit or drop the entire net gun unit to the ground. Failure to do so may result in the net becoming entangled in the tail rotor.

Worn weight tethers are also a concern as they may break during a shot and end up hitting a rotor or other undesirable places if the muzzle is not pointed in a safe direction.

When reloading nets, ensure the net gun is unloaded, the action is open, and the safety is engaged. Also, ensure the action and barrels are kept out of the snow.

Appendix B - Net Gun Inspection and Verification Checklist

COMP	ANY INFORMATION	N .						
Inspect	Inspectors name:							
Addres	SS:							
Teleph	one number:							
Unit ID	(of Gun)							
Date of	f Inspection:							
CHEC	KLIST	Comme	nts/ Deficiencies					
	Gun unit free of cracks	s, loose parts or other damage						
	Action/Breech/Trigger	clean, free of obstruction and operable						
	Safety mechanism in	working order						
	Barrels on all canister	units clean, free of cracks and obstructions						
	Canister release mech	nanism operates smoothly on all canisters						
	Canisters free of cracl	ss or other damage (inside/outside)						
	Canister flaps secure/	adequate Velcro to hold net						
	All nuts/bolts on canis	ters tight						
	Weights undamaged a	and fit smoothly into barrels						
	O-rings capable of hol	ding weights into barrels						
	Weight tethers free of	fraying or wear						

Supplemental Notes:

Nets free of holes/tears

I hereby declare that the above listed unit is serviceable

I hereby declare that the above listed unit is unserviceable based on the noted deficiencies and shall not be used until rectified and re-inspected

Signature of Inspector

Appendix C - Bison Capture and Health Data Sheet																			
Personr	Personnel: Date: (dd-mm-yy)																		
Animal Data:																			
GNWT ID	GNWT ID# Sex: □ M □ F □ Unk. Age class: □ Calf □ Yearling □ Sub-adult																		
Other ID#	‡			I	Lactating? ☐ Yes ☐ No ☐ Adult ☐ Breeding Adult ☐ Old ☐ Very Old)ld					
Recaptur																			
Relocated? ☐ Yes ☐ No Location:																			
Capture	e an	d Lo	cati	on D	ata:														
Capture N						System:		Dart S					Ch	ase S	Start	(hh:r	nm:ss)		
□ Net □	Dart	□ Co	mbo			□ PneuD	rt			, ,									
☐ Other:					ther:			□ Rap			t/end	ports	Ch	ase l	End	(hh:m	ım:ss)		
General L	ocati	oni						□ Oth	lei	r:								1_	
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Drug Da	ata:									V	itals	(5 mi	nut	tes	inte	erval	s):		
-		Injec	ct #1	Injec	t #2	Inject #3	Re	eversal		Time		PR		RR		°C	SpO ₂	02	
Drug/Cor	nbo					,													
Vial ID#																			
Dose(mg))																		
Vol. (ml)																			
Time																			
Inj. site	· /																		
Inducti	on S	equ	ence	e (hh	:mm:	ss)					Rec				nce	e (hh	:mm:	ss)	
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□ Red			Hair	_				ound fl		snea		freq. □ Tat		що			□ Capt		iata
☐ Purple ☐ SST			Feces					ntment nitter n		anota	off	locati		# &			⊐ Loca data	uon	
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Comme	Comments:																		

Appendix C – Bison Capture and Health Data Sheet

Health Assessment Checklist:

Motor Dla	aca talza	nictures o	fante	locione in	addition	to rocording	data in	the checklist
note: Fie	ase lake	pictures of	lally	16210112 111	auuiuoii	to recording	uata III	the thetkiist

			duition to recording t	
Overall	Eyes	Nostrils	Mouth	Teeth
□ Appears healthy	☐ Normal	☐ Normal	☐ Normal	□ Nomal
□ Unhealthy	☐ Abnormal: ☐ L ☐ R	☐ Abnormal: ☐ L ☐ R	☐ Ulcer/Erosion	☐ Broken
☐ Pictures	☐ Redness	□ Ulcer/Erosion	☐ Growths	☐ Abnormal wear
☐ Other:	☐ Swelling	☐ Trauma	☐ Swelling	☐ Pictures
<u></u>	☐ Cloudy	☐ Discharge: ☐ L ☐ R	☐ Trauma	☐ Other:
BCS (1-5)	☐ Trauma	□ Clear	☐ Discharge	Ears
☐ Emaciated (1)	☐ Ulcer/Erosion	□ Pus	□ Pus	☐ Normal
□ Poor (2)	□ Sunken	□ Blood	☐ Blood	☐ Abnormal: ☐ L ☐ R
☐ Satisfactory (3)	☐ Discharge: ☐ L ☐ R	□ Foam	☐ Rumen fluid	□ Trauma
☐ Good (4)	□ Pus	☐ Rumen fluid	☐ Swab(s)	☐ Discharge
□ Obese (5)	□ Blood	☐ Swab(s)	☐ Other:	☐ Swab(s)
	☐ Swab(s)	☐ Other:	1	☐ Other:
	☐ Other:		1	
Skin	Respiratory	Cardiovascular	Gastrointestinal	Musculoskeletal
□ Normal	□ Normal	□ Normal	☐ Normal	□ Normal
☐ Normal	☐ Normal	☐ Abnormal	☐ Bloat	
		□ > 2sec. CPR	☐ Regurgitation	☐ Swollen joint(s)
☐ Poor/dull fur	☐ Blue m.m.	☐ Weak pulses	☐ Diarrhea	□ Abscess
☐ Fur loss	☐ Abnormal sounds	☐ Pale m.m. ☐ Tacky/dry m.m.	☐ Fecal staining ☐ Rectal blood	☐ Growth ☐ Trauma
☐ Growths	☐ Difficulty breathing	☐ Hemorrhage	☐ Rectal blood ☐ Parasites	☐ Hoof lesion(s)
□ Wounds	☐ Aspiration	☐ Other	☐ Swab(s)	☐ Muscle wasting
☐ Parasites	□ Blood		□ Other:	□ Swab(s)
☐ Samples taken	☐ Foam	1	1	☐ Other:
☐ Other:	☐ Swab(s)	1	1	1
	☐ Other:	ĺ'	<u> </u>	<u> </u>
Reproductive	Urinary	Nervous	Horn Age Classification	Body Condition Score (BCS; 1-5)
☐ Normal	☐ Normal	☐ Normal	MALES FEMALES 6 Month Calf 6 Month Calf	BCS Spine
☐ Lactating	□ Blood	☐ Abnormal	6 Month Call	1 Hips
☐ Mastitis	☐ Sampled	☐ Trauma	12 Month Yearling 12 Month Yearling	1) / / / / / / / / / / / / / / / / / /
☐ Vulvar swelling☐ Prepuce swelling	☐ Other:	☐ Paresis ☐ Paralysis	18 Month	1
☐ Discharge	1	☐ Paralysis ☐ Seizure	18 Month C1 Young Cow 2 and 3 Years	
☐ Clear		☐ Other:	A D 0 0 0 0	2
□ Pus	Lymph nodes	1	B1 Juvenile . 2 to 3 Years C2 Prime Cow 3 to 12 Years	1 ' _ `
☐ Blood	☐ Normal	1		3
☐ Swab(s)	☐ Enlarged	1	B2 Subadult 4 to 6 Years C2 Prime Cow 3 to 12 Years	3 }
☐ Other:	☐ Discharge	1		
	☐ Sampled ☐ Other:	1	B3 Adult 7, 8 and Older C2 Prime Cow 3 to 12 Years	
	☐ Other:	1	B3 Adult 7,8 and order	4
	1	1		'
	1	1	B4 Prime Older than B3 C3 Old Cow 12 to 20 Years	\sim
	1	1		5
	1	1	B5 Old Older than B4 C3 Old Cow 15 to 25 Years	/ \
	1	1		BCS adapted from: www.usask.ca/wcvm/herdmed/specialstock/bison/pdf/bc
	1	1	Adapted with GNWT permission, 2018	www.usask.ca/wcvm/nerdmed/specialstock/bison/pdf/bc s-bison.pdf

Comments:	 	

Appendix D – Field Necropsy Data Form

Personnel: Date: Location:

Euthanasia: ☐ Yes ☐ No If Yes: ☐ Management ☐ Capture related ☐ Disease suspect ☐ Other:

Method of euthanasia:

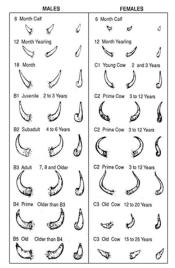
Site Characteristics: * **CAUTION: Anthrax suspect? If Yes, Do Not Open Carcass** * Contact supervisor and if appropriate, initiate NWT Anthrax Response Plan.

Distant Observations	Weather at Carcass	Other Observations/Comments:
□ 1 dead	Temp: ⁰ C	
□ >1 dead*		
☐ Others ill/diseased*	Humidity:	
☐ Blood from orifices*	☐ Rain ☐ Snow	
☐ Saw horse stance*		
☐ Bloat*	Amount: cm	
☐ Predator on site	Other:	
□ Other:		

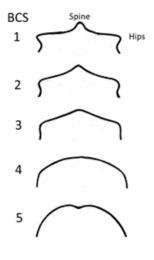
Carcass Information:

ID #s: □ Collar:	□ Ear tag	☐ Other ID:					
Sex: □ M □ F □ Unknown	Pregnant: □ Yes	□ No □ NA	Lactating: ☐ Yes ☐ No ☐ NA				
Age Class: □ Calf □ Yearling □ Sub-adult □ Adult □ Breeding Adult □ Old □ Very Old							
Horn and Incisor Wear: ☐ Pictur	es	BCS: □1 □ 2 □ 3 □ 4 □ 5					
Carcass Condition: □ Intact/Whole □ Partial □ Fresh □ Decomposed □ Predation □ Scavenging							
Pictures of Mortality Site: ☐ Who	ole carcass tality site	Estimated Days Since Death:					

Age Classification



Body Condition Scoring (BCS; 1-5)



BCS adapted from: www.usask.ca/wcvm/herdmed/specialstock/bison/pdf/bcs-bison.pdf

Appendix D – Field Necropsy Data Form

*Caution: Indicates a possible hazardous/zoonotic disease (e.g. Anthrax, TB or Brucellosis)

Overall	Eyes	Nostrils	Mouth	Teeth
☐ Appears healthy	☐ Normal	☐ Normal	☐ Normal	☐ Normal
☐ Unhealthy	□ Abnormal: □ L □ R	□ Abnormal: □ L □ R	□ Ulcer/Erosion	☐ Broken
□ Pictures	☐ Redness	☐ Ulcer/Erosion	☐ Growths	☐ Abnormal wear
☐ Other:	☐ Swelling	☐ Trauma	☐ Swelling	☐ Impacted
	☐ Cloudy	☐ Discharge: ☐ L ☐ R	☐ Trauma	☐ Other:
BCS (1-5)	□ Trauma	□ Clear	☐ Discharge	Ears
☐ Emaciated (1)*	☐ Ulcer/Erosion	□ Pus	□ Pus	□ Normal
☐ Poor (2)*	☐ Sunken	☐ Blood*	☐ Blood*	☐ Abnormal: ☐ L ☐ R
☐ Satisfactory (3)	☐ Discharge: ☐ L ☐ R	□ Foam*	□ Foam*	□ Trauma
☐ Good (4)	□ Pus	☐ Rumen fluid	☐ Rumen fluid	☐ Discharge
☐ Obese (5)	☐ Blood*	☐ Swab(s)	☐ Tonsils swollen	☐ Swab(s)
	☐ Swab(s)	☐ Other:	☐ Swab(s)	☐ Other:
	☐ Other:		☐ Other:	Li otiler.
Skin	Lungs	Heart	Gastrointestinal	Musculoskeletal
□ Normal	□ Normal	□ Normal	□ Normal	□ Normal
☐ Swelling*	☐ Pleural fluid/inflam.	☐ Pericardial effusion	☐ Bloat	
☐ Skin lesion(s)*	☐ Nodules*	☐ Hemorrhage*	☐ Regurgitation	☐ Swollen joint(s)*
☐ Poor/dull fur	☐ Blood*	☐ Scar tissue	☐ Regulgitation ☐ Diarrhea	☐ Abscess*
☐ Fur loss	□ Foam	☐ Necrosis	☐ Fecal staining	☐ Growth(s)
☐ Growth(s)	□ Pus	☐ Thickened valves	☐ Rectal blood*	☐ Trauma
☐ Wound(s)	☐ Aspiration		☐ Nodules*	
	☐ Aspiration ☐ Cranial, middle and	☐ Other:	☐ Nodules* ☐ Thickened*	☐ Hoof lesion(s)
☐ Parasites	,			☐ Muscle wasting*
☐ Other:	caudal sections taken		☐ Parasites	☐ Cooked muscle
	☐ Swab(s)		☐ Swab(s)	☐ Swab(s)
	☐ Other:		☐ Other:	☐ Other:
Reproductive	Kidneys and Urine	Liver & Spleen		imples Checklist
□ Normal	☐ Normal	☐ Normal (L)	Skin	□ Kidneys
☐ Lactating	□ Pale kidneys	☐ Enlarged (L)	☐ Hair ☐ Feces	☐ Adrenals ☐ GI (rumen, SI, LI, C)
☐ Mastitis*	□ Necrosis	☐ Abscess (L)	☐ Incisors	☐ Muscle
☐ Vulvar swelling*	☐ Blood*	☐ Normal (S)	☐ Thyroid	☐ LN (RP, SM, P, Mes)
☐ Prepuce swelling*	☐ Brown urine	☐ Enlarged (S)*	☐ Lung x 3	☐ Ovary/testes
☐ Orchitis*	☐ Sampled	☐ Other:	☐ Heart	☐ Brain
□ Discharge	☐ Other:		☐ Liver x 3 ☐ Spleen	☐ Femur/bone marrow ☐ Blood
☐ Clear			_ opicen	□ Urine
□ Pus*	Lymph nodes (LN)	Endocrine	Other Samp	les If Applicable
☐ Blood*	□ Normal	☐ Thyroid/parathyroid	☐ Fetus	☐ Any tissues with lesions
□ Fetus	☐ Enlarged*	☐ Adrenals	□ Placenta	□ Fat
□ Placenta	☐ Discharge/Abscess*		☐ Thymus ☐ Parasites	
☐ Swab(s)	☐ Sampled multiple		☐ Swabs of abn. fluid,	
☐ Other:	☐ Other:		abscess, swollen joints	
Comments:				

Appendix E - Biological Sample Collection, Handling and Storage Protocols

Please contact the NWT wildlife veterinarian for further information or with questions on sample protocols.

	Live Animal and Post-Mortem Standard Sampling									
Sample	Туре	Amount	Initial storage	Processing	Long-term storage					
	SST	1-3 vials	Keep vials upright at 37 © for 30min or until clotted.	Centrifuge after clotted and within 24 hours	Freeze serum in cryovials at -20 to -80 ℂ					
Blood	Red top	1-3 vials	Keep vials upright at 37 ℂ for 30min or until clotted.	Centrifuge after clotted and within 24 hours	Freeze serum in cryovials at -20 to -80 ℂ					
	EDTA	1-3 vials * Rotate vial for 30s to mix with EDTA	3-5℃	Centrifuge and store buffy coat and plasma in separate cryovials	Freeze at -20 to - 80©					
Hair	Base of tail	~ hand full	Dry coin envelope	None	At room temp.					
Feces	Per rectum/fresh from ground	~ hand full	Whirl-Pak	None	Whirl-Pak					
Skin biopsy	Ear tag sample	6mm punch biopsy	Dry in cryovial or coin envelop	None	Room temp.					
Swabs	Wounds, discharge, etc.	Min. 1/site	3-5.℃	Fresh is best (at vet request) or freeze	Freeze at -20 to - 80 ℂ					
Parasites	Any	Any	70% ethanol in vial	None	Room. temp					
		Additional	Post Mortem Sampli	ing						
Tissues	Standard +/- Others	1cm ³	Formalin 10:1 ratio	None	Room. temp					
Tissues	Standard +/- Others	100-200 grams	Whirl-Pak x 2 (1 fresh + 1 frozen)	Liver – Trace Nutrients Others – None initially	Freeze at -20 to -80 ℂ					
Teeth	Incisors	1-2 or take entire mandible	Whirl-Pak	None	Room. temp					
Urine		5-10ml via cystocentesis	Vial	Fresh is best (at vet request) or freeze	Freeze at -20 to - 80 ℂ					
Misc. abn. fluid	Any	5-10mls	Red top tube	Fresh is best (at vet request) or freeze	Freeze at -20 to - 80 ℂ					